

ABSTRACTS

XXIXth Belgian Week of Gastroenterology 2017

ABSTRACTS

- A01 — A43 Belgian Association for the Study of the Liver (BASL)
- B01 — B18 Belgian Network on Gastrointestinal Mechanisms (GIREM)”
- C01 — C10 Case Reports
- G01 — G18 Belgian Society for Gastrointestinal Endoscopy (BSGIE)
- H01 — H13 Belgian Society for Pediatric Gastroenterology, Hepatology and Nutrition (BeSPGHAN)
- I01 — I47 IBD Research Group (BIRD)
- O01 — O18 Belgian Group for Digestive Oncology (BGDO)
- P02 — P09 Belgian Pancreatic Club (BPC)
- R02 — R15 Radiology, Pathology and Nuclear Medicine

**BELGIAN ASSOCIATION FOR THE STUDY OF THE LIVER
(BASL)**

- A01 -

PLACENTAL GROWTH FACTOR INHIBITION PREVENTS HEPATOPULMONARY SYNDROME IN MICE. S. Raevens (1), A. Paridaens (1), S. Lefere (1), C. Casteleyn (2), B. Jonckx (3), X. Verhelst (1), H. Van Vlierberghe (1), C. Van Steenkiste (1), A. Geerts (1), L. Devisscher (1), I. Colle (1). [1] Ghent University, Ghent, Belgium, Gastroenterology-Hepatology, [2] Ghent University, Ghent, Belgium, Morphology, Veterinary Medicine, [3] ThromboGenics NV, Heverlee, Belgium.

Introduction : Hepatopulmonary syndrome (HPS) is a severe pulmonary complication of liver disease, without medical treatment to date. Pulmonary angiogenesis is known to contribute to HPS pathogenesis and has only been studied in rats till now. However, the majority of experimental interventions requires a mouse model to study disease pathogenesis and future therapeutic options.

Aim : Our aim was to establish a mouse model for HPS and to investigate the potential to counteract pathological angiogenesis in this model as therapeutic strategy for HPS.

Methods : Eight-week-old Swiss mice underwent common bile duct ligation (CBDL) or sham surgery and were sacrificed 6 weeks later. HPS was confirmed by hypoxemia on arterial blood gas analysis and intrapulmonary shunting by arterial detection of intravenously injected fluorescent labeled microspheres. Liver and lung tissue was collected in order to study liver fibrosis, pulmonary angiogenesis and inflammation. Scanning electron microscopy (SEM) was performed on vascular corrosion casts to visualize the pulmonary vasculature during cirrhosis *ex vivo*. Anti-PIGF antibodies (aPIGF Ab, 25 mg/kg 2x/week, ThromboGenics), administered from week 0-6 post-surgery, were tested for their potential to inhibit angiogenesis and ameliorate pulmonary function in CBDL mice.

Results : CBDL significantly induced liver fibrosis 6 weeks post-surgery compared to sham, as previously reported. CBDL mice suffered from hypoxemia (mean PaO₂ 63.2±9.2 mmHg vs. 92.2±6.7 mmHg in control mice, P=0.03) and intrapulmonary shunting as demonstrated by significantly more fluorescent-labeled spheres in their arterial blood compared to sham controls. CBDL resulted in enhanced pulmonary angiogenesis, evidenced by increased pulmonary CD105 and von Willebrand Factor (vWF) immunoreactivity (both P<0.0001) and elevated pulmonary PIGF protein expression (P=0.0072), along with induction of pulmonary monocyte chemoattractant protein 1 (MCP-1) (P=0.0017), compared to control surgery. In addition, SEM revealed regions of disorganized pulmonary angio-architecture in lungs of cirrhotic mice compared to shams. Preventive aPIGF Ab administration effectively neutralized pulmonary PIGF, which resulted in reduced pulmonary CD105 (P=0.007) and vWF immunopositivity (P=0.0049), and decreased pulmonary MCP-1 expression (P=0.06), compared to IgG control treatment in CBDL mice. Moreover, PIGF inhibition led to partial normalization of the pulmonary vascular network as demonstrated by SEM on lung vascular casts. Importantly, mice treated with aPIGF showed normal gas exchange, reflected by physiological PaO₂ levels (P=0.025), compared to IgG-treated CBDL mice which suffered from hypoxemia.

Conclusions : Pharmacological PIGF inhibition counteracts pulmonary angiogenesis and inflammation and is able to inhibit experimental HPS development in mice. Future research will have to reveal if aPIGF Ab might be an attractive therapeutic strategy for human HPS patients.

- A02 -

APOLIPOPROTEIN F AFFECTS HEPATIC PHOSPHATIDYLCHOLINE METABOLISM AND IS REDUCED IN NASH IN HUMANS. J. Haas (1), E. Baugé (1), A. Verrijken (2), S. Quemener (1), N. Hennuyer (1), F. Lalloyer (1), A. Deprince (1), C. Gheeraert (1), R. Paumelle-Lestrelin (1), S. Caron-Houde (1), L. Van Gaal (3), P. Lefebvre (1), D. Dombrowicz (1), S. Francque (4), B. Staels (1). [1] Institut Pasteur, Lille, France, INSERM U1011, [2] Antwerp University Hospital, Edegem, Belgium, Department of Endocrinology, Diabetology and Metabolism, [3] ANTWERP UNIVERSITY HOSPITAL, Edegem, Belgium, Dept. of Gastroenterology and Hepatology, [4] Antwerp University Hospital, Edegem, Belgium, Dept. of Gastroenterology and Hepatology.

Introduction : Nonalcoholic fatty liver disease (NAFLD) is rapidly becoming a major global health problem with its prevalence rising in concert with the epidemic of obesity. Critically, the molecular processes leading to the development of NAFLD and its more severe subtype, non-alcoholic steatohepatitis (NASH), remain poorly understood.

Aim : To identify molecular mechanisms implicated in development and resolution of NASH in humans.

Methods : A cohort of 170 patients covering the spectrum of NAFLD were biopsied at baseline and one year following dietary intervention or bariatric surgery totaling 253 biopsies. The biopsies were scored histologically according to the NAFLD Activity Score (NAS) criteria and each patient received a full clinical workup before and after the intervention. Expression microarray was used to assess gene expression changes in these biopsies and identify genes whose expression was correlated with histological features of NAFLD. In animal studies, acute overexpression of human or murine

ApoF was achieved using hydrodynamic injection via tail vein, and adenovirus-mediated shRNA expression was employed for transient knockdown of endogenous Apof. All experiments were performed using wildtype C57BL6/J mice fed a normal chow diet.

Results : In the human biopsies, we identified Apolipoprotein F (ApoF) as the strongest transcript inversely correlated with steatosis score in males (Spearman rho=-0.675, nominal p=7.6x10⁻⁸). Upon further investigation, we found ApoF also inversely correlated with lobular inflammation scores (Spearman rho=-0.612, nominal p=2.3x10⁻⁶), and hence, significantly reduced in males with NASH. These findings were confirmed using publicly available data from previously published NAFLD cohorts. Following bariatric surgery-mediated resolution of NASH, APOF transcript levels were increased nearly 2-fold (nominal p=3.5x10⁻⁴). To determine whether ApoF may play a causative role in NAFLD development, we assessed intracellular lipid metabolism using radiolabeled ¹⁴C-oleate. We found that transfecting Apof in Hepa1c1c7 cells led to reduced incorporation of the label into phosphatidylcholine (PC) while incorporation into triglycerides (TG) was unaffected. Due to the requirement of PC for efficient VLDL secretion, we suspected that changes in ApoF may affect plasma lipid levels. Indeed, overexpression of either human or murine ApoF in mice led to reduced plasma TG levels, associated with a reduction in hepatic VLDL production. Conversely, acute knockdown of ApoF led to increased plasma cholesterol levels 7 days post injection combined with increased liver weight.

Conclusions : We have identified ApoF as a transcript whose expression is strongly diminished with NAFLD severity and improves following NASH reversion in humans. Through a combination of animal and cell-based studies we demonstrate that reduced ApoF affects phosphatidylcholine homeostasis and leads to increased plasma cholesterol levels and likely elevated hepatic TGs. These findings highlight the importance of hepatic PC metabolism in the development of NASH and suggest ApoF is a novel player in this setting.

- A03 -

AUTOLOGOUS HEMATOPOIETIC STEM CELL TRANSPLANTATION INTO THE LIVER IN ALCOHOLIC HEPATITIS : WHAT IS THE IMPACT ON LIVER HISTOLOGY AND GENE EXPRESSION PATTERNS? N. Lanthier (1), N. Lin-Marq (2), L. Rubbia-Brandt (2), S. Clément (2), N. Goossens (1), L. Spahr (1). [1] University Hospitals of Geneva and Faculty of Medicine, Switzerland, Hepato-Gastroenterology, [2] University Hospitals of Geneva and Faculty of Medicine, Switzerland, Clinical pathology.

Introduction : Liver stem cell therapy (SCT) is suggested as potential means to improve liver regeneration in advanced liver disease. However, data of trials and reports are heterogeneous with no systematic histological evaluation. In a recent randomized controlled trial (Spahr et al., PlosOne 2013), we observed that patients who received SCT had a similar improvement of liver function over time as compared to controls.

Aim : The aims of this study were firstly to specifically analyze the effect of autologous SCT on liver cell proliferation and hepatic macrophages (implicated in liver regeneration) and second to perform an in depth transcriptome analysis in paired liver biopsies before and after transarterial administration of autologous hematopoietic stem cells in patients with alcoholic hepatitis.

Methods : Immunohistochemistry (Ki67, CK7 and CD68), in situ hybridization (SPINK1) and global gene expression analysis were performed on liver biopsies of 58 patients (30 controls and 28 stem cell treated) both at baseline and after 4 weeks of follow-up.

Results : Patients who received SCT did not exhibit any increased proliferative activity in hepatocytes nor in K7 positive liver progenitor cells on the liver biopsy performed at 4 weeks compared to controls. However, on repeat biopsy, patients who received SCT showed a more important CD68+ liver macrophagic expansion as compared to controls (p<0.05). Transcriptome data revealed significant upregulated genes linked with inflammation (CD68, SAA, CXCL6), regeneration (SPINK1, HGF), fibrosis (COL1A1) and stem cells (CD45) in SCT patients compared to controls. No major changes in gene pathways involved in liver growth, and in particular in cell cycle proteins were evidenced between the two groups. SPINK1 mRNA identified as a good baseline prognostic factor (Lanthier et al., J Hepatol 2015) was present by in situ hybridization at week 4 in SCT patients in liver parenchyma areas adjacent to macrophage recruitment and liver cell proliferation.

Conclusions : The analysis of liver tissue after SCT demonstrated an expansion of macrophages concurrent with an upregulated expression of genes involved in inflammatory and regenerative pathways. With the negative results of the clinical trial, it has to be interpreted as a weak impact of the SCT, which is not able to modify the clinical course of this severe liver disease.

RAPID, PERSISTENT HEPATITIS B VIRAL DNA SUPPRESSION PREDICTS NUCLEOS(T)IDE ANALOGUE INDUCED HBeAg SEROCONVERSION IN A BELGIAN, PREDOMINANTLY CAUCASIAN COHORT OF CHRONIC HEPATITIS B PATIENTS. S. Van Hees (1), S. Bourgeois (2), H. Van Vlierberghe (3), T. Sersté (4), P. Michielsen (1), H. Reynaert (5), J. Henrion (6), S. Negrin-Dastis (7), L. Lasser (8), F. Janssens (9), G. Robaey (10), P. Stärkel (11), C. Moreno (12), F. Nevens (13), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology and Hepatology, [2] ZNA Antwerpen, Belgium, Department of Gastroenterology and Hepatology, [3] Ghent University Hospital, Gent, Belgium, Department of Gastroenterology and Hepatology, [4] CHU Saint-Pierre, Brussels, Belgium, Department of Gastroenterology and Hepatology, [5] University Hospital Brussels, Vrije Universiteit Brussel, Belgium, Department of Gastroenterology and Hepatology, [6] Centre Hospitalier de Jolimont-Lobbes, La Louvière, Belgium, Department of Gastroenterology and Hepatology, [7] Grand Hopital de Charleroi, Charleroi, Belgium, Department of Gastroenterology and Hepatology, [8] CHU Brugmann Brussels, Brussels, Belgium, Department of Gastroenterology and Hepatology, [9] Jessa Hospital, Hasselt, Belgium, Department of Gastroenterology and Hepatology, [10] ZOL, Genk, Belgium, Department of Gastroenterology and Hepatology, [11] Cliniques Universitaires St Luc, Brussels, Belgium, Department of Gastroenterology and Hepatology, [12] CUB Hôpital Erasme, Bruxelles, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology, [13] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Start-of-treatment HBV DNA and ALT values have previously been found to be associated with HBeAg seroconversion during nucleos(t)ide analogue (NA) treatment in Asian Chronic Hepatitis B (CHB) patients.

Aim : We studied predictive factors for HBeAg seroconversion in a Belgian, predominantly Caucasian cohort of CHB patients.

Methods : This is a pooled analysis of mono-infected, non-immune-suppressed, start of treatment HBeAg positive CHB patients from 13 hospitals in Belgium treated with different NA for ≥ 3 months. HBeAg seroconversion was defined as loss of HBeAg and appearance of anti-HBeAg at two time points ≥ 1 month apart. Follow-up time was calculated as time from baseline (start of treatment) until HBeAg seroconversion or end of follow-up. A Cox regression model was used to determine predictive factors for HBeAg seroconversion. Upper Limit of Normal (ULN) for ALT was defined as 40 IU/mL.

Results : A total of 326 patients (74.8% male; 63% Caucasian, 17% African) were included. Patients were treated for a median of 3.4 years with lamivudine (n=141); adefovir (n=5); adefovir+lamivudine (n=3); tenofovir (n=87); entecavir (n=84); telbivudine (n=4) or entecavir+lamivudine (n=2). Treatment was switched to another NA in 90 patients. At baseline 17% of the patients were cirrhotic and mean HBV DNA was (7.52 ± 1.52) log IU/mL. Ninety-six patients HBeAg seroconverted after a median treatment duration of 15.5 months. Multivariate COX regression confirmed that baseline ALT levels correlated positively with the chance of HBeAg seroconversion (HR 1.194 per ULN increment; $p=0.001$), whereas baseline HBV DNA levels did not (HR 0.913; $p=0.537$). In addition, time after treatment start to persistent HBV DNA suppression <2000 IU/mL was highly predictive for HBeAg seroconversion (HR 0.972 per treatment month; $p=0.001$). Persistent suppression of viral HBV DNA to <2000 IU/mL within 1 year time after treatment start was associated with an up to two-fold increased chance of HBeAg seroconversion (Log-Rank : $p<0.001$).

Conclusions : Both baseline ALT levels and time to HBV DNA suppression correlate with HBeAg seroconversion rates in a predominantly Caucasian cohort of CHB patients. Persistent HBV DNA suppression < 2000 IU/mL within 1 year increases the HBeAg seroconversion rate up to two-fold compared to suppression later on.

HMGB1-DRIVEN FEEDFORWARD HEPATOCYTE NECROPTOSIS CIRCUIT IN LETHAL ACETAMINOPHEN-INDUCED LIVER INJURY. C. Minsart (1), C. Liefferinckx (1), S. Rorive (2), A. Lemmers (3), C. Dressen (4), E. Quertinmont (5), E. Trépo (6), C. Moreno (6), J. Devière (7), S. Goriely (8), I. Leclercq (9), R. Moreau (10), T. Gustot (7). [1] Université Libre de Bruxelles Faculté de Médecine, Brussels, Belgium, Laboratory of Experimental Gastroenterology, [2] Erasme Hospital, Anderlecht, Belgium, Department of Pathology, [3] Erasme Hospital, Anderlecht, Belgium, Department of Gastroenterology, HepatoPancreatology and Digestive Oncology, [4] Université Libre de Bruxelles Faculté de Médecine, Brussels, Belgium, Laboratory of Physiology and Pharmacology, [5] Université Libre de Bruxelles Faculté de Médecine, Brussels, Belgium, Laboratory of Experimental Gastroenterology, [6] Erasme Hospital, Brussels, Belgium, Department of Gastroenterology, HepatoPancreatology and Digestive Oncology, [7] Erasme Hospital, Brussels, Belgium, Department of Gastroenterology, HepatoPancreatology and Digestive Oncology, [8] Université Libre de Bruxelles Faculté de Médecine, Brussels, Belgium, Institute for Medical Immunology, [9] Université catholique de Louvain (UCL), Brussels, Belgium, Laboratory of Hepato-Gastroenterology, [10] Hôpital Beaujon, Service d'Hépatologie, INSERM U-481, Clichy, France, Centre de Recherche sur l'Inflammation (CRI).

Introduction : Voluntary or accidental acetaminophen (N-acetyl-p-aminophenol, APAP) overdose can induce a hyperacute form of liver failure potentially responsible for multiple organ failures and death. The mechanisms of this hepatotoxicity are incompletely understood. In “normal” conditions, high-mobility group box 1 (HMGB1) is a small nuclear protein who binds DNA and regulate many transcriptional events by modulating transcription factor-DNA interactions. In case of overdose, APAP induces hepatocytes necrosis and thus HMGB1 is released. Extracellular HMGB1 acts like damage-associated molecular pattern (DAMPs) and contributes to APAP-induced liver injury but the mechanisms associated with this activity are incompletely understood or controversial.

Aim : The aim of the present study was to investigate the early effects of HMGB1 in APAP-induced liver injury, its direct effect on hepatocytes and its role in the propagation of necrosis process.

Methods : APAP hepatotoxicity was induced in vivo by intraperitoneal injection in C57Bl/6 mice and in vitro on cultured HepaRG cells. HMGB1 was quantified by ELISA or immuno-staining. Cell death was determined by MTT, ALT, LDH and caspase-3 assays. Glycyrrhizin (GL) and ethyl pyruvate (EP) was used to inhibit HMGB1. Liposomal clodronate was administrated to mice to deplete Kupffer cells (KC). Expression of HMGB1 receptors was assessed by RT-PCR and flow cytometry. Expression of proteins who participate to necroptotic process was demonstrated by western blot. Dabrafenib and necrostatin-1 was used to inhibit receptor-interacting protein (RIP)3 and RIP1 respectively.

Results : We confirmed that, in APAP-challenged mice, inhibition of HMGB1 by glycyrrhizin improved survival and reduced further HMGB1 release. Depletion of Kupffer cells by liposomal clodronate in mice exacerbated APAP-induced hepatocyte necrosis and HMGB1 release suggesting that HMGB1 did not act through Kupffer cells activation. Based on these results, we hypothesized that a feed-forward circuit between HMGB1 and hepatocytes exist. In vitro, addition of APAP on cultured HepaRG cells induced cell necrosis characterized by lactate dehydrogenase release without caspase-3 activation, and HMGB1 release. Inhibition of HMGB1 by glycyrrhizin or ethyl pyruvate reduced APAP-induced HepaRG cell necrosis and further HMGB1 release. Exposure of HepaRG cells to recombinant human HMGB1 (rhHMGB1) resulted in cell death, supporting the hypothesis that HMGB1 acts directly on hepatocytes. Inhibition of RIPK3 by dabrafenib prevented APAP- and rhHMGB1-induced HepaRG cell death but inhibition of RIPK1 by Necrostatin-1 did not, suggesting the contribution of necroptosis. Moreover, inhibition of TRIF by Pepinh- TRIF reduced rhHMGB1-induced HepaRG cell death and Trif mutant mice were partially protected against APAP-induced liver injury.

Conclusions : In conclusion, these data support the hypothesis that HMGB1 contributes to the amplification of APAP-induced liver injury through feed-forward circuit with hepatocytes. This pathway seems to be independent of resident macrophages and, at least partially, dependent of TRIF/RIPK3 necrosis pathway resulting in the propagation of the liver injury.

- A06 -

CHARACTERIZATION OF A TERTIARY CENTER PBC COHORT AND VALIDATION OF PROGNOSTIC RISK SCORES. T. Claes (1), A. Geerts (1), H. Van Vlierberghe (1), X. Verhelst (1). [1] Ghent University Hospital, Gent, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Primary biliary cholangitis (PBC), formerly known as primary biliary cirrhosis is a relatively rare autoimmune liver disease. PBC can result in cirrhosis with decompensation and HCC development. Currently, ursodeoxycholic acid (UDCA) is the only registered and accepted drug for PBC treatment. Responsive patients in which treatment is started in early stages (I and II) of disease show similar survival rates to age and sex matched groups of the general population. Unfortunately, up to 40% of patients respond suboptimally to treatment. Internationally validated scoring systems predict probability of survival and are able to identify non-responders. Non-responsive patients could profit from other treatment options.

Aim : In this work, first we tried to characterize PBC patients in a single center tertiary Hepatology referral clinic at the Ghent University Hospital. Second, we validated two prognostic scores, the GLOBE-score and the Paris-II criteria, in this cohort.

Methods : In this retrospective study 67 PBC patients in the Ghent University Hospital were included between 1985 and 2015. Baseline characteristics, biochemical parameters, and outcome data were collected from the Patient Medical Record. The GLOBE-score and the Paris-II criteria were calculated. The Kaplan-Meier method was used to compute observed and expected event-free survival, transplant-free survival and probability of hepatocellular carcinoma (HCC) occurrence. Log Rank test was performed to compare event-free survival, transplant-free survival and probability of HCC occurrence between responders and non-responders according to the GLOBE-score and Paris-II criteria, AMA status, variant/non-variant presentation.

Results : There were 50 women and 17 men. Thirty patients (44.8%) were symptomatic at the time of diagnosis. Throughout follow-up 62 patients (92.5%) received UDCA treatment. The mean value of the GLOBE-score was -0.29 (SD=1.66). The GLOBE-score identified 26.1% of patients as non-responders. The Paris-II criteria identified 44.0% of patients as non-responders. A total of 30 patients (44.8%) either suffered from liver related death, underwent liver transplantation or had at least 1 occurrence of a clinical event during follow-up. Twenty-four patients (35.8%) developed biopsy-confirmed liver cirrhosis during follow-up. Liver decompensation occurred in 15 of cirrhotic patients (22.4%).

A total of 7 patients (10.3%) developed HCC. During follow-up 9 patients (13.4%) died and 14 patients (20.6%) underwent liver transplantation. The GLOBE-score was able to predict event-free survival ($P < 0.001$) and HCC occurrence ($P \leq 0.01$), but not transplant-free survival ($P = 0.066$). The Paris-II criteria were able to predict both event-free survival ($P < 0.01$) and transplant-free survival ($P < 0.05$).

Conclusions : We characterized a single center cohort of PBC in a tertiary Belgian liver unit. Furthermore, we were able to validate the strong predictive performance of the GLOBE-score and the Paris-II criteria in the prediction of event-free survival in this cohort.

- A07 -

LIPID, FETUIN-A AND MACROPHAGE ZONATION IN HIGH FAT DIET FOZ-FOZ MICE WITH NON-ALCOHOLIC STEATOHEPATITIS. N. Lanthier (1), Q. Etienne (1), V. Lebrun (1), L. Poekes (1), Y. Horsmans (1), I. Leclercq (1). [1] Université Catholique de Louvain, Belgium, Laboratory of Gastroenterology and Hepatology, Institut de Recherche Expérimentale et Clinique.

Introduction : Non-alcoholic fatty liver disease (NAFLD) is characterized by steatosis (accumulation of triglycerides in the liver) and insulin resistance. A subgroup of patients can develop a more serious condition called non-alcoholic steatohepatitis (NASH) with increased risk of fibrosis development. Innate immunity, cell injury, lipid metabolism and severity of insulin resistance constitute potential mechanisms underlying disease progression. Fetuin-A, an emerging player in insulin resistance in type 2 diabetic patients, is described as a liver-derived protein increased in human NAFLD.

Aim : Here, we explore the effect of a high fat diet on the expression of fetuin-A and its relation with the development of steatosis, cell injury and liver macrophage (Kupffer cell) activation in a mouse model of obesity and NASH.

Methods : Male foz/foz mice were fed a normal diet (ND) or a high fat diet (HFD) for 12 (long term HFD or LHFD) or 30 weeks (very long term HFD or VLHFD) ($n = 4/\text{group}$) to induce early or definite fibrosing NASH, respectively. Liver tissue homogenates were prepared for Western blot protein studies and total RNA was extracted for gene expression analysis. Liver paraffin-embedded sections were used for hematoxylin and eosin staining, Sirius red staining and double immunofluorescence detection of F4/80 and fetuin-A.

Results : Compared to foz/foz mice fed a ND, HFD-fed foz/foz mice developed obesity, insulin resistance and either steatosis (LHFD) or steatohepatitis with steatosis, hepatocyte ballooning, inflammation and fibrosis (VLHFD). In ND fed mice, fetuin-A staining was positive in the cytoplasm of zone 3 centrilobular hepatocytes while F4/80+ Kupffer cells were located in the sinusoids of the intermediate lobular zone 2. In LHFD fed mice, lipid deposition occurred in the hepatocytes of the zone 3 centrilobular areas. Fetuin-A protein was also located in the cytoplasm of these zone 3 centrilobular hepatocytes. F4/80+ macrophages distributed mainly in the sinusoids of the intermediate lobular zones 2, as seen in ND fed mice. However, liver m-RNA expression showed a 2-fold increased level of F4/80+ macrophage mRNA compared to ND ($p < 0.05$), suggesting activation. In VLHFD, we observed a loss of zonation of liver steatosis with the presence of fat loaded hepatocytes in all liver lobular zones. Fetuin-A was highest in periportal fat-laden hepatocytes and next to inflammatory infiltrates. There was a 4-fold F4/80 mRNA increased level upon VLHFD compared to ND ($p < 0.05$). Three types of F4/80+ cells were recognized on the morphology : elongated cells located in liver sinusoids compatible with liver resident Kupffer cells, cells forming lipogranuloma together with fat loaded hepatocytes and small inflammatory cells located in inflammatory foci compatible with recruited macrophages. Interestingly, F4/80+ cells from lipogranuloma were positive for fetuin-A protein staining. Liver fetuin-A mRNA levels remained unchanged either in LHFD or VLHFD compared to ND. Similarly, liver fetuin-A protein level was also stable under HFD.

Conclusions : We demonstrate that VLHFD foz-foz mice develop NASH together with zonal changes of steatosis, liver macrophage activation and fetuin-A expression in fatty hepatocytes and macrophages. A shift of steatosis and fetuin-A from the centrilobular region in ND and LHFD to the periportal zone was observed in VLHFD, together with macrophage activation, recruitment and fetuin-A co-localization in macrophages forming the lipogranuloma. The stable liver fetuin-A protein level could be compatible with a redistribution of this protein and/or the profile of a secretory factor. Taken together, we could imagine that lipid deposition and macrophage infiltration may be important factors in the liver tissue remodeling observed during NASH development. Further work is planned to delineate whether fetuin-A presence in macrophages is linked with a production and/or a simple storage in those cells in this model as well as the role of this protein in NASH progression and insulin resistance pathogenesis.

- A08 -

SERUM VASCULAR CELL ADHESION MOLECULE-1 PREDICTS SIGNIFICANT LIVER FIBROSIS IN OBESE PATIENTS WITH NON-ALCOHOLIC FATTY LIVER DISEASE. S. Lefere (1), F. Van De Velde (2), L. Devisscher (1), M. Bekaert (2), S. Raevens (1), X. Verhelst (1), Y. Van Nieuwenhove (3), M. Praet (4), A. Hoorens (4), C. Van Steenkiste (5), H. Van Vlierberghe (1), B. Lapauw (2), A. Geerts (1). [1] Ghent University, Ghent, Belgium, Gastroenterology and

Hepatology, [2] Ghent University, Ghent, Belgium, Endocrinology, [3] Ghent University, Ghent, Belgium, Gastrointestinal Surgery, [4] Ghent University, Ghent, Belgium, Pathology, [5] Maria Middelaers Ziekenhuis, Gent, Belgium, Gastroenterology and Hepatology.

