Study title: Pro-Euro-DILI Registry: Creation of a multicentre and multidisciplinary registry of prospective drug-induced liver injury cases.

Study acronym: Pro-Euro-DILI

Sponsor	UK Site Sponsor: Nottingham University Hospitals NHS Trust
Funder	European Association for the Study of the Liver (EASL)
Chief Investigator	
REC Reference Number	
Internal Reference Number	
Version Number and Date	Template for COST 13th Oct 2019 Jane Grove

Confidentiality Statement

This document contains confidential information that must not be disclosed to anyone other than the Sponsor, the Investigator Team, host NHS Trust (s), regulatory authorities, and members of the Research Ethics Committee.

TABLE OF CONTENTS

STUDY MANAGEMENT GROUP

Chief Investigator (UK) Name: Professor Guruprasad P Aithal Address: Nottingham Digestive Diseases Centre and NIHR Nottingham Biomedical Research Centre, Nottingham University Hospitals NHS Trust and the University of	**Pro-Euro DILI Chief Investigator (SPAIN)** Name: Professor Raul J. Andrade Address: Institute of Biomedical Research (IBIMA), Malaga University Hospital, Malaga University, Malaga, Spain
Statistician	**Study Coordinator:**
Nurse Lead	**Research Coordinator**

Study Coordination Centre

UK: Nottingham Digestive Diseases Centre and NIHR Nottingham Biomedical Research Centre, E Floor, West Block, Queens Medical Centre, Nottingham, NG7 2UH
EUROPE: Institute of Biomedical Research (IBIMA), Malaga University Hospital, Spain.

For general queries, supply of study documentation, and collection of data, please contact:

Study Coordinator:
Address:

Clinical Queries

Clinical queries should be directed to xxx who will direct the query to the appropriate person.

Sponsor:

Funder:

This protocol describes the Pro-Euro-DILI registry and provides information about procedures for entering participants. Every care was taken in its drafting, but corrections or amendments may be necessary. These will be circulated to investigators in the study. Problems relating to this study should be referred, in the first instance, to the Chief Investigator.

This study will adhere to the principles outlined in the NHS Research Governance Framework for Health and Social Care (2nd edition). It will be conducted in compliance with the protocol, the Data Protection Act and other regulatory requirements as appropriate.

AMENDMENT HISTORY

PROTOCOL APPROVAL

Pro-Euro-DILI Registry: Creation of a multicentre and multidisciplinary European registry of prospective drug-induced liver injury cases.

Chief Investigator	Signature	Date
Trial Statistician	Signature	Date
Sponsor Representative	Signature	Date

GLOSSARY OF ABBREVIATIONS

AE	Adverse Event
AIH	Auto-immune hepatitis
AR	Adverse Reaction
ALT	Alanine transaminase
ALP	Alkaline phosphatase
ATC	Anatomical therapeutic chemical
BRC	NIHR Nottingham Biomedical Research Centre
CI	Chief investigator
CMV	Cytomegalovirus
CPI	Check Point Inhibitors
CRF	Case Report Form
CyTOF	Cytometry Time of Flight (immunophenotyping)
DILI	Drug-Induced Liver injury
DIKI	Drug-induced kidney injury
DIPI,	Drug-induced pancreatic injury
DIVI	Drug-induced vascular injury
DINI	Drug-induced neurological injury
EBV	Epstein-Barr virus
EFPIA	European Union and the European Federation of Pharmaceutical Industries & Associations
eCRF	Electronic case report form
EMA	European Medicines Agency
FDA	United States (US) Food and Drug Administration
PMDA	Critical Path Institute's Pharmaceuticals and Medical Devices Agency
GCP	Good Clinical Practise
HIV	Human Immunodeficiency virus
ICH	International Conference on Harmonisation
iDILI	Idiosyncratic drug-induced liver injury
INR	International normalized ratio
IMI	Innovative Medicines Initiative
ISF	Investigator Site File
NDDC	Nottingham Digestive Diseases Centre, University of Nottingham
NIHR	National Institute of Health Research
NUH	Nottingham University Hospitals
PI	Principal Investigator
PPI	Public Patient Involvement
R	Ratio
REC	Research Ethics committee
RUCAM/CIOMS	Roussel Uclaf Causality Assessment/Council of International Organization of Medical Sciences
SAE	Serious Adverse Event
SAFE-T	Safer and faster Evidence based translation Consortium
SAR	Serious Adverse Reaction
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reaction
TBL	Total bilirubin
TMF	Trial Master File
UAR	Unexpected Adverse Reaction
UK-AIH	UK Autoimmune cohort collaboration
ULN	Upper limit of normal
WHO	World Health Organisation

ZeBanC Zentrale Biobmaterialbank der Charité

KEYWORDS
DILI, Liver Injury, Registry, multicentre, phenotyping

STUDY SUMMARY

Title	Pro-Euro-DILI Registry: Creation of a multicentre and multidisciplinary European registry of prospective drug-induced liver injury cases.
Acronym	Pro-Euro-DILI
Short title	Pro-Euro-DILI Registry
Chief Investigator	
Objectives	• To set-up an international European electronic registry of patients with idiosyncratic DILI, (iDILI) enrolled prospectively with the collation of in-depth phenotyping: including details of drug dose, duration, concomitant medications, host demography, comorbidity including insulin sensitivity as well as the course of the event. • To collect and store biological samples (blood, urine, stool and liver biopsy) from patients with suspected idiosyncratic DILI and the symptomatic non-DILI control group through the course (at onset and follow-up) of the event. • To collect and store biological samples (blood, urine and stool) from control patients at a single visit. • To make biosamples available for collaborative research through a dedicated biobank. • To determine and analyse individual phenotypes and biochemical and cellular component levels in patients and control groups in order to develop translatable disease biomarkers
Study Configuration	Case controlled multi-centre study for the collection, analysis and storage of biological samples and data (medical, demographic and clinical).
Setting	Secondary care (hospital based)
Sample size estimate	N/A
Number of participants	UK cohort: • 150 suspected DILI cases (symptomatic) • 20 cases of autoimmune hepatitis with DILI-like symptoms* Total, including European cohort: • 300 **confirmed** DILI cases (50 in the UK)
Eligibility criteria	Symptomatic adults diagnosed with suspected acute DILI (within 8 weeks of diagnosis).

Description of interventions	DILI patients, symptomatic non-DILI patients: Blood, urine and stool samples will be collected at each visit. We plan to additionally collect any available surplus liver biopsy or blood samples taken as part of clinical care (and any archived paraffin sections taken previously as part of clinical care).
Duration of study	Recruitment will be over a 9 year period (to Apr 2024)

INTRODUCTION

Background

Idiosyncratic drug-induced liver injury (iDILI) is an acute adverse hepatic reaction to a medication used in its therapeutic dose and that is unexpected from the known pharmacological action of the agent. The incidence of DILI is between 2.4 and 8.4 per 100,000 person years in population-based studies [1, 2]. Although a recent study reported an annual incidence of 19.1 per 100,000 people [3], the study used a lower threshold for case definition than that was recommended by the 'phenotypic standardization project' [4]. The 'European Commission' on Public Health defines rare diseases as a life-threatening or chronically debilitating disease which affects fewer than 1 in 2,000 people, so that special combined efforts are needed to address them. Despite its rarity, idiosyncratic DILI accounts for 7-15% of the cases of acute liver failure in Europe and is the most frequent reason for the market withdrawal of an approved drug. In addition, DILI occurs in association with a large number of drugs and shows heterogeneity. There are no markers that can effectively pre-empt and prevent DILI or monitor the severity and course of the adverse event. More recently it is emerging that immunotherapy regimins devised for cancer treatment are associated with increased risk of DILI development [5]. Further characterisation and understanding of this is urgently needed to distinguish from other causes and develop more effective treatment. This project seeks to specifically investigate the immunological mechanisms involved in mediating this toxicity.

