Barts Health NHS Trust
The Royal London Hospital
Pathology and Pharmacy Building
80 Newark Street, London E1 2ES

Institute of Cell and Molecular Science
Queen Mary University London

Dr Daniel Hart, Chief Investigator
Dr Drew Provan, Collaborator
Tel: +44 (0) 207 377 7000, Ext 61114

Fax: +44 (0) 203 246 0230
Email: d.hart@qmul.ac.uk
uk-itp.registryteam@nhs.net

Study Protocol
UK Adult ImmuneThrombocytopenic Purpura (ITP) Registry

Purpose:

This study seeks to uncover information regarding the natural progression[1], treatment effectiveness, and co-morbidities of adult ITP in the United Kingdom via establishment of a multi-centre disease registry.

Background:

A previous version of this study was submitted to the London Research Ethics Committee (MREC) in 2002 under the title, "Establishment of a UK Registry for Adults with Immune Thrombocytopenic Purpura (ITP) and Investigation into the Role of Cytokine Genes." The study was ethically approved in August 2002 (MREC/02/2/58) and remained highly active until August 2005, during which time over 600 patients were enrolled.

Due to unforeseen circumstances, which resulted in a temporary switch of positions, the chief investigator was forced to slow analysis of the cohort at this time. He has since returned to The Royal London Hospital as both a consultant haematologist and senior clinical lecturer and is eager to reinitiate the study with minor substantive alterations. He will be aided in this task by a study coordinator, a molecular scientist, and a data manager, full-time positions funded through support from GlaxoSmithKline (GSK) Inc. and The ITP Support Association.

Design & Methodology:

Recruitment & Consent:

The proposed protocol entails investigation of prospective and past study participants. Adult patients presenting at haematology clinics at The Royal London Hospital or collaborating centres with a low platelet count (< 100 × 10^9/L)[2] and no evidence of known thrombocytopenic-inducing disease or treatments following a standard ITP workup will be invited to take part in the study. Any patients who were diagnosed with ITP at any time, even prior to the publication of the latest international consensus in 2010, will continue to be enrolled onto the registry. At The Royal London Hospital, recruitment and informed consent will be conducted by the chief investigator and two clinical investigators.

These tasks will be directed by members of the chief investigator's team[3] at all other external clinics. Consultant haematologists at these sites will be issued a study overview (**UKITP Haematologist Overview 2.4**) and asked to identify all eligible patients. Following a brief introduction to the investigation, consultants will ask patients for their permission to forward their name and address to the data manager.

These patients will subsequently be mailed a comprehensive, lay overview of the study (**UKITP Prospective Participant Overview 3.1**), which will contain means through which to contact both the chief investigator and study coordinator with any pre or post-enrolment questions or concerns. Included with this paperwork will be two informed consent agreements, concerning the study and subsequent tissue storage respectively, returnable to

[1] Natural progression shall be defined as platelet count, bleeding events, and mortality over time.
[2] Following the international consensus on diagnosis of ITP (Provan et al, 2010), the platelet count criterion changed from 150 x 10^9 to <100 x 10^9. All patients on previous diagnosis base on platelet count <150 x 10^9 will continue to be enrolled.
[3] The chief investigator's team shall be comprised of the Chief Investigator, the Lead Epidemiologist, and the Data Manager.

the study coordinator via a pre-paid, self-addressed envelope. Originals of this paperwork will be kept on file at The Royal London Hospital.

Blood Collection & Genetic Testing:

At the time of registration, one blood sample (15 ml, ~3 teaspoonsful) will be drawn during a routinely scheduled venepuncture (or a saliva sample [Oragene® kit]) and mailed to the study coordinator, who will be responsible for its semi-anonymisation via labelling with a uniquely allocated study number. For some patients, especially those who are seen at the Royal London Hospital, an additional 20mls of blood will be collected for T cell analysis. The Chief Investigators team will inform the patients if this is required during their visit at the hospital. All samples collected will be sent to The Molecular Haematology Laboratory at The Royal London Hospital and/or Blizzard Institute for safe storage and analysis.

Genetic contents will be isolated and stored for subsequent single-nucleotide polymorphism (SNP) and gene expression analysis. Polymerase chain reaction (PCR) or other contemporary genetic technology will be used to amplify DNA regions of genes hypothesised to be linked with ITP or other autoimmune diseases. The presence of SNPs within genes will be recorded and assessed for their ability to predict disease severity and patient response to treatments. Genome-wide gene expression will be measured using state of the art mircroarray technology, which will permit snapshots of expression at a particular time in a particular cell type. These expression profiles will be used as an early, hypothesis-generating tool to gauge whether novel genes may be involved in the disease process.