Introduction : Non-alcoholic fatty liver disease (NAFLD) is the most common chronic liver disease worldwide and is strongly associated with obesity, dyslipidemia and insulin resistance. NAFLD often presents as simple steatosis (NAFL) but can progress to non-alcoholic steatohepatitis (NASH) and fibrosis. Current non-invasive biomarkers are not tailored to identify significant ($\geq F2$) fibrosis, although recent guidelines recommend a stringent follow-up of this patient population. We and others have reported on the role of pathological angiogenesis in the pathogenesis of NAFLD, highlighting pro-angiogenic factors as potential diagnostic markers.

Aim : We aimed to investigate the applicability of angiogenic factors as non-invasive diagnostic tools for NASH-associated fibrosis in obese patients.

Methods : Serum protein levels and visceral adipose tissue gene expression of endothelial dysfunction and angiogenic markers were analyzed by multiplex bead-based assay and quantitative RT-PCR, respectively, in sixty-one morbidly obese male patients undergoing bariatric surgery and in thirty-five control patients.

Results : We identified serum vascular cell adhesion molecule-1 (VCAM-1) as an independent predictor for $\geq F2$ fibrosis (median 14.0 vs. 8.7 ng/ml in patients with and without significant fibrosis; $P < 0.0001$) with an area under the receiver operating characteristics curve (AUROC) of 0.80. The cut-off point of 13.2 ng/ml showed a sensitivity of 80% and specificity of 83%. The AUROC increased to 0.86 when VCAM-1 and the presence of type 2 diabetes were combined. These AUROCs were higher than those for the simple fibrosis risk scores FIB-4 and BAAT. In line with these results, VCAM-1 visceral adipose tissue gene expression was also elevated in patients with fibrosis ($P = 0.030$).

Conclusions : Serum VCAM-1 levels were able to accurately predict significant ($\geq F2$) fibrosis in obese men with NAFLD.

- A09 -

UBIQUITIN CARBOXY-TERMINAL HYDROLASE L1 EXPRESSION IS INCREASED IN HEPATOCELLULAR CARCINOMA CELLS AND RENDERS THOSE CELLS MORE SENSITIVE TO ER STRESS-INDUCED CELL DEATH FOLLOWING INHIBITION. A. Vandierendonck (1), F. Fornari (2), O. Govaere (3), D. Laukens (1), X. Verhelst (1), A. Geerts (1), C. Ampe (4), M. Van Troys (4), T. Roskams (5), V. Hans (1), L. Devisscher (1), Y. Vandewynckel (1). [1] Ghent University, Ghent, Belgium, Gastroenterology and hepatology, [2] Azienda Ospedaliero-Universitaria Policlinico S. Orsola-Malpighi e Università di Bologna, Italy, Centro di Ricerca Biomedica Applicata (CRBA), [3] Newcastle University, Newcastle upon Tyne, United Kingdom (the), Fibrosis lab, Institute of Cellular Medicine, [4] Ghent University, Ghent, Belgium, Biochemistry, [5] KU Leuven, Belgium, Translational Cell and Tissue Research, Department of Imaging and Pathology.

Introduction : The development of hepatocellular carcinoma (HCC) and surrounding micro-environment cause cellular stress. This compromises endoplasmic reticulum (ER)-dependent protein folding and results in ER stress and unfolded protein response (UPR) activation. The UPR aims to restore protein homeostasis or induces cell death via CHOP. Protein degradation is enhanced by UPR-induced proteasome stimulation, a process that is fine-tuned by deubiquitinases (DUBs). DUBs are critical in the regulation of proteins involved in cellular processes and are proposed as potential oncotargets. However, their exact role in HCC development and progression is currently unknown.

Aim : Here, we investigated the expression of Ubiquitin carboxy-terminal hydrolase L1 (UCHL1), which is known to be involved in proteasome-dependent pathways, in human and experimental HCC and the effect of UCHL1 inhibition by LDN57444 on liver tumour cell survival during ER stress in vitro.

Methods : Micro-array data of human HCCs and corresponding non-neoplastic liver samples (GSE59259) were used for the expression analysis of the UCH-family of DUBs. Micro-array data and RNA samples of diethylnitrosamine (DEN)-induced HCC livers of rats were provided by dr. Fornari (University of Bologna, Italy). The effect of ER stress and UCHL1 inhibition by LDN57444 on the expression of UPR markers and UCHL1 and cellular viability was analysed in HepG2 cells by using RT-qPCR and MTT assays, respectively.

Results : Micro-array analysis revealed that UCHL1 is the only DUB of the UCH-family to be significantly increased in human HCC ($p < 0.05$). This upregulation was confirmed in a DEN-induced HCC rat model, both by micro-array analysis and RT-qPCR. In vitro, the ER stress inducer thapsigargin upregulated the expression of UCHL1 and reduced cell viability ($p < 0.05$). Interestingly, loss of viability was even more pronounced in addition of LDN57444 ($p < 0.0001$). The observed reduced viability might be UPR-mediated since combined treatment of thapsigargin and LDN57444 resulted in significantly enhanced mRNA upregulation of CHOP and its downstream effector GADD34 compared to each mono-treatment.

Conclusions : UCHL1 is upregulated in both human and experimental HCC. UCHL1 is induced upon ER-stress in HCC cells and renders those cells more sensitive to ER stress-induced cell death following UCHL1 inhibition. Further in vivo studies will have to reveal if UCHL1 inhibition might be an attractive therapeutic strategy for HCCs characterized by ER stress.

DO OR DON'T : HCV SCREENING IN THE BELGIAN BABY BOOM COHORT. R. Botterman (1), E. Glorieus (1), S. Lefere (1), X. Verhelst (1), P. Van De Voorde (2), S. Hachimi Idrissi (2), A. Geerts (1), E. Padalko (3), P. De Paepe (2), H. Van Vlierberghe (1). [1] Ghent University, Ghent, Belgium, Gastroenterology and Hepatology, [2] Ghent University, Ghent, Belgium, Emergency Department, [3] Ghent University, Ghent, Belgium, Clinical Biology.

Introduction : The US Preventive Services Task Force recommends one-time hepatitis C virus (HCV) screening of all baby boomers (born 1945-1965). Since about the half of the HCV patients in Belgium are not aware of their disease status, this study investigated if the baby boom cohort effect could be present in our country. It is investigated if age or other variables are predictive factors for HCV.

Aim : The aim is to investigate if HCV screening is opportune in the baby boom cohort in Belgium.

Methods : A cross-sectional study was performed from 05/09/2016 until 30/11/2016 at the emergency unit in the Ghent University Hospital. In 1106 patients admitted at the emergency department, after signing informant consent and in need for a blood sample, a HCV ELISA antibody test was performed. A questionnaire on general risk factors for HCV infection was presented to those patients able to fill in this questionnaire.

Results : Data of 1106 patients (606 men/500 women) were collected, 376 (34%) were born between 1945-1965, 494 (44.7%) after and 236 (21.3%) before. There was a HCV positive prevalence of 1.9% in the entire patient population. In the baby boom cohort, the prevalence of HCV was 1.6%. In the younger and older tested population, respectively 2.02% and 2.12% were positive. A significant correlation was found between positive HCV results and a history of IV drug use. There was no significant correlation between a positive HCV test and age, sex, blood transfusion before 1990 or tattoos and piercings.

Conclusions : There is a prevalence of 1.9% of HCV positive patients in a Belgian University hospital. This is higher than has been published previously. 44.7% of the tested patients were part of the baby boom cohort. However, there was no significant correlation between HCV positive results and age or age groups. Only screening in the baby boom group seems not opportune and in contrast to what is seen in the US study, a substantial number of HCV positive patients could be missed by the application of this recommendation. Other approaches need to be studied (eg. screening of the entire population, screening of populations at risk such as previous IV drug use).

ECV-ASSOCIATED MIRNA LEVELS AS NON-INVASIVE BIOMARKERS FOR EARLY-STAGE HBV/HCV-INDUCED LIVER FIBROSIS. J. Lambrecht (1), P. Poortmans (1), H. Reynaert (2), I. Mannaerts (1), L. Van Grunsven (1). [1] Vrije Universiteit Brussel, Jette, Belgium, Basic Biomedical Sciences, Liver Cell Biology Lab, [2] UZ Brussel, Jette, Belgium, Department of Gastro-enterology and Hepatology.

Introduction : Liver fibrosis is the pathological condition of the liver, resulting from sustained scar formation in response to chronic liver injury, such as chronic infection with hepatitis B (HBV) and hepatitis C (HCV) virus. The main effector cell in this pathology is the hepatic stellate cell (HSC) which will undergo a myofibroblastic transdifferentiation process towards an activated state, in which it will excessively produce and deposit extracellular matrix (ECM). Till date, the diagnosis of liver fibrosis occurs by several non-invasive techniques, which are however insensitive and unable to detect early disease stages, and liver biopsy, known to be invasive and associated with some minor and major complications. Novel non-invasive diagnostic scoring systems are being developed, but none of these have yet proven their sensitivity for the detection of early-stage liver fibrosis, nor their potential to discriminate between the various fibrotic stages.

Aim : Our aim was to analyze the potential of circulating miRNAs, with emphasis on extracellular vesicle (ECV)-associated miRNAs as novel biomarkers for early-stage liver fibrosis.

Methods : This study included patients with liver fibrosis by chronic HBV (n=19) or HCV (n=20) infection, which were identified as early-stage fibrotic (\leq F2) by transient elastography (Fibroscan). Relative expression levels of miRNAs in circulating ECVs and total plasma were analyzed by use of qRT-PCR. MiRNAs were selected based on their significant dysregulation during HSC activation (miRNA-192, -200b, -150), or their known presence in the circulation (miRNA-21, 92a, -122). To analyze the potential of these circulating ECVs to represent the presence or absence of activated HSCs in the liver, primary murine HSCs were cultured in vitro to induce spontaneous activation. ECVs were extracted from the culture medium of quiescent- (day 0 till day 2) and activated HSCs (day 8 till day 10).

Results : Analysis of miRNA levels in total plasma confirmed the up-regulation of miRNA-122 during early-stage fibrosis. With the exception of up-regulated levels of miRNA-192 in early-stage HBV-induced fibrosis, no other miRNAs showed significant changes in early-stage fibrosis by HBV or HCV infection. In contrast, miRNA-analysis of circulating ECVs identified significant changing levels of miRNA-150, -192, -200b and -92a during early-stage liver fibrosis by HBV and HCV infection. Especially the down-regulated levels of ECV-associated miRNA-192 (HBV AUC : 0.9802; HCV AUC : 0.9762) and miRNA-200b (HBV AUC : 0.9699; HCV AUC : 0.9841) seem to have an inherent diagnostic potential for early-stage fibrosis. Comparison of the miRNA levels from circulating ECVs with the ECVs extracted from

in vitro activating HSCs showed a similar trend in the down-regulation of miRNA-192, suggesting that ECV-associated miRNA-192 levels might represent the activation status of HSCs in the liver.

Conclusions : Circulating ECV-associated miRNAs could be used as novel tools for the diagnosis of early-stage liver fibrosis, through their potential to identify the absence or presence of activated HSCs in the liver.

- A12 -

PRETRANSPLANT GLYCOMIC ANALYSIS OF PERFUSATE IS PREDICTIVE OF PRIMARY NON FUNCTION AFTER LIVER TRANSPLANTATION. X. Verhelst (1), A. Geerts (2), D. Vanderschaeghe (3), X. Rogiers (4), A. Vanlander (4), F. Berrevoet (4), N. Callewaert (3), R. Troisi (4), H. Van Vlierberghe (2). [1] Ghent University Hospital, Ghent, Belgium, Department of Gastroenterology and Hepatology, [2] Ghent University Hospital, Ghent, Belgium, Gastroenterology and Hepatology, [3] VIB, Gent, Belgium, Medical Biotechnology Center, [4] Ghent University Hospital, Ghent, Belgium, Hepatobiliary and Transplant Surgery.

Introduction : Primary non function (PNF) is a rare but major complication after liver transplantation requiring urgent retransplantation. It is associated with the use of extended-criteria donors. The donor risk index is a clinical score that can guide the estimation of graft quality but lacks the power to predict PNF risk in individual patients. Perfusate analysis is an attractive tool for assessment of donor liver function before implantation. Glycomic assessment of serum has proven useful in the diagnosis of liver disease.

Aim : The aim of this study was to identify a specific glycomic signature in perfusate that is associated with PNF after liver transplantation.

Methods : In this prospective monocentric study 66 consecutive liver transplantations between October 2011 and July 2013 were included. Perfusate samples were collected after flushing of the hepatic veins before implantation of the liver graft. All donor grafts were transported using cold static storage. Based on an optimized DNA sequencer technology we performed glycomic analysis of these perfusate samples and searched for glycomic alterations in PNF patients.

Results : One single glycan, an agalacto core-alpha-1,6-fucosylated biantennary glycan (NGA2F) was significantly increased in the perfusate of the 3 patients that developed PNF after liver transplantation. It could identify PNF patients with 100% accuracy. This glycomarker was the only predictor of PNF in a multivariate analysis including donor risk index and perfusate AST/ALT levels ($p < 0.0001$).

Conclusions : In this cohort, patients who developed PNF after liver transplantation showed a specific glycomic signature in perfusate (before liver transplantation) that could distinguish them from non-PNF patients with 100% accuracy. This approach could guide the removal of donor grafts at risk for PNF from the donor pool, especially when the use of highrisk organs is considered.

- A13 -

A GLYCOMIC SERUM MARKER ANALYSED AT ONE WEEK AFTER LIVER TRANSPLANTATION IS AN INDEPENDENT PREDICTOR OF GRAFT LOSS DURING THE FIRST YEAR AFTER LIVER TRANSPLANTATION. X. Verhelst (1), A. Geerts (2), X. Rogiers (3), A. Vanlander (3), F. Berrevoet (3), N. Callewaert (4), R. Troisi (3), H. Van Vlierberghe (2). [1] Ghent University Hospital, Ghent, Belgium, Department of Gastroenterology and Hepatology, [2] Ghent University Hospital, Ghent, Belgium, Gastroenterology and Hepatology, [3] Ghent University Hospital, Ghent, Belgium, Hepatobiliary and Transplant Surgery, [4] VIB, Gent, Belgium, Medical Biotechnology Center.

Introduction : Graft failure after liver transplantation (LT) remains a challenge for transplant professionals and sometimes requires retransplantation. Pretransplant estimation of graft function using scores like donor risk index has limited use in individual patients. Diagnosis of early allograft dysfunction after liver transplantation by clinical criteria can predict graft survival. However, biomarkers that reliably identify patients at risk for graft failure after LT are lacking. Analysis of N-glycans in serum (glycomics) has shown to reflect the underlying liver function in liver disease but has never been assessed after liver transplantation.

Aim : The aim of this study was to assess the potential of serum glycomics as predictive markers for graft and patient survival after liver transplantation.

Methods : Serum glycomic profiles were collected before and after liver transplantation in 127 consecutive liver transplant recipients between 1 December 2012 and 31 December 2014 and analysed using an optimized glycomic technology on a DNA sequencer. The major outcome parameters (graft and patient survival) were related to the observed glycomic alterations.

Results : The assessment of 2 serum glycans NG1A2F (an agalacto, core-alpha-1,6-fucosylated biantennary glycan structure) and NA3 (a triantennary glycan), combined as $\log(\text{NA3}/\text{NG1A2F})$ on day 7 after liver transplantation was strongly associated with graft loss (hazard ratio = 7.222; $p < 0.001$; 95% CI 2.352-22.182) and patient death (hazard ratio

= 3.885; p=0.30; 95% CI 1.127-13.276) during the first year after liver transplantation (cox regression analysis). In a multivariable cox regression model including early allograft dysfunction (according to Olthoff) and Donor Risk Index, this glycomic marker, called GlycoTransplantTest, was the only independent predictor of graft survival (p=0.003).

Conclusions : Assessment of GlycoTransplantTest, a glycomic serum marker, on day 7 post liver transplantation is a strong and independent predictor of graft survival during the first year after liver transplantation.

- A14 -

RELAPSE RATES AND CLINICAL OUTCOMES AFTER NUCLEOS(T)IDE ANALOGUE THERAPY STOP IN A BELGIAN, PREDOMINANTLY CAUCASIAN COHORT OF CHRONIC HEPATITIS B PATIENTS. S. Van Hees (1), S. Bourgeois (2), H. Van Vlierberghe (3), T. Sersté (4), P. Michielsen (1), H. Reynaert (5), J. Henrion (6), S. Negrin-Dastis (7), L. Lasser (8), F. Janssens (9), G. Robaey (10), P. Stärkel (11), C. Moreno (12), F. Nevens (13), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology and Hepatology, [2] ZNA Antwerpen, Belgium, Department of Gastroenterology and Hepatology, [3] Ghent University Hospital, Ghent, Belgium, Department of Gastroenterology and Hepatology, [4] CHU Saint-Pierre, Brussels, Belgium, Department of Gastroenterology and Hepatology, [5] University Hospital Brussels, Vrije Universiteit Brussel, Belgium, Department of Gastroenterology and Hepatology, [6] Centre Hospitalier de Jolimont-Lobbes. La Louvière, Belgium, Department of Gastroenterology and Hepatology, [7] Grand Hôpital de Charleroi, Charleroi, Belgium, Department of Gastroenterology and Hepatology, [8] CHU Brugmann Brussels, Brussels, Belgium, Department of Gastroenterology and Hepatology, [9] Jessa Hospital, Hasselt, Belgium, Department of Gastroenterology and Hepatology, [10] ZOL, Genk, Belgium, Department of Gastroenterology and Hepatology, [11] Cliniques universitaires St-Luc, Brussels, Belgium, Department of Gastroenterology and Hepatology, [12] CUB Hôpital Erasme, Bruxelles, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology, [13] University Hospital Leuven, KU Leuven, , Belgium, Department of Gastroenterology and Hepatology.

Introduction : Cessation of Nucleo(s)ide analogues (NA) therapy after HBeAg seroconversion is associated with a high degree of relapse, but evidence in Caucasian patients is scarce.

Aim : We investigated relapse rates and clinical outcomes after NA stop in a Belgian cohort of HBeAg positive Chronic Hepatitis B (CHB) patients.

Methods : This is a pooled analysis of non-immune-suppressed HBeAg-positive, mono-infected CHB patients from 13 hospitals in Belgium, treated with different NA for ≥ 3 months. Data were collected between 1998 and 2016. HBeAg seroconversion was defined as the loss of HBeAg and the appearance of anti-HBeAg on two time points ≥ 1 month apart. Virological relapse was defined as HBV DNA > 2000 IU/mL; biochemical relapse as ALT $> 2 \times$ ULN (with ULN defined as 40 IU/mL). Clinical events were defined as the appearance of hepatic decompensation, HCC or liver-related death. Cox regression model was used to identify predictive factors for relapse. Follow-up time was calculated as time from HBeAg seroconversion until relapse or end of follow-up.

Results : A total of 326 patients (74.8% male; 63% Caucasian; 17% African) were included; 96 of whom showed HBeAg seroconversion. Treatment was stopped in 57/96 patients (of whom 8 were cirrhotic at baseline) after HBeAg seroconversion with a subsequent median consolidation therapy of 7.5 months. The median follow-up after treatment stop was 2.9 years during which 25 patients showed relapse (14 solely virological, 11 combined biochemical and virological), necessitating retreatment in 15 cases. HBeAg reversion was observed in 3/25 (12%) relapsed patients. Cox regression model showed that neither the presence of cirrhosis (HR 3.386; p=0.116) at start of treatment, nor Caucasian ethnicity (HR 0.509; p=0.133) were significantly associated with relapse after treatment stop. Relapse was accompanied by hepatic failure in two cases leading to liver-related death. Treatment was continued after HBeAg seroconversion in 26 patients (of whom 9 were cirrhotic at baseline) for a median of 4.1 years. Three patients (all cirrhotic) developed ascites in the latter group, but recovered thereafter. No patient died. **Conclusions** : Treatment cessation after HBeAg seroconversion led to relapse in 44% of predominantly Caucasian patients within a median follow-up 1056 days. Two relapsed patients showed severe clinical events leading to liver-related death.

- A15 -

LIVER FIBROSIS PROMOTES HEPATOCARCINOMA GROWTH THROUGH INFILTRATION BY TUMOR-ASSOCIATED INFLAMMATORY CELLS. B. Delire (1), P. Henriët (2), P. Lemoine (2), I. Leclercq (1), P. Stärkel (3). [1] Institut de Recherche Expérimentale et Clinique (IREC), Catholic University of Louvain (UCL), Belgium, Laboratory of Hepato-Gastroenterology, [2] De Duve Institute, Belgium, Cell Biology Unit, [3] Saint-Luc Academic Hospital and Institute of Clinical Research, Catholic University of Louvain, Belgium, Department of Gastroenterology.

Introduction : Liver fibrosis is the main risk factor for hepatocarcinoma (HCC). Mechanisms linking fibrosis and hepatocarcinogenesis remain however poorly understood. In many malignant diseases, inflammatory cells that infiltrate the tumor are key players in cancer development.

Aim : Our aim was to study in a mouse orthotopic transplantation model the impact of fibrosis on HCC development and local tumor infiltration and explore potential roles of macrophages and neutrophils.

Methods : The HCC cell line Hepa 1-6 is syngenic with the C57Bl/6 mouse strain. Hepa 1-6 cells were injected into non-fibrotic livers (normal liver group-NLG) and in severe fibrotic livers (severe fibrosis group-SFG) without immunosuppressive therapy. Severe fibrosis was induced by CCl₄ for 7 weeks. Mice were sacrificed 2 weeks post HCC cell injection. The liver was sliced and examined for the presence of tumor (nodule \geq 1mm). The tumor volume and the liver to body weight ratio (LW/BW) were used as parameters of tumor burden. A part of each tumor was used for histological analysis, proteins and RNA preparation.

Results : A tumor nodule was observed in 60% of animals in the NLG but in 100% of them in SFG. The tumor volume and the LW/BW were significantly higher in the SFG ($p < 0.0001$; $p = 0.005$) compared to the NLG. Tumor macrophages infiltration was evaluated by F4/80 immunohistochemistry : while F4/80 positive cells were mainly located around the tumor in NLG livers, macrophages infiltrated deeper the HCC nodules in SFG livers. F4/80 mRNA expression ($p < 0.0003$) as well as CD11b expression ($p < 0.0003$), a marker of recruited macrophages, were higher in SFG than in NLG tumors. Similarly, we observed a higher NIMP-R14+ neutrophils infiltration in tumors that developed in SFG compared to those in NLG ($p = 0.0289$). Many tumor-associated macrophages and neutrophils-derived molecules such as matrix metalloproteinase (MMP-2) and MMP-9 are involved in tumor progression and invasiveness. Compared to NLG, tumors in SFG livers expressed higher levels of Mmp2 ($p = 0.0019$) and Mmp9 ($p = 0.0047$). Mmp2 mRNA was significantly higher in the tumor compared to adjacent liver parenchyma in both groups (NLG : $p = 0.0006$; SFG : $p = 0.0002$) while high Mmp9 expression in tumor compared to adjacent parenchyma was only seen in SFG livers ($p = 0.0006$) but not in NLG livers. Furthermore, tumor volume positively and significantly correlated with intra-tumor Mmp2 ($r = 0.571$, $p = 0.026$) as well as with intra-tumor Mmp -9 ($r = 0.741$, $p = 0.002$) mRNAs. Zymography evaluates pro- and active MMP-2/ -9 : pro- and active MMP-2 and -9 were significantly higher in SFG tumors compared to NLG tumors (pro-MMP-2 : $p = 0.0007$, active MMP-2 : $p = 0.008$; pro-MMP-9 : $p = 0.008$, active MMP-9 : $p < 0.05$). Similarly to gene expression, MMP-2 and -9 enzymes were significantly more active in tumor than in adjacent parenchyma (MMP-2 : $p < 0.05$; MMP-9 : $p < 0.05$). MMP-2 and -9 are known activators of transforming growth factor β (TGF β), an inflammatory cytokine that promotes tumor cells growth. TGF β mRNA expression was higher in SFG than in NLG tumors ($p = 0.0012$). Moreover, there were higher amounts of active (cleaved) TGF β protein, measured by ELISA, in the SFG tumors compared to the NLG tumors.

Conclusions : Liver fibrosis promotes HCC development in a mouse orthotopic transplantation model. Our results suggest that a fibrotic liver background favors a higher infiltration of tumor associated macrophages and neutrophils in the developing tumor. These secrete and activate molecules such as MMP-2, MMP-9 and TGF β that promote tumor progression.

- A16 -

PAIRED BIOPSY ANALYSIS OF HUMAN LIVER TRANSCRIPTOME BEFORE AND 1 YEAR AFTER BARIATRIC SURGERY IDENTIFIES A RESTRICTED SET OF INFLAMMATION- AND EXTRACELLULAR MATRIX-RELATED GENES AS PIVOTAL IN NASH AND FIBROSIS PATHOGENESIS. S. Francque (1), P. Lefebvre (2), F. Lalloyer (2), M. Pawlak (2), E. Baugé (2), C. Gheeraert (2), H. Dehondt (2), J. Vanhoutte (2), N. Hennuyer (2), C. Claire Mazuy (2), B. Derudas (2), A. Driessen (3), G. Hubens (4), L. Vonghia (1), W. Kwanten (1), T. Vanwolleghem (1), P. Michielsens (1), J. Eeckhoutte (2), A. Verrijcken (5), L. Van Gaal (5), B. Staels (2) / [1] Antwerp University Hospital, Edegem, Belgium, Gastroenterology Hepatology, [2] Univ. Lille, CHU-Lille, Institut Pasteur de Lille, Lille, France, Inserm, [3] Antwerp University Hospital, Edegem, Belgium, Pathology, [4] Antwerp University Hospital, Edegem, Belgium, Abdominal Surgery, [5] Antwerp University HOSPITAL, Edegem, Belgium, Endocrinology, Diabetology and Metabolic Diseases.

Introduction : Pathogenic mechanisms leading to progression from simple steatosis towards active non-alcoholic steatohepatitis (NASH) and fibrosis are poorly defined.

Aim : We investigated the liver transcriptome in a human cohort of histologically staged NASH patients both at baseline and follow-up to identify key components of progression of disease and hence potential targets for therapy.

Methods : Obese patients were prospectively screened for presence of NASH and if suspected, liver biopsy was proposed. Patients entered a weight management program, including bariatric surgery (BarSur) in some, and were re-evaluated after 1 year including biopsy. Liver biopsy was scored using the NASH CRN scoring system. Gene profiling (Affymetrix GeneChip arrays + functional annotation and enrichment) was performed. Paired analysis of the liver transcriptome before and 1 year after BarSur identified genes dysregulated in NASH and fibrosis and whose expression was normalized upon regression of lesions. A meta-analysis with publicly available datasets with comparable histology was carried out to even more stringently identify genes dysregulated in NASH and fibrosis. Data were further crossed with transcriptomic data from NASH and fibrosis mouse models.

Results : Analysis was performed in 87 patients with paired biopsies. Progressive baseline histological damage from steatosis to NASH to NASH+fibrosis were characterized by gene expression patterns successively reflecting altered functions in metabolism, inflammation and epithelial-mesenchymal transition. The molecular signature for active NASH+fibrosis contained 193 upregulated genes (immune responses and ECM homeostasis) and 58 downregulated (metabolic pathways). Of these, 103 and 36 were normalized after BarSur, leading to a 139-gene signature of NASH+fibrosis normalized upon resolution. Comparison with existing databases led to a 24 BarSur-sensitive human NASH+fibrosis signature strongly enriched with ECM matrix formation and inflammatory responses. Comparison with NASH and fibrosis gene signatures of MCD and CCl4 mouse models respectively resulted in a 16-gene set of NASH+fibrosis with normalisation upon regression. This analysis pointed towards dermatopontin (DPT) as an important player.

Conclusions : Liver damage during NASH progression is characterized by deregulated expression of a restricted set of inflammation- and ECM-related genes. Targeting DPT may be a valuable strategy to reverse the hepatic fibrotic process.

- A17 -

LIVER PROGENITOR CELLS SIGNIFICANTLY CONTRIBUTE TO HEPATOCYTE POOL IN CHRONIC LIVER INJURY AND CIRRHOSIS : A KINETIC STUDY IN MICE. R. Manco (1), L. Clerbaux (1), I. Leclercq (1). [1] Université Catholique de Louvain, Brussels, Belgium, Laboratory of Hepato-Gastroenterology.

