HEPATOLOGY RESEARCH GROUP
INSTITUTE OF TRANSLATIONAL AND STRATIFIED MEDICINE, PLYMOUTH UNIVERSITY PENINSULA SCHOOLS OF MEDICINE AND DENTISTRY

Hepatology Research Group (HRG) structure:

Head
Prof Matthew Cramp, Professor of Hepatology and Consultant Hepatologist

Senior Clinical Academics:
Dr David Sheridan, Associate Professor (Senior Lecturer) of Hepatology and Honorary Consultant Hepatologist,
Dr Ashwin Dhanda, Honorary Lecturer and Consultant Hepatologist

Senior Scientists:
Dr Daniel Felmlee, Lecturer,
Dr Doha Hegazy, HRG Manager

Clinical Research Fellows:
Dr Ollie Rupar – PhD student
Dr Kris Bennett - NIHR Academic Clinical Fellow,
Dr Asma Ahmed (writing up)

PhD Fellows:
Paula Boeira
Justyna Lopatecka

Hepatology Research Group Annual Report 2018

Overview:
2018 was a good year for the Hepatology Research Group (HRG) with continued growth and diversification of our research portfolio and the first full year of working in the excellent laboratories in the new Derriford Research Facility.

The HRG published a good number of high impact papers as well as presenting our findings at national and international meetings. The group focuses on 4 main areas of research interest:
1) hepatitis C virus infection and protection from infection
2) non-alcoholic fatty liver disease and metabolic liver diseases
3) alcohol related liver disease
4) liver cancer development and its prevention.

A short summary of the work in each of these areas is included in this report.

2018 was the first full year of Matthew Cramp's BASL Presidency and saw a busy year with a national survey of clinical services, the biggest ever BASL annual meeting in terms of attendance, and the setting up of a number of BASL special interest groups to facilitate liver research, grant applications and clinical developments at a national level in a wide range of liver conditions.

The group were successful in appointing Dr Queenie Tan as Hepatology Clinical Research Fellow. Dr Tan will join the HRG in March 2019 for a 3 year PhD project working with Dr Dhanda. Dr Kris Bennett continues to develop his PhD proposal in NAFLD as well as making good progress with his clinical training in gastroenterology and hepatology.

The HRG inflammatory liver disease biobank gained full ethics and HTA approval in 2017 and has continued to grow in 2018. This long term project allows us to prospectively collect and archive blood and tissue specimens from consenting patients with any underlying cause of liver disease and will underpin much future work with stored samples from the biobank expected to be used for a wide variety of scientific studies. The biobank is already helping our research programmes on non-alcoholic fatty liver disease and liver cancer.

The research group, together with the wider clinical team of the South West Liver Unit, continue to be actively involved in clinical trials and are major recruiters to many interventional studies (including clinical trials of new treatments for non-alcoholic fatty liver disease, primary biliary cholangitis, alcoholic hepatitis, and advanced decompensated liver disease) as well as additional observational studies.

A useful ongoing collaboration with Bath University saw another excellent undergraduate complete his year with the HRG in August and another student has joined us from September and is making good progress. We hope to continue with this successful link.

Hepatitis C Virus infection and Protection from Infection
Leads: Matthew Cramp, Dan Felmlee

The HRG studies on our two cohorts of HCV exposed but uninfected cases continue to uncover new findings and shed light on mechanisms of viral resistance. In the injection drug user cohort the work started by Dr Isaac Shawa continues. Isaac was awarded his PhD at the end of 2017 and his thesis included work describing significant differences in the lipidomics of HCV resistant cases from others in the IV drug using cohort he identified and recruited. Further analysis of this very large dataset in collaboration with Kings College Hospital (Dr Mark McPhail) is ongoing but points towards a potential ink with alterations in the lipid profile and innate immune responses. This is further supported by the work Dan and Kris are undertaking to follow on from Isaacs finding of major upregulation of in-vitro innate responses to viral RNA stimulation in EU cases, compared to infected cases.