Rationale for current study:

Case of need: One of the biggest hurdles in DILI studies is the limited number of identified cases. Due to the relative rarity of this condition it is unlikely that a single hospital will be able to identify a sufficient number of cases to perform well-powered studies. Hence, a collaborative effort is practically a prerequisite for these kinds of studies. National multicentric DILI registers, such as the Spanish DILI Registry and the US Drug-induced liver injury network (DILIN) have demonstrated the advantage of collaborations [6,7]. Furthermore, international consortia, such as SAFE-T (www.imi-safe-t.eu) and iDILIC / iSAEC (www.saeconsotrium.org) have also emerged with the goal of developing new more sensitive and specific DILI markers than the routine liver profile markers currently used in clinical practice to determine liver injury, as well as genetic variations that can predict DILI susceptibility.

To address this challenge, the Pro Euro DILI consortium of leading European research institutions and SMEs has been established.

The consortium will generate exploratory and confirmatory data enabling regulatory qualification of new safety biomarkers for application in drug development.

Given the significant expertise available across the consortium, the group will be able to tackle the key challenges related to successful biomarker qualification. A key driving principle of the consortium is cross-linking via existing networks of top profile research institutions, as well as capitalizing on existing data and resources. The Consortium is embedded into a network of international research collaborations such as IMI Transbioline, TransQST, eTRANSAFE, the i2b2 tranSMART Foundation, the CIOMS DILI working group, EPoS, LITMUS, and BBMRI.

In addition, majority of cases of DILI in the published clinical and genetic studies have been enrolled retrospectively; hence, the data set is reliant on clinical investigations and the setup of health services which are incomplete. In addition, the majority of the studies have used healthy populations as control groups (rather than those exposed to the drugs). Despite the discovery of genetic factors such as the association of specific HLA genotype with DILI, a low positive predictive value of the genotype in predicting DILI is limiting its use in clinical practice [8,9]. Age, smoking, metabolic syndrome, co-morbidity and other yet unidentified factors may generate an environment of oxidative stress that contributes to DILI. Therefore, we need ‘in-depth phenotyping’ together with data from an exposed control group to develop refined algorithms incorporating drug-related factors, host genetic and environmental risk factors that would enable us to pre-empt DILI. Deeply phenotyped cohorts with biological samples are essential for the development and validation of novel diagnostic/ prognostic markers [10].

STUDY OBJECTIVES AND PURPOSE

Purpose

The purpose of this study is to set up an international interdisciplinary consortium to obtain a better understanding of the mechanisms underlying drug-induced liver injury (DILI) and to develop methods of preventing DILI and its consequences. The consortium including the clinicians and scientists from European countries covering a wide area of expertise will allow the development of mature hypotheses that can be tested using a robust study design. The consortium will facilitate the development of strong translational research proposals based on our hypothesis.

Primary Objective

To, collect, store and catalogue biological samples and clinical data, to form a large international patient/control cohort available for subsequent detailed analysis by consortium members which can also be used in current and future epidemiologic and mechanistic studies.

Proposal Aims:

We hypothesise that the severity of DILI is determined by factors that modulate cellular response to oxidative stress and the transition of adaptive response to the development of serious DILI can be recognised by a combination of markers that reflect the balance

between pro- and anti-inflammatory responses to oxidative stress. So, we will develop a panel of markers that assess the severity of DILI. As the prospective in-depth phenotyped DILI cohort and controls are being recruited, the network infrastructure can also support any evaluation and validation of novel tests with potential application in DILI.

There are no evidence-based treatments for DILI; rarity of the condition and heterogeneity of the manifestations prevents the conception and successful completion of clinical trials. Multi-centre Pro-Euro-DILI registry will provide both the infra-structure to conduct clinical trials, and also a sufficient number of well characterised patients to evaluate the efficacy of novel anti-cholestatic agents in the resolution of DILI [11].

STUDY PLAN

Biomarker Identification & Evaluation:

Participants will be asked to give consent for their anonymised samples and anonymised data to be stored and used for the purposes of this International study and to be stored in a research tissue bank and used for future research.

Clinical and phenotypic data, outlined in the CRF, will be collected by the study centre. Each participating study centre will collect and process biological samples as per SOP. Samples will be kept at -80°C and shipped in regular batches to the NIHR Nottingham Biomedical Research Centre (BRC). To determine other biomarkers of interest, samples will be analysed by researchers at Nottingham or collaborating institutions or by commercial suppliers.

Upon completion of the study, where consent is given, samples will be transferred to an appropriate ethically approved research tissue bank and made available for ethically approved research programs with approval of partners as specified in collaboration agreements. Any samples stored at Nottingham University Hospitals NHS Trust will be transferred to the custodianship of University of Nottingham (HTA Licence No. 12265; Designated Individual: Dr William Dunn) and stored in the NDDC-BRU Research Tissue Bank for up to 10 years.

<u>Outline of Laboratory Analyses</u>

Appendix 1 gives an overview of the patient data and clinical analyses to be included (where possible) which are part of clinical care.

Plasma and serum will be prepared from blood samples after collection and stored at -80°C for subsequent analysis of circulating biomarkers identified in other studies. This resource will allow subsequent studies to validate new markers as potential indicators for diseases diagnosis or prognosis.

Whole blood will be collected and stored to be available for future DNA extraction and analysis of genetic biomarkers. Although recent studies have identified some genetic factors for DILI [13], genetic tests are not yet available for DILI. Any findings of genetic tests for these patients will only be revealed some time after the DILI event has resolved so is not likely to be of significant clinical value to their condition and will not be conveyed to the patient.

The total volume of blood taken at one visit will not exceed 80ml.

Stool and Urine samples will also be collected where possible. These will be stored for analysis to investigate urinary biomarkers of disease and for gut microbiome and metabolite analysis.

Any surplus liver tissue available where a liver biopsy is performed as part of clinical care will be collected and stored for research use. This will facilitate analysis of tissue markers and correlation with circulating biomarkers and will enable specific staining to identify clinical features. Extracted RNA will be utilised for expression analysis to investigate possible disease mechanisms.

Similarly where available residual blood from routine clinical testing may be collected for analysis of biomarkers where available.

STUDY DESIGN

Study Configuration:

This is a case-controlled multi-centre study for the collection and storage of biological samples and data (medical, demographic and clinical). We will prospectively develop a cohort of patients with liver injury suspected to be induced by prescription drugs, over-the-counter medications, herbal remedies or dietary supplement intake identified by the participating clinical centres. Each patient will be followed from the identification of hepatotoxicity until normalization of the analytical liver profile.

Coordinating Centres:

This is a European multicentre collaboration consisting of members of the Pro-Euro-DILI COST Consortium. This is coordinated by 2 centres: The University of Nottingham Nottingham Digestive Diseases Centre (NDDC) within the NIHR Nottingham BRC in the UK. and The Malaga Scientific Research Institute, Spain. Both coordinating centres will be responsible for the adjudication of each potential DILI case entered into the Pro-Euro-DILI Registry database. Both will share responsibility for the overall data monitoring, causality assessment of cases and the organisation/scheduling of regular conference call meetings and annual meetings.

Participating European Centres: (Incl both coordinating centres)

Each Centre will obtain ethical approval for the study, nationally and locally and form links with their local regional liver transplant unit. Each centre will be responsible for the collection of biological samples and the shipment of samples for analysis or storage at Nottingham BRC or Charite, or to an approved research tissue bank for long term storage (when the study ends). Samples will be stored at -80°C and shipped in batches.

Each Centre will also be responsible for entering the corresponding data into the Pro-Euro DILI Registry database (developed by Coresoft Clinic S.L, http://www.coresoft.es), available online. Tutorials on data entry and database implementations will be organized by the co-ordinating centres (Málaga and Nottingham) if required.

The web-based application and data will be hosted at Malaga University servers, with automated data backups on a daily basis, a report system for successful backup monitoring and reliable network connectivity. The software strictly follows the regulations imposed by the European Union ensuring that registries are secure and protected by the law. Furthermore, the software follows the Declaration of Helsinki requirements to ensure protection of patient's rights. No data that can reveal the patients identity will be entered into the database. Internet access to the registry platform is password protected and all stored passwords will be encrypted. Subsequent transfer of datasets for analysis will follow agreed data transfer procedure and adhere to data protection standards.