Introduction : Self-renewal of mature hepatocytes supports homeostasis and regeneration of adult liver. Recent studies indicate that liver progenitor cells (LPC) are recruited upon injury as a facultative reservoir for generation of hepatocytes, although only a small number of mature hepatocytes were shown to derive from LPC in vivo. Models used for these studies do not recapitulate long lasting chronic hepatocellular damage and fibrosis seen in human chronic liver disease and cirrhosis.

Aim : Our aim is therefore to follow the dynamics of ductular reaction (DR) and the LPC's fate during chronic liver injury in mice.

Methods : We used tamoxifen-inducible Osteopontin-Cre (OPN-CreERT2) mice crossed with yellow fluorescent protein (YFP) reporter mice to follow the fate of LPC and biliary cells with an efficiency >85%. Long-term chronic injury was induced by repeated carbon tetrachloride (CCl4) injections 3x/week for 4, 6, 8, 16 and 24 weeks, resulting in chronic fibrosis and eventually cirrhosis. Livers from 8 and 16 weeks were also analyzed after 4 weeks and 2 and 4 weeks of CCl4-free recovery period, respectively.

Results : After 4 weeks CCl4, DR is minimal with few ck19+/YFP+ positive cells in periportal area and LPC-derived hepatocytes (traced as YFP+ hepatocytes) are inconspicuous. After 6 weeks, DR is similar in intensity but small foci of YFP+ hepatocytes adjacent to portal area are readily seen; these have a median size of $3010\mu\text{m}^2$. As fibrotic disease increases in severity, the DR is negligible while patches of YFP+ hepatocytes become larger (median size of $3850\mu\text{m}^2$ and $7040\mu\text{m}^2$ at 8 and 16weeks, respectively) and extend to into the parenchyma. In the cirrhotic liver (24 weeks CCl4) some regenerative nodules are entirely composed of YFP+ hepatocytes. The number of YFP+ hepatocytes does not rise accordingly to the size of the patches as they represent $4.2 \pm 2.4\%$ of the lobule area in 6 weeks' samples, increases up to $11.5 \pm 3.8\%$ in 8 weeks' samples and stabilizes around 5% thereafter, suggesting that not all YFP+ hepatocytes expand into growing patches. At 6 weeks, YFP+ hepatocytes are significantly smaller cells than YFP- native hepatocytes (750 vs $981\mu\text{m}^2$) but in 16 weeks' samples YFP+ and YFP- hepatocytes have the same size (996 and $1001\mu\text{m}^2$). The dynamic of the YFP+ hepatocytes was also evaluated upon recovery : in the 4 weeks following 8 weeks of CCl4, the area occupied by YFP+ hepatocytes has a tendency to decrease from $11.5 \pm 3.8\%$ to $5.03 \pm 3.8\%$ ($p=\text{NS}$), while in the 2 and 4 weeks of recovery after 16 weeks of CCl4 the area significantly increases from $4.58 \pm 1.7\%$ to $7.7 \pm 3.3\%$ ($p=\text{NS}$), up to $13.8 \pm 0.7\%$ ($p<0.001$), respectively. Whereas, upon recovery the size of the YFP+ hepatocytes, in all the different time points, is the same of the native hepatocytes.

Conclusions : Our data demonstrate a significant contribution of LPC to the hepatocytes regeneration in a model of chronic liver injury leading to cirrhosis. The kinetic study supports that when DR is present, LPC differentiate into small hepatocytes, some of these subsequently increase in number, to form growing patches, and in size, becoming undistinguishable from the native hepatocytes. Upon recovery the growth of the patches of the LPC-derived hepatocytes depends on the severity of the underlying injury. Clonality studies are ongoing to test this hypothesis.

- A18 -

EARLY TIPS PLACEMENT AS A FEASIBLE AND SAFE STRATEGY FOR VARICEAL BLEEDING IN HIGH RISK LIVER PATIENTS : A 5-YEAR MONOCENTRE EXPERIENCE. E. Verly (1), X. Verhelst (1), H. Van Vlierbergh (1), A. Geerts (1). [1] Ghent University Hospital, Ghent, Belgium, Department of Hepatology & Gastroenterology.

Introduction : Variceal bleeding is a severe complication of cirrhosis. The treatment of variceal bleeding is based on proper supportive care, vasoactive medication and endoscopic therapy. Since 2010, early TIPS placement has shown improved survival in patients variceal bleeding with a high risk for rebleeding as defined by Garcia Pagan et al. (NEJM 2010).

Aim : The aim of this study was to retrospectively review the use of TIPS in variceal bleeding in our centre.

Methods : This retrospective monocentric study was performed in a tertiary referral centre for liver disease and liver transplantation (Ghent University Hospital). All patients admitted with variceal bleeding between January 2010 and December 2014 were included. Clinical data and results were retrieved from the medical files. Outcome was assessed at hospitalisation, 3 and 12 months after variceal bleeding. Statistical analysis was performed using SPSS (version 23).

Results : In this cohort 56 patients were identified with variceal bleeding, 16 female and 40 male patients between the ages of 22 and 84. Forty-nine (87,5%) patients survived the hospitalisation, 48 (85,7%) were alive after 3 months and 1-year survival was 73,2% (41 patients). 17 patients had a CHILD-PUGH classification of A, 24 CHILD-PUGH B and 12 were CHILD-PUGH C. Of 3 patients, the CHILD-PUGH score could not be calculated due to missing variables. All patients received supportive care, vasoactive medication and endoscopy within 12 hours of admission. In this cohort, 20 patients were treated with TIPS placement. 6 of these patients were classified as CHILD-PUGH A, 9 as CHILD-PUGH B and 5 as CHILD-PUGH C. Eleven patients (19,6%) received TIPS in the early-TIPS strategy after initial bleeding, 7 (12,5%) due to a rebleeding episode. In two patients (3,6%) TIPS placement was postponed after the 72 hours' time window but was given as an early-TIPS placement, and not due to rebleeding. In the early-TIPS group, 3 month and one year survival was respectively 92,3% and 84,6%. Transient encephalopathy after TIPS placement was observed in 7 patients (35,0%). In the early TIPS group, 4 patients (30,8%) had transient encephalopathy.

Conclusions : The implementation of the early TIPS protocol for variceal bleeding is safe and shows excellent one-year survival rates in this high-risk population. Serious Adverse events were rare and manageable in the majority of patients.

- A20 -

ASSOCIATING LIVER PARTITION AND PORTAL VEIN LIGATION FOR STAGED HEPATECTOMY (ALPPS) : ESTABLISHMENT OF A UNIQUE, INNOVATING ANIMAL MODEL WITH INSUFFICIENT LIVER REMNANT. A. Dili (1), V. Lebrun (2), C. Bertrand (3), I. Leclercq (2). [1] CHU UCL Namur and Laboratory of Gastroenterology, Istitut de Recherche Expérimentale et Clinique, UCL, Brussels, Belgium, Surgery and Laboratory of Hepato-Gastroenterology, Institut de Recherche Expérimentale et Clinique, Université catholique de Louvain, Brussels, Belgium, [2] Laboratory of Hepato-Gastroenterology, Institut de Recherche Expérimentale et Clinique, Université catholique de Louvain, Brussels, Belgium, Brussels, Belgium, Laboratory of Hepato-Gastroenterology, Institut de Recherche Expérimentale et Clinique, Université catholique de Louvain, Brussels, Belgium, [3] CHU UCL Namur, Yvoir, Belgium, Surgery.

Introduction : ALPPS is a surgical technic that combines portal vein ligation (PVL) and parenchymal transection followed by resection of the deportalized liver within 2 weeks. ALPPS achieves rapid hypertrophy of the future liver remnant (FLR) protecting patients from liver failure after extended otherwise non-viable hepatectomy (small for size syndrome-SFSS). SFSS is related to portal hyperperfusion of a very small hepatic parenchyma, with a compensatory constriction of the common hepatic artery (hepatic arterial buffer response-HABR) believed lied to desarterialisation of FLR and postoperative liver failure. In ALPPS, PVL and parenchymal transection redirect the whole portal flow through a small FLR. Despite a growing use of the ALPPS procedure in clinics, consequences on arterial flow and underlying mechanisms for accelerated regeneration and protection from SFSS are still unknown.

Aim : There are reports on animal models for ALPPS, but none accurately mimics the human procedure : rodent models either do not achieve liver resection leaving a small, insufficient for survival, FLR or propose hepatic resection during the first step of ALPPS. Differences in volume of FLR and in surgical events may introduce bias in our understanding of pathophysiological mechanisms. This study aims to develop a model mimicking ALPPS with minimal FLR and to analyze hepatic hemodynamics.

Methods : In rodents, the left median lobe (LML), represents 10% of the liver volume. Px90 represents a total hepatectomy except LML, transection (T) a hepatotomy between the right and left segment of median lobe and PVL a ligation of all portal branches except those that perfuse LML. PVL followed by Px90 is a strict copy of conventional human ALPPS. The first experiment (group A) studied the volume hypertrophy of LML after a unique procedure (T, PVL, PVL and sham); rats were harvested at 6hours,1,2,3,7days. The second experiment (group B) analyzed mortality and volume hypertrophy after Px90 and two step procedures, PVL-Px90 and PVL-T-Px90. Flow rate in portal trunc and common hepatic artery (HA) were measured by US-Doppler in Sham, PVL, PVL-T and Px90.

Results : In group A, hypertrophy of FLR was greater at day 2 and 3 after PVL-T compared to PVL ($p<0,05$) but not at day 7, suggesting that PVL-T accelerated initial hypertrophy. Hepatocyte proliferation, assessed by Ki67 and BrdU IHC, was significantly higher at day 2 and 3 in PVL-T remnants ($p<0,05$). We observed no hypertrophy after T. In group B, ALPPS was associated with a low seven day mortality rate (29,41%) compared to Px90 (77,7%) or PVL-Px90 (38,46%) ($p<0,05$). Acceleration in regeneration was confirmed by a significantly higher kinetic growth ratio in 1st and 2nd stage

ALPPS (PVL, PVL-Px90) compared to PVL and PVL-Px90 ($p < 0.005$). Total portal vein flow was similarly reduced after PVL, PVL and Px90 compared to sham ($p < 0.001$). However, because 90% of the liver parenchyma was excluded from the portal circulation in PVL, PVL and Px90, the portal flow in the FLR was increased by a factor 4 to 5 compared to flow reaching LML in sham animals ($p < 0.0001$). A decrease in HA flow occurred after PVL and PVL compared to sham ($p < 0.001$) and was further lowered after Px90 ($p < 0.5$ vs PVL; $p < 0.01$ vs PVL) suggesting a HABR concomitant to portal hyperperfusion in all 3 procedures. While arterial blood is distributed in the entire liver in PVL and PVL, it only enters the 10% FLR in Px90, in consequence, effective arterial flow into FLR is increased after Px90, but is halved after PVL ($p < 0.05$) and decreased in a lesser extent after PVL ($p = ns$). Immunohistochemistry using pimonidazole (an ischemia marker) demonstrated a significantly higher ischemia at day 1 in PVL compared to sham, PVL and Px90 ($p < 0.05$).

Conclusions : We describe the first animal model with minimal FLR, leading to high mortality due to SFSS unless ALPPS is applied. The degree of liver growth and kinetic growth ratio confirm that ALPPS boosts liver hypertrophy more than PVL. Hemodynamic study suggests that even if HABR exists in Px90, the SFSS consecutive to this kind of marginal hepatectomy is not related to parenchymal desarterialisation; on the contrary, reduction of arterial parenchymal perfusion as observed in PVL (the first step in ALPPS procedure) may protect the FLR from hepatocellular failure and stimulate regeneration. This model reproduces the objectives intended in human conventional ALPPS and should be valuable for study of physiological mechanisms.

- A21 -

THE RISK OF EARLY OCCURRENCE AND RECURRENCE OF HEPATOCELLULAR CARCINOMA IN HEPATITIS C INFECTED PATIENTS TREATED WITH DIRECT ACTING ANTIVIRALS WITH AND WITHOUT PEGYLATED INTERFERON : A BELGIAN EXPERIENCE. R. Bielen (1), C. Moreno (2), H. Van Vlierberghe (3), S. Bourgeois (4), J. Mulkay (5), S. Francque (6), W. Verlinden (6), C. Brixko (7), J. Decaestecker (8), C. De Galocsy (9), F. Janssens (10), M. Cool (11), L. Van Overbeke (12), C. Van Steenkiste (13), F. D'heygere (14), K. Wuycckens (1), F. Nevens (15), G. Robaey (16) / [1] University Hasselt, Hasselt, Belgium, Faculty of medicine and life sciences, [2] Erasme Hospital, Brussels, Belgium, Gastro-Enterology and Hepatopancreatology, [3] UZ Gent, Gent, Belgium, Gastro-Enterology and Hepatology, [4] ZNA Stuivenberg Hospital, , Belgium, Gastro-Enterology and Hepatology, [5] Hôpital Saint-Pierre, Belgium, Gastro-Enterology and Hepatology, [6] Antwerp University Hospital, Edegem, Belgium, Gastro-Enterology and Hepatology, [7] CHR La Citadelle, Belgium, Gastro-Enterology and Digestive Oncology, [8] AZ Delta, Roeselare, Belgium, Gastro-Enterology and Hepatology, [9] Hôp. Iris Sud Bracops, Bruxelles, Belgium, Gastro-Enterology and Hepatology, [10] Jessa Hospital, Hasselt, Belgium, Gastro-Enterology and Hepatology, [11] AZ Damiaan, Oostende, Belgium, Gastro-Enterology and Hepatology, [12] AZ Sint-Maarten, Mechelen, Belgium, Gastro-Enterology and Hepatology, [13] AZ Maria Middelaes, Ghent, Belgium, Gastro-Enterology and Hepatology, [14] AZ Groeninge, Kortrijk, Belgium, Gastro-Enterology and Hepatology, [15] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Gastro-Enterology and Hepatology, [16] Ziekenhuis Oost-Limburg Genk, Genk, Belgium, Gastro-Enterology and Hepatology.

Introduction : Direct antiviral agents (DAA) have made HCV treatment very effective and safe these last years. Recently, concerns were raised of high rates of HCC recurrence in patients treated with DAA.

Aim : We investigated the HCC occurrence and recurrence rates within six months after treatment with DAA with or without Pegylated Interferon (PEG-IFN).

Methods : This is a national, retrospective, multicenter cohort trial, executed in 15 hospitals distributed across Belgium. Data were available from two earlier trials investigating the outcome of treatment with DAA with and without PEG-IFN. A new data collection based on the patient files was executed by medical doctors. Populations were matched based on fibrosis score starting from F3. Patients with a Child-Pugh score \geq B were excluded. In total, 472 patients were included in this trial, of whom 72 were treated with DAA with PEG-IFN from 2008 to 2013 and 400 with DAA without PEG-IFN from 2013 until November 2015. In this cohort, also an analysis of the rates of follow up by radiographic analysis was performed.

Results : Patients treated with DAA with PEG-IFN ($53y \pm 8$) were younger than patients treated with DAA without PEG-IFN ($59y \pm 12$) ($p = 0.001$). 48% (38/72) of patients treated with DAA with PEG-IFN were in the F4 stage versus nearly 65% (259/399) of patients treated with DAA without PEG-IFN ($p = 0.004$). The rates of radiographic follow up were 77.8% ($n = 56/72$) in patients treated with DAA with PEG-IFN, and 78.0% ($n = 312/400$) in patients treated with DAA without PEG-IFN. The early occurrence rate of HCC in patients treated with DAA with PEG-IFN was 3.6% ($n = 2/55$) and 1.1% ($n = 3/277$) in patients treated with DAA without PEG-IFN. The early recurrence rate was 0% ($n = 0/1$) in patients treated with DAA with PEG-IFN, and 20.0% ($n = 7/35$) in patients treated with DAA without PEG-IFN.

Conclusions : There is no difference in early occurrence of new HCC between patients treated with DAA with and without PEG-IFN. We did observe a high early recurrence rate of HCC in patients treated with DAA without PEG-IFN. However, we cannot state that this difference is significant to patients treated with DAA with PEG-IFN, especially since there were significant differences in patient characteristics such as age and fibrosis stage. In 20%, screening for HCC was inadequate. More efforts are necessary as we need to remain vigilant when treating high risk patients.

PEGYLATED INTERFERON ALPHA TREATMENT RAPIDLY CLEARS HEPATITIS E VIRUS INFECTIONS IN HUMANIZED MICE. M. Van De Garde (1), S. Pas (2), G. Van Oord (3), L. Gama (4), Y. Choi (5), R. De Man (3), A. Boonstra (3), T. Vanwolleghem (6). [1] Erasmus Medical Center, Rotterdam, Netherlands (the), Gastroenterology and Hepatology, [2] Erasmus Medical Center, Rotterdam, Netherlands (the), Department of Viroscience, [3] Erasmus Medical Center, Rotterdam, Netherlands (the), Department of Gastroenterology and Hepatology, [4] Johns Hopkins University, Baltimore, United States (the), Department of Molecular and Comparative Pathobiology, [5] Center for Disease Control and Prevention, Atlanta, United States (the), Division of Viral Hepatitis, [6] Erasmus Medical Center, Rotterdam, Netherlands (the), Gastroenterology & Hepatology.

Introduction : Safe and effective antiviral options are needed for ribavirin intolerant, immunocompromised patients with chronic Hepatitis E Virus (HEV) genotype (gt) 3 infections. Pegylated interferon (pegIFN) has been used extensively to treat chronic viral hepatitis infections and baseline intrahepatic IFN-stimulated gene (ISG) expression has been linked to treatment success.

Aim : We studied the antiviral potency of pegIFN against HEV gt3, HEV gt1 and HBV gtA infections in an immunocompromised small animal model and modelled intrahepatic ISG responses pre- and post-treatment.

Methods : 65 uPA+/+Nod-SCID-IL2R γ ^{-/-} mice were transplanted with one of three human hepatocyte donors. Human liver-chimeric mice were challenged with HEV gt3, HEV gt1 or HBV gtA. Infected mice received either a single or twice weekly injections with pegIFNa-2b for 2 or 4 weeks. Quantification of HEV RNA was performed in liver, bile and feces using RT-qPCR. Human gene expression of human-chimeric mouse livers was analyzed using RT-qPCR and the nanostring nCounter® human-immunology panel for respectively 10 and 578 genes. 5 Non-chimeric mice were used as controls. Human CXCL10 was measured in mouse serum.

Results : HEV gt3 infections were cleared from liver and feces after 8 and 4 pegIFN doses, but relapsed in 2/4 mice after a single pegIFN injection. PegIFN anti-HEV activity was confirmed in HEV gt1 infected mice with complete clearance from liver and feces after 4 injections. In contrast, HBV gtA infected mice showed a <1 log IU/ml drop in serum HBV DNA and had high intrahepatic HBV DNA levels (>6 log IU/gr liver) at the end of a 2 week pegIFN treatment course. Baseline pre-treatment ISG expression was evaluated in 20 HEV gt3 and 10 HEV gt1 infected chimeric-mouse livers and revealed no ISG induction compared to 8 control chimeric mice. An in-depth gene expression array on 14 HEV gt3 infected chimeric-mice confirmed the absence ISG induction, irrespective of time point after inoculation, hepatocyte donor or HEV strain. Post- pegIFN treatment a clear human specific ISG induction was observed in liver (>10-fold CXCL10 mRNA increase), which led to increased circulating human CXCL10 levels in mouse serum.

Conclusions : HEV gt1 and gt3 infections do not induce innate intrahepatic immune responses and are extremely sensitive to pegIFN in immunocompromised humanized mice. This might inform treatment strategies for ribavirin resistant HEV.

PERSONALIZED SUBCUTANEOUS ADMINISTRATION OF HEPATITIS B SURFACE ANTIBODIES WITHOUT NUCLEOS(T)IDE ANALOGUES IS HIGHLY EFFECTIVE AND REDUCES COST FOR HEPATITIS B PROPHYLAXIS AFTER LIVER TRANSPLANTATION. R. Bielen (1), G. Robaey (2), S. Schelfhout (3), D. Monbaliu (4), S. Van Der Merwe (3), J. Pirenne (4), F. Nevens (3). [1] University Hasselt, Hasselt, Belgium, Faculty of medicine and life sciences, [2] Ziekenhuis Oost-Limburg Genk, Genk, Belgium, Gastro-Enterology and Hepatology, [3] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Gastro-Enterology and Hepatology, [4] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Abdominal Transplant Surgery.

Introduction : Intravenous Hepatitis B Immunoglobulins (HBIG) in combination with nucleos(t)ide analogues (NAs) are the cornerstone of prophylaxis against Hepatitis B recurrence after liver transplantation (LT). Long-term use of IV HBIG has a high cost and the regular admission in the hospital is inconvenient. NAs alone does not always prevent HBsAg recurrence and can be nephrotoxic. SC HBIG can be self-administered. The optimal dose of SC HBIG without concomitant use of NAs has never been studied.

Aim : To study the optimal dose of SC HBIG without concomitant use of NAs.

Methods : This is an investigator driven, prospective trial. All patients receiving IV HBIG were switched to SC HBIG (Zutectra®) without NAs. The doses and interval of SC HBIG administration were aimed to keep HBsAg and HBV DNA undetectable. First dosage of Zutectra® was based on the guidelines of the manufacturer (< 75 kg : 500 IU/week; ≥ 75 kg : 1.000 IU/week). Thereafter, the titer of HBsAb was monitored regularly and if the titer was higher than the target levels at 2 successive occasions, a dose reduction was executed. In patients with low risk of recurrence (pts with undetectable HBV without antiviral therapy before LT, pts with acute liver failure and Delta hepatitis co-infected pts), the targeted titer was ≥ 100 IU/l and in the other patients ≥ 200 IU/l. The tolerance of the patients (IV or SC) was assessed by a specific questionnaire.

Results : 44 patients were included in this trial. One patient preferred to switch again to IV HBIG, all the others (n=43) preferred SC HBIG, they did not report side effects and the compliance was 100%. The mean time after LT was 9 ± 6 years. Mean follow up time was 2 years \pm 7 months. None of the patients had a relapse of HBsAg or HBV DNA. The mean HBsAb titer before the study was 332 ± 173 IU/l. The mean HBsAb titer at the end of the follow up period was 253 ± 121 IU/l in the low risk group (n=14) and 281 ± 91 IU/l in the high risk group (n=21). In 76% (n=33) doses reductions were possible. The total combined dose at the start was reduced from 118.000 IU /month to 68.135 IU/month. The median frequency of injections reduced from 1/w to 1/2w (range 2/w -1/3 w).

Conclusions : All except one patient preferred subcutaneous HBIG. SC HBIG without NAs had a 100% success rate in the long-term prevention of HBsAg and HBV DNA reappearance. Doses adaptation based on pre LT risk factors for HBV recurrence resulted in the vast majority of the pts in reduction of doses and/or prolongation of the interval and together with the self-administration and the no use of NAs induced a significant reduction of cost.

- A24 -

PPAR α -REGULATED DERMATOPONTIN IS AN IMPORTANT CONTRIBUTOR TO THE LIVER FIBROTIC RESPONSE IN MOUSE MODELS AND HAS RELEVANCE TO FIBROSIS PROGRESSION IN NAFLD PATIENTS. P. Lefebvre (1), S. Francque (2), F. Lalloyer (3), E. Baugé (4), A. Verijken (5), L. Van Gaal (5), B. Staels (3). [1] INSERM, France, Institut Pasteur de Lille, [2] UZA, Edegem, Belgium, Department of Gastroenterology and Hepatology, University Hospital Antwerp, [3] Univ Lille 2, France, Institut Pasteur de Lille, [4] Institut Pasteur, Lille, France, N/A, [5] UZA, Edegem, Belgium, Department of Endocrinology, Diabetology and Metabolism, University Hospital Antwerp,

Introduction : Non-Alcoholic Fatty Liver Disease (NAFLD) is associated to obesity and predisposes to liver- and extrahepatic-related morbidities such as cirrhosis, hepatocarcinoma and cardiovascular diseases. A key step in NAFLD progression is fibrosis, whereby abnormal deposition of extra-cellular matrix (ECM) components occurs in the space of Disse.

Aim : Identifying molecular mechanisms leading to increased ECM deposition, and defining molecular pathways amenable to pharmacological manipulation would be decisive in fighting NAFLD progression.

Methods : A comparative analysis of liver transcriptome from NASH patients and murine models of nonalcoholic steatohepatitis (NASH) was carried out. Candidate genes whose expression was correlated to the severity of NAFLD (NASH CRN score) were selected. A gene whose expression increased in NASH/fibrosis and was normalized by the activation of hepatic peroxisome proliferator activated receptor alpha (PPAR α) was identified. Its contribution to the fibrotic response was studied by gene deletion studies in mice.

Results : Comparative transcriptomic studies in NASH patients and murine models of NASH or fibrosis identified a response characteristic of hepatic stellate activation. A subset of genes was identified as a potential target of the TGF β , CTGF or the PPAR pathway and involved in ECM homeostasis using data mining strategies in ChIP-Seq databases and gene ontology term enrichment. Among them, dermatopontin (Dpt) was identified as a novel contributor to the fibrotic response. Gene deletion showed decreased ECM deposition in Dpt KO mice submitted to a pro-fibrotic insult (CC14). In various models of rodent NASH, Dpt expression was lowered by PPAR α activation. Furthermore, Dpt expression was normalized by bariatric surgery in human NASH patients.

Conclusions : Dpt is an important contributor to the fibrotic response and its expression is amenable to pharmacological control.

- A26 -

KUPFFER CELL POOL IS MAINTAINED BY LOCAL PROLIFERATION AND THE DIFFERENTIATION OF BONE MARROW MONOCYTES INTO SHORT-LIVED MONOCYTE-DERIVED KUPFFER CELLS DURING NON-ALCOHOLIC STEATOHEPATITIS AND RECOVERY. L. Devisscher (1), C. Scott (2), S. Lefere (1), S. Raevens (1), E. Bogaerts (1), A. Paridaens (1), X. Verhelst (1), A. Geerts (1), M. Guilliams (2), H. Van Vlierberghe (1). [1] Ghent University, Ghent, Belgium, Gastro-enterology and Hepatology, [2] Ghent University, Ghent, Belgium, Department of Biomedical Molecular Biology.

Introduction : Kupffer cells (KCs) and liver infiltrating bone-marrow (BM) monocyte-derived macrophages (mo-Mf) have been denoted as key players in the pathogenesis of non-alcoholic steatohepatitis (NASH). Despite this, to date it has not been possible to accurately discriminate between these two populations due to the lack of specific markers. Additionally, KCs were believed to be derived solely from embryonic progenitors, which are maintained by self-renewal, however, it has recently been demonstrated that BM monocytes can differentiate into bona fide KCs (mo-KCs) when required. To date, it is also unclear if mo-KCs are present during NASH. Understanding which of the distinct macrophage populations are present and the roles they play in NASH is crucial to furthering our understanding of NASH pathogenesis and the development of novel therapies.

Aim : By using newly defined specific markers including Clec4F and Tim4 alongside BM chimeras, we aimed at accurately characterize the dynamic changes and origins of the distinct liver macrophage subsets in experimental-induced NASH and recovery.

Methods : Immunohistopathology and flow cytometry were used to determine the level of steatosis, steatohepatitis and recovery in methionine and choline deficient (MCD) diet fed mice. Flow cytometric analysis including the specific markers Clec4F and Tim4 and BM chimeras were applied to identify the distinct liver macrophage subsets and their origins.

Results : Mice fed the MCD diet for 8 weeks gradually developed severe steatohepatitis while replacement of MCD diet by normal chow resulted in full recovery after 4 weeks. Ly6CloClec4F-Tim4⁻ infiltrated mo-Mf were observed from week 2 of MCD feeding, further increased during MCD feeding and returned to baseline during recovery. The absolute number of KCs, characterized as Ly6CloClec4F⁺Tim4⁺ cells, did not differ significantly between mice fed either MCD or the control diet (CD) over the duration of feeding or during recovery. However, an increased proportion of Ki-67⁺ proliferating KCs were observed in mice fed MCD diet compared with mice fed control diet. In line with this, we observed the development of a new population of Ly6CloClec4F⁺Tim4⁻ KCs, only typically present in minor numbers in steady state, previously identified as mo-KCs. Mo-KCs developed from week 4 on the MCD diet and remained present during recovery. As lack of Tim4 expression is only a temporal marker of mo-KCs, with mo-KCs gradually gaining Tim4 expression after their differentiation, we utilised BM chimeras to both validate the presence of mo-KCs and determine their longevity. Interestingly, while these cells do develop from monocytes during MCD feeding and peak during initial recovery, they do not have the capacity to self-renew as their numbers are reduced by week 4 recovery.