In 2018 the work on the blood transfusion exposed but uninfected cohort made good progress. We have previously identified a number of rare genetic polymorphisms associated with high level resistance to HCV infection and have developed a cell culture model to functionally validate the genes associated. By using the CRISPR /Cas-9 system to knock out the identified genes, Dan with help from the Bath University students has developed a number of knock-out cell lines that are now being challenged with HCV pseudo-particles to determine if they are resistant to infection. This work will be greatly facilitated once the containment level lab in the DRF is open and we can start using replicating hepatitis C virus, but for now the work is continuing using the pseudoparticles. We are exploring the possibility that resistance to HCV infection may also confer protection from viruses using similar mechanisms of cell entry to HCV – these include Dengue and Zika virus both of which pose major public health problems.

The next 12 months will see Dan submit an MRC grant bid to take this work forward and the opening of the Containment Level 3 facility at the DRF needed for the next stages of the laboratory work.

On the clinical front, Matthew Cramp as clinical lead for the Peninsula HCV Operational Delivery Network oversaw more than 400 patients across Devon Cornwall and Somerset successfully treated and cured of their HCV infection in 2018 – a record number.

Non-alcoholic fatty liver disease and metabolic liver diseases
Lead: David Sheridan, Dan Felmlee

Research developments in the area of NAFLD and metabolic liver disease continued apace in 2018. Dr Sheridan as clinical lead for NAFLD has designed and implemented a new local referral pathway. Work on the themes of streamlining NAFLD management care pathways with primary care, links to weight management

services, and with a research focus on the utility of non-invasive diagnostics within clinical pathways plus motivation / behaviour changes for lifestyle interventions.

Dr Sheridan has a growing reputation in the study of metabolic liver diseases, and was an invited speaker at the international inherited disorders of metabolism affecting the liver meeting in Leuven, Belgium in March 2018. He was co-chair and organiser of the metabolic liver disease symposium at the BASL Basic Science meeting held in June 2018 in Derbyshire. He has provided peer review for grants and publications from Wellcome, MRC, NIHR and Cancer Research UK in 2018 .

On the clinical research front, Dr David Sheridan was Principal Investigator for the M118 study evaluating the diagnostic utility of the Fibroscan XL probe and controlled attenuation parameter for NAFLD. This has reported several abstracts presented as both oral and poster presentations at EASL and AASLD, and accepted for publication in Gastroenterology, the most prominent journal in the field of gastrointestinal disease (Impact Factor 20.773)

Increasing interest and collaboration with Prof Adrian Taylor, Tom Thomson and the PenCTU / RDS South West (Peninsula Clinical Trials Unit / Research Development Service) on the role of lifestyle interventions and behaviour change as first line treatment for NAFLD has culminated in a NIHR multicentre grant proposal, prioritised by the BASL BSG NAFLD special interest group.

Paul Boeira successfully transferred to the PhD programme for her project investigating mitochondrial dysfunction in NASH in collaboration with CN Bio Innovations. We are very grateful to Northcott Devon Medical Foundation (TB/MG/N05002) for a consumables grant of £9987 to support this work.

Successful recruitment into global phase 2 and 3 clinical trials for novel therapeutic targets for NASH continues, with Plymouth Hospitals being a major recruiting centre. We are using Multiparametric MRI scanning technology from Perspectum Diagnostics Ltd in clinical trials sponsored by Intercept and Novartis, and are exploring further utility of this non-invasive diagnostic technology in clinical diagnostic pathways as an alternative to liver biopsy.

The Inflammatory Liver Diseases Biobank (ILB) is growing and has focused on collection of blood samples and clinical data as well as liver tissue. The expectation is that this resource will continue to grow and in the future support a wide range of transitional liver disease research projects.

Alcohol related liver disease
Lead: Ashwin Dhanda

The key development in 2018 was Dr Ashwin Dhanda's appointment as a Consultant Hepatologist at University Hospitals Plymouth NHS Trust. This permanent position includes funded University time to enable Ashwin to build on the strong links with the

South West Liver Unit and to continue his growing research into alcohol-related liver disease.