Collaborating centres include (but will not be limited to):

Primary endpoint

Acquisition of clinical samples and clinical data collected from a total of (across all sites in Europe):

- 430 suspected DILI cases (symptomatic)
    - Of which 300 **confirmed** DILI cases

Secondary endpoints

We will collate detailed and comprehensive data using an electronic case report form (eCRF) to achieve in-depth phenotyping. (A paper version for data collection is available as a separate document: Annex. 1 CRF.)

Levels of putative biomarkers in samples will be assessed and evaluated to determine correlation with diseases phenotype and likely value in diagnosis or prognosis.

Causality assessment: Causal relationships between the event and the drug will be assessed using the Roussel Uclaf Causality Assessment/Council of International Organization of Medical Sciences (RUCAM/CIOMS). DILI will be phenotyped and severity graded as recommended by the phenotyping standardization project [4].

Standard variables in DILI, such as type of liver damage, severity and chronicity, will be classified as recommended by the phenotyping standardisation project [4]. The type of liver damage will be defined as hepatocellular, cholestatic or mixed based on the ratio (R) between alanine transaminase (ALT) and alkaline phosphatase (ALP) expressed in times the upper limit of normal (ULN), R = (ALT/ULN) / (ALP/ULN). Hepatocellular damage is defined as R≥5, cholestatic damage ≤2 and mixed damage as 2<R<5. The R value will be calculated from the first available analytical values after DILI initiation.

Severity of DILI will be graded as follows:
Mild: Elevated ALT/ALP concentration reaching criteria for DILI but total bilirubin (TBL) concentration <2x ULN
Moderate: Elevated ALT/ALP concentration reaching criteria for DILI and TBL ≥2x ULN, or symptomatic hepatitis
Severe: Elevated ALT/ALP concentration reaching criteria for DILI, TBL ≥2x ULN and one of the following:

- international normalized ratio (INR) ≥1.5

- ascites and/or encephalopathy, disease duration <26 weeks and absence of underlying cirrhosis
- other organ failure considered to be due to DILI

Fatal or transplantation: Death or liver transplantation due to DILI

Disease duration will be graded as follows:

- **Persistent DILI:** Evidence of continued liver injury >3 months for hepatocellular or mixed type of liver damage and >6 months for cholestatic liver damage

- **Chronic DILI:** Evidence of persistent liver injury at >1 year after the onset of DILI

Causal DILI agents will be classified according to World Health Organisation (WHO) anatomical therapeutic chemical (ATC) classification system. Associated diseases will be classified according to the WHO tenth edition of the international classification of diseases, ICD-10.

STUDY POPULATION

Recruitment will be over a nine year period (up to April 2024).

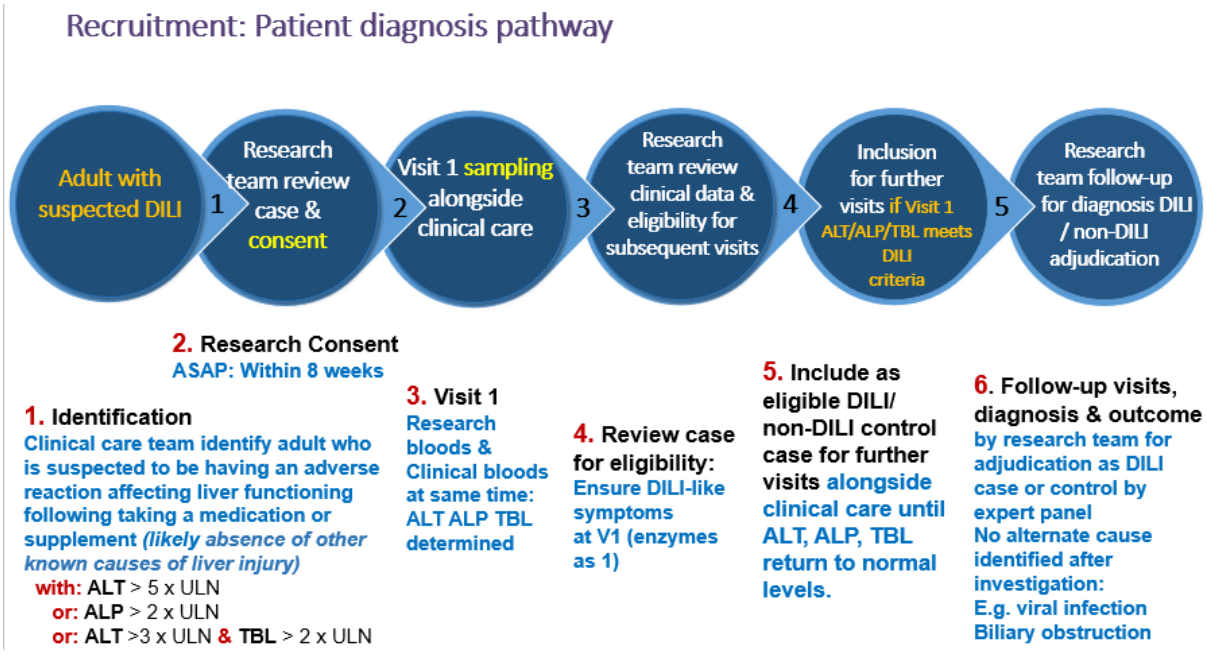
-**For suspected DILI patients :** 3-7 visits, (each visit lasting for up to 30mins). Each patient will be in the study for up to 6 months duration.

Follow-up: Each patient will have a clinical follow up until the event resolves or 12 weeks after the first event whichever occurs later. We will also follow-up outcomes in clinical notes over a 10 year period after recruitment where consent is given.

Enrolment:

We expect to enrol each case at the earliest opportunity possible, following the suspected DILI event (or AIH event) (up to a maximum of 8 weeks from diagnosis) when research team anticipates that they will meet inclusion criteria below. Each patient will be followed from the identification of hepatotoxicity until normalization of the analytical liver profile.

Enrolment will begin once a patient has been clinically diagnosed with suspected DILI and gives consent to take part in the study. In all instances the visit schedule should be adapted to routine hospital procedures in order to facilitate patient participation as well as avoid additional hospital costs (see diagram below).



- Control participants (on medication or untreated but without DILI symptoms): A single study visit of up to 30 minutes duration. Controls will be required to provide biological samples, demographic and medical data will also be collected.

Study Duration:

Enrolment will end at 9 years from the start of the study (Apr 2024) or once the recruitment target has been met, whichever is soonest.

INCLUSION CRITERIA

All patient and control groups:

- Aged 18 years and over
- Able to give written informed consent OR consent provided by consultee (personal or nominated) for potential participants with suspected DILI who lack capacity to give written informed consent.

A. Suspected DILI Patients:

Adults diagnosed with suspected acute DILI (within 8 weeks of diagnosis) and meeting the following criteria will be included:

1. Meets one of the following analytical thresholds at enrolment (visit 1)

- alanine transaminase (ALT) exceeding 5 times upper limit of normal (ULN) OR
- alkaline phosphatase exceeding 2 times ULN OR
- ALT exceeding 3 times ULN plus bilirubin exceeding 2 times ULN

Patients who are found to not meet these thresholds on date of recruitment when clinical test results are returned should not be invited for subsequent visits.

2. Exposure to drugs including any prescription drug, over-the-counter drug, recreational drug, herbal remedies or dietary supplements prior to the DILI onset.

3. Clinical suspicion of having possible DILI.

EXCLUSION CRITERIA

Anyone with any definite diagnosis of the following conditions **identified prior to recruitment** will not be included in the study as a suspected DILI case:

- Acute viral hepatitis due to hepatitis A, B, C, E, CMV, EBV, HIV.
- Acute presentation of auto-immune hepatitis unrelated to the drug.
- Confirmed acute liver injury that explains the clinical manifestation e: ischemic hepatitis, acute ascending cholangitis.
- Acute exacerbation/ decompensation of known chronic liver disease that explains the acute event.
- Biliary obstruction explaining cholestasis.
- On the judgement of CI that the person has certain alternative explanation to the acute event (rather than DILI).