Conclusions : Our findings demonstrate that during NASH pathogenesis and recovery the KC pool is maintained by proliferation and the differentiation of short-lived mo-KCs in the MCD diet model.

- A27 -

CESSATION OF NUCLEOS(T)IDE ANALOGUE THERAPY AFTER HBEAG SEROCONVERSION IS ASSOCIATED WITH A DECREASED CHANCE OF HBSAG LOSS IN A BELGIAN, PREDOMINANTLY CAUCASIAN COHORT OF CHRONIC HEPATITIS B PATIENTS. S. Van Hees (1), S. Bourgeois (2), H. Van Vlierberghe (3), T. Sersté (4), P. Michielsen (1), H. Reynaert (5), J. Henrion (6), S. Negrin-Dastis (7), L. Lasser (8), F. Janssens (9), G. Robaey (10), P. Stärkel (11), C. Moreno (12), F. Nevens (13), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology and Hepatology, [2] ZNA Antwerpen, , Belgium, Department of Gastroenterology and Hepatology, [3] Ghent University Hospital, Ghent, Belgium, Department of Gastroenterology and Hepatology, [4] CHU Saint-Pierre, Brussels, Belgium, Department of Gastroenterology and Hepatology, [5] University Hospital Brussels, Vrije Universiteit Brussel, , Belgium, Department of Gastroenterology and Hepatology, [6] Centre Hospitalier de Jolimont-Lobbes., La Louvière, Belgium, Department of Gastroenterology and Hepatology, [7] Grand Hopital de Charleroi, Charleroi, Belgium, Department of Gastroenterology and Hepatology, [8] CHU Brugmann Brussels, Brussels, Belgium, Department of Gastroenterology and Hepatology, [9] Jessa Hospital, Hasselt, Belgium, Department of Gastroenterology and Hepatology, [10] ZOL, Genk, Belgium, Department of Gastroenterology and Hepatology, [11] Cliniques Universitaires St Luc, Brussels, Belgium, Department of Gastroenterology and Hepatology, [12] CUB Hôpital Erasme, Bruxelles, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology, [13] University Hospital Leuven, KU Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : High relapse rates are seen when Nucleos(t)ide Analogue (NA) treatment is discontinued after HBeAg seroconversion, but this might be accompanied by significant rates of HBsAg loss.

Aim : We studied whether NA stop after HBeAg seroconversion is associated with increased HBsAg loss rates in a Belgian, predominantly Caucasian cohort of Chronic Hepatitis B patients.

Methods : This is a pooled analysis including mono-infected, non-immune-suppressed patients treated with different NA for ≥ 3 months from 13 hospitals in Belgium. All patients were HBeAg positive at start of NA treatment. HBeAg seroconversion was defined as the loss of HBeAg and the appearance of anti-HBeAg on two time points ≥ 1 month apart. Follow-up time was calculated as time from baseline (date of HBeAg seroconversion) until HBsAg loss or end of Follow-up. A Cox regression model was used to determine predictive factors for HBsAg loss.

Results : A total of 326 NA treated patients (74.8% male; 63% Caucasian; 17% African) were included. Patients were treated for a median of 3.4 years using either lamivudine, adefovir, tenofovir, entecavir or telbivudine. Ninety six patients (median age at HBeAg seroconversion 38 years) showed HBeAg seroconversion after a median treatment duration of 15.5 months. NA were stopped in 57/96 patients after a median consolidation therapy of 7.5 months. HBsAg loss was observed in 10 patients on-treatment and 8 patients off-treatment. COX model revealed that stopping NA was significantly associated with a decreased chance of HBsAg loss (HR 0.263; $p=0.006$), whereas presence of cirrhosis at start-of-treatment (HR 0.478; $p=0.147$), age at HBeAg seroconversion (HR 0.795; $p=0.380$) and length of consolidation therapy (HR 1.608; $p=0.409$) were not. Results remained unchanged when adjusted for time to response. Stopped patients had a longer follow-up time after HBeAg seroconversion (median 4 years vs. 2 years; $p=0.003$) and had less cirrhosis (40.6% vs 12.3%; $p=0.001$) compared to continuously treated patients. There was no difference in age at time of HBeAg seroconversion (median 43 vs 35 years; $p=0.567$).

Conclusions : Cessation of NA treatment post-HBeAg seroconversion was associated with a decreased chance of HBsAg loss. In addition, longer consolidation therapy had no significant effect on the chance of HBsAg loss.

- A28 -

A NEW CLASSIFICATION OF CHRONIC PORTAL VEIN OCCLUSION FOR ASSESSING THE FEASIBILITY OF RECANALIZATION IN NON-CIRRHOTIC PATIENTS. A. Marot (1), J. Vieira Barbosa (2), A. Denys (3), P. Deltenre (2). [1] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Gastro-enterology, [2] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Division of Gastroenterology and Hepatology, [3] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Division of Radiology.

Introduction : Chronic portal vein occlusion (PVO) can be associated with gastrointestinal bleeding (GIB), portal biliopathy or intestinal ischemia. Portal vein recanalisation (PVR) is a technique able to treat or prevent complications related to portal hypertension (PH) by addressing PVO itself. However, failure of PVR and stent thrombosis are challenging.

Aim : Identify factors associated with PVR failure and evaluate short and long-term stent patency in non-cirrhotic patients with chronic PVO.

Methods : The charts of patients with chronic PVO in which placement of a stent has been attempted and using a trans-hepatic approach were reviewed. Extension of occlusion was assessed by portography before PVR.

Results : 15 patients were included (12 men, median age 49 years [95% CI : 39-57]). Indications for PVR were GIB (n=5), portal biliopathy (n=2), the need for reducing PH before surgery (n=5) and other reasons (n=3). A procoagulate state was identified in 36% and a local prothrombotic factor in 47%. Occlusion involved the main portal vein, either without (n=8) or with (n=7) the mesenteric and/or the splenic veins. Regarding the intra-hepatic extension of PVO, patients were classified into 3 groups : "type 1" with occlusion limited to the main portal vein (n=6), "type 2" with involvement of portal bifurcation and extension to segmental branches (n=7), and "type 3" with extension to distal branches (n=2). PVR was successful in 13 cases (87%). Failure of PVR occurred in 2 patients : one with type 2 and one with type 3 PVO. The second patient with type 3 developed stent thrombosis 24 hours after PVR. Overall, failure of PVR or stent occlusion within the first 24 hours occurred in 100% in patients with type 3 PVO vs. 8% in those with types 1 or 2 (p=0.002). One patient suffered from liver capsule perforation. The median follow-up was 38 months (95% CI : 12-60). Anticoagulation was given to 10 patients after PVR (77%, median duration : 105 days [95% CI : 57-1000]). In per-protocol analysis performed in the 13 patients in which PVR was feasible, the actuarial probability of stent permeability was 82% at 2 years (95% CI : 59-100, 100% vs. 60% in patients who received and who did not receive anticoagulation, respectively, p=0.1). Ninety percents of the patients had resolution of manifestations related to PH.

Conclusions : PVR is feasible in most patients with PVO unless there is no extension to distal branches. Most patients in which PVR was successful have a permeable stent at 2 years. Anticoagulation seems to prevent secondary thrombosis. PVR has a place in the management of complications related to PVO.

- A29 -

THE PREVALENCE OF CARDIAC INDUCED LIVER DISEASE IN PATIENTS WITH A CHRONIC RIGHT HEART DISORDER. N. Opdeweegh (1), W. Budts (2), J. Van Cleemput (2), T. Roskams (3), J. Pirenne (4), B. Meyns (5), F. Nevens (6). [1] University Hospital Leuven, KU Leuven, Belgium, Department of Gastroenterology & Hepatology, [2] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Cardiology, [3] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Pathology, [4] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Abdominal transplant surgery, [5] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Cardiac surgery, [6] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology & Hepatology.

Introduction : The survival of patients with chronic heart disease has significantly improved, especially in children. Chronic elevated right heart pressure might provoke cardiac induced liver disease and finally cardiac cirrhosis, which is overall a rare condition.

Aim : We studied the prevalence of cardiac induced liver disease in patients with longstanding elevated right heart pressure.

Methods : The study population consists of 120 patients. The suspicion of a cardiac induced liver disease was based on lab tests and abdominal ultrasound and the risk of liver related mortality was assessed by the VAST-score (0-3). The first study group were 98 young adult patients who underwent a Fontan procedure during childhood. Mean time post-Fontan was 17 ± 6 years. The second study group contained of 22 patients of middle age with end staged cardiac disease who were possible candidates for heart transplantation and with suspicion of associated liver disease based on lab tests. The presence of cardiac cirrhosis in this group was investigated with transjugular liver biopsy.

Results : In the Fontan patients 4/98 (4%) needed a heart transplantation; 9/98 (9%) had a VAST-score ≥ 2 . In the majority, the Fontan intervention was performed > 16 year before. None of these patients needed a combined heart-liver transplantation. In the second group, 9/22 (36%) received a heart transplantation; 8/22 (36%) had a VAST-score ≥ 2 and 4/22 (18 %) patients had histological proven cardiac cirrhosis. A combined heart-liver transplantation was necessary in 5/9 (55%).

Conclusions : Cardiac induced liver disease is not uncommon in patients with chronic elevated right heart pressure. It occurred in 9% of our adult Fontan patients after long-term follow-up. In patients with end stage cardiac failure and disturbed liver test, the incidence of cardiac cirrhosis was 18% and combined heart-liver transplantation should be considered in these patients.

- A30 -

BEYOND MILAN CRITERIA IN HEPATOCELLULAR CARCINOMA : DOES FLUORINE-18-FLUORODEOXYGLUCOSE (FDG-PET) HOLD THE KEY. V. AGARWAL (1), S. PANDE (2) / [1] MEDANTA THE MEDICITY HOSPITAL, Gurgaon, India, NUCLEAR MEDICINE DEPARTMENT, [2] MEDANTA THE MEDICITY HOSPITAL, Gurgaon, India, Nuclear Medicine Department.

Introduction : Worldwide, the Milan criteria has been the standard for selection of patients with hepatocellular carcinomas (HCC) for liver transplantation, with reported 5-year survival of over 70% after transplantation. However, it is deemed to be too restrictive, and many strategies like including serum tumour marker assays or tumour biopsy to assess histological grade are being attempted to include patients who would have otherwise been denied potentially curative liver transplant without compromising survival. A modest expansion of this criteria now seems justified, in the setting of availability of living donors.

Aim : The aim of our study was to assess the usefulness of positron emission tomography with fluorine 18-fluorodeoxyglucose (FDG-PET) in modification of recipient selection in more advanced tumour stage.

Methods : FDG PET scans of 23 patients on the liver transplant service with primary tumour exceeding Milan Criteria (single tumor >5 cm and multiple tumors >3) were retrospectively reviewed. FDG positivity was defined as tumour / non tumour background ratio more than 1.5 (group A, n=13), while non-avid tumours were categorized into group B (n=11). The largest single tumour measured was 8.1 cm, and 8 cases were multicentric. Results were correlated after transplant with clinical, biochemical and radiological follow up (mean period 48 months).

Results : Of the 13 patients with FDG positivity at baseline, 11 (84.6%) presented with recurrence (intra and extra hepatic). In 8 patients (61%), the primary tumour was solitary, suggesting that metabolic information is independent of macromorphology. None of the patients with tumour to background ratio more than 3 or absolute SUV of more than 9 had tumour free survival exceeding 8 months. In group B (n=10), there were only 2 (20%) recurrences. Overall, the tumour free survival in group B was 100%, 90% and 80% at 2, 3 and 4 years, respectively. There was a significant association between recurrence and FDG avidity ($p = 0.0003$), whereas absence of FDG uptake in spite of tumour size >5cm correlated with favorable outcome.

Conclusions : In the effort to offer curative liver transplant to maximum number of patients with HCC, merely considering tumour macromorphology appears insufficient. Instead, a multidisciplinary approach including factors providing reliable insight into tumour biology would be more appropriate. In this regard, the metabolic information provided by FDG-PET could be the single most important factor in guiding the decision making process in advanced tumors.

- A31 -

INCREASED NON-SHIVERING THERMOGENESIS HAD PREVENTIVE BUT NO THERAPEUTIC EFFECTS ON NON-ALCOHOLIC STEATOHEPATITIS. L. Poekes (1), Y. Horsmans (2), G. Farrell (3), I. Leclercq (1). [1] Université Catholique de Louvain, Brussels, Belgium, Laboratory of Hepato-Gastroenterology, [2] Cliniques Universitaires Saint-Luc, Belgium, Gastroenterology Unit, [3] Australian National University, Australia, Liver Research Group.

Introduction : Non-alcoholic steatohepatitis (NASH) is the progressive form of non-alcoholic fatty liver disease spectrum. No treatment has been proven efficacious except for lifestyle modifications coupling physical exercise with weight reduction. We recently identified defective adaptive thermogenesis as a contributing factor to obesity and metabolic syndrome in *foz/foz* mice.

Aim : We now aim to test whether increased non-shivering thermogenesis prevent and/or improve pre-existing NASH in mice.

Methods : A HFD for 4 or 8 weeks induced a metabolic syndrome with fatty liver or NASH, respectively in male *foz/foz* mice. Mice were randomized and treated with a beta 3-adrenergic receptor (B3AR) agonist (CL-316,243 - 1mg/kg/day) to enhance thermogenic capacities or with vehicle (untreated) together with HFD for 2 or 4 weeks, respectively. C57B16

and db/db mice were fed a methionine and choline deficient (MCD) diet to induce NASH and treated with B3AR agonist for 4 additional weeks (n=6-8/group).

Results : In foz/foz mice with metabolic syndrome and liver steatosis, B3AR agonist improved brown adipose tissue (BAT) function assessed by increased cAMP and UCP1 BAT contents and upregulation of thermogenic genes (UCP1, DIO2). It also caused browning of white adipose tissue. All this resulted in increased tolerance to cold exposure and was associated with a better glucose tolerance ($p<0.05$), a decreased NAS score (2 ± 1.3 vs 3.7 ± 1.6 ; $p<0.05$) and decreased transaminases levels ($p<0.05$) with no change in body weight. When treatment was initiated after the onset of NASH (NAS score = 5 ± 1.15) in foz/foz mice, B3AR agonist treatment restored BAT function, induced a slight 2% weight loss ($p<0.05$), increased glucose tolerance ($p<0.001$) but had no impact on liver pathology (NAS score 5.6 ± 2.1 vs 6.7 ± 1.3 ; ALT 286 ± 117 vs 396 ± 190 U/L) compared to untreated mice. Similarly, B3AR agonist has no therapeutic effect when administered for 4 weeks on MCD-induced NASH whether in C57Bl6 or in obese and diabetics db/db mice.

Conclusions : B3AR agonist treatment improved BAT function and glucose tolerance, prevented the progression of a simple steatosis to NASH but was not sufficient to cure a pre-established NASH, supporting previous observation that control over metabolic syndrome is insufficient to treat NASH. In our study, B3AR agonist caused no major weight loss and therefore, it will be of interest to evaluate whether BAT stimulation offers an additional advantage over weight loss therapy in NASH management.

- A32 -

SYSTEMATIC REVIEW WITH META-ANALYSIS : LIVER STIFFNESS AND PLATELET COUNT FOR IDENTIFYING PATIENTS WITH COMPENSATED LIVER DISEASE AT LOW RISK OF VARICEAL BLEEDING. A. Marot (1), E. Trepo (2), C. Doerig (1), A. Schoepfer (1), C. Moreno (2), P. Deltenre (1). [1] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Division of Gastroenterology and Hepatology, [2] Erasme Hospital, Brussels, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology.

Introduction : The 2015 Baveno VI guidelines recommend against performing upper gastrointestinal endoscopy in patients with compensated cirrhosis who have a liver stiffness <20 kPa and a platelet count $>150'000/\text{mm}^3$ because of a low prevalence of varices at risk of bleeding in this population.

Aim : Synthesize the available evidence on the usefulness of the combined use of liver stiffness and platelet count to identify patients without esophageal varices.

Methods : Meta-analysis of trials evaluating the usefulness of a given cut-off for liver stiffness and platelet count to rule out the presence of esophageal varices.

Results : Fifteen studies were included. There were 997 patients with low liver stiffness and normal platelet count and 2'367 patients with either high liver stiffness or low platelet count. All studies excepting 5 used the Baveno VI criteria. Compared to patients with either high liver stiffness or low platelet count, those with low liver stiffness and normal platelet count had a lower risk of varices (OR=0.23, 95% CI=0.17-0.32, $p<0.001$) with moderate heterogeneity between studies ($I^2=28\%$). They also had a lower risk of varices at risk of bleeding (OR=0.22, 95% CI=0.13-0.39, $p<0.001$) with low heterogeneity between studies ($I^2=21\%$). In patients with low liver stiffness and normal platelet count, the pooled estimate rates for varices at risk of bleeding was 0.040 (95% CI=0.027-0.059) with low heterogeneity between studies ($I^2=3\%$). In the sensitivity analyses excluding studies that did not use a liver stiffness cut-off of 20 kPa or a platelet cut-off of $150'000/\text{mm}^3$, the pooled estimate rate for varices at risk of bleeding was 0.031 (95% CI=0.017-0.055) with no heterogeneity between studies ($p=0.5$, $I^2=0\%$). In the subgroup analysis including only published studies, the pooled estimate rate for varices at risk of bleeding was 0.025 (95% CI=0.012-0.052) for patients with low liver stiffness and normal platelet count, with no heterogeneity between studies ($p=0.8$, $I^2=0\%$).

Conclusions : Patients with low liver stiffness and normal platelet count have a lower risk of varices than those with either high liver stiffness or low platelet count. Varices at risk of bleeding are found in no more than 4% of patients when liver stiffness is <20 kPa and platelet count is normal.

- A33 -

LARGE-SCALE SCREENING IS NOT USEFUL TO IDENTIFY INDIVIDUALS WITH HEPATITIS B OR C VIRUS INFECTION : RESULTS OF AN INTERIM ANALYSIS. A. MAROT (1), A. TRABELSI (2), C. ANDRÉ (3), P. DELTENRE (2). [1] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Gastro-enterology, [2] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Division of Gastroenterology and Hepatology, [3] Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland, Division of Immunology and allergology.

Introduction : Current treatments are able to control HBV replication and to eradicate HCV in almost all cases. Further improvements in the management of HBV and HCV infections will be possible by focusing on treatment impact at a

population level for which screening is an essential step. As most patients with HBV or HCV infection are still undiagnosed, large-scale screening could be useful.

Aim : To investigate whether large-scale screening for HBV or HCV infection (e.g. risk-based vs. age-based) could identify infected individuals.

Methods : Individuals between 18 and 80 years attending the pre-operative consultation prior to minor surgery in a general surgical outpatient clinic are being tested for HBsAg, anti-HBc and anti-HCV since November 2014. The presence of anti-HCV was confirmed by an Immunodot test. HBV DNA and HCV RNA were determined in HBsAg- and anti-HCV-positive individuals.

Results : Among 1345 individuals tested so far, two were positive for HBsAg (0.2%) and one of these had detectable HBV DNA. Five individuals were positive for anti-HCV (0.4%). Two of these had detectable HCV RNA and 3 had undetectable HCV RNA (1 spontaneously and 2 after a successful antiviral treatment). When compared to those without, people with anti-HCV antibodies had already been screened more frequently for HCV (100% vs. 12%, $p<0.001$) as well as for HBV (80% vs. 20%, $p<0.001$), had more frequently anti-HBc antibodies (40% vs. 4.5%, $p<0.001$), and had used intravenous drugs (80% vs. 0.2%, $p<0.001$), nasal drugs (80% vs. 6.4%, $p<0.001$), or cannabis (80% vs. 7.3%, $p<0.001$) more frequently. None of them were immigrant from an endemic area. The median age of individuals with anti-HCV antibodies was not different from those without (49 years [range : 39-62] vs. 44 years [95% CI : 43-45], $p=0.6$).

Conclusions : In the study population investigated, this interim analysis indicates that a large-scale screening is not useful to identify individuals with undiagnosed HBV or HCV infection. Screening for HBV and HCV infection should focus on individuals with well-known risk factors.

- A34 -

ISOLATION AND CHARACTERISATION OF HEPATIC PROGENITOR CELLS FROM HUMAN ALCOHOLIC LIVERS IDENTIFY A NEW PLAYER : IL-17A. A. Ceulemans (1), S. Verhulst (2), M. Van Haele (1), L. Van Grunsven (2), T. Roskams (1). [1] Katholieke Universiteit Leuven, Leuven, Belgium, Department of Imaging & Pathology, Translational Cell & Tissue Research Group, [2] Vrije Universiteit Brussel, Brussels, Belgium, Department of Basic Biomedical Sciences, Liver Cell Biology Lab.

Introduction : Hepatic progenitor cells (HPCs) are small cells with a relative large oval nucleus and a scanty cytoplasm situated in the canals of Hering. Phenotypically, HPCs express both markers of (immature) hepatocytes (e.g. alpha-fetoprotein) and markers of cholangiocytes (e.g. cytokeratin K7 and K19). The mechanisms facilitating proliferation and differentiation of human HPCs are still poorly understood.

Aim : In this study, we aimed to characterise human HPCs through isolating and comparing, on both protein and RNA level, HPC-enriched cell populations from adult human liver tissue using different isolation methods.

Methods : Fresh human liver tissue was collected from alcoholic steatohepatitis explant livers and HPC-enriched cell populations were obtained via three different isolation methods : side population (SP) which is based on the cell's efflux capacity of Hoechst-33342 and isolation based on the membrane markers EpCAM and TROP-2. FACS-sorted cells and whole liver extracts were evaluated at both protein level (immunohistochemical staining) and RNA level (RNA sequencing). Pathway analysis was performed using Ingenuity Pathway Analysis.

Results : Immunohistochemical evaluation of the isolated fractions indicated the enrichment of HPCs in the SP, EpCAM-positive and TROP-2-positive cell populations. Pathway analysis of the RNA sequencing data from the different isolated HPC fractions shows an enrichment and activation of known HPC pathways like the Wnt/beta-catenin pathway, but also a pathway thus far not linked to HPC activation : IL-17A signalling. Upregulation of downstream targets like IL-8, CXCL1 and CCL20 indicate activation of the IL-17A pathway in HPCs. Interestingly, chemoattractants like IL-23A and IL-1b are upregulated in HPCs, possibly to recruit and activate IL-17A producing cells in the liver. IL-17A has already been linked with fibrogenesis through activation of stellate cells and with inflammation through inducing the production of immune cell chemoattractants. Upregulation of TGFbeta, VEGF, IL6 and PDGFB in HPCs is correspondent with the known link of IL-17A with fibrogenesis.

Conclusions : Our analysis indicates an important role for IL-17A signalling during HPC activation, associated fibrogenesis and inflammation in human alcoholic liver disease.

- A35 -

INTRAPORTAL INFUSION OF LIVER-MESENCHYMAL STEM CELLS IN RATS LEAD TO TRANSIENT INTERRUPTION OF THE HEPATIC BLOOD FLOW : INTRAVITAL MICROSCOPY AND ANAPATHOLOGICAL ANALYSIS. L. Coppin (1), P. Van Der Smissen (2), P. Dollet (3), C. Lombard (3), I. Scheers (1), F. Smets (1), E. Sokal (1), X. Stephenne (1). [1] Cliniques Universitaires St Luc, Brussels, Belgium, Pediatric gastroenterology and hepatology, [2] Université catholique de Louvain (UCL), Brussels, Belgium, Institut de Duve, [3] Université catholique de Louvain (UCL), Brussels, Belgium, Pediatric Gastroenterology and Hepatology.

Introduction : Previous studies showed that liver Mesenchymal Stem Cells (MSC) express a procoagulant activity (PCA) that can be controlled in vitro by an anticoagulant cocktail, combining an antithrombin activator (Heparin) and a thrombin inhibitor (Bivalirudin).

Aim : First we would like to confirm in vivo the PCA of human liver MSCs in a rat model, then study its inhibition by a specific anticoagulant cocktail and finally assess the effect of this cocktail on cell implantation. The main aim of this study is to control the PCA induced by infused liver MSCs without reducing the implantation of cells.

Methods : Wistar rats were transplanted with 50x10⁶/kg fluorescent (cell tracker red) human liver MSCs by intraportal infusion with (n=12) or without (n=13) anticoagulant drugs. Using an intra-femoral catheter we injected FITC-dextran and Hoechst to visualise liver vasculature and cell nuclei. By intravital microscopy (IVM) we analysed the left liver lobe at different time points, 1h-24h-48h and 7days post-infusion to study liver MSCs localisation and their effect on liver microvasculature. By pathological examination, cell localisation, cell implantation, vascular and tissue alterations were studied at the same time points. Serial slides were performed for a standard haematoxylin eosin staining and a human cell staining (human B-1-integrin).

Results : By IVM we observed that the transplanted cells in the first hour formed aggregates in the larger liver vessels, then cells migrate to the sinusoids after 24h, to be quickly cleared after 7days. After 24h, large defects of perfusion were observed in both groups, but normal hepatic vascularisation was restored after 48h. These observations were confirmed by pathological examination. Large necrotic zones surrounding the infused cells were observed after 24h, with respectively 14.7% and 19.5% of the liver tissue in the non-anticoagulated and anticoagulated group. This necrosis decreased rapidly after 48h to 0.5% and 1.9%. The anticoagulant drugs didn't prevent this necrosis, no difference has been observed between the 2 groups (p<0.05). We confirmed that the infused cells are rapidly cleared after 24h from the liver over time, with respectively after 1h, 24h, 48h and 7days a decrease from 1.3%, 0.3%, 0.07% till 0.03% of infused cells. No difference in cell implantation has been observed between the groups with or without anticoagulation (p>0.05).

Conclusions : Intraportal infusion of human liver MSCs to Wistar rats induces a transient alteration in liver vascular flow after 24h. This could be explained by the temporary localisation of liver MSCs in large portal veins and sinusoids up to 1hour after the infusion, in addition to a possible xenotransplantation acute reaction. After 24h more than 70% of cells are cleared and cells are surrounded by transient necrotic-hemorrhagic regions that regress almost completely after 48h. After 7days no necrosis and very few cells are seen. We observed no difference between the two groups, with or without anticoagulation.

- A36 -

LIVER TRANSPLANTATION DOES NOT IMPACT THE RENAL FUNCTION OUTCOME IN ALAGILLE SYNDROME. T. DEMARET (1), S. VARMA (1), Y. VAINILOVICH (1), U. HALAC (1), D. EL BIZRI (1), J. AMBROISE (2), I. SCHEERS (1), X. STEPHENNE (1), F. SMETS (1), E. SOKAL (1). [1] Cliniques Universitaires Saint-Luc, Belgium, Pediatric gastroenterology & hepatology, [2] Université catholique de Louvain, Belgium, Centre for applied molecular technologies (CTMA).

Introduction : Alagille syndrome (AS) is an autosomal dominant multi-systemic disorder caused by pathogenic variants in JAG1 and NOTCH2. Characteristic findings include hepatic involvement with bile duct paucity and 20-50% eventually need a liver transplantation. Post-LT Tacrolimus induced nephropathy is well recognised and 40% of AS patients have an underlying renal anomaly.

Aim : In the current study, we analysed the impact of LT and Tacrolimus on the evolution of renal function (RF) in children with AS.

Methods : Retrospective study including 50 children that satisfied 3 of 5 major Alagille syndrome criteria and under regular follow-up at our centre between 1984 and 2016. Clinical, biochemical and radiological data were collected at similar time points of follow-up among the transplanted and non-transplanted children. The time points were at diagnosis or at LT and after 1-2 years, 2-3 years, 3-5 years, 5-7 years and 7-10 years of follow-up. The RF was estimated by glomerular filtration rate (eGFR) using the updated Schwartz formula. The RF outcomes of children with AS having undergone LT were compared with those without LT and also with children having undergone LT for non-AS related indication but without associated nephropathy.