Blood Collection for Immune Profiling:

A subset of patients will be invited to contribute up to four blood samples of 50mls (10 teaspoons) over a six month period for detailed analysis of anti-platelet antibodies, B and T cell profiling, and B and T cell responses to autoantigens. Existing registry patients or those who have expressed an interest in participating as outlined in the section on Recruitment & Consent above, will be given the updated patient information leaflet (UKITP Prospective Participant Overview 3.1) with a verbal explanation of the substudy, by their usual physician, during the course of a routine consultation. Donations will be obtained at a subsequent routine visit, when patients have had a minimum of 24 hours to consider their involvement in the study.

The first bleed of 50 mls will be used for analysis of anti-platelet antibodies. This analysis is performed in patients as part of routine service and if such an analysis has been undertaken within the previous three months it will not be repeated.

The subsequent blood samples will be used for analysis of leucocyte surface markers using flow cytometry, for B cell and T cell functional assays to determine responses to autoantigens found in ITP, autoantibody assays and analysis of gene and protein expression. During the course of the cellular assays, blood samples will be transported to GSK laboratories and other 3rd parties at GSKs discretion, for completion of analysis. All samples will be stored in accordance with the HTA guidelines and tracked using GSKs human biological samples management database. Any remaining samples will be destroyed at the end of the study.

Medical Record Extraction Procedure:

During the defined ten-year study period, standard proforma will be used to extract information from participant hospital medical records at registration and on a schedule determined by the local centres but it will be done at least once annually. Some centres may wish to collect information more frequently than yearly because the study-related data are readily available soon after participants are seen by their haematology teams. Some may find it more manageable to perform data collection on a case by case basis than several at a time. Others may wish to schedule specific points in time to collect data on all their participants. The registry can assist any centres decide on the best data collection schedule that suits them. Whatever schedule a particular centre wishes to adopt will be entirely its own decision. For study participants registered at The Royal London Hospital, these extractions will be undertaken by the registry's team. The data detailed under the heading 'Field for Extraction'

will be directly entered into an electronic study database and cross-verified. Should inconsistencies arise during this check, a subsequent extraction will be performed and similarly cross-verified. At all other collaborating centres, a member of the patient's clinical care team will be responsible for performing the scheduled extractions and for forwarding the abstracted data to the chief investigator's team using a secured server.

Although we expect that most information that we require will be available from the medical records at the referral hospital (i.e. collaborating centre), on certain occasion some may not be available. This may be attributed to several potential reasons:

At presentation or registration

1. Not all the information which is relevant to the study may have been sent during referral to RLH or collaborating centres, especially for participants who were diagnosed quite some time before their registration onto the study.

During follow up

1. After some time, certain patients may have been discharged to other healthcare centres for monitoring or ITP management, e.g. general practitioners.
2. Some patients may migrate to other regions within the UK and require to be followed up.
3. A minority of patients may have unfortunately passed away which require to be recorded.

To address these situations, we will search new and existing NHS services. Some of these services were introduced to improve access to data and allow follow up of patients for various clinically important reasons and to facilitate research.

At the Royal London Hospital, just like many other NHS healthcare providers, the highly secure Spine portal will come into place to allow access to patients' data under the 'Connecting for Health' scheme. Summary Care Records is the service that we will use to obtain up-to-date key health information on our participants. Only data that are required for our study will be extracted from this source. However, this service is still being updated nationally (mainly by general practice) and data may not be available for many patients yet.

We recognised that general practitioners are more likely to be up-to-date about the patients' overall conditions and care. They may also be involved in monitoring ITP patients and may also be involved in their management. We will therefore contact them to request certain information. The data that we will ask GPs to provide are the same that we will be gathering from the collaborating centres. What we require from general practitioners is described in the proforma (version 1.0) which will be accompanied by a letter (GP letter version 1.0) that outlays our research aims and the reasons behind our request.

The Data Linkage Services provided by Health and Social Care Information Centre is the NHS research resource that we will engage. This is a highly secure service which can trace patients records for certain study-related data, such as demographic status and mortality (including cause of death or whether certain patients is not in the UK) and hospital episode information (e.g. certain diagnoses or operations). Therefore, this valuable tool will aid our effort to reduce lost to follow up and obtain more up-to-date study-related data about our participants.

These resources are important to this study and will ensure that our data is as complete and up-to-date. Below is a list of the fields on which we wish to extract data from the various sources mentioned.