It is possible that detailed investigations of suspected DILI cases my reveal one or more of these conditions, this will not exclude the participants from continuation with study visits but these cases will be adjudicated as 'non-DILI' and analysed as a separate study group.

WITHDRAWAL CRITERIA

It will be explained to the potential participant that entry into the study is entirely voluntary and that their treatment and care will not be affected by their decision. It will also be explained that they can withdraw at any time from the study but attempts will be made to avoid this occurrence. In the event of their withdrawal it will be explained that their data collected so far cannot be erased and we may still use the anonymised data in the final analyses. Remaining tissue samples can be destroyed/disposed of if the participant so wishes. Similarly, the information about them will be deleted so that it cannot be used again. We will explain to the participant that if they decide to withdraw after a long period of time, the samples may already have been used and we cannot recall samples or information from researchers if this is the case.

The Principal Investigator may also remove a subject if, in his / her opinion, it is in the best interests of the subject. If a patient permanently withdraws from the study, or is lost to follow-up, the reason will be recorded.

PARTICIPANT SELECTION AND ENROLMENT

IDENTIFYING PARTICIPANTS

Potential participants will be recruited from a secondary care setting, from the acute and in-patient services as well as outpatient clinics at the treating hospital. Patients who may have DILI event or auto- immune hepatitis will be identified and approached. The initial approach will be from a member of the patient's usual care team (which may include the investigator and his research team), and information about the study will be on display in the relevant clinical areas (A study Info leaflet for patients and study info poster for clinicians are provided as separated files).

Patients will be identified as having suspected DILI from their medical records and laboratory test results from samples taken as part of their standard clinical care. The study will be explained and potential participants will be given a copy of the Participant Information Sheet

to read and asked if they would be interested in taking part in the study. Invitation letters will also be sent out to potential participants with their results letter and or clinic appointment letter.

If a patient agrees to take part in the study, they will be asked to sign a copy of the consent form. Once signed, the patient will have a blood sample taken, the results of this blood test taken at enrolment will be used to assess/determine eligibility for subsequent visits; if eligible, the patient will continue in the study. In all instances the visit schedule should be adapted to routine hospital procedures in order to facilitate patient participation as well as avoid additional hospital costs.

The investigator or their nominee, e.g. from the research team or a member of the participant's usual care team, will inform the participant (or their nominated representative if they are unable to give informed written consent) of all aspects pertaining to participation in the study.

If needed, the usual hospital interpreter and translator services will be available to assist with discussion of the trial, the participant information sheets, and consent forms, but the consent forms and information sheets for UK sites will not be available printed in other languages.

Healthy Control Group:

Adults who are exposed to the same drugs (as those responsible for DILI) without developing DILI will be identified. The Chief Investigator will access hospital pharmacy databases to identify patients taking specific drugs of interest. Then patients exposed to particular drugs without developing DILI will be invited to participate in the study as a control group. In the UK this will be done at the Nottingham site initially, extending to other sites where appropriate.

We will also advertise through BRC website (https://nddcbru.org.uk/pro-euro-dili) (appendix 2 shows screen shot) and social media pages (Facebook and Twitter) to invite people who have taken the particular drugs to contact us if they are interested in participating in the study (as control group).

A proportion of patients who are suspected to have DILI and enrolled as cases will be found to have an alternative explanation for their clinical manifestation found on subsequent investigation. The data and samples taken from these patients will be retained in the study and termed 'symptomatic controls' for analysis purposes. Sample and data collection will be continued in line with confirmed DILI participants.

INFORMED CONSENT

The process for obtaining participant informed consent will be in accordance with the REC guidance, and Good Clinical Practice (GCP) and any other regulatory requirements that might be introduced. Since DILI symptoms are acute and patients often present in emergency departments with sudden onset of severe symptoms, a long period (e.g. 24h) to consider consent is often difficult to provide in order to recruit patients while symptomatic and without asking patients to make additional hospital visits. Therefore, where appropriate, patients will be given a reasonable time period (ideally1 hour without disturbance), to read the patient information sheet and consider enrolment before being approached for consent by the research team. Appropriate steps will be taken to ensure that the patient is able to discuss participation in the study with those not involved in the research. The investigator or

their nominee and the participant or other legally authorised representative shall both sign and date the Consent Form before the person can participate in the study.

The participant will receive a copy of the signed and dated forms and the original will be retained in the Study records. A second copy will be filed in the participant's medical notes and a signed and dated entry made in the notes that informed consent was obtained for the study.

The decision regarding participation in the study is entirely voluntary. The investigator or their nominee shall emphasize to them that consent regarding study participation may be withdrawn at any time without penalty or affecting the quality or quantity of their future medical care, or loss of benefits to which the participant is otherwise entitled. No study-specific interventions will be done before informed consent has been obtained.

The investigator will inform the participant of any relevant information that becomes available during the course of the study, and will discuss with them, whether they wish to continue with the study. If applicable they will be asked to sign revised consent forms.

If the Consent Form is amended during the study, the investigator shall follow all applicable regulatory requirements pertaining to approval of the amended Consent Form by the REC and use of the amended form (including for on-going participants).

Where a participant later withdraws consent, the site PI should be informed. They will be responsible for identification of the participant unique study code and notification of the study CI. The CI (or substitute) will then notify the sample coordinators at University of Nottingham and University of Malaga so that all samples and data can be removed from the project if requested. The eCRF will record whether consent is given for DNA studies and future storage under biobank custodianship after the study closes. It will also record withdrawal of consent enabling samples and data to be tracked and removed accordingly.

Participants who lack capacity to consent to research

Due to the nature of the study some potential participants may lack capacity to give informed consent. This is especially likely in the case of hepatic encephalopathy, which is the occurrence of confusion, altered level of consciousness, and coma as a result of liver failure. It is important that we try to enrol such patients where possible into this study, as research into this area may be of benefit to future patients with this condition. Similarly, some patients with DILI may be suspected to have or have a prior diagnosis of dementia or other pre-existing conditions affecting functioning of the brain. It would be important to include these patients in the study, but appropriate consideration must be made regarding their consent to participate. It is more appropriate in this instance to request consent via a consultee. As the study has minimal risk to the patient, we believe that the benefits of enrolling such patients outweigh the potential risk.

We have referred to the Mental Capacity Act (2005) in our decision to enrol patients who lack capacity. This states that a person lacks capacity and is unable to make a decision for himself or herself due to an impairment of disturbance in the functioning of the mind or brain. Reasonable steps must be taken to see whether the patient/participant lacks capacity. Any research must have the potential benefit to the participant without imposing a burden that is

disproportionate to the potential benefit. Similarly, the research must be intended to provide knowledge of the causes or treatment of their condition.

We have also referred to Clinical Trials Directive 2001/20/EC. This states that inclusion in clinical trials of incapacitated adults who have not given informed consent before the onset of their incapacity shall be allowed only if:

- informed consent of the legal representative has been obtained
- such research is essential to validate data obtained in clinical trials on persons able to give informed consent or by other research methods and relates directly to a life-threatening or debilitating clinical condition from which the incapacitated adult concerned suffers
- the trial has been designed to minimise pain, discomfort, fear and other foreseeable risk in relation to the disease

Therefore, there are a number of provisions specific to research that we have taken into account. It is noted that the research in question will be connected with the impairing condition affecting the participant. We will seek consent from those who lack capacity and have severe liver injury diagnosis. We will not seek consent from those with encephalopathy who would have been likely to lack capacity before any liver injury has occurred. We will recruit patients who have pre-existing conditions affecting brain function only when the suspected drug-induced liver injury has had no observed effects on mental capacity and when no drug- related neurological symptoms are present. We will therefore not be enrolling patients who are believed to have a "double impairment". Secondly, we believe that research of equal effectiveness cannot be carried out if confined to participants with capacity, as, the most severe cases of the condition we are researching, can cause impairments and can affect people with existing conditions affecting mental capacity. We believe that it is important to include these cases in our research. Thirdly, although our research does not directly benefit the patient, this research will provide knowledge of the causes or treatment of others with the same condition, and involves negligible risk to the participant, does not interfere significantly with freedom of action or privacy, and is not unduly invasive or restrictive. We will only take a minimal amount of blood from these patients alongside clinical care.