Results : 28 of 50 (56%) included AS children underwent LT and were compared with 77 children transplanted for non-AS indications. Mean eGFR post-LT in AS patients and non-AS patients were 93.8 mL/min and 143.2 mL/min, respectively (difference : 49.4 mL/min, p<0.0001). Among children with AS mean eGFR observed in those who did not receive LT was 87.9 mL/min, -5.9 mL/min compared to those who received LT though this was statistically insignificant (p=0.32). Presence of renal ultrasound abnormalities was correlated to RF impairment in AS patients, with or without LT : -14.6 mL/min (98.5 mL/min vs 83.9 mL/min, p=0.03) and -40.9 mL/min (97.8 mL/min vs 56.9 mL/min, p<0.0001), respectively.

Conclusions : Post-LT renal function outcomes are significantly worse in children with AS being the primary disease. Among the children with AS, the RF outcome is not worse after LT.

ANTIBIOTICS INDUCE REMISSION IN PEDIATRIC PSC-AIH OVERLAP SYNDROME ALLOWING CORTICOSTEROID-FREE THERAPY. P. Sambon (1), S. Varma (2), M. Komuta (3), P. Clapuyt (4), E. Sokal (1). [1] Cliniques Universitaires St-Luc, Brussels, Belgium, Pediatric Gastroenterology and Hepatology, [2] Cliniques universitaires St-Luc, Brussels, Belgium, Pediatric Gastroenterology and Hepatology, [3] Cliniques Universitaires St-Luc, Brussels, Belgium, Service of anatomical pathology, [4] Cliniques Universitaires St Luc, Brussels, Belgium, Pediatric Radiology Unit.

Introduction : Concomitant presence of autoimmune hepatitis (AIH) and primary sclerosing cholangitis (PSC) is labelled as AIH-PSC overlap syndrome or autoimmune sclerosing cholangitis (ASC). Treatment of AIH with corticosteroids and azathioprine; and of the PSC component with ursodeoxycholic acid (UDCA) is the standard practice. Antibiotics are increasingly being shown to have benefit in PSC but their role in paediatric ASC is not well evaluated.

Aim : We investigated the response to oral antibiotics as initial or subsequent therapy in children with ASC.

Methods : Patients diagnosed with ASC on basis of biochemical, liver biopsy and radiology findings were included. They received metronidazole or vancomycin for 14 days [10-220] either at diagnosis (i.e. initial therapy) or during their maintenance period. When antibiotics were administered as initial therapy, steroid free induction regime was adopted. In children during the maintenance phase antibiotics were administered if they had not achieved biochemical remission with their standard treatment of steroids, azathioprine and UDCA. The outcome parameters to assess the efficacy of antibiotics were achievement of biochemical remission and additionally steroid avoidance when given in the initial therapy.

Results : Ten children with ASC were included, of which 6 received oral antibiotics (4 metronidazole, 2 vancomycin) at diagnosis and 4 received metronidazole during the maintenance period. All patients showed a significant decrease in their AST (-55%, $p=0,005$), ALT (-84%, $p=0,003$) and GGT (-53%, $p=0,003$), without significant difference across the two groups. All six children in the initial therapy group did not need corticosteroids and continued to be in remission until last follow up duration of 400 days [216-888]. Among the four children administered antibiotics in the maintenance phase, two showed biochemical remission and steroids could be tapered; while two did not show any benefit. There was transient biochemical relapse after stopping antibiotics in one responder, for which they were restarted and continued until last follow up while continuing to be in remission.

Conclusions : We demonstrate the benefit of antibiotics in ASC by achieving steroid free treatment when given at diagnosis as induction regime. When given in the maintenance phase they assist in achieving long term biochemical remission in an otherwise uncontrolled ASC.

HAIR ETHYL GLUCURONIDE IS A HIGHLY ACCURATE AND OBJECTIVE BIOMARKER OF CONTINUED ALCOHOL USE IN PATIENTS WITH ALCOHOLIC CIRRHOSIS. J. Verbeek (1), C. Crunelle (2), P. Michielsens (3), M. De Doncker (4), S. Van Der Merwe (1), D. Cassiman (1), H. Neels (2), F. Nevens (1). [1] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology & Hepatology, [2] University of Antwerp, Antwerp, Belgium, Toxicological Center, [3] University Hospital Antwerp, Antwerp, Belgium, Department of Gastroenterology & Hepatology, [4] ZNA Campus Stuivenberg, Belgium, Department and Toxicology Laboratory.

Introduction : There is no golden standard to prove chronic alcohol abuse in patients with cirrhosis. Hair ethyl glucuronide (hEtG) is an objective biomarker of alcohol use in patients without cirrhosis and it reflects the amount of alcohol intake over several months.

Aim : We tested the diagnostic accuracy of hEtG in a cohort of patients with alcoholic cirrhosis who minimalized their alcohol use.

Methods : The validation cohort ($n=101$) consisted of 43 healthy volunteers and 58 patients with alcoholic cirrhosis. Abstinence in this group was defined as the absence of any clinical sign of alcohol use during a 1 year follow-up. The clinical application group consisted of 43 random patients with alcoholic cirrhosis who minimalized or ignored ongoing excessive alcohol use (≥ 60 g/d). A detailed questionnaire, proximal 3 cm scalp hair strand to measure hEtG and blood to assess conventional biomarkers were collected and in the application group transjugular liver biopsies were performed to assess of satellitosis.

Results : There was no difference in the characteristics of the patients with cirrhosis in the validation ($n=50$) and the application group ($n=43$). Gamma-glutamyl transferase and AST were excellent indicators (sensitivity 94%) of abstinence but lacked specificity (respectively 26% and 33%). The correlation between hEtG and the histologic signs of ongoing alcohol use was modest. In contrast, hEtG value ≥ 50 pg/mg was highly sensitive (100%) and specific (100%) of chronic excessive alcohol use over the last 3 months in the validation group. Hair EtG levels correlated with the amount of alcohol intake in the previous 3 months ($R^2 = 0.62$, $p=0.0087$) and were not influenced by the stage of liver or renal impairment. Of the patients in the application group who minimalized their alcohol use, 35% had values ≥ 50 pg/mg.

Conclusions : Also in patients with cirrhosis, hEtG is a highly accurate and objective biomarker of chronic excessive alcohol use. Where gamma-glutamyl transferase and AST stay elevated, it offers the clinician an objective proof of alcohol abuse during the preceding 3 months.

- A39 -

HBsAg KINETICS DURING LONG-TERM NUC THERAPY IN A PREDOMINANTLY CAUCASIAN AND AFRICAN COHORT. E. Ho (1), S. Van Hees (2), M. Ieven (3), V. Matheeußen (3), S. Francque (4), P. Michielsen (4), T. Vanwolleghem (4). [1] Universitair Ziekenhuis Antwerpen, Edegem, Belgium, Gastroenterology and Hepatology, [2] University of Antwerp, Antwerp, Belgium, Laboratory of Experimental Medicine and Pediatrics, Faculty of Medicine and Health Sciences, [3] Antwerp University Hospital, Edegem, Belgium, Laboratory Medicine, [4] Antwerp University Hospital, Edegem, Belgium, Gastroenterology and Hepatology.

Introduction : Background and Aims : Quantitative HBsAg levels (qHBsAg) have been shown to decrease slowly in virally suppressed chronic HBV (CHB) patients treated with nucleos(t)ide analogues (NUCs). The kinetics of qHBsAg has been extensively studied in Asian cohorts, but evidence in Caucasian or African patients is scarce.

Aim : This study aims to describe long-term HBsAg kinetics in a single centre with NUC treated patients of predominantly Caucasian and African ethnicity in Belgium.

Methods : qHBsAg was analysed using the HBsAg II quant assay of Roche Diagnostics in banked sera from CHB patients followed at the Antwerp University Hospital between 2010 and 2016 (2-7 visits).

Results : Sixty-two patients were included; 77.4% (48/62) were male. Mean age was 46.4 years (25.4-76.8); 58.1% were of Caucasian, 21.0% of Sub-Saharan African and 21.0% of Asian ethnicity. Treatment consisted of lamivudine (n=8), adefovir (n=3), entecavir (n=26) and tenofovir (n=25). 77.4% (48/62) of patients were HBeAg negative at inclusion. One person became HBeAg negative during follow-up. Mean follow-up time was 32.5 months (6.3-65.8) and mean treatment duration 71.2 months (7.0-197.9). Mean duration of viral suppression (defined as time passed between first occurrence of HBV DNA < 15 IU/mL and the end of follow-up period) was 38.4 months (0-136.7). Overall qHBsAg decline was significant (p<0.001). Medians at start and end of follow-up were 2444.5 (9.9-52,000.0) IU/mL and 2231.0 (20.0-27,375.0) IU/mL, respectively. qHBsAg levels and qHBsAg evolution over time did not differ significantly according to ethnicity and NUC used, but qHBsAg levels were lower in HBeAg negative patients compared to HBeAg positive patients throughout follow-up (medians at start : 2381 (9.9-15,539.0) vs. 11566.5 (78.1-52,000.0), p=0.039, medians at end : 1774.0 (20.0-13,142.0) vs. 8551.0 (92.9-27,375.0), p=0.003). In HBeAg negative patients, qHBsAg decline was significant (p<0.001) but no significant difference was found in HBeAg positive patients (p=0.8). qHBsAg levels at the end of follow-up correlated inversely with the length of treatment and of viral suppression : respectively r=-0.310, p=0.014 and r=-0.331, p=0.009.

Conclusions : In a cohort of predominantly Caucasian and African HBeAg negative CHB patients, qHBsAg decreases slowly during continuous NUC-induced HBV DNA suppression. During a mean follow-up of 32.5 months, no significant qHBsAg changes were seen in virally suppressed NUC-treated HBeAg positive patients.

- A40 -

“CHINA ON THE SCHELDT” : HEPATITIS B AND C IN THE CHINESE POPULATION IN BELGIUM AND COMPARISON OF SCREENING METHODS. E. Ho (1), P. Michielsen (1), P. Van Damme (2), S. Francque (1), M. Ieven (3), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Gastroenterology and Hepatology, [2] University of Antwerp, Antwerp, Belgium, Centre for the Evaluation of Vaccination, Vaccine & Infectious Disease Institute, Faculty of Medicine and Health Sciences, [3] Antwerp University Hospital, Edegem, Belgium, Laboratory Medicine.

Introduction : Recent reports suggest a considerable fraction of the viral hepatitis burden in Western countries is introduced by immigrants from high endemic countries such as China.

Aim : This study aims to screen for HBV and HCV infection by testing HBsAg, anti-HBc and anti-HCV in the Chinese community in Belgium. Three screening methods are compared : serum, saliva and dried blood spot (DBS).

Methods : On-site screening with the cooperation of volunteers of the Chinese community was performed starting 25/10/2014 and is ongoing. Minimal clinical and personal information were obtained. Saliva testing was performed using the OraSure Intercept 2 Oral Collection Device. PBS-Tween 0.05% solution was used to acquire eluates from DBS on Whatman Protein Saver 701 cards. All sample types were tested on the Roche Elecsys serology platform.

Results : 460 persons were screened. HBsAg negative persons were slightly older (mean 52 years (18-87) vs. 45 (26-66)), had higher self-reported levels of vaccination (27.3% vs. 9.4%) and less likely received treatment (1.9% vs. 18.8%) Serum results were available from 456 persons. Serological testing : HBsAg was positive in 32/456 persons (7.0%). Almost half of them were unaware of their HBV infection (19/32, 59.4%). Anti-HBc antibody was positive in 244/456 persons

(53.5%). Nobody was positive for anti-HCV. Comparative serologic testing is ongoing and pending validation : saliva testing was positive for HBsAg 18/25 (72.0%) and for DBS testing 29/32 (90.6%). For anti-HBc testing, positivity was 16/175 (9.1%) and 42/244 (17.2%) for saliva and DBS testing, respectively. 16/32 persons have reported for outpatient follow-up. In the patients in follow-up, 2 main groups were found : inactive carriers and immune active, chronic hepatitis B patients. In the immune active patients, HBV DNA was higher (756 vs. log 6.9), ALT was higher (92.6 vs. 29.3) and mean kPa values in transient elastography was higher (20.5 vs. 4.9).

Conclusions : HBsAg seroprevalence in Chinese immigrants in Belgium is high (7.0%) and HBV status was unknown to many (59.4%). Most come from Eastern and Southern Chinese regions.

- A41 -

NON-SENSE MEDIATED RNA DECAY REGULATES THE UNFOLDED PROTEIN RESPONSE DURING HEPATIC STELLATE CELL ACTIVATION. I. Mannaerts (1), L. Thoen (1), F. Cubero (2), I. Colle (3), C. Trautwein (2), I. Coldham (4), L. Van Grunsven (1) / [1] Vrije Universiteit Brussel, Jette, Belgium, Bmwe-Livr, [2] UH Aachen, Belgium, Internal Medicine III, [3] UZ Gent, Gent, Belgium, Hepatology and Gastroenterology, [4] University of Sheffield, Sheffield, United Kingdom (the), Synthetic Organic Chemistry.

Introduction : Liver fibrosis or scarring of the liver is the consequence of prolonged hepatocytic damage that results in persisting hepatic stellate cell (HSC) activation. This makes stellate cells the primary targets for anti-fibrotic therapy and emphasizes the need to understand how HSCs contribute to fibrosis development. The unfolded protein response (UPR) is a cellular response related to ER stress. Chemical induction of ER stress has been shown to affect HSC activation. The nonsense-mediated RNA decay (NMD) pathway functions in RNA quality control. Aberrant mRNAs are rapidly degraded, and a subset of normal mRNAs is regulated by NMD.

Aim : To evaluate whether the endogenous UPR is essential for the earliest phases of mouse HSC (mHSC) activation and how this UPR is regulated.

Methods : In vitro and in vivo activated HSCs were analyzed for ER stress by qPCR, western blot and immunohistochemistry, in WT and JNK-Knock-out mice. ER stress inducers and NMD inhibitors are used in HSC cultures.

Results : The ER stress markers, XBP1spliced, Bip and Chop, showed an early peak in mRNA and protein expression already 10h after seeding primary mouse HSCs on plastic culture dishes, followed by a decreased expression at 24h. This temporarily increased ER stress is also seen in freshly isolated HSCs from mice 10h after 1 CCl₄ injection, suggesting that ER stress is an early event of HSC activation also in vivo. HSCs cultured as 3D spheroids showed prevention of early ER stress and inhibition of HSC activation compared to HSCs plated on plastic. In 3D HSC cultures, chemical induced ER-stress is not sufficient to induce HSC activation. Treatment of HSCs with JNK inhibitors prevents the initial ER stress and reduces culture-induced activation of primary mouse HSC. This role for JNK was confirmed using JNK1 KO mice where decreased ER stress and activation were observed when isolated HSCs were plated. NMD inhibitors induce UPR and enhance HSC activation in vitro suggesting an active role for NMD in the regulation of the UPR and HSC activation.

Conclusions : ER stress induction is an early event during HSC activation in vitro and in vivo. In vitro, this acute ER stress is JNK1 dependent, but is not sufficient to drive the activation process. Ongoing work strongly suggests a potential role for NMD in the regulation of the UPR termination and HSC activation.

- A42 -

THE INFLUENCE OF DIRECT ACTING ANTIVIRALS ON EXTRAHEPATIC MANIFESTATIONS OF THE HEPATITIS C VIRUS. S. Van Hees (1), P. Michielsen (1), L. Vonghia (1), S. Francque (1), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology and Hepatology.

Introduction : With the recent introduction of direct acting antivirals (DAA), eradication of the Hepatitis C Virus (HCV) has become possible in almost all patients. The effect of DAA's on liver-related manifestations of HCV has been extensively studied, but their effect on extrahepatic manifestations remains unknown.

Aim : We studied the effect of DAA on extrahepatic manifestations of the Hepatitis C Virus.

Methods : This is a retrospective analysis of all DAA treated patients from the Antwerp University Hospital showing extrahepatic manifestations of HCV. Patients were included if they showed extrahepatic manifestations at the time of DAA treatment start. Wilcoxon-rank and chi-square tests were used to test for associations between treatment-related factors and clinical disease improvement of HCV extrahepatic manifestations.

Results : A total of 10 patients were included. They showed either non-Hodgkin B-cell lymphoma (NHL) (n=4), cryoglobulinemia/vasculitis/neuropathy (n=1), arthritis (n=2), or cryoglobuline-associated nephropathy (n=3). Patients were treated with sofosbuvir-daclatasvir±ribavirine (n=5), simeprevir-sofosbuvir (n=1), ledipasvir-sofosbuvir (n=1) or

Peg-interferon-Ribavirine combined with either sofosbuvir (n=2) or simeprevir (n=1). All patients achieved an end-of-treatment response. Four patients had genotype 1a, two genotype 1b, one genotype 2, three genotype 3. Following end-of-treatment response, partial or complete clinical remission of NHL was observed, whereas no clinical improvement was observed in case of vasculitis/neuropathy or nephropathy. Cryoglobulins disappeared from the blood after HCV treatment. Signs of arthritis improved slightly in 1/2 patient. Wilcoxon signed ranks test confirmed that overall end-of-treatment response was not associated with clinical disease improvement (p=0.739). Chi-square test revealed that clinical disease improvement upon end-of-treatment response significantly depended on the type of extrahepatic manifestation (p=0.030). There was no association between HCV genotype and clinical improvement of extrahepatic manifestations after treatment stop (chi-square : p=0.267).

Conclusions : Clinical improvement of HCV-related extrahepatic manifestations after DAA treatment largely depends upon the type of manifestation. Remission was observed in case of HCV-associated NHL and cryoglobulinemia, but not in case of neuro- and nephropathy.

- A43 -

CONTRIBUTION OF HCV RESISTANCE ASSOCIATED MUTATIONS MEASUREMENT TO TREATMENT DECISION MAKING : BLIND OR DOCUMENTED? S. Van Hees (1), S. Pas (2), L. Vonghia (1), P. Michielsen (1), S. Francque (1), T. Vanwolleghem (1). [1] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology and Hepatology, [2] Erasmus Medical Center, Rotterdam, Netherlands (the), Department of Viroscience.

Introduction : The EASL Clinical Practice Guidelines 2016 on Chronic Hepatitis C (CHC) propose resistance screening by deep sequencing prior to start of elbasvir-grazoprevir in gt1a infection and restart of Direct Acting Antivirals (DAA) in case of a previous relapse post DAA treatment. However, screening for mutations using deep sequencing brings a high cost, is not available in every European country and its contribution to therapeutic decision making, remains unsure.

Aim : To determine whether the measurement of resistance associated substitutions (RAS) could change the subsequent antiviral treatment decision.

Methods : This is a retrospective analysis of blood samples from DAA treated patients from the Antwerp University Hospital. Patients were included if they showed relapse after DAA treatment or if they were treated with elbasvir-grazoprevir. Samples were taken before start of respectively retreatment and elbasvir-grazoprevir treatment and were retrospectively screened for mutations in the NS3 and/or NS5A protein by deep sequencing.

Results : A total of 7 patients were included; 6 of whom (5 genotype 1a, 1 genotype 4) showed relapse post treatment with simeprevir-sofosbuvir (n=2), sofosbuvir-daclatasvir (n=3), peg-interferon-ribavirine-daclatasvir (n=1); one patient (genotype 1a) was treated with elbasvir-grazoprevir. Retreatments were started in a blind manner in 5 relapsed patients using a NS3-NS5A class switch approach on a sofosbuvir backbone (using simeprevir for daclatasvir failures and vice-versa) Sustained Viral Response (SVR) at 12 weeks post treatment was achieved in three retreated patients; retreatment was still ongoing in two, but both showed an on-treatment response. Retrospective sequencing analysis learned that 2/3 retreated patients who previously relapsed after daclatasvir treatment showed NS5A RAS (28T or 93C) but not NS3 RAS. The third patient, though SVR12 post-treatment was achieved using simeprevir-sofosbuvir retreatment, showed a NS3 RAS (80K). None of the patients (0/2) who were retreated with simeprevir-daclatasvir, showed NS5A RASes known to have lower daclatasvir susceptibility. One HIV-HCV coinfecting patient with a documented sexually transmitted infection at time of HCV gt 1a relapse had 56 substitutions in the NS5A gene, confirming re-infection rather than relapse. He was not retreated waiting for potential spontaneous clearance. Baseline viral load of the patient started on elbasvir-grazoprevir was 5.76 log IU/ml and did not reveal any NS5A RAS predictive for elbasvir resistance. His treatment regimen is therefore curtailed at 12 weeks instead of 16 weeks.

Conclusions : Measuring Resistance Associated Substitutions pro-actively in patients showing relapse under DAA treatment or patients in which elbasvir-grazoprevir treatment is started, yield results that impact therapeutic decision making in 2 out of 7 patients. However, blind retreatment did not affect final SVR of the patients after retreatment.

BELGIAN NETWORK ON GASTROINTESTINAL MECHANISMS (GIREM)

- B01 -

AN IMPORTANT ROLE FOR SERINE PROTEASES SUCH AS TRYPTASE IN A POST-INFLAMMATORY RAT MODEL FOR VISCERAL HYPERSENSITIVITY. H. Ceuleers (1), J. De Man (2), J. Joossens (3), K. Augustyns (4), S. Francque (5), I. De Meester (6), B. De Winter (2). [1] University of Antwerp, Antwerp, Belgium, Laboratory of Experimental Medicine and Pediatrics, Division of Gastroenterology, [2] University of Antwerp, Antwerp, Belgium, Laboratory of Experimental Medicine and Pediatrics - division Gastroenterology and Hepatology, [3] University of Antwerp, Antwerp, Belgium, Research affairs and innovation - valorisation, [4] University of Antwerp, Antwerp, Belgium, Laboratory of Medicinal Chemistry, [5] Antwerp University Hospital, Edegem, Belgium, Gastroenterology & hepatology, [6] University of Antwerp, Antwerp, Belgium, Laboratory of Medical Biochemistry.

Introduction : Serine proteases such as tryptase are thought to play an important role in the pathogenesis of irritable bowel syndrome (IBS). Previously, we showed a beneficial effect of nafamostat mesilate (a non-selective serine protease inhibitor) on visceral pain. However, the exact nature of the serine proteases implicated in visceral hypersensitivity needs further study.

Aim : The aims of this study were (1) to quantify tryptase expression in the colon of post-inflammatory rats at protein and mRNA level and (2) to investigate the effect of a novel irreversible serine protease inhibitor UAMC-1162 (patent WO2007045496 (A1), Joossens et al., J. Med. Chem. 2007).

Methods : Male Sprague-Dawley rats (200-225g) received an intrarectal administration with 2,4,6-trinitrobenzenesulfonic acid (TNBS) or 0.9% NaCl on day 0. Subsequently, a colonoscopy was performed on day 3 to confirm the presence of colitis and from day 10 onwards every 4 days until complete healing of the mucosa was observed. Afterwards, visceral sensitivity was quantified in all animals using the visceromotor responses (VMRs) to colorectal distension. The serine protease inhibitor UAMC-1162 (1-2.5mg/kg) or vehicle was injected intraperitoneally (ip), 30 min prior to the VMR experiment. Furthermore, colonic compliance was assessed to investigate the elastic properties of the colon. Finally, the inflammatory parameters (colonoscopy, macroscopy, microscopy, MPO activity) were scored to confirm the post-inflammatory status of the rats. Upon sacrifice, colonic samples were collected and used to quantify mast cell tryptase by immunohistochemistry. Furthermore, colon and DRG L1-6 were sampled to determine the mRNA expression of tryptase, matriptase, KLK4, KLK8 and uPA by means of qPCR.

Results : All TNBS rats demonstrated a significantly higher colonoscopic score at day 3 compared to controls (6.9 ± 0.5 vs 0.0 ± 0.0 $n=8$ each; $p < 0.05$). At the day of the VMR, the post-inflammatory status of all TNBS rats was confirmed (day 13-21, data not shown). Mucosal tryptase expression was significantly increased in the colon of post-colitis compared to control rats (% positive area mucosa : 0.46 ± 0.07 vs 0.15 ± 0.02 $n=9$ each; $p < 0.05$). The mRNA expression of tryptase was significantly increased in post-colitis rats compared to controls at the colonic level (rel mRNA expression : 4.6 ± 1.1 vs 1.3 ± 0.4 ; $p < 0.05$). We could not detect significant differences for the other proteases at the level of the colon or DRGs (data not shown). The vehicle-treated post-colitis rats showed significantly higher VMRs compared to controls, indicating visceral hypersensitivity (total AUC : 2134 ± 124 vs $1099 \pm 80 \mu V$ $n=8$; $p < 0.05$). UAMC-1162 (1-2.5 mg/kg) dose-dependently reversed visceral hypersensitivity, achieving complete reversal with the highest dose used (total AUC : 1066 ± 78 vs 2134 ± 124 ; $n=8$ each; $p < 0.05$). UAMC-1162 significantly decreased visceral sensitivity in controls at 40 mmHg, without affecting VMRs at other pressures (data not shown). Colonic compliance was not affected in control nor post-colitis rats upon the administration of UAMC-1162.

Conclusions : Our results strongly support an important role for colonic serine proteases and more precisely for tryptase in post-inflammatory visceral hypersensitivity. Regarding further research into serine protease inhibitors as a possible treatment strategy for visceral pain in IBS patients, a compound with a high specificity for tryptase seems mandatory.

- B02 -

EVIDENCE FOR TRP CHANNEL SENSITIZATION IN IBS WITH HISTAMINE 1 RECEPTOR ANTAGONISM AS EFFECTIVE TREATMENT. D. Balemans (1), J. Aguilera-Lizarraga (1), M. Florens (1), S. Theofanous (1), E. Perna (1), S. Van Der Merwe (2), M. Wouters (1), G. Boeckxstaens (1) / [1] KU, Leuven, Belgium, TARGID, [2] KU, Leuven, Belgium, Department of Gastroenterology & Hepatology.

Introduction : Increased abdominal pain perception or visceral hypersensitivity (VHS) is the hallmark of irritable bowel syndrome (IBS). We previously showed histamine-1-receptor (Hrh1)-mediated sensitization of TRPV1, TRPA1 and TRPV4 in patients with IBS.

Aim : In the present study, we investigated the prevalence of TRP channel sensitization and its correlation with symptoms in IBS patients versus healthy volunteers (HV). In addition, we evaluated the effect of treatment with the Hrh1 antagonist ebastine on TRP channel sensitization and symptom scores in IBS patients.

Methods : 25 HV (11M, 28 yrs IQR [23-53]) and 34 IBS patients (9M, 35 yrs IQR [25-53]) were invited to provide rectal biopsies for live Ca²⁺ imaging. Submucosal neurons were loaded with Fluo-4 and their response to the TRP channel agonists capsaicin (1nM, TRPV1), cinnamaldehyde (1 μ M, TRPA1) and/or GSK1016790A (1nM, TRPV4) was assessed. Thereafter, a subgroup of IBS patients (n=22) was treated (open label) with ebastine (20-40 mg) and invited to provide rectal biopsies for live Ca²⁺ imaging after 8-12 weeks of treatment. Finally, all subjects filled out validated symptom questionnaires (ROME III, GSRS and numeric scale for pain (0 : no pain, 10 : worst possible)). Correlations between symptoms and neuronal data were calculated using Spearman correlations and Fishers exact tests.

Results : Sensitization of TRPV1, TRPA1 or TRPV4 was significantly more prevalent in rectal submucosal neurons of IBS patients compared to those of HV (Fig. 1A). TRP channel sensitization of one or more TRP channels was detected in 68% of IBS patients compared to 7% in HV (p<0.001, Fishers exact test). 60% of IBS patients with positive symptom scores for pain, bloating, urgency, flatulence and diarrhea had sensitized TRP channels compared to 30% of patients with normal TRP sensitivity. Within the IBS group, no significant correlation was detected between the response of submucosal neurons and abdominal symptoms, including pain, urgency, bloating, flatulence and diarrhea. Ebastine however significantly improved IBS symptoms resulting in 63% pain responders (>30% reduction in pain score). Of note, patients responding to ebastine had a normalization of their neuronal Ca²⁺ response to capsaicin, cinnamaldehyde or GSK1016790A (n=3).

