ISRCTN88417663 https://doi.org/10.1186/ISRCTN88417663

Study investigating metabolomics (analysis of molecules produced from cell activity) as a new measurement method for liver enzyme function

Submission date 18/05/2021	**Recruitment status** Stopped	[ ] Prospectively registered [ ] Protocol [ ] Statistical analysis plan [ ] Results [ ] Individual participant data [ ] Record updated in last year
Registration date 21/05/2021	**Overall study status** Stopped	
Last Edited 27/01/2023	**Condition category** Other	

Plain English summary of protocol

Background and study aims
Medications that are taken by mouth need to get to their site of action (e.g. the liver). They then need to be removed from the body - a combination of metabolism (breakdown of the drug by the body) and elimination (removal of the drug, often in the urine). Most of this drug metabolism is undertaken by a group of chemicals called the CYP enzymes. These CYP enzymes vary between people due to differences in genetics, and also in response to other drugs and a persons diet. The activity of these CYP enzymes can make a big impact on how well a particular drug works, or indeed how likely it is to cause harm.
It would be very helpful to know the status of an individual's CYP enzymes before prescribing a drug as we might choose a higher dose or use one drug in preference of another. The gold standard test to work out how well someone's CYP enzymes are working is a complex and long (>24 hr) study called a pharmacokinetic study. As this is long and complex it is never undertaken before prescribing a drug. If we could develop a simple blood test that can tell us the CYP enzyme activity for an individual we could measure this before prescribing.
In this study we will undertake pharmacokinetic studies to accurately determine activity of one of the CYP enzymes (called CYP2C9). We will do this in people with a particular genetic variant of CYP2C9, and also after they take a tablet that reduces the enzyme activity. We will measure blood chemicals called 'endogenous metabolites' as we have prior evidence that these might give us a snapshot of CYP activity. As proof of principle, we hope to find particular metabolites that are a proxy of CYP2C9 activity.

Who can participate?
A healthy, not-pregnant, non-diabetic person, aged 18 - 60 years, where the CYP2C9 genotype is known.

What does the study involve?
A screening visit (45 min).
Two study days with overnight fast and one tablet at 9 pm the night before (placebo /fluconazole). The study day is about 7 hours. During the study day, the person will be challenged with tolbutamide, and 7 blood samples will be taken and urine will be collected for

2x 12 hrs. At t=0 a blood sample for metabolomics will be drawn. Tolbutamide will be determined in the following blood samples.

What are the possible benefits and risks of participating?
Participants will attend the Clinical Research Centre for the three visits as detailed above.
The tests performed during visits to the centre will be of no clinical benefit to participants. On two visits, participants will be given a single low dose of tolbutamide by one of our qualified nurses. There is no clinical reason for participants to take this drug; it is not being given as a medical treatment but as a model compound for determining the CYP2C9 function. It will be only given twice as part of the research study. There will be no direct clinical benefit to participants from taking part in this study. However, study participation will aid our team in our research. With the results from this study, we hope to improve our understanding of drug-drug interaction in large scale in population studies and as a tool in precision medicine for the individual patient. This may be of benefit to participants, but not in the immediate term.
Our CYP model compound tolbutamide is licensed for use in the treatment of diabetes mellitus. It is currently in use for treatment for specific patients with genetic alterations involving a potassium channel in their beta cells, therefore its efficacy and safety profile is well established. An expected rare risk with use of tolbutamide is hypoglycaemia. During this study, participants will be monitored closely and will be offered a glucose drink and we have glucose infusion prepared in the unlikely event of hypoglycemia. The dose given in this study will be 500 mg in the first instance, which is the lowest suggested starting or maintenance dosing regimen in national guidelines. In addition, tolbutamide dosing in this study will be as a single oral dose in study visit 2 and 3. Tolbutamide is contraindicated in those who have hypersensitivity to sulphonylureas or sulphonamides, type 1diabetes mellitus, severe hepatic or renal insufficiency, pregnancy and lactation. The pharmacokinetics and/or pharmacodynamics of tolbutamide may be altered in those with severe hepatic insufficiency or renal failure.
The patient's medical history and baseline blood tests will be assessed during their screening visit to ensure no interactions with tolbutamide, and that renal and hepatic function meet inclusion/exclusion criteria. Tolbutamide is contraindicated with significant alcohol consumption, however, doses will be on two isolated occasions as a single 500 mg tolbutamide dose in a controlled study environment. Due to the contraindication with alcohol, patients will be asked to make a temporary lifestyle change and abstain from alcohol for 24 hours prior to the study visit. If alcohol is consumed within 24 hours prior to the study, the visit will be rescheduled. This lifestyle change is to maintain safety during and following the procedure and is explained in the PIS. Fluconazole or placebo will be given the night before study visit 2 and 3. Fluconazole is an antifungal treatment that is an inhibitor of CYP2C9 and to a lesser extend CYP3A4. Single dose fluconazole administration inhibits CYP2C9 and thus increases the AUC for tolbutamide or slows the removal of tolbutamide from the blood. Thus, treatment with fluconazole allows for us to measure changes in CYP2C9 and makes us able to determine if the metabolites in plasma changes accordingly (X-11787, X-11247, X-12063, X-02249). At the screening visit all medication the patient takes will be screened for interaction with fluconazole.
If significant interaction is detected, the patient will be excluded from participating in the study.
Intravenous Cannulation Visits 2-3 will require intravenous cannulation. Intravenous cannulas will be managed in accordance with local NHS Tayside protocols for infection control and clinical governance. The cannula will be managed in accordance with infection control and clinical governance protocols set out by NHS Tayside. Risks of cannulation are discussed in the Participant Information Sheet and will be discussed fully at the screening visit prior to informed consent. One of the main risks associated with cannulation is leakage of drip fluid into the surrounding skin. This is known as extravasation. To avoid this, the cannula will be checked to ensure it is working properly following insertion. Nursing and medical staff will also be present throughout each test visit, they will regularly check the cannula to ensure comfort and safety. Should leakage occur, this will be managed in accordance with local NHS Tayside protocol. The