If the PI or Investigator has reason to suspect that a person may lack capacity, that individual will be assessed according to normal local hospital procedures. If the person is found to lack capacity then the research team will ask someone close to that person if they would consider acting as their personal consultee, e.g. next of kin, a relative, spouse, close friend. The personal consultee should be someone the person who lacks capacity would trust with important decisions about their welfare.

If the PI or Investigator and research team are unable to identify/appoint a suitable personal consultee then efforts will be made to nominate a health professional who is unconnected with the research project to act as a professional consultee. We will firstly ask a member of the standard medical care team of the person who lacks capacity if they would consider taking on the role. The professional consultee will be required to perform the same role as a personal consultee in advising the research team but they may not know the person who lacks capacity and therefore may be required when attempting to determine what the person's wishes and feelings about the research would be if they had capacity, seek the views of any family, friends/carers of the person who lacks consent, or seek other

professional colleagues with an interest in the person's welfare or condition. We will continue to assess capacity throughout the research and will seek consent as soon as feasible.

All consultees (personal and professional) will receive a copy of the consultee information sheet to read. If and when the participant regains capacity, consent will be sought from the participant retrospectively, and will be withdrawn from the study if consent is not given.

We currently have no sites or plans to recruit from sites in Northern Ireland. If this was to occur, we are aware that Northern Ireland follow different legislation and a different process would need to be followed regarding the documentation. We would ensure that this process was followed correctly. We are currently seeking approval to recruit these patients in England and Wales only. We plan to put a future application to a Scotland A REC in order to allow us to recruit patients who lack capacity in Scotland.

STUDY PROCEDURES

SAMPLE COLLECTION
Suspected DILI patients

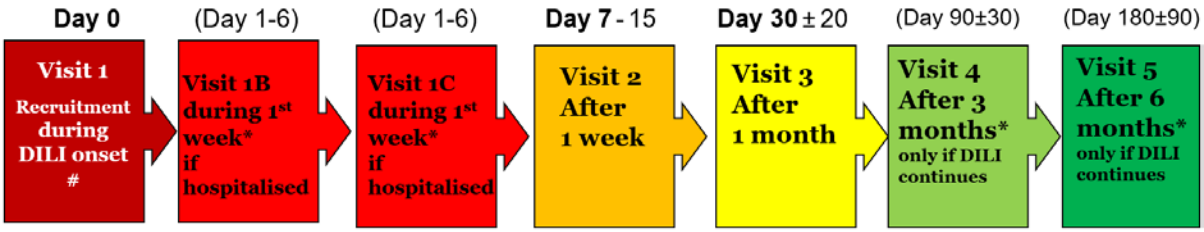
Biological samples will be collected from each enrolled patient on up to 7 occasions (see diagram below). These visits will take place as soon as possible following acute presentation; there will be a time lag of up to 8 weeks between identification of an episode of DILI by the team responsible for the clinical care of the patients and the patient giving consent to take part in the study. Patients' blood tests must meet the eligibility inclusion criteria on the day they are enrolled (Day 0, visit 1) in order to continue to participate in the study.

As far as possible, research bloods will be collected at the same time as bloods for clinical care of the patient and visits arranged to coincide with hospital appointments. Preferred visit times are given below but visits outside the times will not be considered as a protocol deviation. Appendix 1 lists the standard clinical care analyses (including ultrasound, viral hepatitis screen, autoantibody screen, haematology and blood chemistry), and research assessments which may be carried out at each visit.

Patients with suspected DILI who are recruited and subsequently found to not have drug-related injury will continue to provide further samples for this research (as detailed in the PIS). Visit 2 will be 7 ± 8 days from Visit 1. Visit 3 will be 30 ± 20 days after the Visit 1

For those who are in-patients (hospitalised at the time of acute DILI) we will secure additional data and samples on up to 2 further time points (termed visit 1B and Visit 1C) during the period of the patient's hospitalization. In such cases samples will be collected as part of clinical care and additional samples collected for research purposes. If patients continue to have DILI symptoms with raised liver enzymes (i.e. ALT, ALP or total bilirubin levels exceeding the normal range) at Visit 3, a further visit after approximately 3 months (Visit 4) will be arranged to collect the same set of samples. If enzymes remain above normal at this visit, samples will be collected again approximately 3 months later at Visit 5.

Sampling Schedule



*optional

- At each visit a delegated member of the research team will collect:
- Up to 80ml of blood. See most recent study presentation and/or sample collection SOP for details.
- Up to **50 ml of urine** (if possible)
- A Stool sample will also be collected (if possible).

Patients may be consented and samples collected at their home if appropriate and with permission of the PI. Nursing staff would be required to follow their Trust's lone working policy and code of practice.

All biological samples will be processed in each collaborating centre according to the agreed standard operating procedures, labelled and stored at -80°C until sent to the BRC. If a liver biopsy is obtained for clinical reasons any samples available (when the liver biopsy core is >25 mm) will be collected and stored for research purposes. Fresh liver tissue will be snap frozen in liquid nitrogen or placed in RNA later prior to storage at -80°C. (We will also aim to obtain and store any archived liver sections that were taken as part of clinical care). The liver biopsy specimens will be reviewed centrally and classified according to the terminology described in the Appendix 2.

ADVERSE EVENT

DEFINITIONS

Adverse Event (AE): any untoward medical occurrence in a patient or clinical study subject.

Serious Adverse Event (SAE): any untoward and unexpected medical occurrence or effect that:

- **Results in death**
- **Is life-threatening** – *refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe*
- **Requires hospitalisation, or prolongation of existing inpatients' hospitalisation**
- **Results in persistent or significant disability or incapacity**
- **Is a congenital anomaly or birth defect**

Medical judgement should be exercised in deciding whether an AE is serious in other situations. Important AEs that are not immediately life-threatening or do not result in death or hospitalisation but may jeopardise the subject or may require intervention to prevent one of the other outcomes listed in the definition above, should also be considered serious.

REPORTING PROCEDURES

All adverse events (as outlined below) should be reported to the PI and CI. Depending on the nature of the event the reporting procedures below should be followed. Any questions concerning adverse event reporting should be directed to the Chief Investigator in the first instance.

Non serious AEs

All such events, whether expected or not, should be recorded.

This is a case control study and doesn't include any clinical or experimental interventions other than blood sampling. In addition, natural history of DILI includes progressive worsening of the liver injury even after offending agent/ medication/ drug has been withdrawn. Acute liver failure and death are recognised complication. Therefore, while we will record accurately the severity of DILI formally based on the international consensus criteria [4],

AE doesn't include a/ an:

1. exacerbation of a pre-existing illness.
2. increase in frequency or intensity of a pre-existing episodic event or condition.
3. condition detected or diagnosed after medicinal product administration even though it may have been present prior to the start of the study.
4. continuous persistent disease or symptoms present at baseline that worsen following the start of the study.
5. medical or surgical procedure (e.g., surgery, endoscopy, tooth extraction, transfusion); but the condition that lead to the procedure is an AE.
6. pre-existing disease or conditions present or detected at the start of the study that did not worsen.
7. situations where an untoward medical occurrence has not occurred (e.g., hospitalisations for cosmetic elective surgery, social and / or convenience admissions).
8. disease or disorder being studied or sign or symptom associated with the disease or disorder unless more severe than expected for the participant's condition.
9. overdose of concurrent medication without any signs or symptoms.

Serious AEs

An SAE form should be completed and faxed to the Chief Investigator within 24 hours.

A Serious Adverse Event (SAE) is any adverse event occurring following study mandated procedures, having received the treatment or intervention that results in any of the following outcomes. However, acute liver failure, liver transplantation and death are recognised consequences of DILI and these will be recorded as part of the outcome of DILI as recommended by consensus criteria [4]. Therefore, following will NOT be considered as SAE:

1. Liver related death
2. Acute liver failure
3. Liver transplantation
4. Inpatient hospitalisation or prolongation of existing hospitalisation
5. A disability / incapacity

In the current study we will be performing venepunctures for blood sampling. So, we will report major bleeding leading to hypotension, drop in Hb, requirement of resuscitation, transfusion or hospital admission will be reported as adverse events. All SAEs should be

reported to the **Research Ethics Committee** where in the opinion of the Chief Investigator, the event was:

- 'related', i.e. resulted from the administration of any of the research procedures; and
- 'unexpected', i.e. an event that is not listed in the protocol as an expected occurrence

Reports of related and unexpected SAEs should be submitted within 15 days of the Chief Investigator becoming aware of the event, using the COREC SAE form for non-IMP studies.