Conclusions : Our data demonstrate that sensitization of submucosal neuronal TRP channels is prevalent and can be detected in up to 68% of IBS patients. Treatment with the Hrh1 antagonist ebastine improved symptoms, a finding that was associated with normalization of neuronal responsiveness to TRP channel activation. These findings indicate 1. that sensitization of submucosal neurons could be an interesting biomarker and 2. that pronociceptive changes in the gut wall involving Hrh1 receptors represent a major pathophysiological mechanism in IBS and thus an interesting target for treatment.

- B03 -

FOOD ANTIGEN-SPECIFIC ANTIBODIES AND MAST CELL ACTIVATION IN POST-INFECTIOUS VISCERAL HYPERSENSITIVITY. M. Florens (1), J. Aguilera-Lizarraga (2), S. Theofanous (2), G. Bosmans (2), D. Balemans (2), E. Perna (2), H. Rodewald (3), M. Wouters (2), G. Boeckxstaens (2). [1] KU, Leuven, Belgium, TARGID, [2] KU, Leuven, Belgium, Clinical and Experimental Medicine, [3] German Cancer Research Center (DKFZ), Heidelberg, Germany, Division for Cellular Immunology.

Introduction : Infectious gastroenteritis is a risk factor to develop irritable bowel syndrome (IBS) but the exact mechanism involved remains unclear. We previously showed that mice infected with *Citrobacter rodentium* develop a bystander immune response against ovalbumin (OVA) during infection. Re-exposure to OVA resulted in visceral hypersensitivity (VHS) and increased colonic permeability.

Aim : Here, we investigated the hypothesis that an adaptive immune response generating OVA-specific antibodies leads to mast cell activation, VHS and increased colonic permeability. In addition, as we previously showed improvement of IBS patients by treatment with the histamine 1 receptor antagonist ebastine, we further evaluated the role of this receptor in our model of post-infectious IBS.

Methods : Balb/C mice (n=6-10/group) were infected with *C. rodentium* in the presence of OVA and re-exposed to OVA 5-7 weeks post-infection. Experiments were performed in mast cell deficient mice (Cpa3-Cre/WT), histamine 1 receptor knock-out mice (Hrh1 KO) and mice treated with the histamine 1 receptor antagonist pyrilamine (5mg/kg) or saline before re-exposure to OVA. Development of VHS was assessed with electromyography recordings during colorectal distention before and at 2, 4, 6 and 7 weeks post-infection. Mice were considered VHS when the area under the curve was >5.04 (95th percentile). After sacrifice, colons were collected to study mucosal permeability in Ussing chambers. Finally, to assess the role of antigen-specific antibody-induced mast cell activation, OVA-specific IgE, IgG1 and IgG2a were measured in serum and homogenized colonic tissue using ELISA.

Results : Infection with *C. rodentium* induced VHS in all groups of mice at 2 weeks post-infection, returning to normal after 4-6 weeks. VHS was re-installed by OVA re-exposure in WT mice but not in Cpa3-Cre and Hrh1 KO mice (p<0.05 and p<0.001 respectively, 2-way ANOVA + multiple comparisons test). In line, pyrilamine reversed VHS while mice treated with saline remained hypersensitive (p<0.01, 2-way ANOVA + multiple comparisons test). Colonic mucosal permeability was increased in infected WT mice re-exposed to OVA but not in Cpa3-Cre and Hrh1KO mice (data not shown). Of note, colonic IgE and IgG2a but not IgG1 antibodies were significantly increased (p<0.001 and p<0.0001 respectively, non-parametric Mann Whitney tests) in infected compared to non-infected mice exposed to OVA while serum levels were comparable in both groups (IgE below detection limit).

Conclusions : Infection with *C. rodentium* triggers an adaptive immune response to OVA with generation of increased levels of local OVA-specific IgG2a and IgE antibodies, most likely leading to mast cell activation upon re-exposure to OVA and histamine 1 receptor-mediated VHS. These data suggest that an adaptive immune response to food antigens may be involved in the pathogenesis of post-infectious IBS and provide evidence for treatment of post-infectious IBS with histamine 1 receptor antagonists.

MAST CELLS MEDIATE STAPHYLOCOCCAL ENTEROTOXIN B-TRIGGERED VISCERAL HYPERSENSITIVITY : POTENTIAL LINK BETWEEN SUPERANTIGENS AND IRRITABLE BOWEL SYNDROME (IBS). J. Aguilera-Lizarraga (1), M. Florens (1), D. Balemans (1), S. Theofanous (1), E. Perna (1), H. Rodewald (2), M. Wouters (1), G. Boeckxstaens (1). [1] KU, Leuven, Belgium, Clinical and Experimental Medicine, [2] German Cancer Research Center (DKFZ), Heidelberg, Germany, Cellular Immunology.

Introduction : Superantigens trigger an aberrant immune response by a polyclonal activation of T-cells, and are mainly produced by *Staphylococcus aureus* and *Streptococcus pyogenes*. Previously, we showed that in mice, similar to an infection with *Citrobacter rodentium*, Staphylococcal enterotoxin B (SEB) administered with ovalbumin (OVA) resulted in visceral hypersensitivity upon re-exposure to OVA, indicating the involvement of an aberrant bystander immune response to OVA.

Aim : In the present study, we wanted to further explore the role of mast cells, B and T cells in SEB-induced visceral hypersensitivity, and investigate to what extent *S. aureus*, *S. pyogenes* and their superantigens are present in fecal samples of IBS patients.

Methods : Wild type (WT) Balb/C, mast cell deficient CPA3-Cre, T and B cell deficient SCID and T cell deficient nude mice received SEB in the presence of OVA for 3 consecutive days. 5 weeks later, mice were re-exposed to OVA by oral gavage every other day. Visceral pain was assessed prior to SEB/OVA administration (baseline) and after 4 OVA challenges by recording of the visceromotor response to colorectal distension using abdominal muscle electromyography. SCID and nude control groups did not receive SEB or OVA. To evaluate the presence of *S. aureus* and *S. pyogenes*, fecal samples of 84 well-characterized IBS patients (66 females, 38 years IQR [25-50]) and 64 healthy subjects (35 females, 49 years IQR [32-58]) were collected and DNA was extracted. *S. aureus* and *S. pyogenes* (*nuc* and *spy1258* genes, respectively) and genes encoding superantigens (*sea*, *seb*, *sec*, *sed*, *seg*, *seh*, *sell*, *selk*, *selm*, *selo*, *selp*, *selq*) were assessed by qPCR.

Results : Balb/C WT mice receiving SEB in the presence of OVA developed visceral hypersensitivity upon OVA re-exposure, however no increase in pain perception was observed in mast cell deficient mice (CPA3-Cre) (Mann Whitney test, $p < 0.01$). SCID and nude mice receiving SEB/OVA did not develop increased visceromotor response upon OVA re-exposure compared to control mice. 19 of the 84 fecal samples of IBS patients (23%) were positive for *S. aureus*, compared with 6 of the 64 healthy subjects (9%) (Fisher's exact test, $p < 0.05$). 9 of the 19 positive IBS samples were positive for one or more superantigen (47%) compared to 1 of the 6 of the healthy subject (17%). No samples were positive for *S. pyogenes*.

Conclusions : SEB-induced visceral hypersensitivity in mice re-exposed to OVA is mediated by T and B cells indicating the involvement of an adaptive immune response to OVA, most likely leading to antigen-specific activation of mast cells. Of note, *Staphylococcus aureus*, including superantigen-producing strains, are more frequently present in fecal samples of IBS patients compared to healthy controls, suggesting that similar to a bacterial infection, superantigens may be involved in installing visceral hypersensitivity in IBS.

EXPRESSION OF A NOVEL FAMILY OF PAIN-RELATED G PROTEIN-COUPLED RECEPTORS IN THE HUMAN INTESTINE : A COMPARISON BETWEEN HEALTHY AND IRRITABLE BOWEL SYNDROME-AFFECTED ILEUM. S. Van Remoortel (1), R. Arora (2), H. De Schepper (3), J. Timmermans (2), R. Buckinx (2). [1] University of Antwerp, Antwerpen, Belgium, Veterinary Sciences, [2] University of Antwerp, Antwerp, Belgium, Veterinary Sciences, [3] Antwerp University Hospital, Edegem, Belgium, Gastroenterology and Hepatology.

Introduction : G protein-coupled receptors (GPCR's) are leading targets in drug discovery because of their extensive regulatory functions and profound molecular diversity. In this respect, Mas-related G protein-coupled receptors (MRGPR) are a promising family of GPCR's that are involved in peripheral pain perception and mast cell physiology. Our previous results show that the expression of multiple murine MRGPR members in enteric neurons and mucosal mast cells markedly changes during intestinal inflammation. This suggests a role in GI neuro-immune communication, which is involved in both pathological and functional gastro-intestinal (GI) disorders (Avula et al. 2011; 2013).

Aim : Because nothing is known about MRGPR expression in the human GI tract, our aim was to characterize their expression in mucosal biopsies and evaluate expressional changes in the GI tract of patients with irritable bowel syndrome (IBS).

Methods : Mucosal biopsies from the terminal ileum were collected during colonoscopy and stored in the RNA stabilizer RNAlater® until further processing. Individuals were categorized as healthy control (n=10) or IBS (n= 18) according to the Rome III criteria. We selected and evaluated a panel of five genes as candidate reference genes, in compliance with MIQE qPCR quality standards (Bustin et al., 2009), and then determined the expression for MRGPRD, MRGPRE, MRGPRF, MRGPRX1-4 and MAS1L.

Results : GeNorm analysis (Vandesompele et al. 2002) showed that reference genes HPRT1 and PPIA, with a combined M-value of 0.255, are expressed most stable in the mucosal biopsies. Among the target genes, MRGPRD, MRGPRES, MRGPRF, MRGPRX2 and MAS1L are clearly expressed in the GI mucosa, while the other target genes are not expressed. Moreover, MRGPRD and MRGPRES expression showed a trend towards downregulation in IBS individuals compared to healthy controls, with less variability between patient samples. This trend is mainly attributed by a reduced expression of MRGPRD and MRGPRES in the IBS-C subtype.

Conclusions : From a selected panel of candidate reference genes, we show that HPRT1 and PPIA are most suitable to use as reference genes for expression normalization in mucosal biopsies from the terminal ileum. We provide first evidence for the expression of multiple MRGPR members in the human ileum, which warrants further work on the (sub) cellular localization of these receptors. Moreover, the plasticity of MRGPR expression suggests a possible role for these receptors in GI (patho)physiology.

- B07 -

PHOSPHODIESTERASE 3A IS MORE THAN AN ICC MARKER. P. Vandenberghe (1), P. Hague (1), C. Erneux (2), J. Vanderwinden (1). [1] ULB Faculty of Medicine, Anderlecht, Belgium, Laboratory of Neurophysiology, [2] ULB Faculty of Medicine, Anderlecht, Belgium, IRIBHM.

Introduction : The cGMP inhibited cAMP phosphodiesterase 3A (PDE3A) downregulates the levels of cyclic nucleotides and thus controls biological responses in several tissues and cell types. We have previously shown that PDE3A is a marker for the interstitial cells of Cajal (ICC) in adult mouse gut (Thys et al., 2015) and that its expression is upregulated in the mouse KITK641E GIST model (Gromova et al., 2009). However, little is known about the role and expression of PDE3A during mouse gut development.

Aim : To unravel the temporal expression profile of PDE3A during mouse gut development and to assess the ICC phenotype in PDE3A deficient transgenic mice (Masciarelli et al., 2004).

Methods : Small intestine, colon and stomach from embryos (E12.5, E14.5 and E17.5) and young wild type mice (P2 and P24) were fixed with paraformaldehyde, cryopreserved in graded sucrose solutions, embedded in OCT and cut on a cryostat. Fixed tissues from adult PDE3A^{-/-} mice were kindly provided by Dr. Steven Hockman, National Heart, Lung and Blood Institute, Bethesda, MD, USA. Immunofluorescence was performed using specific antibodies for PDE3A, Kit and smooth muscle actin (α -SMA). Embryonic and postnatal samples were imaged by confocal microscopy. ICC quantification was performed on the whole tissue circumference as described previously (Thys et al., 2015).

Results : Our observation showed that PDE3A expression in the gut appeared as early as E14.5, when mesenchymal cells begin to differentiate. PDE3A expression was present in both ICC and longitudinal smooth muscle cells until the postnatal period. Later, PDE3A expression persisted in ICC only. We also observed a significant decreased ICC density in adult PDE3A^{-/-} mouse antrum and colon compared to WT littermates.

Conclusions : PDE3A is expressed early during development of the gut mesenchyme in precursor cells forming the future longitudinal muscle layer and ICC-MP. Its expression changed over time while ICC/SMC differentiation occurs. Furthermore, adult mice lacking PDE3A expression exhibit a reduced density of KIT⁺ ICC along the gut without other gross anomaly. Taken altogether, our observations suggest that PDE3A plays an important role in the development of the KIT⁺ ICC lineage during differentiation of the gut mesenchyme and in mature KIT⁺ ICC.

- B08 -

INFLUENCE OF PHOSPHODIESTERASES ON BASAL AND 5-HT₄ RECEPTOR-FACILITATED CHOLINERGIC CONTRACTILITY IN THE MURINE GASTROINTESTINAL TRACT. V. Pauwelyn (1), E. Van Deynse (1), R. Lefebvre (1). [1] Ghent University, Ghent, Belgium, Department of Pharmacology - Heymans Institute.

Introduction : The 5-HT₄ receptor, a G-protein coupled receptor linked to adenylate cyclase and cAMP, is present on cholinergic neurons innervating gastrointestinal (GI) smooth muscle cells in several species including mouse, pig and man. Activation of these receptors with a selective 5-HT₄ receptor agonist, such as prucalopride, generates cAMP which facilitates the ongoing acetylcholine release resulting in increased smooth muscle contraction. In porcine stomach and colon the facilitating effect of prucalopride is enhanced by selective phosphodiesterase (PDE) 4 inhibition, indicating that the signalling pathway of the 5-HT₄ receptor within the cholinergic neurons is controlled by PDE4. At the level of the smooth muscle cell, cAMP induces relaxation and its turnover is mainly controlled by PDE3 in the porcine GI tract. The gastroprokinetic effect of prucalopride could thus be enhanced by combining it with a selective PDE4 inhibitor, without counteracting interference at the muscular level.

Aim : The aim of the present study was to investigate the influence of cAMP-metabolizing PDEs in the circular smooth muscle activity and in the signal transduction pathway of the 5-HT₄ receptors on the cholinergic neurons of the murine fundus, jejunum and colon.

Methods : Circular smooth muscle strips from murine fundus, jejunum and colon were mounted at optimal load for isometric tension recording. Submaximal cholinergic contractions were repetitively induced by either activating the cholinergic neurons with electrical field stimulation (EFS) in the presence of guanethidine, L-NAME and for colon also MRS 2500, or by directly activating the smooth muscle cells with the muscarinic receptor agonist carbachol. The influence of the PDE inhibitors IBMX (non-selective), vinpocetine (PDE1), EHNA (PDE2), cilostamide (PDE3) and rolipram (PDE4) was tested on (1) carbachol-induced contractions (to investigate the involvement of PDEs in the cAMP turnover in the smooth muscle cell), (2) EFS-induced contractions (to estimate the effect of the PDE inhibitors on EFS-induced contractions necessary for the selection of the concentrations used in the third set of experiments), and (3) on the facilitating effect of prucalopride on EFS-induced contractions (to check whether the 5-HT₄ receptor signaling pathway is controlled by one or more PDEs).

Results : In the 3 tissues IBMX and cilostamide concentration-dependently inhibited the carbachol-induced cholinergic contractions when compared to their corresponding solvent. This suggests that at the level of the smooth muscle PDE3 contributes to the cyclic nucleotide turnover. The latter was confirmed in the experiments with EFS where, in comparison to their corresponding solvent, IBMX and cilostamide again concentration-dependently decreased the EFS-induced contractions in all tissue types. Rolipram mildly reduced carbachol-induced contractions in the 3 tissue types and EFS-induced contractions only in colonic tissue; some contribution of PDE4 in the cAMP turnover in the smooth muscle cells can thus not be excluded. Based on the effects in the experiments with EFS, one concentration for each PDE inhibitor was selected to study its influence on the facilitating effect of prucalopride on EFS-induced cholinergic contractions (for PDE inhibitors that reduced the EFS-induced contractions, a concentration inducing maximally 35 % reduction; for vinpocetine and EHNA 10 μ M). In this concentration, none of the PDE inhibitors significantly enhanced the effect of a submaximal concentration of prucalopride. This suggests that the 5-HT₄ receptor signalling pathway in the cholinergic neurons of the murine GI tract is not controlled by PDEs.

Conclusions : In murine GI smooth muscle cells PDE3, preferentially metabolising cAMP, contributes to the cyclic nucleotide turnover, with a supportive role for the cAMP specific PDE4. In contrast to the porcine GI tract, there is no evidence for a PDE-mediated control of the 5-HT₄ receptor pathway in myenteric cholinergic neurons in the murine GI tract. This murine model is thus not suitable for in vivo testing of the combination of a 5-HT₄ receptor agonist with a PDE inhibitor.

- B09 -

VAGUS NERVE STIMULATION AND PRUCALOPRIDE HAVE ANTI-INFLAMMATORY PROPERTIES AND IMPROVE POSTOPERATIVE ILEUS IN HUMAN. N. Stakenborg (1), P. Gomez-Pinilla (1), R. Aerts (2), I. Appeltans (1), K. Van Beek (1), A. Wolthuis (2), A. D'hoore (2), G. Bosmans (1), E. Labeeuw (1), S. Verheijden (1), G. Matteoli (1), G. Boeckxstaens (1). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), KULeuven, Leuven, Belgium, Clical and Experimental Medicine, [2] Abdominal Surgery, University Hospital of Leuven, Belgium, Abdominal Surgery.

Introduction : We previously showed in a model of postoperative ileus (POI) that activation of the anti-inflammatory pathway by vagus nerve stimulation (VNS) or administration of the 5-HT₄ receptor agonist prucalopride prior to surgery inhibits surgery-induced intestinal inflammation and improves gastrointestinal (GI) transit, an effect mediated by inhibition of resident muscularis macrophages.

Aim : In this study, we evaluated if also in human VNS or prucalopride administered prior to surgery has anti-inflammatory properties and improves POI.

Methods : Thirty patients (50% male, 64 \pm 12 yrs) undergoing pancreaticoduodenectomy for carcinoma were included in a randomized double-blind pilot study comparing the effect of sham/placebo (n=10), stimulation of the abdominal vagus nerve (n=10; 20 Hz, 1 ms) and prucalopride (n=10; 2x2 mg prior to surgery). The release of IL6, IL8 and TNF α was measured in LPS-stimulated whole blood taken prior to and 24 hours after surgery. Duodenal full thickness samples were taken at the beginning (t=0h) and end of surgery (t=2h) to assess the degree of inflammation. Clinical recovery was assessed by nasogastric tube (NGT) output at day 3, time to removal of NGT, first solid food and defecation and length of hospital stay (LOS). Differences between groups were determined by 1-way ANOVA with post-hoc correction. Differences in time and groups were analyzed by repeated 2-way ANOVA with Bonferonni correction.

Results : IL8 (p<0.05), IL6 (p<0.05) and TNF α (p<0.05) levels of LPS stimulated whole blood (t=24h postop) were reduced in prucalopride-treated patients compared to sham/placebo. VNS decreased the production of IL8 (p<0.05). IL6, IL8 and TNF were upregulated in the muscularis of the 3 groups at t=2h compared to t=0h. Of note, IL6 expression was reduced in patients treated with prucalopride (0.7 \pm 0.2; p<0.01) or VNS (1.0 \pm 0.2; p<0.05) compared to those treated with sham/placebo (1.8 \pm 0.4), while IL8 expression tended to be decreased in the prucalopride group. Five patients developed a surgical site-specific complication (e.g. abdominal infection); 1 in the sham/placebo, 2 in the VNS and 2 in the prucalopride group, and were excluded from further analysis. Prucalopride improved recovery compared to sham/placebo, as shown by a reduction in time to removal of NGT (p<0.05), time to first solids (p<0.01) and an overall reduced LOS.

Conclusions : VNS and prucalopride have anti-inflammatory properties, as shown by reduced IL6 expression in the muscularis and a reduction in whole blood cytokine release. Of note, pre-operative administration of prucalopride significantly improved clinical recovery compared to sham/placebo treatment. Based on these findings, we propose that patients should be pre-treated with prucalopride to prevent surgery-induced intestinal inflammation and thus reduce POI, but larger clinical trials are required to confirm these results.

- B10 -

DIMETHYL FUMARATE IMPROVES MURINE POSTOPERATIVE ILEUS INDEPENDENTLY OF HEME OXYGENASE-1. J. Van Dingenen (1), R. Lefebvre (1). [1] Ghent University, Ghent, Belgium, Department of Pharmacology - Heymans Institute.

Introduction : Heme oxygenase (HO) catalyzes the degradation of heme into ferrous iron, carbon monoxide (CO) and biliverdin, which is subsequently reduced to bilirubin; bilirubin and CO have anti-oxidative and anti-inflammatory properties. The inducible isoform HO-1 is expressed in reaction to oxidative stress and inflammation, contributing to tissue protection in these conditions. Postoperative ileus (POI), the impairment of gastrointestinal motility after abdominal surgery, is mainly due to intestinal muscular inflammation triggered by surgical handling. It was already shown that CO-releasing compounds exert an anti-inflammatory effect in murine POI partially through induction of HO-1. Dimethyl fumarate (DMF) is currently used to treat patients with multiple sclerosis or psoriasis. In vitro it is a very effective inducer of HO-1 and preclinical studies suggest that its immunosuppressive and neuroprotective effects are related to induction of HO-1.

Aim : The aim of this study was therefore to investigate the effect of DMF on the intestinal inflammation and on the delay in gastrointestinal transit caused by POI.

Methods : C57Bl6J mice were anesthetized (isoflurane) and after laparotomy, POI was induced by compressing the small intestine with cotton applicators (intestinal manipulation; IM) for 5 min. DMF was administered intragastrically (100 mg/kg) via gavage or intraperitoneally (30 mg/kg) 24 h before IM. Intestinal transit was assessed 24 h postoperatively using fluorescent imaging 90 min after fluorescein gavage (geometric centre [GC] of intestinal fluorescein progression). The small intestine was divided in 6 equal parts; mucosa-free muscularis segments were prepared and stored at -80° C for later analysis of myeloperoxidase (MPO) activity as an index of leukocyte infiltration, of the inflammatory cytokine interleukin 6 (IL-6) and of HO-1 protein expression. IL-6 and HO-1 were assayed by enzyme immunoassay and MPO by spectrophotometric monitoring.

Results : Pre-treatment with DMF via both oral (GC : 7.4 ± 0.4 ; mean \pm s.e.m. of $n = 6$) and intraperitoneal (GC : 7.7 ± 0.9) administration prevented the delayed transit seen after IM (GC : 4.6 ± 0.7 ; in controls 7.5 ± 0.3). Concomitantly peroral and intraperitoneal DMF significantly reduced the increased IL-6 levels in the intestinal muscularis caused by POI. However, it only reduced the elevated leukocyte infiltration (MPO) significantly when administered intraperitoneally. Furthermore, IM per se caused a significant increase in intestinal HO-1 protein expression as previously shown; this effect was not magnified by pre-treatment with DMF. Likewise, 12 h and 24 h after administration of DMF in non-operated animals, no increase in HO-1 levels was measured.

Conclusions : The present study indicates that pre-treatment with DMF prevents delayed intestinal transit and reduces inflammation upon IM independently of intestinal HO-1 induction. To further investigate the possible mechanism of action of DMF in POI, the role of the nuclear factor (erythroid-derived 2)-like 2 (Nrf2)-antioxidant response element signaling pathway and of the Nrf2-independent inhibition of the proinflammatory NF-kappaB pathway will be studied.

- B12 -

EFFECT OF OBESITY ON THE BITTER AND SWEET CHEMOSENSORY SIGNALLING PATHWAYS THAT REGULATE GHRELIN RELEASE IN THE HUMAN GUT. Q. Wang (1), E. Deloose (2), L. Vancleef (1), E. Canovai (3), L. Ceulemans (3), S. Steensels (1), R. Farré Marti (4), J. Tack (2), J. Pirenne (3), M. Lannoo (5), I. Depoortere (1). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), KULeuven, Leuven, Belgium, Gut Peptide Research Lab, [2] Translational Research Center for Gastrointestinal Disorders (TARGID), KULeuven, Leuven, Belgium, GI motility and sensitivity research group, [3] Abdominal Transplant Surgery, University Hospitals Leuven, Leuven, Belgium, University hospital of Leuven, [4] Translational Research Center for Gastrointestinal Disorders (TARGID), KULeuven, Leuven, Belgium, laboratory of digestion and absorption, [5] Abdominal Surgery, University Hospital of Leuven, Belgium, University hospital of Leuven.

Introduction : In health, extra-oral taste receptors on enteroendocrine cells sense nutrients and transmit signals to control the secretion of gut hormones involved in appetite regulation. In disease, disturbances or adaptations in the expression and sensitivity of these receptors may affect metabolism. In obese patients levels of the hunger hormone ghrelin are lower and food fails to suppress plasma ghrelin levels.

Aim : This study aimed to investigate the effect of obesity on 1) the expression of bitter and sweet taste chemosensory signalling pathways in the human gut and 2) on bitter and sweet-induced ghrelin release in ex-vivo gut segments/isolated crypts.

Methods : Gastric and small intestinal tissue was collected from lean brain-death organ donors (BMI : 24.5±1.0 kg/m²) (fundus, n=6 and distal duodenum, n=5), and from obese patients (BMI : 41.2±1.1 kg/m²) who underwent sleeve gastrectomy (fundus, n=6) or Roux-en-Y gastric bypass surgery (proximal jejunum, n=7), respectively. Mucosal segments or isolated crypts were incubated for 2 hours in Krebs-Ringer buffer (11mM glucose) with denatonium benzoate (DB) (1-20 mM), phenylthiocarbamide (PTC) (1-10 mM), chloroquine (1-5 mM) or glucose (25-200 mM). Octanoyl ghrelin levels were determined by radioimmunoassay in the tissue culture supernatant and corrected for tissue weight (segments) or ghrelin cell content (crypts). The expression of the bitter taste receptors TAS2R4, TAS2R10, TAS2R3 and TAS2R38, sweet taste receptor subunit TAS1R3, sodium dependent glucose transporter 1 (SGLT1) and the glucose transporter 2 (GLUT2) was demonstrated by real-time PCR.

Results : In both lean and obese subjects, basal ghrelin release in mucosal segments from the fundus was respectively 6.1 fold and 2.3 fold higher than in the small intestine. In obese patients, basal ghrelin release was 3.3 fold lower in the fundus but not in the small intestine compared to lean subjects. Bitter taste receptors TAS2R4 (DB), TAS2R10 (DB), and TAS2R3 (chloroquine) were expressed in mucosal tissue from the fundus and small intestine, while TAS2R38 (PTC) was only present in the small intestine. Obesity down-regulated the expression of TAS2R4 and TAS2R3 in a region-independent manner. In lean subjects, DB increased ghrelin release in a dose-dependent manner (overall P<0.01) with maximal effects at 5mM in the fundus (20.3±10.5 (basal) vs 35.0±18.0 (DB) pg/mg tissue) and in the small intestine (3.2±1.1 vs 7.7±2.8 pg/mg tissue). In obese subjects, DB was as effective as in lean subjects to increase ghrelin release in the fundus (overall P<0.05) (7.8±4.3 vs 10.5±5.9 pg/mg tissue at 5mM). In contrast, small intestinal mucosal segments from obese patients did not respond to DB. In agreement with the receptor expression, both DB (5 mM) and chloroquine (5 mM) but not PTC (10 mM) increased ghrelin release in isolated crypts from the fundus. Obesity tended to down-regulate the expression of the sweet taste receptor subunit TAS1R3, and up-regulated the expression of SGLT1 (3.5 fold) and GLUT2 (3.8 fold) in the small intestine. Glucose (50 mM) decreased octanoyl ghrelin secretion in the fundus of lean (19.8±9.0 vs 13.2±6.5 pg/mg tissue (P<0.01)) and obese subjects (4.9±2.5 vs 3.2±1.7 pg/mg tissue (P<0.01)) to the same extent. Similar effects were observed in the small intestine.