Ashwin completed two small projects in 2018. The first, funded by Guts UK (formerly Core), investigated whether an established immune blood test could predict outcome of patients with alcoholic hepatitis. The preliminary results are positive and have led to further funding to evaluate this blood test in another 200 patients. The second project, funded by Plymouth Hospitals Charity and performed in collaboration with the Department of Chemistry in the University, measured a range of important trace elements in blood samples from over 300 patients with alcohol-related liver disease. A report is currently being prepared.

Ashwin is a work-stream lead in the MRC Stratified Medicine Consortium "Minimising mortality from alcoholic hepatitis" and work continues in getting this set up. The first clinical trial has now opened to recruitment and the cohort study is due to commence in Spring 2019. Ashwin's diagnostic evaluation study, part of the cohort study, will start in Summer 2019 after recruitment of a research technician.

Ashwin also secured European funding from ERAB: the European Foundation for Alcohol Research. In collaboration with Dr Garry Farnham, Prof Matthias Futschik and Prof Mat Upton from the Faculty of Medicine, he will investigate whether a circulating bacterial or fungal DNA 'signature' can predict development of subsequent infection or death in patients with alcohol-related liver disease.

2019 holds much promise for alcohol-related liver disease in Plymouth. A new PhD student, Justyna Lopatecka, started in October 2018 working together with Dr Gyorgyi Fejer investigating innate immune responses in macrophages. A clinical PhD student is due to start with Ashwin in the Spring to work on investigating the regulation of steroid signalling in patients with alcoholic hepatitis. Together with the new research technician due to start in the Summer, Ashwin will be able to continue his laboratory research into immune dysfunction in alcohol-related liver disease.

Liver cancer development and its prevention
Lead: Matthew Cramp, Jemimah Denson, Michael Jarvis

Dr Ollie Rupar continues his work in this area studying universal tumour associated antigen (U-TAA) expression in various types of liver tumour using a combination of histological and genetic methods. He has managed to successfully extract and then sequence DNA from a large number of archived tissue samples. He has sequenced U-TAA from both tumour and non-tumour tissue with known polymorphisms shown to be expressed in differing patterns according to tumour type. He is now working on tissue staining of U-TAA using immunohistochemistry and will then move on to seek potential biomarkers of liver tumour development in stored sera held in the biobank. We hope our findings will help better refine the identification of early malignancy and variant tumours that progress more rapidly.

HRG Management and teaching
Lead: Doha Hegazy

The move from the John Bull Building to the Derriford Research Facility in September 2017 was a big step forward for us and the DRF has proved to be an excellent environment for networking as well as providing fantastic laboratory facilities. We now regularly see and meet with scientists with different areas of expertise – this has enhanced meetings, seminars and the whole research work experience.

My role within the group is mainly management and teaching. At the management level in 2018, I completed multiple tasks with the general lab managers to provide a safer and healthier working environment. The Containment Level 3 laboratory is nearly complete and the HRG are looking forward to being able to use this facility once all the regulatory approvals are in place.
The general data protection regulations (GDPR) were changed in May 2018, and we ensured that all team members were trained to meet all required standards in both information governance and data security as well as those required by the Human Tissue Act regulations which remains especially important with the growth of the inflammatory liver diseases biobank.
Inspiring the next generation of researchers is key, and the HRG led by Doha, ran a number of successful Special Study Units in 2018 to introduce undergraduate medical students to the laboratory and to foster interest in translational research. In particular the healthcare environment SSU for years 1 and 2 proved very successful and the students submitted excellent essays about the efforts and progress towards the WHO goal of elimination of viral hepatitis as a public health threat by 2030.

I attended European Association for the Study of the Liver (EASL), Paris, 2018, the conference was a great opportunity to enrich and update my teaching materials.

Current and future plans

2018 was the first full year in the Derriford Research Facility (DRF) adjacent to the main Plymouth University Peninsula Schools of Medicine and Dentistry (PUPSMD) building and right beside Derriford Hospital. The DRF has brought medical, dental and biomedical laboratory-based researchers together and has significantly improved interdisciplinary working and clinical collaborations.