Local investigators should report any SAEs as required by their Local Research Ethics Committee and/or Research & Development Office.

Sponsor Contact Details for SAEs:

1. email to

Any queries please contact a member of staff in the Research & Innovations department:

Telephone:

ASSESSMENT AND FOLLOW-UP

Follow-up: There are no protocol-driven follow up visits beyond Day 180 (Visit 5).

End of the Study

This study involves the collection and storage of data and tissue. We will be seeking consent to collate, store and use samples and data for a minimum of 10 years.

Collated biological samples will be stored in a REC-approved tissue bank located on HTA licenced premises for subsequent analysis by members of the consortium and collaborators in future projects for at least 10 years.

Data collection

Data to be collected from each patient are described in Appendix 1. Data to be collected from controls will follow the same protocol, with the exclusion of information directly relating to a DILI episode.

The PI in each collaborating hospital will be responsible for entering the corresponding patient/control data into the Pro-Euro DILI Registry database (developed by Coresoft Clinic S.L, http://www.coresoft.es), available online. Tutorials on data entry and database implementations will be organized by the co-ordinating centres (Málaga and Nottingham) during the pilot phase and later if required.

Each participating institute will only be able to access its corresponding patient/data entries in the database, with the exception of the national coordinator, who will have access to all cases from the corresponding country when there are multiple national institutes involved. The two coordinating centres, Málaga and Nottingham will have full access to all patient/control entries. The master database will be held by Malaga in a password encrypted file.

Documents relating to the study that contain personal data that may disclose the identity of the subject will be stored in the Investigator Site File securely, with restricted access. The Investigator should not provide any personal data that may identify the subject to any third party at any time during or after the study. Subject confidentiality will be further assured by utilising unique subject identification code number. The link between the patient's name and

code will be broken and the sample completely anonymised when the study is completed and all clinical data have been obtained.

The following identifiers along with the trial ID will be detailed in the study recruitment log at site to allow identification of the participant's tissue samples or when chasing data queries with participating remote sites.
Each participant will be assigned a trial identity code number, allocated at randomisation if appropriate, for use on CRFs other trial documents and the electronic database.

CRFs will be treated as confidential documents and held securely in accordance with regulations. The investigator will make a separate confidential record of the participant's name, date of birth, local hospital number or NHS number, and Participant Study Number (the Study Recruitment Log), to permit identification of all participants enrolled in the trial, in accordance with regulatory requirements and for follow-up as required.

CRFs shall be restricted to those personnel approved by the Chief or local Principal Investigator and recorded on the 'Study Delegation Log.'

All paper forms shall be filled in using black ballpoint pen. Errors shall be lined out but not obliterated by using correction fluid and the correction inserted, initialled and dated.

The Chief or local Principal Investigator shall sign a declaration ensuring accuracy of data recorded in the eCRF.

STATISTICS AND DATA ANALYSIS

All routine laboratory analysis will be done by appropriate hospital as part as normal patient care. Blood will be processed for storage of serum and plasma as outlined in the SOP. Data and all appropriate documentation will be stored for a minimum of 25 years after the completion of the study, including the follow-up period.
We will collate detailed and comprehensive data using an electronic case report form (eCRF) to achieve in-depth phenotyping.

The procedure for data removal in the event of withdrawal of consent is described on pg 25.

Statistical Analyses

Researchers will be blinded to patient type/group being investigated – samples will be anonymized and labelled only with study ID code. Study groups will be compared using T-tests and non-parametric testing to identify biomarkers which distinguish DILI.

STUDY MANAGEMENT

The day-to-day management of the study will be co-ordinated through Nottingham Digestive Diseases Centre and NIHR Nottingham Biomedical Research Centre.

SPONSOR

Nottingham University Hospitals NHS Trust will act as the main sponsor for UK based sites for this study. Delegated responsibilities will be assigned to the NHS trusts taking part in this study.

FUNDING

<u>Participant stipends and payments</u>

Participants will not be paid to participate in the study. Travel expenses will be offered for any hospital visits in excess of usual care.

AUDITS

The study may be subject to inspection and audit by Nottingham University Hospitals under their remit as sponsor and other regulatory bodies to ensure adherence to GCP and the NHS Research Governance Framework for Health and Social Care (2nd edition).

STUDY CONDUCT

Study conduct will be subject to systems audit for inclusion of essential documents; permissions to conduct the study; CVs of study staff and training received; local document control procedures; consent procedures and recruitment logs; adherence to procedures defined in the protocol (e.g. inclusion / exclusion criteria, timeliness of visits); accountability of study materials and equipment calibration logs.

STUDY DATA

Study data is collected on paper CRFs (an overview of variables collected is provided in Appendix 1 of this document and paper version Pro Euro DILI CRF to collate required data for electronic database is available as separate file) and would therefore be considered source data. This is inputted onto web site http://proeurodili.eu/. Although this study will not be routinely monitored, standard error checking and remote monitoring will be conducted by Nottingham and Malaga teams. Monitoring of study data may include confirmation of informed consent; source data verification (where applicable) and data storage procedures according to NUH SOP_RES 013.

CRFs may be spot-checked on a regular basis for error-checking. In addition, the subsequent capture of the data on the study database will be checked. Where corrections are required these will carry a full audit trail and justification.

Study data and evidence of monitoring and systems audits will be made available for inspection by the REC as required.

TRANSPORT AND STORAGE OF THE TISSUES

Serum, plasma, whole blood (EDTA), urine, and stool samples will be stored in aliquots at -80°C in the BRC within Nottingham University Hospitals NHS Trust. When the study closes, where consent is given, samples will be transferred to an appropriate ethically approved research tissue bank and made available for ethically approved research programs with approval of partners as specified in collaboration agreements to facilitate completion of the research towards the stated aims. Residual samples stored at Nottingham University Hospitals NHS Trust will be transferred to the custodianship of University of Nottingham and stored in the NDDC-BRU Research Tissue Bank.

The Biobank Managers (or Research Tissue Bank Administrator) will be responsible for removal and destruction of samples belonging to participants who withdraw approval. The procedure for this will be that participant should contact recruiting site team intitially so that they can determine the participant individual study code. (Where this team is no longer available, the site manager responsible for R&I should provide an alternative contact person or the Study CI may be contacted to advise on access to the confidential study data). The CI (or substitute) should be notified of the study ID and be responsible for contacting the Biobank teams in order to locate the relevent samples and request removal of data from the associated databases (Pro Euro DILI database via the Investigators stated in this document).

We will also receive samples of plasma, serum, whole blood, urine and stool from the respective national collaborators in batch shipments by courier as frequently as required.

All shipments will comply with HTA/EUTCD and will contain a complete inventory of all samples (see separate sample processing and shipping log template document), along with the name of the person responsible for sending the samples and a copy of the signed MTA. Samples will be stored within the Research Tissue Bank for future research.

Since this study is concerned with collection of samples to form a research tissue bank resource and includes research/analysis, patients who do not wish samples to be used for future research will not be recruited. Where participants do not agree to the future use of the samples, they will be destroyed in accordance with the Human Tissue Act, 2004 when the study ends.

DISCONTINUATION OF THE TRIAL BY THE SPONSOR

The Sponsor reserves the right to discontinue this study at any time for failure to meet expected enrolment goals, for safety or any other administrative reasons. The Sponsor shall take advice as appropriate in making this decision.