drip fluid in this study is isotonic saline, if leakage occurs, it could cause pain, redness, swelling and in rarer instance, infection around the cannula site. Should this occur, this will be documented in the study folder by the research team and follow up arranged to ensure resolution.
The other risk of cannulation is of infection around the cannula site. All cannulas will be inserted and managed in accordance with infection control and clinical governance protocols set out by NHS Tayside. Should infection occur, this will be documented by the research team. Infection may require a course of antibiotics from the participant's GP. Should infection occur, follow up will be arranged with NHS services to ensure resolution.

Where is the study run from?
University of Dundee, Ninewells Hospital (UK)

When is the study starting and how long is it expected to run for?
March 2020 to March 2023

Who is funding the study?
Wellcome Trust (UK)

Who is the main contact?
Professor Ewan Pearson, e.z.pearson@dundee.ac.uk
Associate Professor Mads Kjolby, mkjoelby001@dundee.ac.uk

Contact information

Type(s)
Public

Contact name
Dr Mads Kjolby

ORCID ID
http://orcid.org/0000-0002-1043-6137

Contact details
Department of Molecular and Clinical Medicine
Level 5, Mailbox 12
Ninewells Hospital and Medical School
Dundee
United Kingdom
DD1 9SY
+44 (0)7476 010515
mkjoelby001@dundee.ac.uk

Type(s)
Public

Contact name
Prof Ewan Pearson

ORCID ID

http://orcid.org/0000-0001-9237-8585

Contact details
Department of Molecular and Clinical Medicine
Level 5, Mailbox 12
Ninewells Hospital and Medical School
Dundee
United Kingdom
DD1 9SY
+44 (0)1382 383387
e.z.pearson@dundee.ac.uk

Additional identifiers

EudraCT/CTIS number
Nil known

IRAS number
285840

ClinicalTrials.gov number
Nil known

Secondary identifying numbers
2-014-20, IRAS 285840

Study information

Scientific Title
Impact of CYP2C9 genotype and CYP2C9 inhibiting drugs on an individual's metabolomics and metabolism of tolbutamide for interrogation of drug interactions

Acronym
MetCYP

Study objectives
We wish to determine if endogenous metabolites are a good measure of current CYP2C9 function and thus can be used in large-scale studies for drug-drug interactions.
To determine whether metabolomic screening of endogenous metabolites reflects current CYP function, we will interrogate the CYP function with gold standard PK determination of a single dose of tolbutamide in patients with known CYP2C9 genotype and followed by inhibition of CYP2C9, using a single dose of fluconazole. We have identified >50 other ADME metabolite proxies, and the above study will be a proof of concept study, that will allow determination of the value of metabolomic screening in drug-drug interaction for use in precision medicine, ultimately allowing to choose the most efficacious drug with lowest amount of adverse effects.

Ethics approval required
Old ethics approval format

Ethics approval(s)

Approved 17/02/2021, West Midlands - Solihull Research Ethics Committee (The Old Chapel Royal Standard Place, Nottingham, NG1 6FS, UK; +44 (0)207 1048310; solihull.rec@hra.nhs.uk), ref: 21/WM/0002

Study design
A recruitment-by-genotype randomized and placebo-controlled double-blind cross-over study

Primary study design
Interventional

Secondary study design
Randomised cross over trial

Study setting(s)
Hospital

Study type(s)
Other

Participant information sheet
See additional file ISRCTN88417663_PIS_v4_21Apr2021 (added 01/06/2021)

Health condition(s) or problem(s) studied
Impact of CYP2C9 genotype and CYP2C9 inhibiting drugs in healthy volunteers

Interventions
Participants are recruited based on their CYP2C9 genotype. This is blinded to the participant and the research team by a third party. The study is a crossover design, so participants will be treated with placebo or Fluconazole before each study day. The order of placebo and fluconazol is blinded to the participant and the researcher. The medication is packed and randomized by the Pharmacy (third party).