The DRF significantly expanded the laboratory facilities of the HRG at the medical school site but we have still not got the containment level 3 (CL3) laboratory fully equipped and approved. The approval of CL3 laboratories is an involved process, and this is taking longer than hoped as well as requiring further investment. We are now close to getting the final bits of equipment needed and hope to have the DRF CL3 lab

fully commissioned in 2019. This will at long last allow the HRG to undertake the viral challenge work here in Plymouth rather than at our collaborators laboratories in Glasgow. This work is a key part of the studies to better understand the mechanism of HCV resistance identified in the blood transfusion cohort of cases.

Collaborations, both within University of Plymouth and outside the university, remain a major part of the groups long term viability strategy. The relocation of many of our internal collaborators to the DRF was a major step in the right direction. Our collaborations with Imperial College London as part of the successful MRC stratified medicine bid in alcoholic hepatitis is going well as is our ongoing work with the MRC Centre for Virology in Glasgow. We have recently developed stronger links with the Institute of Hepatology at Kings College London and hope to further develop these in 2019.

Work with the SW Academic Health Science Network has helped us access new funding streams to develop alcohol services. In 2019 the resource allocated by commissioners will further strengthen the assertive alcohol outreach service in Plymouth, to try and reduce hospital admissions from alcohol related harm.

Our involvement in clinical trials continues. The HRG and SWLU are the largest recruiter to trials in liver disease in the southwest and have brought a range of new treatments to patients from across Devon Cornwall and Somerset. We are currently setting up clinical trials in NAFLD, primary biliary cholangitis, primary sclerosing cholangitis, hepatic encephalopathy, severe alcoholic hepatitis and in decompensated liver disease.

A key marker of the groups success will be increasing our success in obtaining grant support. Dr Dhanda has started work on our first Medical Research Council funded project. Ensuring the financial sustainability of the group at a time of increasing pressure on university and academic funding remains a key priority and in 2019 we hope to hear the outcome of a current application to NIHR and will be submitting a number of new applications to the MRC, NIHR and others.

The generosity and financial support provided by the Mary Kinross Trust for 2018 and for 2019 has been instrumental in keeping the HRG viable and to allow us to continue working towards greater financial stability from external grant support. We are extremely grateful to them for all their support over many years and their ongoing commitment to help us in 2019.

We remain indebted to all the patients and their families who have supported us by getting involved in clinical trials and consenting to the research work, to the clinical team at the SWLU and to all our collaborators both in the UK and abroad.

Research Outputs for 2018:

Publications:

Accuracy of FibroScan Controlled Attenuation Parameter and Liver Stiffness Measurement in Assessing Steatosis and Fibrosis in Patients With Nonalcoholic Fatty Liver Disease.
Eddowes PJ, Sasso M, Allison M, Tsochatzis E, Anstee QM, **Sheridan D**, Guha IN, Cobbold JF, Deeks JJ, Paradis V, Bedossa P, Newsome PN.
Gastroenterology. 2019 Jan 25: S0016-5085(19)30105-2

Gathering momentum for the way ahead: fifth report of the Lancet Standing Commission on Liver Disease in the UK.
Williams R, Alexander G, Aspinall R, Batterham R, Bhala N, Bosanquet N, Severi K, Burton A, Burton R, **Cramp ME**, Day N, Dhawan A, Dillon J, Drummond C, Dyson J, Ferguson J, Foster GR, Gilmore I, Greenberg J, Henn C, Hudson M, Jarvis H, Kelly D, Mann J, McDougall N, McKee M, Moriarty K, Morling J, Newsome P, O'Grady J, Rolfe L, Rice P, Rutter H, Sheron N, Thorburn D, Verne J, Vohra J, Wass J, Yeoman A.
Lancet. 2018 Dec 1;392:2398-2412.