GOOD CLINICAL PRACTICE
ETHICAL CONDUCT OF THE STUDY

The study will not be initiated before the protocol, consent forms and participant information sheets have received approval / favourable opinion from the Research Ethics Committee (REC), and the respective National Health Service (NHS) Research & Development (R&D) department. Should a protocol amendment be made that requires REC approval, the changes in the protocol will not be instituted until the amendment and revised informed consent forms and participant information sheets (if appropriate) have been reviewed and received approval / favourable opinion from the REC and R&D departments. A protocol amendment intended to eliminate an apparent immediate hazard to participants may be implemented immediately providing that the REC are notified as soon as possible and an approval is requested. Minor protocol amendments only for logistical or administrative changes may be implemented immediately; and the REC will be informed.

The study will be conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki, 2013; the principles of Good Clinical Practice, and the Department of Health Research Governance Framework for Health and Social care, 2005.

CONFIDENTIALITY

All laboratory specimens, evaluation forms, reports, and other records will be identified in a manner designed to maintain participant confidentiality. All records will be kept in a secure storage area with limited access to study staff only. Clinical information will not be released without the written permission of the participant, except as necessary for monitoring and auditing by the Sponsor or its designee. The CI and study staff involved with this study will not disclose or use for any purpose other than performance of the study, any data, record, or other unpublished, confidential information disclosed to those individuals for the purpose of the study. Prior written agreement from the Sponsor or its designee will be obtained for the disclosure of any said confidential information to other parties.

DATA PROTECTION

The CI and study staff involved with this study will comply with the requirements of the Data Protection Act 1998 with regard to the collection, storage, processing and disclosure of personal information and will uphold the Act's core principles. The CI and study staff will also adhere, if appropriate, to the current version of the NHS Scotland Code of Practice on Protecting Patient Confidentiality. Access to collated participant data will be restricted to the CI and appropriate study staff.

Computers used to collate the data will have limited access measures via user names and passwords.

Published results will not contain any personal data that could allow identification of individual participants.

Pro-Euro DILI Software strictly follows the regulations imposed by the European Union, ensuring all data collected is secure and protected throughout the trial process. Only pseudonymised data is collected in the study database

Study data is only stored within the eCRF system. At study closure a pdf of each patient's eCRF is created and forwarded to the responsible physician. At any stage, a patient can request a copy of his own data compiled in user-friendly, easily readable pdf format.

INDEMNITY

Standard NHS indemnity applies.

STUDY CONDUCT AND RESPONSIBILITIES

PROTOCOL AMENDMENT, DEVIATIONS AND BREACHES

The CI will seek approval for any amendments to the Protocol or other study documents from the Sponsor, REC and NHS R&D Office(s). Amendments to the protocol or other study docs will not be implemented without these approvals.

In the event that a CI needs to deviate from the protocol, the nature of and reasons for the deviation will be recorded in the CRF, documented and submitted to the Sponsor. If this necessitates a subsequent protocol amendment, this will be submitted to the Sponsor for approval and then to the appropriate REC and lead NHS R&D Office for review and approval.

In the event that a serious breach of GCP is suspected, this will be reported to the Sponsor immediately.

RECORDS

Sample Labelling Samples will be stored in linked anonymised format in the BRC laboratories. Each participant will be assigned a trial identity code number for use on the samples, consent forms and other study documents and the electronic database. All samples (whether from Nottingham or elsewhere in Europe) will be labelled according to an agreed system compatible with the database developed by Coresoft as outlined in separate document: SOP DILI Biological Samples Collection. Samples for clinical care clinical analysis will be labelled in accordance with local procedures.

Source documents

Source documents shall be filed at the investigator's site and may include but are not limited to, consent forms, current medical records, laboratory results and records. CRF's may also completely serve as its own source data. Only study staff as listed on the Delegation Log shall have access to study documentation other than the regulatory requirements listed below.

Direct access to source data / documents

Study documents, including progress notes and copies of laboratory and medical test results shall be made available at all times for review by the Chief Investigator, Sponsor's designee and inspection by relevant regulatory authorities.

The Principal investigator in each collaborating hospital will be responsible for entering the corresponding patient/control data into the Pro-Euro DILI Registry database (developed by Coresoft Clinic S.L, http://www.coresoft.es), available online. Tutorials on data entry and database implementations will be organized by the co-ordinating centres (Málaga and Nottingham) during the pilot phase and later if required.

The web-based application and data will be hosted at Malaga University servers, with automated data backups on a daily basis, a report system for successful backup monitoring and reliable network connectivity. The software strictly follows the regulations imposed by the European Union ensuring that registries are secure and protected by the law. Furthermore, the software follows the Declaration of Helsinki requirements to ensure protection of patient's rights. No data that can reveal the patients identity will be entered into the database. Internet access to the registry platform is password protected and all stored passwords will be encrypted. A separate document entitled 1- SOP PED-Data Entry & Management 1.0, describes the procedures for electronic data entry and management including verification and quality control. Subsequent transfer of data subsets to collaborators partners will be subject to same regulations and follow standard secure transfer procedures.

All study staff and investigators will endeavour to protect the study participants' rights to privacy and informed consent, and will adhere to the General Data Protection Regulation laws. Our policy is available publically via www.nuh.nhs.uk/GDPR. The sponsor, Nottingham University Hospitals Trust will be the data controller. Only the minimum required information for the purposes of the study shall be collected. Documents will be held securely, in a locked room, or locked cupboard or cabinet. Access to the information will be limited to the study staff and investigators and relevant regulatory authorities (see above). Computer held data including the study database will be held securely and password protected. All data will be stored on a secure dedicated web server. Access will be restricted by user identifiers and passwords (encrypted using a one way encryption method). Identifiable personal data will be securely kept and confidential destruction arranged 12 months after the study ends.

Information about the study in the participant's medical records / hospital notes will be treated confidentially in the same way as all other confidential medical information.

RECORD RETENTION AND ARCHIVING

In compliance with the ICH/GCP guidelines, regulations and in accordance with the Nottingham University Hospital SOP-RES-028 and Research Ethics, the Chief or local Principal Investigator will maintain all records and documents regarding the conduct of the study. These will be retained for 25 years. If the responsible investigator is no longer able to maintain the study records, a second person will be nominated to take over this responsibility.

The study documents held by the Chief Investigator on behalf of the Sponsor shall be finally archived at secure archive facilities. This archive shall include all anonymised audio recordings, study databases and associated meta-data encryption codes.

END OF STUDY

The end of study is defined as last patient last visit. The Sponsor or CI have the right at any time to terminate the study for clinical or administrative reasons.

The end of the study will be reported to the Sponsor and REC within 90 days, or 15 days if the study is terminated prematurely. The CI will ensure that any appropriate follow up is arranged for all participants.

A summary report of the study will be provided to the Sponsor and REC within 1 year of the end of the study.

This study involves the collection and storage of data and tissue to form a biorepository for future studies. When the study ends, collected biological samples will be stored in a REC-approved research tissue bank located on HTA licenced premises for subsequent analysis by members of the consortium and collaborators in future projects for at least 5 years. The research tissue bank project will be reviewed and renewal requested from the REC every 5 years. We will be seeking consent to collate, store and use samples and data for a minimum of 10 years.

REPORTING, PUBLICATIONS AND NOTIFICATION OF RESULTS

AUTHORSHIP POLICY

Ownership of the data arising from this study resides with the study team and their respective employers and with collaborating partners as set out in contracts of collaboration executed by either the sponsor R&I department (Nottingham University Hospitals NHS Trust or study team employer's legal representatives where appropriate (from University of Nottingham). On completion of the study, the study data will be analysed and tabulated, and a clinical study report will be prepared.

PUBLICATION POLICY

The clinical study report will be used for publication and presentation at scientific meetings. Investigators have the right to publish orally or in writing the results of the study.

Summaries of results will also be made available to Investigators for dissemination within their clinical areas (where appropriate and according to their discretion).

All potential manuscripts deriving from Pro-Euro-DILI Network samples and patient data must be presented to the scientific committee for acceptance prior to submission. Publication authorship will be dependent on study participation and case recruitment, with centres having enrolled a greater number of cases appearing earlier in the list of authors than those with fewer case enrolments. In the event of patent applications resulting from the Pro-Euro-DILI Network data the authorship will be determined based on the same strategy as outlined for publications.