Fields for Extraction

The fields that are for data collection at registration are listed below. Some of them are <u>also</u> for collection during follow up and are marked 'FU' accordingly.

Demographic, anthropometric and healthstyle information:

- Patient Name and Contact Address [+FU]
- Patient Gender
- Patient Date of Birth
- Ethnicity
- Patient Weight at Diagnosis
- Patient Height
- BMI at diagnosis
- Smoking status [+FU]
- Alcohol consumption [+FU]

Referral centre information at diagnosis and registration (if different)
- Name of referrer
- Specialty of referrer
- Centre name and contact address
- Centre phone and fax numbers

Current Hospital and Haematologist
- Name [+FU]
- Centre Name and Contact details[+FU]
-

Key information and dates:
- Registration Date
- NHS number
- Name, specialty and address of healthcare practitioner(s) who made ITP diagnosisDate of Diagnosis
- Date of Last Clinic Visit [+FU]

Bleeding Events[+FU] (yes/no, severity, date)
- Cutaneous Bleeds
- Bleeds from the Oral Cavity
- Epistaxis
- Uterine Bleeds
- Haematuria
- Gastrointestinal Bleeds
- Intracranial Haemorrhage
- Muscle Bleeds
- Joint Bleeds
- Subconjunctival Haemorrhage
- Retinal Bleeds

Treatment [+FU](Yes/No; Date(s), Dosage(s) & Duration(s) where applicable)
- Prednisolone
- IVIg
- Splenectomy (Laparoscopic/Open Technique)
- Anti-D
- Methylprednisolone
- Dexamethasone
- Danazol
- Dapsone
- Azathioprine
- Cyclophosphamide
- Vinca Alkaloids
- Mycophenolate
- Eltrombopag

- Romiplostim
- Plasmapheresis
- Protein A Immunoadsortion
- Interferon
- Cyclosporine
- Rituximab
- Platelet Transfusion
- Red Blood Cell Transfusion
- *H. pylori* Treatment
- Vitamin C Supplements

Co-therapies

- Anti-lipid therapy
- Antihypertensive therapy
- Anticoagulation therapy
- Thrombolysis therapy
- Antifibrinolytic therapy
- Other

Co-Morbid Conditions^{+FU} (Yes/No, Date of diagnoses)

- Cataracts
- Osteoarthritis
- Type I Diabetes
- Type II Diabetes
- Hypertension
- Peptic Ulcers
- *H. pylori* Infection
- Renal Failure or Impairment
- Chronic Liver Disease
- Hypercholesterolemia
- Myocardial Infarction
- Unstable Angina
- Revascularisation procedure
- Sudden cardiac death
- Ischaemic Stroke
- Transient Ischaemic Attack
- Deep Vein Thrombosis
- Pulmonary Embolism
- Splenomegaly
- Thyroid Disease
- Depression/Anxiety
- Miscarriage
- Cushing's Syndrome
- *Candida* Infection
- Pneumonia
- Other Autoimmune Disease
- Haematological Malignancy
- Solid Tumour/Malignancy
- Photoxicity
- Other

Family history^{+FU}

- Cancer
- Ischemic Heart Disease
- Stroke
- ITP or other autoimmune disease
- Any other relevant family history

Biochemical Fields (Levels at Diagnosis)

- Alanine Transaminase (ALT)
- Aspartate Transaminase (AST)
- Alkaline Phosphatase (ALP)
- Bilirubin

Haematological Fields

- Platelet Count(s) (Count & Date) $^{+FU}$
- Haemoglobin $^{+FU}$
- White Blood Cells (Level at Diagnosis) [Neutrophils $^{+FU}$
- Red Blood Cells Count (Level at Diagnosis) Mean Platelet Volume (MPV) [Volume at Diagnosis]
- Blood Group (A, B, AB & O; Rh Positive/Negative) Marrow Aspirate (Yes/No, Conclusions)
- Trephine Biopsy (Yes/No, Conclusions) Direct Agglutination Test (DAT) (Level at Diagnosis)

Immunological Fields (Levels at Diagnosis) Immunoglobulin

  - IgG
  - IgM
  - IgA

- Anti-Nuclear Antibodies

- Coagulatory Fields (Levels at Diagnosis) Prothrombin Time (PT)
- Activated Partial Prothrombin Time (APPT)
- Lupus Anticoagulant (LA)
- Anticardiolipin Antibody (aCl)
- Reticulocyte Percentage

Diagnostic Information:

- Change of ITP Diagnosis (Yes/No) $^{+FU}$
- Indium Scanning ($t_{80\%}/t_{30\ minute}$ spleen/liver ratio, date) $^{+FU}$

Clinical contacts and resource utilisation (mainly at RLH and some centres[4])

- Hospitalisation
- Clinic visits

Other

- Mortality and cause of death

Data Protection:

The collection of information from medical records at The Royal London Hospital will be performed by the chief investigator's team. For participants at all other centres, the data retrieval process will be conducted by a member of the local clinical care team. Access to hospital medical records at these sites will be extended to the chief investigator's team but will be limited to monitoring and quality assurance purposes only. To note, our current data protection safeguards which are in place also match the requirements expressed by the HSCIC for using its Data Linkage Services.