Bacterial and fungal infections in acute-on-chronic liver failure: prevalence, characteristics and impact on prognosis.
Fernández J, **Acevedo J**, Wiest R, Gustot T, Amoros A, Deulofeu C, Reverter E, Martínez J, Saliba F, Jalan R, Welzel T, Pavesi M, Hernández-Tejero M, Ginès P, Arroyo V; European Foundation for the Study of Chronic Liver Failure.
Gut. 2018 Oct;67(10):1870-1880

Elbasvir/grazoprevir and sofosbuvir for hepatitis C virus genotype 3 infection with compensated cirrhosis: A randomized trial.
Foster GR, Agarwal K, **Cramp ME**, Moreea S, Barclay S, Collier J, Brown AS, Ryder SD, Ustianowski A, Forton DM, Fox R, Gordon F, Rosenberg WM, Mutimer DJ, Du J, Gilbert CL, Asante-Appiah E, Wahl J, Robertson MN, Barr E, Haber B.
Hepatology. 2018 Jun;67(6):2113-2126

Primary Prophylaxis of Variceal Bleeding Is Combined Prophylaxis More Effective?
Bennett K, Vine L, Lim Y, Bendall O and **Juan Acevedo**
Gastroenterology Medicine & Research. 2018 May;1(4)

Disease burden and costs from excess alcohol consumption, obesity, and viral hepatitis: fourth report of the Lancet Standing Commission on Liver Disease in the UK.
Williams R, Alexander G, Armstrong I, Baker A, Bhala N, Camps-Walsh G, **Cramp ME**, de Lusignan S, Day N, Dhawan A, Dillon J, Drummond C, Dyson J, Foster G, Gilmore I, Hudson M, Kelly D, Langford A, McDougall N, Meier P, Moriarty K, Newsome P, O'Grady J, Pryke R, Rolfe L, Rice P, Rutter H, Sheron N, Taylor A, Thompson J, Thorburn D, Verne J, Wass J, Yeoman A.

Lancet. 2018 Mar 17;391:1097-1107

Enhanced natural killer cell activity is found in exposed uninfected recipients of hepatitis C-contaminated blood.
Ow MM, Hegazy D, Warshow UM, Cramp ME.
J Viral Hepat. 2018 Mar;25(3):245-253.

Conference Presentations:

International Meetings:

European Association for the Study of the Liver meeting
Paris April 2018:

Upregulated innate immune responses in a hepatitis C virus exposed uninfected cohort.
Shawa I, Bennett K, Sheridan D, Felmlee D, Hegazy D, Ahmed A, Wood C, Jackson S, Fejer G, **Cramp ME**

Performance of controlled attenuation parameter (CAP) to assess steatosis in a large prospective multicentre UK study of patients with non-alcoholic fatty liver disease (NAFLD)
P. Eddowes, M. Allison , E. Tsochatzis , Q. Anstee , **D. Sheridan** , I.N. Guha , J. Cobbold , V. Paradis, P. Bedossa, P. Newsome

Treatment Efficacy and Safety of Seladelpar, a Selective Peroxisome Proliferator-Activated Receptor Delta agonist, in Primary Biliary Cholangitis Patients: 12-and 26-Week Analyses of an Ongoing, International, Randomized, Dose Ranging Phase 2 Study
G. Hirschfield, P. Boudes , C. Bowlus, N. Gitlin, G. Michael, S. Harrison, S.C. Gordon, R. Aspinall, Y. Doerffel, A.E. Kremer, **D. Sheridan**, B. Bacon, C. Berg, B. Borg, T. Hassanein, J. Odin, M. Shiffman, P.J. Thuluvath, D. Thorburn, D. Bernstein, P. Buggisch, L. Corless, C. Levy, M.J. Mayo, M.G. Swain, J. Vierling, M.-A. Wörns, A.(S.) Steinberg, S. Bergheanu, Y.-J. Choi , M. Varg, R. Martin, C. Mcwherter, D. Jones

Pharmacokinetics and pharmacodynamics of seladelpar, a potent and selective PPAR-delta, in patients with primary biliary cholangitis
P. Boudes , G. Michael, C. Bowlus, G. Hirschfield, D. Jones, J. Odin, **D. Sheridan**, N. Gitlin, S. Harrison, T. Hassanein, C. Levy, M. Shiffman, D. Thorburn, P.J. Thuluvath, J. Vierling, Y.-J. Choi, M. Varga, A.(Sasha) Steinberg, C. Mcwherter, R. Martin.