<u>Plan of diffusion</u>

The Pro-Euro-DILI Network (https://proeurodilinet.eu/) website has been developed in order to disseminate the presence of the consortium and its work. The coordinating centres (Málaga and Nottingham) will be responsible for the initiation and maintenance of the web site. All collaborating centres are encouraged to present the Pro-Euro-DILI Network at national conferences, but any potential work to be presented must be provided to the

scientific committee prior to submission. If a group wants to present multinational data, i.e. data relating to cases from other countries, permission must be obtained from the scientific committee.

USER AND PUBLIC INVOLVEMENT

We will disseminate findings to the public and provide study information and updates on our website and newsletter and at public engagement events such as the BRC open day. We held a meeting of representatives from the BRC patient advisory group in June 2018 to provide feedback about study dissemination. In particular advice was provided to develop strategies for patient engagement and publicity to improve recruitment. As a consequence we developed improved patient information and website. The advisory group were also supportive of our plan to extend study through securing funding via a European consortium in order to develop biomarker tools for use in clinical care. They felt that the purpose of a biorepository would be to share samples and develop collaborations worldwide since this is a rare condition and this would enable specialize analyses to be included.

REFERENCES

1. de Abajo FJ. et al., Br J Clin Pharmacol. 2004;58:71-80.
2. Sgro C. et al., Hepatology. 2002;36:451-5.
3. Björnsson ES. et al., Gastroenterology. 2013;144:1419-25.
4. Aithal GP. et al., Clin Pharmacol Ther. 2011;89:806-15.
5. Marie-Lea Gauci et al., 2018 J Hepatology; 69 j 547–558
6. Andrade RJ, Lucena MI, Fernández MC, et al. Gastroenterology 2005;129:512-51.
7. Chalasani N, Fontana RJ, et al.. Gastroenterology 2008;135:1924-1934.
8. Aithal GP. Nat Rev Rheumatol. 2011;7:139-50.
9. Lucena MI. et al., Gastroenterology. 2011;141:338-47.
10. Antoine DJ, Dear JW, Lewis PS, et al. Hepatology 2013;58:777-87.
11. Wree A. et al., Digestion. 2011;84:54-9.
12. Urban TJ, Daly AK, Aithal GP. Seminars in Liver Disease 2014; 34:122-33.
13. Foureau, D. 2014, Clinical and experimental immunology 180:40-51.
14. Taubert R. 2014 J Hepatology 61;1106-1114.
15. Benesic A et al. Gut. 2015; 0,1-9.
16. Kay AW et al. Methods Mol Biol. 2016; 1441: 13–26.

APPENDICES:

Appendix 1. Overview of Patient assessment schedule

	Day 0	Day 1-6	Day 2-6	Day 7 -15	Day 30 ±20	Day 90 ±30	Day 180 ±90
	Visit 1#	Visit 1B Optional§	Visit 1C Optional§	Visit 2	Visit 3	Visit 4 Optional §§	Visit 5 Optional §§§
Inclusion/Exclusion criteria (LFTs reviewed after visit completed)	X						
Medical history comorbidity	x						
current medical condition	X	X	X	X	X	X	X
Demography inc pregnancy and ethnicity	X						
Body height and weight	X					X	X
Drug history in past 6 months or longer, including OTC, herbal, dietary supplement, recreational drugs (include at least 10 most recently taken drugs).Subsequent visits only need to document new drugs	X	X	X	X	X	X	X
Smoking history	X						
Alcohol, quantitative assessment	X						
Viral hepatitis screen (where appropriate for clinical care)	X						
Autoantibody* screening (where appropriate for clinical care)	X				(X)**		
Liver ultrasound(where appropriate for clinical care)	X						
Liver ultrasound(where appropriate for clinical care)	X						
Blood sample collection (see separate SOP for detail) up to 80 ml in total inc clinical bloods	X	X	X	X	X	X	X
Urine sample collection if possible	X	X	X	X	X	X	X
Liver biopsy tissue where done as part of clinical care (surplus tissue only)	X						
Stool sample collection if possible	X	X	X	X	X	X	X

*Autoantibodies: antinuclear (ANA), antismooth muscle (ASMA), antimitochondrial (AMA) and liver kidney microsomal type 1 (LKM-1)

**Repeat autoantibody screening if positive in week 1

For 'past DILI' patient group and control groups there will be just a single visit, visit 1 – clinical blood tests will not be done.

§Only if patient is hospitalized, §§Only if patient has elevated liver profile values at visit 3, §§§Only patients with elevated liver profile values at previous visit (visit 4)

Appendix 2 Histopathological terms to be used for liver biopsy reports in patients with suspected drug-induced hepatotoxicity.

CODE	DESCRIPTION
1	HEPATOCELLULAR NECROSIS
11	FOCAL NECROSIS
12	BRIDGE NECROSIS
13	ZONAL NECROSIS
14	MASSIVE NECROSIS
2	STEATOSIS / FATTY LIVER
21	ACUTE STEATOTIC CHANGES
22	STEATOHEPATITIS (NASH)
3	GRANULOMATOUS REACTION
4	ACUTE COLESTASIS
41	CHOLESTASIS WITHOUT HEPATITIS
42	CHOLESTASIS WITH HEPATITIS
43	CHOLESTASIS WITH DUCTAL LESION
5	CHRONIC CHOLESTASIS
51	PROLONGED CHOLESTASIS
52	DUCTOPENIA ("VANISHING BILE DUCT SYNDROME")
53	SCLEROSING CHOLANGITIS
6	CHRONIC HEPATITIS
61	CHRONIC ACTIVE HEPATITIS
62	HEPATIC FIBROSIS AND CIRRHOSIS
7	VASCULAR ALTERATIONS
71	SINUSOIDAL DILATATION (FLARING) AND PELIOSIS
72	NON-CIRRHOTIC PORTAL HYPERTENTION
73	OBSTRUCTION TO THE VENOUS HEPATIC FLOW (Budd-Chiari
74	OTHERS
8	HEPATIC TUMORS
81	HEPATOCELLULAR ADENOMA
82	HEPATOCELLULAR CARCINOMA
83	OTHER CARCINOMAS
84	ANGIOSARCOMA
9	UNSPECIFIC CHANGES

Appendix 3

Drug-induced Liver Injury

Introduction to DILI

- DILI is an unexpected injury to the liver that can be caused by prescribed medications, over-the-counter medications, recreational drugs and supplements.
- Some drugs such as paracetamol are known to be harmful to the liver in high doses but other drugs, taken at a normal dose can unpredictably damage the liver of certain people.
- DILI is a rare, unpredictable disease and it is not known why certain individuals are more likely to develop drug-induced liver injury than others. This can be life-threatening.
- Specific genetic risk factors can contribute to individual susceptibility but they do not fully explain why people get DILI.

Study Aims

> To create a unique International Biobank resource to enable research into the causes and characteristics of drug-induced liver injury (DILI) so that new, non-invasive diagnostic tests can be developed

> To predict and prevent drug-induced liver injury (DILI) so patients can be safely treated with medications they need.

This research will:

- Improve our understanding of how medications cause DILI
- Identify risk factors for DILI development
- Develop and evaluate new non-invasive tests to specifically diagnose DILI.

Who can participate?

Men & women aged over 18, who appear to have had an adverse reaction to a medication or supplement which affected liver functioning (patients); or who have had no problematic response to taking specific medications or supplements (healthy volunteers)

Participation involves:

Patients:

Several visits/appointments with our research nurse (coinciding with your medical care where possible) to provide:

- a blood sample,
- a urine sample (optional)
- a stool sample (optional)
- Medical history

Healthy volunteers:

One visit/appointment with our research nurse to provide:

- a blood sample,
- a urine sample (optional)
- a stool sample (optional)
- Medical history

Study contacts:

Dr Jane Grove, Study Coordinator, jane.grove@nottingham.ac.uk 0115 9249924 ext 31151

Amy Ward, Research/Specialist Nurse, amy.ward2@nuh.nhs.uk 0115 9249924 ext 60612

Prof Guru Aithal, Principal Investigator 0115 8231149