Importantly, all information collected will be kept strictly confidential. Any published data will be anonymised so that participants cannot be identified from it. Annually during the study, fully-anonymised data will be shared with medical researchers at GlaxoSmithKline Research Ltd. and the Paediatric & Adult Intercontinental Registry on Chronic Idiopathic Thrombocytopenic Purpura (PARC-ITP) Study in Basel, Switzerland. These partnerships will enhance resources with which to investigate the natural progression, causes, and treatment of adult ITP while strengthening analysis of the study's findings. The information submitted to

[4] Centres which are able to provide this data will be invited to do so in the next registry update due at the end of 2013 and beginning of 2014.

these two organisations will contain no personally-identifiable material, and all planned analyses utilising it will require favourable review from a research oversight body.

Past Study Participants:

Owing to the non-sensitive nature of the proposed revisions, The London MREC determined that it would not be necessary to re-consent past study participants. The same information will be extracted from the hospital records of past participants as from those of prospectively enrolled participants.

Participant Medical Record Extraction Analysis:

Information obtained from participants' medical records, general practices and NHS research services will be used to conduct the qualitative and quantitative analyses highlighted in Appendix A1, which includes investigating the natural progression of the disease and the effectiveness of currently implemented therapies. These analyses will be carried in stages from 2013 until the conclusion of the study in 2018; results from these investigations will be published in peer-review journals, in our study newsletter, and in *The Platelet*, the official newsletter of the ITP Support Organisation.

Communication of Study Progress:

Though study participants will not be actively involved in the investigation following registration, they will be kept closely appraised of study progress through an electronically posted bi-monthly newsletter on our study website: **www.ukitpregistry.com**. They, too, will be provided with multiple means to contact the chief investigator's team regarding any questions or concerns that they may have throughout the duration of the study.

References:

1. McDowell et al. *Arthritis Rheum.1995;* 38, 221-8
2. John, et al. *Annals of the Rheumatic Diseases*. 1998;57, 361-5
3. Cantagrel, et al. *Arthritis Rheum. 1999;* 42, 1093-100
4. Hackstein et al. *Tissue Antigens. 1999;* 54, 471-7
5. Fishman, et al. *J Clin Invest. 1998 ;* 102, 1369-76
6. Olomolaiye et al. *Eur J Immunogenet. 1998;* 25, 267
7. Bornscheuer et al. *Dermatology. 1999.* 198, 118-121
8. Turner, et al. 1997; *Eur J Immunogenet* 24, 1-8
9. Lympany, et al.1998; *Tissue Antigens* 52, 573-8
10. Wilson et al. *Proc Natl Acad Sci USA. 1997;* 94, 3195-9
11. Siegmund, et al. *Thyroid. 1998;* 8, 1013-7
12. Jiang, et al., *Immunogenetics. 2000;* 51, 429-35
13. Singal et al. 2000; *Tissue Antigens* 55, 44-7
14. Heward, et al., *J Clin Endocrinol Metab. 1999;* 84, 2398-401
15. Liu, et al., *Proc Natl Acad Sci USA. 2001;* 98, 9960–9964
16. Provan et al. Blood. 2010;115:168-186
17. Yusuf et al. Lancet 2004; 364: 937–52
18. ASH Fact sheet. 2011. http://ash.org.uk/files/documents/ASH_111.pdf
19. Hearnshaw et al. Gut. 2011;60(10):1327-35.
20. Patra et al. BMC Public Health. 2010;10:258
21. Stampfer et al. JAMA. 1996 Sep 18;276(11):882-8
22. Orchard et al. Diabetes Care. 2006;29(11):2528-38.
23. Ridker et al. Lancet. 2012;380(9841):565-71.
24. Sundstrom et al. BJOG 2009;116:91–97.
25. Bachmann et al. Circulation. 2012;125(25):3092-8

Laio et al. Stroke. 1997;28(10):1908-12.

Chief Investigator Signature:

Dr Daniel Hart 22 February 2017