Algorithm to identify patients with an activity grade > 2 in type 2 diabetic patients with non-alcoholic fatty liver disease (NAFLD)- development in a large prospective multicenter UK study

P. Eddowes, M. Allison, E. Tsochatzis, Q. Anstee, **D. Sheridan**, I.N. Guha, J. Cobbold , Valérie Paradis, P. Bedossa, P. Newsome

American Association for the Study of Liver Disease meeting
San Francisco November 2018:

Fibroscan-Based Score (FS3) to Identify Nash Patients with NAS≥4 and F≥2: Development in a NAFLD UK Cohort - External Validation in a Malaysian NAFLD Cohort, a US Screening Cohort and a French Bariatric Surgery Cohort
abstract 0140 oral
M Sasso, WK Chan, SA. Harrison, S Czernichow, MED Allison, EA. Tsochatzis, QM. Anstee, **DA. Sheridan**, IN Guha, JFL Cobbold, M Raihan, V. Paradis, P Bedossa, Mahadeva, C Barsamian, AH. Paredes, CD. Fournier, V Miette, L Sandrin PN. Newsome

Expanding the Use of the VCTE XL Probe in Morbid Obese Patients: Validation of a New Automated Adaptive Measurement Depths Algorithm in a Large UK Multicenter Cohort; abstract 2299 poster
M Clet, P Eddowes, MED Allison, EA. Tsochatzis, QM. Anstee, **DA. Sheridan**, INeil Guha, JF L Cobbold, V Paradis, P Bedossa, CD. Fournier, L Sandrin, V Miette PN. Newsome

Cost Analysis of Triage Algorithms to Identify Patients with Nash+NAS≥4+F≥2Abstract 1682 poster
M Sasso, PN. Newsome, AH. Paredes, MED Allison, EA. Tsochatzis, QM. Anstee, **DA. Sheridan**, INeil Guha, JFL Cobbold, J Whitehead, V Paradis, P Bedossa, C Fournier, V Miette, SA. Harrison

Does the Inter-Quartile Range (IQR) of Controlled Attenuation Parameter (CAP) Serve As a Reliability Criterion for the Evaluation of Steatosis? - Evaluation in a Large Prospective Multicenter UK Study on Non-Alcoholic Fatty Liver Disease Patients. Abstract 1688 poster
P Eddowes, MED Allison, EA. Tsochatzis, QM. Anstee, **DA. Sheridan**, IN Guha, JFL Cobbold, V Paradis, P Bedossa, PN. Newsome

Efficacy and Safety of Seladelpar, a Selective Peroxisome Proliferator-Activated Receptor Delta Agonist, in Primary Biliary Cholangitis: 52-Week Analysis
Late Breaker 3.
CL. Bowlus, GW. Neff, R Aspinall, MR. Galambos, A Goel, G Hirschfield, A Kremer, MJ. Mayo, MG Swain, B Borg, Y Dörffel, SC Gordon, SA. Harrison, D Jones, P J. Thuluvath, C Levy, **DA Sheridan**, CM. Stanca, BR. Bacon, C Berg, TI Hassanein, J Odin, ML. Shiffman, D Thorburn, JM. Vierling, D Bernstein, P Buggisch, L Corless, CS Landis, AL. Peyton, HA. Shah, MA Woerns, N Gitlin, A Steinberg, S Bergheanu, G Amato, YJ Choi, S Rosenbusch, M Varga, C McWherter, P Boudes

Corticosteroid treatment is associated with restoration of altered CD4+ T cell phenotype in patients with severe alcoholic hepatitis
A Dhanda, E Yates, M Cramp. Presented as a poster

National meetings:

BASL Annual Meeting, York, Sept 2018

Trace element deficiency is associated with immune dysfunction in patients with alcohol-related liver disease
E Yates, R Clough, A Fisher, M Cramp, A Dhanda. Presented as a poster

Lipid Interactions and HCV Resistance
Dr David Sheridan Invited Oral presentation

Thinking Ahead in advanced cirrhosis
Anthony Moffat and David Sheridan (Poster)