Conclusions : Bitter and sweet tastants have an opposite effect on ghrelin release in the human gut. The expression of bitter and sweet chemosensory signalling pathways in the gut is altered in obese patients. Obesity impairs ghrelin release but does not change the effect of glucose on ghrelin release, although bitter-induced ghrelin release is less effective in the small intestine.

- B13 -

EFFECT OF DIET-INDUCED OBESITY ON SEROTONERGIC ENTERIC NEURONS AND INTESTINAL TRANSMIT IN THE ZEBRAFISH. L. Uyttebroek (1), S. Van Remoortel (2), L. Jongman (1), V. Wijtvliet (1), B. De Winter (3), J. Timmermans (2), G. Hubens (1), L. Van Nassauw (1). [1] Universiteit Antwerpen, Antwerpen, Belgium, Laboratory Human Anatomy and Embryology, dept. ASTARC, Faculty of Medicine and Health Sciences, [2] Universiteit Antwerpen, Antwerpen, Belgium, Laboratory of Cell Biology and Histology, dept. Veterinary Sciences, Faculty of Pharmaceutical, Biomedical and Veterinary Sciences, [3] Universiteit Antwerpen, Antwerpen, Belgium, Laboratory of Experimental Medicine and Pediatrics, Faculty of Medicine and Health Sciences.

Introduction : Obesity is a worldwide epidemic and a major risk factor for numerous diseases including cardiac failure, diabetes and cancer. The regulation of feeding behavior and body weight depends on a wide range of neuronal pathways influencing satiety and hunger. Serotonin (5-HT) is one of those players identified to have a profound effect on energy homeostasis and correlates positively with body weight. Furthermore, an altered expression of 5-HT receptor subtypes, including the 5-HT₄ receptor, has been reported in the central nervous system of obese patients. In the enteric nervous system (ENS), 5-HT initiates peristalsis and is involved in secretion. The effect of obesity on the 5-HT metabolism in the intestine and its underlying mechanisms still needs to be further elaborated.

Aim : The aim of the present study was to investigate the effect of diet-induced obesity (DIO) on enteric 5-HT expression and on GI transit in the zebrafish.

Methods : Zebrafish were kept in small colonies and fed either a high caloric diet (DIO : 150 cal/day/fish) or a normal diet (ND : 20 cal/day/fish) for 4 weeks. The proportion of serotonergic neurons in the GI tract was analyzed using a multiple immunofluorescence staining method and antibodies directed against 5-HT and the pan-neuronal marker, HuC/D. In addition, quantitative PCR (qPCR) was performed on homogenates of brain and intestine to analyze the expression of tryptophan hydroxylase (TPH) 1a, 1b and 2, the rate limiting enzymes in 5-HT metabolism, and the 5-HT₄ receptor. GI transit was measured by gavaging glass beads into the proximal intestine and calculating the geometric centre (GC) after 5 hours.

Results : After 4 weeks, the body mass index (g/cm²) of DIO fish was significantly increased. Overfeeding increased the proportion of serotonergic neurons in the proximal and first part of the mid intestine. qPCR revealed significant elevated

levels for TPH2 in brain and intestine, but not for TPH1a/b. Furthermore, a significant increase in the expression of the 5-HT4 receptor was observed in brain, but not in the intestine. Preliminary experiments showed an increase in the GC after 5 hours in DIO fish compared to ND.

Conclusions : In the present study, analysis of the GI tract of DIO fish revealed an increase of 5-HT expression in enteric neurons in the proximal part of the intestine, which is probably due to an increased TPH2 expression in the intestine, resulting in increased GI transit. Furthermore, DIO revealed increased expression of 5-HT4 receptor in the brain but not in the gut, suggesting other receptors to be involved. These data obtained from zebrafish are in line with earlier findings in (some) mammalian models. Given the faster developmental features, the zebrafish offers additional perspectives for obesity research.

- B14 -

LIVE CALCIUM AND MITOCHONDRIAL IMAGING IN THE ENTERIC NERVOUS SYSTEM OF PARKINSON PATIENTS AND CONTROLS. A. Desmet (1), C. Cirillo (1), J. Tack (2), W. Vandenberghe (3), P. Vanden Berghe (1). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), Lab for Enteric Neuroscience (LENS), KU Leuven, Leuven, Belgium, Experimentele Geneeskunde, [2] Translational Research Center for Gastrointestinal Disorders (TARGID), KU Leuven, Leuven, Belgium, experimentele geneeskunde, [3] University Hospitals Leuven, Leuven, Belgium, Laboratory for Parkinson Research.

Introduction : Parkinson's disease (PD) is a neurodegenerative disease with motor and non-motor symptoms that severely affects quality of life. Besides alpha-synuclein aggregation, mitochondrial dysfunction and dysregulation of intracellular calcium concentration probably contribute to the pathogenesis of PD. Because dysphagia and constipation are frequent symptoms of PD, several studies have investigated the gastrointestinal tract (GI), and more specifically the enteric nervous system (ENS), in search of a biomarker of PD. It has even been proposed that PD may start in the ENS and spread from there to the brain.

Aim : Here we assessed neuronal and mitochondrial functioning in primary enteric neurons of PD patients and their healthy partners as controls.

Methods : Using a unique combination of live microscopy techniques applied to neurons in the submucous plexus, we were able to image neuronal calcium (Ca²⁺) responses and mitochondrial membrane potential in routine duodenal biopsies of PD patients.

Results : We found that submucous neurons and mitochondria were not affected in PD patients compared to healthy controls, which suggests that these neurons are not involved in the pathogenesis or the gastrointestinal symptoms of PD.

Conclusions : Our study provides unprecedented functional information on live PD neurons and paves the way for testing the involvement of enteric neurons in specific subgroups of PD and in other neurodegenerative diseases.

- B15 -

FOETAL INTRAPERITONEAL AAV8 INJECTIONS RESULTS IN THE SPECIFIC TARGETING OF MYENTERIC NEURONS IN THE MOUSE GASTROINTESTINAL TRACT. K. Winckelmans (1), R. Buckinx (1), S. Waddington (2), J. Timmermans (1). [1] University of Antwerp, Antwerp, Belgium, Veterinary Sciences, [2] University College London, London, United Kingdom (the), Gene Transfer Technology.

Introduction : Adeno-associated viral vectors (AAV) are a promising and versatile tool for gene transfer to manipulate neuronal function in preclinical research and might have great potential in gene therapy. Previously, it was shown that neonatal and postnatal AAV injections resulted in *in vivo* transduction of the mouse enteric nervous system (ENS). Although the full development of the mouse ENS also continues after birth, the prenatal phase starting from E9,5 till E15 spans the period of the main colonization of the mouse gastrointestinal (GI) tract by vagal and sacral crest cells. In humans the equivalent period corresponds to week 4-7 of gestation. Defects in ENS development result in congenital gastrointestinal phenotypes, such as Hirschsprung's disease. Therefore, genetic studies performed at prenatal stage are essential for a better understanding of the underlying mechanisms of ENS development and related dysfunctions.

Aim : Use foetal intraperitoneal AAV8 injections as a gene transfer tool to transduce the murine enteric nervous system at prenatal stage.

Methods : E14,5 mouse foetuses were intraperitoneally injected with AAV8 carrying a cassette encoding eGFP as a reporter under the control of the human cytomegalovirus promoter. At postnatal day 35, the transduction of cell types in the GI tract was evaluated using fluorescence microscopic imaging of the transgene-encoded eGFP in combination with a set of neuronal and glial markers.

Results : In the distinct GI segments, i.e. oesophagus, gastric corpus, ileum and distal colon, eGFP was exclusively found in HuC/D-immunoreactive (-ir) cells, indicating a highly neuron-specific transduction. Neurochemical analysis showed

the expression of viral-encoded eGFP in nitrergic, calbindin-ir, calretinin-ir as well as CGRP-ir neurons, indicating that several functional neuronal subpopulations were susceptible to transduction. Neuronal fibers also showed a strong eGFP signal, either of intrinsic or possibly also of extrinsic origin as systemic administration of AAVs at foetal stage leads to extrinsic transduction of the nervous system as well. No eGFP expression could be observed in enteric glial cells.

Conclusions : These results demonstrate the specific and successful transduction of the prenatal ENS and strengthens the validity of AAV vectors in transducing murine enteric neurons. However, compared to earlier results from the neonatal and postnatal injections, the transduction efficiency in the myenteric plexus in prenatal injections is still significantly lower (6% versus 20-30%). Therefore, further research is indicated to improve the technique (e.g. exploring other AAV serotypes and promoters), but the results presented in this study clearly show the potential of this technique for in vivo enteric neuron manipulation in addition to the transgenic animal breeding.

- B16 -

INFLUENCE OF ACUTE TRYPTOPHAN DEPLETION ON OESOPHAGEAL SENSITIVITY AND VISCERAL PAIN PERCEPTION IN HEALTH. C. Broers (1), B. Van Houtte (2), P. Vermeersch (3), N. Peersman (3), J. Tack (2), A. Pauwels (2). [1] KU Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, [2] KU Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, [3] UZ Leuven, Leuven, Belgium, Laboratoriumgeneeskunde.

Introduction : Proton pump inhibitors (PPIs) are effective in healing oesophagitis, however 30% of non-erosive gastro-oesophageal reflux disease (NORD) patients remain symptomatic while taking PPIs. In these patients, oesophageal hypersensitivity is considered an important pathophysiological mechanism. Serotonin (5-HT) is predominantly found in the central nervous system and in the gastro-intestinal (GI) tract. 5-HT plays a major role in the regulation of GI secretion, motility and sensitivity, and has been associated with emotion regulation. Acute tryptophan depletion (ATD) temporarily reduces the availability of tryptophan (TRP), thereby decreasing central and peripheral 5-HT synthesis. From previous studies, ATD is known to affect GI physiology and enhance visceral pain perception in the colon.

Aim : To study the effect of ATD on oesophageal sensitivity in healthy volunteers (HV).

Methods : Oesophageal multimodal sensitivity was assessed after intragastric infusion of an amino-acid mixture (AA-mix) containing 15 AAs with TRP (control condition) or without TRP (ATD condition). After an incubation period of 5 hours, a probe with a balloon was positioned in the distal oesophagus. Thermal (recirculating a heated saline solution through the balloon), mechanical (increasing balloon volume), electrical (2 stimulation electrodes) and chemical sensitivity (modified Bernstein) were tested. Stimulus intensities were evaluated for first perception, pain perception threshold (PPT) and pain toleration threshold (PTT). At 3 time points blood samples were collected for biochemical analysis. General mood was assessed by the Positive and Negative Affect Schedule (PANAS) and the State-Trait Anxiety Inventory (STAI) questionnaires. Results were analyzed using paired t-tests and two-way ANOVA repeated measures and corrected for multiple testing (Bonferroni correction). A p-value of <0.05 was considered significant.

Results : We compared control condition with ATD in 11 HV (4m/7f, mean age 24y [range 21y-33y]). ATD significantly reduced plasma levels of TRP, 5 and 7 hours after administration of the AA-mix (5h : 139.0 $\mu\text{mol/L}$ [97.1-175.1] vs. 6.1 $\mu\text{mol/L}$ [3.3-21.1], 7h : 65.6 $\mu\text{mol/L}$ [53.0-133.1] vs. 6.1 $\mu\text{mol/L}$ [4.8-12.2], $p < 0.0001$). ATD significantly decreased the PPT during chemical stimulation ($p = 0.03$) with a pronounced effect size (Cohen's $d = 0.75$) (Table 1). However, ATD had no significant influence on sensitivity to the other stimulation modalities. There was no significant difference in PANAS and STAI-State scores before, during and after the stimulations for the 2 conditions. Table 1 : Results of oesophageal multimodal stimulation for control condition and acute tryptophan depletion (ATD). Results are presented as median [25th -75th percentile]. $n = 11$, unless indicated otherwise since only HV reaching the sensitivity thresholds were taken into account for analysis.

Temperature stimulation ($^{\circ}\text{C}$) :

PPT control : 42.66 $^{\circ}\text{C}$ [40.56-46.71] vs. PPT ATD : 43.71 $^{\circ}\text{C}$ [41.22-46.65], $p = 0.53$

PTT control : 46.43 $^{\circ}\text{C}$ [45.00-49.04] vs. PTT ATD : 46.32 $^{\circ}\text{C}$ [44.42-51.88], $p = 0.56$

Mechanical stimulation (ml) :

PPT control : 17.70ml [16.00-19.40] vs. PPT ATD : 16.65ml [11.70-21.50], $p = 0.23$

PTT control ($n = 9$) : 22.10ml [20.35-24.35] vs. PTT ATD ($n = 9$) : 25.00ml [17.90-30.80], $p = 0.17$

Electrical stimulation (mA) :

1st perception control : 6.33mA [4.17-6.83] vs. 1st perception ATD : 6.00mA [4.33-13.33], $p = 0.17$

PPT control : 11.83mA [8.50-14.33] vs. PPT ATD : 10.00mA [8.00-19.83], $p = 0.41$

Chemical stimulation (ml) :

1st perception control : 10.00ml [7.00-18.00] vs. 1st perception ATD : 8.00ml [6.00-11.00], $p = 0.27$

PPT control : 25.00ml [14.00-29.00] vs. PPT ATD : 14.00ml [13.00-24.00], $p = 0.03$

PTT control : 34.00ml [24.00-42.00] vs. PTT ATD : 26.00ml [16.00-36.00], $p = 0.23$

Conclusions : To our knowledge, this is the first study to address the effect of ATD on esophageal multimodal sensitivity in health. ATD significantly decreased pain perception threshold during chemical stimulation, without affecting sensitivity to mechanical, thermal or electrical stimulation. The findings may have implications for the understanding and treatment of refractory GORD or functional heartburn.

- B17 -

A DOUBLE-BLIND, PLACEBO-CONTROLLED TRIAL WITH BACLOFEN FOR THE TREATMENT OF REFRACTORY GASTRO-ESOPHAGEAL REFLUX DISEASE. A. Pauwels (1), V. Boecxstaens (1), C. Broers (1), J. Iven (1), D. Zhao (1), T. Vanuytsel (1), J. Tack (1). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), KU Leuven, Leuven, Belgium, clinical and experimental medicine

Introduction : A significant proportion of patients with gastro-esophageal reflux disease (GERD) remains symptomatic while on proton pump inhibitor (PPI) therapy. PPIs significantly reduce the proportion of acid reflux, however they have no effect on non-acid reflux which can provoke symptoms in GERD patients. Baclofen, a γ -aminobutyric acid agonist, is able to decrease both acid and non-acid reflux. To date, studies with baclofen focused on mechanistic aspects in patients with proven ongoing weakly acidic reflux, but the symptomatic outcome of patients with refractory GERD symptoms has not received much attention.

Aim : The aim of this study was to assess the efficacy of baclofen 10mg t.i.d. vs placebo as add-on therapy in GERD patients with insufficient response to PPI therapy, in a randomized, parallel, double-blind, placebo-controlled study.

Methods : Patients with an incomplete control of typical GERD symptoms (heartburn/regurgitation) in spite of PPI therapy were randomized to baclofen or placebo for 4 weeks. Patients were taking PPIs (b.i.d) throughout the entire study. Prior to the study and at the end of the treatment, patients underwent a 24h impedance-pH monitoring and were asked to fill out the ReQuest questionnaire to assess symptoms of GERD. Reflux parameters post-treatment were compared to baseline using Wilcoxon signed rank test and were compared between baclofen and placebo using Mann-Whitney test. The differences in wellbeing and acid complaints ReQuest domains after treatment were compared using mixed models.

Results : 60 patients were included (age 47.5y (range 19-73), BMI 24.99kg/m², 41f/19m). One patient decided not to start with the medication and 5 patients did not complete the study due to side effects (headache, nausea, drowsiness); all of them were taking baclofen at that time. 26 patients were randomized in the baclofen arm, while 28 patients were randomized in the placebo arm. There was a decrease in total number of reflux episodes, the number of non-acid reflux episodes and the number of reflux episodes with a high proximal extent in the baclofen condition compared to baseline, while no differences were observed in the placebo condition (Table 1). There was a significant main effect of time on wellbeing in all patients ($p=0.018$), but we found no difference between both arms. Similar results were found for the scores for acid complaints, with a significant time effect (0.016), a borderline condition effect ($p=0.048$) but no interaction effect.

Conclusions : Although several reflux parameters decrease with baclofen treatment, there was no gain in symptom control or wellbeing from add-on baclofen therapy over placebo in patients with persisting typical GERD symptoms on PPI. These findings argue against add-on baclofen therapy based on symptom evaluation alone.

- B18 -

THE WAITING ROOM QUESTIONNAIRE : VALIDATION OF A NOVEL PATIENT REPORTED OUTCOME QUESTIONNAIRE FOR THE DIAGNOSIS OF FUNCTIONAL GASTROINTESTINAL DISORDERS. N. Goelen (1), F. Carbone (1), L. Holvoet (2), A. Vandenberghe (3), J. Arts (2), P. Caenepeel (2), H. Piessevaux (4), J. Tack (1). [1] KU Leuven, Leuven, Belgium, TARGID, [2] UZ Leuven, Leuven, Belgium, Gastroenterology, [3] Medical Research Consultant, Chaumont-Gistoux, Belgium, [4] Cliniques Universitaires St Luc, Brussels, Belgium, Gastroenterology.

Introduction : Functional gastrointestinal disorders (FGID) are a heterogeneous group of chronic conditions without a known organic etiology, although with a significant socio-economic impact. Despite the upgraded Rome IV criteria, several challenges remain to interfere with the appropriate assessment of FGI symptoms. Accurate interpretation and communication of the symptom pattern is imperative for a correct diagnosis. Hence, our group recently developed a Rome III criteria-based Waiting Room Questionnaire (WRQ) with pictograms as a visual aid to improve symptom identification.

Aim : The aim of this study is to validate the WRQ for symptom assessment and diagnosis of frequent FGID.

Methods : Ambulatory patients completed the WRQ which includes 40 questions with pictograms used to diagnose gastroesophageal reflux disease (GERD), functional dyspepsia (FD) subgroups PDS and EPS, irritable bowel syndrome (IBS) and chronic idiopathic nausea (CIN). Cohen's kappa statistic was used to assess inter-rater concordance, defined as the fraction of cases where the predominant symptomatic profile identified by the clinician's interview matched with the profile identified by the patient on the WRQ which was completed in absence of a clinician. Secondary endpoints were the clarity and relevance of the drawings and cognitive validity.

Results : In total, 138 ambulatory patients from 10 Belgian gastroenterology clinics presenting with FGID symptoms (75% female, 42 ± 1.3 years) filled out the WRQ while waiting for their consultation. After the consultation and based on their expert opinion, clinicians diagnosed 20 patients with predominant GERD, 45 PDS, 22 EPS, 37 IBS and 14 CIN. Pictograms entailed an added value for 77% of the patients and 87% of the patients understood all questions. Clinicians indicated that $> 90\%$ of the written answers were confirmed during their interview in 75% of the cases. Patients were highly reliable in 81% of the cases with regard to meal-related symptoms and in 83% with regard to bowel movements. Overall, concordance for the predominant symptomatic profile identified by the WRQ and by the clinician (inter-rater agreement) was 'good' (72.3%, $\kappa = 0.65 \pm 0.05$). Concordance was the highest for CIN (93%) and EPS (86%), followed by IBS (76%), PDS (64%) and GERD (50%) (Figure 1). The concordance was 80% for the pooled FD population. Solely based on the answers in the WRQ 45% of GERD patients would have been misdiagnosed with PDS or EPS. PPI usage was a major confounder, probably through its effect on heartburn : 78% of the patients with a physician-based diagnosis of GERD not confirmed by the WRQ received PPI therapy. The prevalence of PPI therapy was also higher in the GERD patients (80%) compared to the other groups (32 - 64%, $p = 0.02$).

Conclusions : The new WRQ is a reliable, accurate and easy to handle tool that can provide diagnostic guidance in (non-)specialist settings and clinical research. Implementation in clinical practice would standardize and enhance the diagnostic approach for a vast population of FGID patients.

CASE REPORTS

- C01 -

A RARE COMPLICATION OF TOTALLY EXTRAPERITONEAL HERNIA REPAIR DETECTED BY COLONOSCOPY. L. THIJIS (1), M. ASBASBASSI (2), G. LAMBERT (3), M. COOL (3), G. DEBOEVER (3). [1] Gastroenterology, University Hospitals Leuven, Leuven, Belgium, Gastroenterology and Hepatology, [2] AZ Damiaan, Oostende, Belgium, General Surgery, [3] AZ Damiaan, Oostende, Belgium, Gastro-enterology.

Introduction : Laparoscopic hernia repair is one of the most frequently performed procedures in general surgery. The preferred method is the ‘totally extraperitoneal’ (TEP) hernia repair [1]. Mesh migration into the gastro-intestinal tract is a rare complication of this procedure. The presenting symptom of mesh migration can be abdominal pain, obstruction, fistula, abscess or hematochezia. Diagnosis is made by CT scan. We present a case mesh migration into the caecal wall detected by colonoscopy.

Case report : A 44-year-old male patient with a family history of colon cancer was referred for a screening colonoscopy. His previous medical history includes a bilateral totally extraperitoneal inguinal hernia repair in 2014. A lightweight polypropylene mesh was used to repair the hernia. Clips were placed during the surgery to seal a gap in the peritoneum. Since this operation he complained of mild chronic lower abdominal pain, without blood loss or change in bowel habits. On a computed tomography (CT) scan performed in February 2015 no explanation for his symptoms was found. During colonoscopy a mesh-like structure, which could not be removed from the colonic wall, was observed in the caecal region. Further investigation by CT scan of the abdomen confirmed the migration of the mesh through the wall of the caecum. There was also a small amount of free air in the peritoneal cavity, but no abscess was found. The patient was referred for laparoscopic exploration and a caecal resection was performed. The unmigrated portion of the mesh was left in situ and the peritoneal defect was closed.

Discussion : There are two groups of groin hernia repair techniques : the open tension-free techniques with mesh and the laparoscopic repair techniques. Laparoscopic inguinal repair techniques involve the transabdominal preperitoneal technique (TAPP) and the totally extraperitoneal technique (TEP). With the TAPP technique the abdominal cavity is entered, leading to the possibility of injury to the intraperitoneal contents [7]. In TEP, the peritoneal cavity is not entered during the surgery and the hernia is sealed from outside the peritoneum [1]. In English literature only 5 cases of mesh migration into the large intestine after laparoscopic inguinal hernia repair were reported [9-10]. Mesh migration into the intestine can lead to fistula, intestinal obstruction and intra-abdominal abscess. [10]. It can cause chronic abdominal pain, change in bowel habits or hematochezia, but sometimes remains completely asymptomatic. Migration of the mesh can be explained by two mechanisms. Primary mesh migration happens when an inadequately connected mesh traverses the adjacent anatomical paths of least resistance or when a relatively connected mesh is shifted by external forces [2,4,6]. Secondary mesh migration occurs through anatomical borders and is caused by chronic inflammation due to foreign body reaction [5,6]. There are multiple factors that determine the risk of mesh migration [2, 6, 10], such as : - The type of mesh materials. There is more chronic inflammation and fibrosis with the polypropylene meshes [10]. - Inadequate fixation of the prosthetic device can also lead to migration of the mesh [11]. - Placement of the mesh plug too deep within the inguinal canal also predisposes to migration of the mesh [11]. - Direct contact between the mesh and viscera [9-10]. - Chronic inflammation accompanying diverticular disease could support erosion through the bowel wall. Mesh migration into the colon is generally managed by resection of the involved bowel [9]. Only one report of endoscopic mesh removal was found in literature (table 1). The mesh was intraluminal and adherent to the splenic flexure. An alligator forceps was used to remove the mesh[12]. Endoscopic attempts of mesh removal increase the risk of damage to the colonic wall and surrounding structures and could lead to intra-abdominal infection.

Conclusion : Mesh migration into the gastro-intestinal tract is a rare complication of laparoscopic inguinal hernia repair. The presenting symptom can be abdominal pain, obstruction, fistula, abscess or hematochezia. It can also occur completely asymptomatic. Diagnosis is made by CT scan or colonoscopy. The treatment of choice is surgical resection of the affected bowel.

- C02 -

POST-LIVER TRANSPLANTATION FOLLOW-UP OVER 17 YEARS FOR MILD ZELLWEGER SPECTRUM DISORDER AND ADDITIONAL CASES. T. Demaret (1), S. Varma (1), X. Stephenne (1), F. Smets (1), I. Scheers (1), L. Van Maldergem (2), R. Reding (3), E. Sokal (1). [1] Cliniques Universitaires Saint-Luc, Belgium, Pediatric Gastroenterology & Hepatology, [2] Centre Hospitalier Régional Universitaire de Besançon, Besançon, France, Center for human genetics, [3] Cliniques Universitaires Saint-Luc, Belgium, Pediatric Surgery and Abdominal Transplantation.

Mild Zellweger spectrum disorder, also described as Infantile Refsum disease, is attributable to mutations in PEX genes. Its clinical course is characterized by progressive hearing and vision loss, and neurodevelopmental regression. Supportive management is currently considered the standard of care, since plasmalogen supplementation, low phytanic acid

diet, cholic acid, and docosahexaenoic acid have not shown clinical benefits. Liver transplantation (LT) was shown to correct levels of circulating toxic metabolites, partly responsible for chronic neurological impairment, with LT survival currently being >95%. Of three patients having undergone LT for mild ZSD, one died after LT, while the other two displayed significant neurodevelopmental improvement on both the long- (17 years post-LT) and short-term (9 months post-LT) follow-up. We documented a sustained improvement in the biochemical profile, with a complete normalization of plasma phytanic, pristanic and pipecolic acid levels. This was associated with improved clinical evolution, puberty achievement, as well as stabilization of hearing and visual functions, and neurodevelopmental status, which has enabled the older patient to lead a relatively autonomous lifestyle on the long-term. The psychomotor acquisitions have been remarkable. Specially seen in comparison to their affected siblings who did not undergo LT and exhibited a poor neurological outcome with severe disabilities. Based on our short- and long-term follow-up experience, we speculate that LT performed before the onset of severe sensorineural defects in mild ZSD, enables partial metabolic remission and improved long-term clinical outcomes.

- C03 -

ENDOSCOPIC GASTROJEJUNOSTOMY USING THE HOTAXIOS LUMEN APPOSING METAL STENT TO TREAT MALIGNANT GASTRIC OUTLET SYNDROME. C. Mouradides (1), I. Borbath (1), P. Deprez (1), T. Moreels (1). [1] Cliniques Universitaires Saint-Luc, Belgium, Hépto-Gastro-Entérologie.

We report the case of a 71-year-old female with a pancreatic head adenocarcinoma, previously treated with Whipple resection with pyloric conservation and adjuvant chemotherapy. However, progression to peritoneal carcinomatosis and local recurrence lead to upper GI obstructive symptoms. Initially the obstruction was treated by means of an uncovered metallic enteral stent, which ended up in the afferent limb, obstructing the alimentary limb, leading to gastric outlet syndrome. The uncovered metallic enteral stent could not be removed because of tumor ingrowth. Peritoneal carcinomatosis rendered the patient not suitable for surgical treatment. There was no endoscopic access to the alimentary limb via or beside the metallic enteral stent because of tumor ingrowth. An endoscopic gastrojejunostomy was then performed, using the HotAxios system (XLumina Axios, Boston Scientific), under EUS guidance (linear array echoendoscope GF-UCT 180, Olympus, Japan) and fluoroscopy : the alimentary limb was identified by transgastric EUS, accessed with a 19G needle, and confirmed fluoroscopically by injection of intraluminal contrast dye. Next, a 0.035inch guidewire was placed, allowing smooth introduction of the HotAxios stent deployment system into the alimentary limb. The fully covered HotAxios stent was then deployed under fluoroscopic and endoscopic guidance, with the distal part in the intestinal lumen, and the proximal flange in the stomach. However, upon release, it immediately migrated outside the stomach. So a second metallic esophageal covered stent (Taewoong Medical Niti, 8cm, 22mm) was placed over the guidewire inside the HotAxios stent, with the proximal part protruding into the gastric lumen. After the procedure, the patient was able to resume oral feeding. 3 months later, she was readmitted for recurrent vomiting due to a migration of the esophageal stent into the stomach, whereas the HotAxios stent was still in place. A new fully covered Nagi stent (Taewoong Medical, 20mm and 16mm diameter) was placed inside the HotAxios stent. 3 months later, a control gastroscopy showed the stents were in correct position and the endoscopic gastrojejunostomy was patent. She finally died due to progression of her malignant disease, 7 months after the endoscopic creation of the gastrojejunostomy. This case illustrates the feasibility of the HotAxios system to create an endoscopic gastrojejunostomy to treat malignant gastric outlet syndrome in altered anatomy. Endoscopic ultrasound-guided gastrojejunostomy is an alternative to surgical gastrojejunostomy to treat benign and malignant gastric outlet syndrome, in normal and surgically altered anatomy. Lumen apposing metallic stent placement reduces the risk of stenosis recurrence classically described in uncovered metallic stents, and prevents migration encountered with covered metallic stents. Technical success rate has recently been reported to be 92%, and clinical success 85%.