This is a recruitment-by-genotype randomised and placebo controlled double-blind cross-over study design. For those recruited from Generation Scotland (GS), GoDARTS and GoSHARE, study visits will take place in the Clinical Research Centre (CRC), Ninewells Hospital & Medical School, Dundee. Each volunteer will be on study for two weeks in total. This includes two distinct treatment periods of each 2 days duration, separated by at least one week wash-out period (Debruyne and Ryckelynck 1993).
Participants will receive their participant information sheet a few days prior to the screening visit. At the baseline / screening visit, participants will have vital signs, height, weight, and BMI measured. Blood and urine samples will be obtained. Medical history including current prescriptions, history of smoking and alcohol use. The outline and purpose of the experiment will be explained, and information will also be supplied in writing, and consent will be obtained. Participants will be supplied with one fluconazole capsule for study day 1, at the end of the screening visit. If a participant is excluded from the study, then the drugs will be returned to the relevant CRC.
Participants will undertake 2 study days. On study day 1 (tolbutamide w/o fluconazole), a baseline blood sample for metabolomics will be drawn, and then an oral dose of tolbutamide will be administered. Blood samples will be drawn according to schema below for pharmacokinetic profile, and plasma will be stored for later UHPLC-MS/MS.
Prior to study day 2 (tolbutamide w fluconazole), the patients will ingest one capsule orally of

fluconazole (200 mg) the night before (21:00) at home. On study day 2, an oral dose of tolbutamide will be administered, and blood samples will be drawn as on study day 1. The sequence of study day 1 and 2, or 2 and 1 will be randomised. The washout period between study day 1 and 2, will be >7 days.

Intervention Type
Drug

Phase
Not Applicable

Drug/device/biological/vaccine name(s)
Fluconazole, tolbutamide

Primary outcome measure
1. Level of tolbutamide, and in the blood measured with UHPLC-MS/MS using blood samples taken at 0, 0.5, 1, 2, 3, 4, 6 hrs, derive pharmacokinetic parameters from measures (CL, T1/2, AUC)
2. Level of tolbutamide, 4-hydroxytolbutamide and carboxytolbutamide in the urine measured with UHPLC-MS/MS using urine samples collected from 0 to 12 hours and from 12 to 24 hours
3. Metabolomics (Metabolon, MS/MS) will measured on T=0 hr samples

Secondary outcome measures
There are no secondary outcome measures

Overall study start date
01/03/2020

Completion date
01/03/2023

Reason abandoned (if study stopped)
Participant recruitment issue

Eligibility

Key inclusion criteria
1. Aged 18 - 60 years
2. Caucasian (to limit genetic variation as much as possible)
3. Not known to have diabetes
4. No daily treatment with antidiabetic medication, statins, amitriptyline or other drugs that inhibit or require CYP2C9 or CYP3A4 metabolism
5. No cognitive impairment or visual impairment
6. Known CYP2C9 genotype – either normal (“wild type”) or two reduced function alleles

Participant type(s)
Healthy volunteer

Age group
Adult

Lower age limit
18 Years

Sex
Both

Target number of participants
24

Total final enrolment
6

Key exclusion criteria
1. Heterozygous CYP2C9 genotype i.e. only one reduced function allele
2. Use of any antifungal drug within the last month
3. Recent involvement (<30 days) in a clinical trial
4. Pregnancy or planning to conceive
5. Inability/unwillingness to comply with the protocol
6. Anaemia

Date of first enrolment
09/05/2021

Date of final enrolment
01/03/2023

Locations

Countries of recruitment
Scotland

United Kingdom

Study participating centre
Ninewells Hospital and Medical School
Department of Molecular and Clinical Medicine
Level 5, Mailbox 12
Dundee
United Kingdom
DD1 9SY

Sponsor information

Organisation
University of Dundee

Sponsor details
TASC (Tayside Medical Science Centre),
Ninewells Hospital & Medical School
TASC Research & Development Office
Residency Block, Level 3
George Pirie Way
Dundee
Scotland
United Kingdom
DD1 9SY
+44 (0)1382 383900
TASCgovernance@dundee.ac.uk

Sponsor type
University/education

Website
http://www.dundee.ac.uk/

ROR
https://ror.org/03h2bxq36

Funder(s)

Funder type
Charity

Funder Name
Wellcome Trust

Alternative Name(s)
Wellcome, WT

Funding Body Type
Private sector organisation

Funding Body Subtype
Trusts, charities, foundations (both public and private)

Location
United Kingdom

Results and Publications

Publication and dissemination plan

The study is expected to be published in peer-reviewed journals with focus on precision medicine. A build up of a bio repository is also expected to be associated with follow up articles.

Intention to publish date
01/05/2024

Individual participant data (IPD) sharing plan
The current data sharing plans for this study are unknown and will be available at a later date.

IPD sharing plan summary
Data sharing statement to be made available at a later date

Study outputs

Output type	Details	Date created	Date added	Peer reviewed?	Patient-facing?
Participant information sheet	version v4	21/04/2021	01/06/2021	No	Yes
HRA research summary			28/06/2023	No	No