- C04 -

ESOPHAGEAL INTRAMURAL PSEUDODIVERTICULOSIS COMPLICATED BY A PNEUMOMEDIASTINUM. M. Struyve (1), C. Langmans (2), G. Robaey (3) / [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Gastroenterology and Hepatology, [2] UZ Leuven Gasthuisberg, Leuven, Belgium, Internal Medicine, [3] Ziekenhuis Oost Limburg, Genk, Belgium, Gastroenterology and Hepatology.

A 64-year-old man was seen at the outpatient clinic complaining about progressive dysphagia since the last week. He suffered from a pronounced intent to vomit, an excessive salivation and a vague chest pain. Physical examination was unremarkable. Prior medical history revealed intermittent dysphagia the past two years with an episode of acute dysphagia one year ago due to a food impaction that was endoscopically removed. The esophagogastroduodenoscopy (EGD) at that time showed a benign esophageal stricture in the distal esophagus. Biopsy specimens that were taken from the

stricture at that time showed active esophagitis and the presence of mycosis. The patient had been successfully treated with proton pump inhibitors and antimycotics for respectively four and two weeks and there was no recurrence of severe dysphagia until today. Because of a recurrent episode of severe dysphagia today, a new EGD was performed as next step in the evaluation of this case. Endoscopic evaluation showed a stenosis of the distal esophagus that could be passed by the scope. Because of the vague chest pain already present before the procedure, we performed a standard chest X-ray that revealed free air as a sign of a pneumomediastinum (Fig. 1A). We consequently performed a computed tomography (CT) scan of the chest which confirmed the presence of a pneumomediastinum with dominant localization around the esophagus from the level of the gastroesophageal junction to the cervical base (Fig 1B). Small intramural air bubbles in the proximal part of the esophagus were seen on these CT images suggestive for intramural esophageal pseudodiverticulosis (EIPD). The next diagnostic step was to perform an X-ray esophagogram with Telebrix gastro® (joxitalamaat, meglumine) which could not reveal a leakage of contrast but revealed pathognomonic signs of EIPD. It also showed a linear to circular loss of filling probably due to the stenosis in the distal esophagus that was visualized on the EGD (Fig. 2A,B,C,D,E). We admitted the patient in the hospital for transient fasting, proton pump inhibitors and treatment with intravenous antibiotics and total parenteral nutrition. Radiographic control after one week no longer showed the presence of free air as a sign of a pneumomediastinum. Oral feeding was restarted without problems and the patient was discharged out of the hospital. Follow-up consultation was scheduled and a balloon dilatation of the stenosis will be considered. Dysphagia is a common complaint of patients seen at the outpatient clinic as well as at the emergency room. The differential diagnosis is broad and includes common causes such as gastroesophageal reflux disease, strictures, webs, tumors, foreign bodies, functional disorders etc. that are well known by physicians. We report here EIPD as a cause of dysphagia that is less known by physicians and it is rarely described in the literature. EIPD is characterized by multiple, segmental or diffuse, flask-like outpouchings in the esophageal wall corresponding to dilated and inflamed excretory ducts of the submucosal esophageal glands. The underlying etiology still remains unclear. Symptoms can be various, but the predominant symptom is dysphagia. Esophageal strictures, esophageal candidiasis and gastroesophageal reflux disease are often associated. The diagnosis can be made by upper gastrointestinal endoscopy, but barium esophagography is the modality of choice. Complications of EIPD are rare and include broncho-esophageal and esophagomediastinal fistula, pleural and pericardial effusion, abscesses, gastrointestinal bleeding from a web-like stenosis or esophageal perforation with pneumomediastinum like in our case. The treatment for EIPD should be directed towards treating underlying associated conditions and relieving symptoms rather than the pseudodiverticulosis itself.

- C05 -

MECKEL'S ENTEROLITH : A RARE CAUSE OF SMALL BOWEL OBSTRUCTION. L. Wauters (1), K. Peeters (2), A. Van Hootegem (3), P. Goetstouwers (3), P. Delvaux (2), J. Callens (3). [1] UZ Leuven, Leuven, Belgium, Gastroenterology, [2] AZ-KLINA, Brasschaat, Belgium, General Surgery, [3] AZ-KLINA, Brasschaat, Belgium, Gastroenterology.

Small bowel obstruction due to an enterolith expelled from Meckel's diverticulum is a rare condition. Diagnosis may be challenging when calculi are not radio-opaque and delayed treatment can lead to complications of bleeding or perforation. We present a case of small bowel obstruction resulting from a Meckel's enterolith. A 42-year-old man presented to the emergency department (ED) with exacerbation of intermittent and colicky abdominal pain, which he had been experiencing for the past 10 days. His last bowel movement the previous day was normal and he reported nausea with bilious vomiting. He was recently admitted at the same hospital for similar complaints with an episode of non-bloody diarrhea 48h after a barbecue. At that time, there was mild abdominal distention and tenderness without peritoneal signs and a temperature of 38.1°C. C-reactive protein (CRP) level was 119 mg/L (normal range 0 - 5 mg/L) with normal white blood cell (WBC) count and there was radiological evidence of dilated small bowel loops with air fluid levels. The patient was discharged after his symptoms had quickly resolved with intravenous analgesia. An empirical treatment with Azithromycin was prescribed for suspected *Campylobacter* enteritis, but stool culture was negative. He had a history of *Salmonella* enteritis with normal ileocolonoscopy and biopsies and no prior abdominal surgery. Upon presentation, physical examination revealed diffuse abdominal pain on palpation. CRP level was 70.7 mg/L and WBC count 25.07 x1000/ μ L (normal range 3.45 - 9.76 x1000/ μ L) with no further abnormalities. Abdominal X-rays suggested small bowel obstruction with absence of gas in the colon and rectum. Abdominal CT-scan confirmed a mechanical obstruction due to a well-defined structure (35mm in diameter) with a central high-density component proximal to the caliber change located in the ileum and suggestive of an impacted food bolus. There was no evidence of appendicitis, ileitis or pneumobilia. Following surgical consult, laparoscopy was performed with resection of a large Meckel's diverticulum located 50cm proximally from the ileocecal valve. Histopathological evaluation showed a Meckel's diverticulum of 6.0x4.5x1.5 cm with intestinal mucosa and no evidence of malignancy. On the second post-operative day, the patient experienced recurrent abdominal pain and vomiting, with clinical suspicion and radiological evidence of small bowel obstruction. Abdominal CT-scan showed the round structure as previously described that was displaced more distally and similarly located proximally to the transition point. On laparotomy, multiple loops of distended ileum surrounded by non-purulent fluid were seen and an impacted stone or enterolith was removed by longitudinal enterotomy. Biochemical analysis showed a composition of 60% cholesterol, 30% bilirubin and some calcite. A final diagnosis of small bowel obstruction

secondary to an expelled Meckel's enterolith was made. The postoperative course was uneventful and the patient was discharged home with no further episodes of abdominal pain.

- C06 -

A RARE CASE OF A TRACHEOESOPHAGEAL FISTULA. L. De Waele (1), J. Maus (1), I. Ruytjens (1). [1] ZNA Middelheim, Antwerpen, Belgium, Gastroenterology.

A 28-year-old woman presented at the outpatient clinic of gastro-enterology with hoarseness, halitosis, intermittent high dysphagia since two years and a squeaking sound during expiration. Medical history revealed appendectomy, meningitis and recurrent respiratory infections at childhood. She denies smoking or alcohol abuse. She doesn't take any medication. Clinical examination showed a good performance with normal abdominal examination. A CAT scan of the neck was normal. Gastroscopic findings were normal, biopsies of the oesophagus showed an aspecific oesophagitis without eosinophilia. A visit to the ORL doctor and a spirometry at the pneumology department were normal. A HR-CAT scan revealed a small congenital high tracheoesophageal fistula and a small diverticulum in the esophagus. A barium swallow test showed and atonic proximal part of the esophagus, which was also seen with an esophageal manometry that showed 100% aperistalsis in the whole oesophagus. Tracheoesophageal fistula (TEF) is a rare congenital anomaly of the respiratory tract. It typically occurs with esophageal atresia and is diagnosed short after birth. Esophageal atresia and TEF are classified according to their anatomic configuration. We describe an H-type TEF which is a fistula without esophageal atresia which accounts for approximately 4% of the esophageal malformations. Due to more subtle symptoms and physician unfamiliarity diagnosis can be missed and postponed until adulthood. Patients mostly present with chronic cough and frequent respiratory infections. Treatment consists of surgery. Due to lack of respiratory infections and subtle symptoms, our patient didn't receive any surgery so far and is doing well until present.

- C07 -

RECURRENT SEPSIS AND LIVER HAMARTOMA'S (VON MEYENBURG'S COMPLEXES). S. Rizzi (1), P. Warzee (2), A. Sibille (2), N. De Visscher (3), X. Holemans (3). [1] Grand Hôpital de Charleroi, Charleroi, Belgium, Gastroenterology Unit, [2] Notre Dame, Charleroi, Belgium, Gastroenterology Unit, [3] Notre Dame, Charleroi, Belgium, Infectious Diseases Unit.

Introduction : Von Meyenburg complexes is a rare anatomic entity. It consists in small multiple and diffuse nodular cystic lesions, occupying the liver, corresponding to bile duct hamartomas. They are usually discovered accidentally during radiologic or post-mortem evaluation. Their presence is not considered as pathologic, since no symptoms nor biologic or functional abnormalities are usually present. We report the case of a patient with VMC presenting with recurrent episodes of sepsis.

Case report : A 71-year-old Caucasian woman referred to our unit for evaluation of recurrent septic episodes of unknown cause. Since 2003 she has been admitted twelve times with high fever, nausea and vomiting and upper abdominal discomfort. Laboratory values at admission often showed transient and slight cholestatic and cytolytic abnormalities and hyperneutrophilia. Urine cultures, chest XRay or CT were always normal. Various microorganisms were identified on blood cultures (Escherichia Coli (n=7), Klebsiella planticola (n=1) , Klebsiella pneumoniae (n=2), Klebsiella Oxytoca (n=1)). Initial endoscopic retrograde cholangiopancreatography (ERCP) showed a papilla located within a duodenal diverticulum and a slight dilated common bile duct with little amount of sludge evacuated after papillotomy. Subsequent elective cholecystectomy was performed, but did not prevent new septic episodes. In every hospitalization we excluded other causes of recurrent gram-negatives sepsis, such as urinary tract infection, diverticulitis, pneumonia, osteitis. Repeated ERCP cannot prove biliary infection maybe due to the fact that early antibiotics were given soon after bacterial mapping in emergency unit. During these episodes we repeat the ERCP and Magnetic resonance cholangiopancreatography (MRCP) exams and they didn't show any abnormality in the common bile duct. The first ERCP showed micronodular aspect of the liver parenchyma with microcysts containing liquid. Abdominal CT scan showed multiple hypodense nodules in the liver. MRCP allowed to confirm the von Meyenburg complexes diagnosis, showing multiple sub-centimeter hyperintense liver cystic lesions, disseminated in the whole of hepatic parenchyma and compatible with bile ducts. A percutaneous liver biopsy realised in 2012 did not confirm the presence of hamartomas. We gave the ciprofloxacin prophylaxis for five days a week during the second and the fourth week each month and we reached good clinical results with a diminution of the septic episodes.

Conclusion : In our case of VMC patient presented recurrent unexplained septic episodes. Only few cases of septic episodes in this condition were described in literature, [5,6] suggesting that cholangitis may be associated to VMC as a result of biliary obstruction, due to excess mucus secretion. The VMC may exceptionally results in recurrent life threatening bacteraemia with potentially chronic consequences. Early wide spectrum antibiotics usually leads to quick favour-

able outcome. Intermittent AB prophylaxis may reduce frequency and severity of septic events but don't eliminate them. The clinical follow-up it is very important in these patients to avoid secondary chronic septic consequences. Several cases of cholangiocarcinoma have been described in patients with von Meyenburg complexes, but an association it has not yet been proven.

- C08 -

PRIMARY SYPHILITIC PROCTITIS. M. Struyve (1), W. Meersseman (2), W. Van Moerkereke (3). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Gastroenterology and Hepatology, [2] UZ Leuven Gasthuisberg, Leuven, Belgium, Internal Medicine, [3] AZ Groeninge, Kortrijk, Belgium, Gastroenterology and Hepatology.

A 36-year-old man was seen at the outpatient clinic after referral by his general practitioner to exclude underlying perianal abscess since the patient was complaining about mushy stools mixed with blood and mucus during the last four days. At the same time, he mainly suffered from a perianal discomfort, abdominal pain and a fever (38.9 degrees Celsius). He had no prior important medical history. Familial history revealed a sister with severe Crohn's disease. Further systemic history didn't reveal any important issues. A physical examination showed a mild abdominal tenderness in the right lower quadrant of the abdomen. Anal inspection showed no signs of an abscess or fistula. Adenopathies could not be clinically discovered. Laboratory analyses demonstrated a normal white blood cell count ($8.69 \times 10^9/l$) with a normal differentiation, but an elevated C-reactive protein level (40 mg/l [0.0 – 5.0]) was seen. The remainder of the blood analysis was satisfying. We performed a stool analysis that could not identify any pathogens. Analysis of fecal calprotectin turned out to be highly positive (814 microgram / g feces, whereas a normal range is below 50). The patient was admitted the same day in the hospital for a semi-urgent diagnostic work-up. We performed a magnetic resonance imaging of the lower abdomen and pelvis for the purpose of excluding hidden abscesses and fistulae, also because this was the reason of patient referral by the general practitioner since the patient had a sister with severe Crohn's disease with perianal manifestations. The images showed an edematous rectal abdominal wall with inflammatory infiltration of the mesorectal adipose tissue and the presence of enlarged lymphatic nodules. An ileocolonoscopy was performed the day after. There was a normal appearing mucosa of the terminal ileum and colon, except for the inspection of the rectal mucosa, which showed multiple atypical mucosal ulcerations with adjacent edematous mucosa. Multiple biopsies were taken as well of the ileum, the colon as the rectum. The anatomopathological findings of the terminal ileum and colon were normal. Those of the rectum showed a mild chronic inflammatory reaction suggestive of an infectious colitis with also an excess of plasma cells in the inflammatory infiltrate. There was no evidence for underlying inflammatory bowel disease. Taking into account that the rectal ulcers had atypical characteristics, we asked for sexual behavior. Because the patient admitted having unprotected anal intercourse, we performed testing for sexually transmitted disease (STD). The analyses of Chlamydia trachomatis and Neisseria gonorrhoeae on a rectal swab and on an urine sample (to exclude urethritis as a co-infection) were negative. We also performed a more general diagnostic work-up for other STD by screening for hepatitis B, hepatitis C and HIV by serologic tests. These results turned out to be negative. Laboratory analyses demonstrated a positive Treponema pallidum hemagglutination assay (TPHA) (index 15.8, positive when > 1.1) and a positive Venereal Disease Research Laboratory test (VDRL). We established the diagnosis of an early primary syphilis infection of the anorectum, also described as a syphilitic proctitis, and treated the patient and his sexual partner with one intramuscular injection of 2.4 million units of benzathine penicillin G. An endoscopic reassessment was performed six weeks after initiation of the treatment. This showed a proper endoscopic healing with remaining small stellate scars. Control of serologic tests after treatment showed a negativation of the VDRL test while the TPHA test remained positive. Rectal ulcerations are an uncommon presentation of a primary syphilis infection or primary syphilitic proctitis. It is difficult to diagnose because of its often asymptomatic or atypical clinical presentation. It is important to consider sexually transmitted diseases in all patients presenting with rectal symptoms. A history of anal sexual intercourse should be made, especially in men having sex with men (MSM). Moreover, the possibility of a primary syphilis infection of the rectum should be considered. Endoscopic findings might be diverse, whereas a typical chancre can present as an anorectal ulcer associated with regional lymphadenopathy. It is important to consider other causes of anorectal ulcers, like other STD, IBD or even malignant causes. The diagnosis of anorectal syphilis is based on the combination of the clinical presentation, serology tests, endoscopic findings and biopsies. The cornerstone of the treatment is based on an intramuscularly administration of a long-acting preparation of penicillin (benzathine penicillin G).

- C09 -

TRANSARTERIAL EMBOLIZATION OF HEPATIC ARTERIOVENOUS MALFORMATIONS IN A NEWBORN. T. Rousseff (1), R. De Bruyne (2), K. De Coen (3), K. Smets (3), W. Decaluwe (4), A. Heyneman (5), J. Panzer (6), K. Vandekerckhove (6), E. Dhont (7), L. Defreyne (8). [1] Ghent University, Ghent, Belgium, Pediatrics, [2] Ghent University, Ghent, Belgium, Pediatric Gastroenterology and nutrition, [3] Ghent University, Ghent, Belgium, Neonatology, [4]

AZ Sint-Jan Brugge-Oostende, Brugge, Belgium, Neonatology, [5] Sint Vincentius Deinze, Deinze, Belgium, Pediatrics, [6] Ghent University, Ghent, Belgium, Pediatric Cardiology, [7] Ghent University, Ghent, Belgium, Pediatric Intensive Care Unit, [8] Ghent University, Ghent, Belgium, Interventional Radiology.

Introduction : Hepatic arteriovenous malformations (AVMs) are rarely encountered congenital lesions. Complications include high-output cardiac failure, embolism and haemorrhage. The pathogenesis of AVMs is not well known, most frequently they are seen in hereditary haemorrhagic teleangiectasia patients, rarely as an isolated disease. We describe the clinical and diagnostic findings and subsequent treatment in a neonate presenting with isolated hepatic AVMs.

Methods : A male neonate was admitted to NICU after an urgent caesarean section at postmenstrual age of 37 weeks because of a preterminal cardiocography (CTG). Due to respiratory failure and difficult oxygenation, mechanical NO-assisted ventilation was necessary. Diagnostic work-up revealed profound cardiomegaly on chest X-ray. Echocardiography demonstrated severe pulmonary arterial hypertension and right heart failure. Laboratory examination showed thrombocytopenia (35 000/ μ L) and coagulopathy and liver failure (prothrombin time 50%, bilirubin 7.5 mg/dL, AST 108 U/L, ALT 93 U/L, gamma-glutamyltransferase 118 U/L and LDH 922 U/L). Color Doppler ultrasound of the liver revealed well defined areas with multidirectional high flow and low resistance (mosaic pattern), 4 in the right and 2 in the left lobe, supplied by the hepatic artery and draining into dilated hepatic veins. Flow was reversed in the portal vein. Cerebral ultrasound was normal. Physical examination showed hyper dynamic cor, no teleangiectasias.

Results : After 2 sessions of transarterial embolization, performed via the umbilical artery and with glue, the child clinically improved with recovery of the right heart failure and pulmonary hypertension on echocardiography. Subsequent further spontaneous resolution of the lesions was observed. The patient was easily weaned from ventilation. After one month the baby was discharged from hospital with only some mild cholestasis remaining. Liver ultrasound demonstrated small residual nidi in the right lobe with hepatofugal flow and normal hepatopetal flow in the left lobe and at the liver hilum.

Conclusions : Isolated hepatic AVMs are rare congenital lesions associated with significant morbidity and mortality. We report the case of a newborn with multiple hepatic AVMs presenting with severe heart failure and pulmonary hypertension. We emphasize the importance of a multidisciplinary approach towards diagnosis and treatment. In this case, the patient was successfully treated by percutaneous transarterial embolization performed via the umbilical artery with good recovery of the high-output heart failure.

- C10 -

AUTO-BREWERY SYNDROME IN A PATIENT AFTER ROUX-EN-Y GASTRIC BYPASS SURGERY : A CASE REPORT. R. Spitaels (1), F. Janssens (2), K. Vande Kerckhove (3), K. Corthouts (3), V. Brabers (4), D. Cassiman (5). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Gastroenterology, [2] Jessa Hospital, Hasselt, Belgium, Gastroenterology, [3] UZ Leuven Gasthuisberg, Leuven, Belgium, endocrinology, [4] Jessa Hospital, Hasselt, Belgium, A&E, [5] UZ Leuven Gasthuisberg, Leuven, Belgium, Gastroenterology/Hepatology.

Introduction : The auto-brewery syndrome is a condition where carbohydrates in the food are fermented in the gut by yeasts or bacteria to ethanol and subsequently absorbed in the circulation. If sufficient amounts of ethanol enter the systemic circulation, it can cause symptoms of alcohol intoxication. It is a clinical syndrome which can occur when there is an abnormal proliferation of microorganisms, ingestion of carbohydrate rich food, abnormalities in the luminal gastrointestinal flow or alterations of the alimentary tract. The most cited culprit microorganisms are *Candida* species. We report a case of auto-brewery syndrome in a patient with prior Roux-en-Y gastric bypass surgery (RYGB).

Methods : We searched Pubmed and Google Scholar for articles between 1984 and 2016, written in English, using the following keywords : “gastric bypass”, “ethanol”, “auto-brewery syndrome”, “abnormal gut fermentation” “gut fermentation syndrome” and “endogenous ethanol”. Abstracts from cross references were reviewed when relevant. Blood alcohol concentration (BAC) was determined using an enzymatic oxidation assay (Cobas 8000) with a sensitivity of 0.101g/L and error margin of 4.1%.

Case Report : A 46-year-old Caucasian male was admitted to the emergency room. He reported symptoms of drunkenness for the previous few weeks. He also experienced occasional abdominal discomfort. He denied drinking alcohol in the days before. Four weeks prior to hospitalisation he received antibiotics for upper respiratory tract infection. The past gastroenterological history was relevant for RYGB surgery 14 years ago. During physical examination an alcohol breath odor was noted; neurologic examination showed mild altered consciousness. The BAC was 0.84g/L. The abdominal ultrasound showed an homogenous hyper reflective liver parenchyma compatible with liver steatosis. After a short observation the patient was discharged with the diagnosis ‘alcohol intoxication’. A few weeks later the patient was readmitted due to persisting symptoms of drunkenness. The possibility of auto-brewery syndrome was first considered. The diagnostic test proposed by Hunnisett et al. was performed : 5g glucose was given after fasting for at least 3 hours and after restraining from alcohol for at least 24 hours. The test results were inconclusive. In order to rule out surreptitious drinking, the patient was strictly and continuously monitored during a second hospitalisation. Fasting BAC was 0.28g/L. One hour after ingestion of 100g glucose, the BAC was 0.10g/L. After two hours of fasting a carbohydrate rich meal

was offered. Four hours later, we noted a significant rise in BAC to 0.41 g/L. A coproculture showed significant growth of *Candida glabrata*. The patient was treated ambulatory with fluconazole followed by nystatin; he was also given the advice to implement a carbohydrate poor diet. This treatment didn't resolve the patient symptoms and elevated BAC were still documented. A coproculture, cultured after the first antifungal treatment, still showed significant growth of *Candida glabrata*. Subsequently he was treated with amphotericin B. This resulted in a partial amelioration of symptoms but elevated BAC was still documented on one occasion. The patient was then referred to a tertiary center where a strict carbohydrate-restricted diet was initiated (modified Atkins diet). The symptoms resolved completely and random BAC remained under the detection level.

Conclusion : This is the first report of auto-brewery syndrome in a patient after RYGB surgery. A trial of fluconazole, nystatin and amphotericin B was only partially successful. After putting the patient on a strict carbohydrate poor diet, the symptoms resolved and subsequent random measurements of the BAC remained under the detection limit. This rare syndrome has to be taken in consideration in the evaluation of patients who are believed to refrain from alcohol ingestion and report drunken-like symptoms, especially after the use of antibiotics or with a known alteration of the alimentary tract. Management includes both antifungal treatment and special diet modification. We believe that this is a case of auto-brewery syndrome based on the rise in BAC after ingestion of glucose and a carbohydrate rich meal, the associated abdominal symptoms and the improvement of symptoms after initiating a ketogenic diet. Since this condition poses legal issues, more stricter monitoring, screenings, and effective methodologies must be implemented.

**BELGIAN SOCIETY FOR GASTROINTESTINAL ENDOSCOPY
(BSGIE)**

- G01 -

THE IMPACT OF ANTITHROMBOTICS ON IMMUNOCHEMICAL FECAL OCCULT BLOOD TESTING FOR COLORECTAL CANCER SCREENING. L. Wauters (1), V. Van Der Voort (2), P. Dobbels (2), K. Hendrickx (2), V. Casneuf (2), J. Vandervoort (2). [1] UZ Leuven, Leuven, Belgium, Gastroenterology, [2] OLV Aalst, Aalst, Belgium, Gastroenterology.

Introduction : The impact of antithrombotics on immunochemical fecal occult blood testing (iFOBT) for colorectal cancer (CRC) screening in the general population remains unclear.

Aim : To study the rate of false positive iFOBT and detection of CRC or advanced adenoma's in patients with and without antithrombotics or Aspirin alone.

Methods : A prospective cohort of patients undergoing endoscopy for positive iFOBT in 2015 at 3 affiliated centers in Belgium was analyzed. Medical records were reviewed for demographic and clinical variables, including lower GI symptoms, family history of polyps or CRC and antithrombotics (Aspirin and/or Clopidogrel, Dipyridamole, Ticagrelor, novel anticoagulants and vitamin K antagonists). Endoscopy reports were checked for colorectal pathology. Significant findings were defined as CRC or advanced adenoma's (sessile serrated adenoma and tubular adenoma of >1cm or with high-grade dysplasia). Rates of false positive iFOBT and detection of CRC or advanced adenoma's were compared in patients with and without antithrombotics or Aspirin alone.

Results : A total of 524 patients (64% male, median (IQR) age 63.2 (60.2 – 66.4) years) with positive iFOBT were included. Colorectal pathology was confirmed in 379/524 (72%) patients and more commonly in males (70% vs. 48%; $p = .03$) and with positive family history (16% vs. 8%; $p = .02$). Significant findings were present in 222/379 (59%) patients with colorectal pathology and more frequently with lower GI symptoms (15% vs. 5%; $p = .002$). Antithrombotics were prescribed in 129/524 (25%) patients and associated with male gender (78% vs. 59%; $p = .0001$), older age (65.2 (62.2 – 70.3) vs. 62.3 (58.5 – 66.3) years; $p = .0001$) and lower GI symptoms (18% vs. 11%; $p = .04$). Aspirin was used in 105/524 (20%) patients and also associated with male gender (81% vs. 60%; $p < .05$) and older age (64.6 (61.8 – 69.6) vs. 62.4 (58.8 – 66.3) years; $p = .0002$). The rate of false positive iFOBT (26% vs. 28%; $p = .70$) and detection of CRC (6% vs. 6%; $p = .79$) or advanced adenoma's (40% vs. 35%; $p = .32$) were similar in patients with or without antithrombotics. The rate of false positive iFOBT (28% vs. 28%; $p = 1$) and detection of CRC (6% vs. 6%; $p = .98$) or advanced adenoma's (42% vs. 35%; $p = .21$) were similar in patients with or without Aspirin.

Conclusions : Although antithrombotics were mostly prescribed in male and older patients with an inherent higher cancer risk, detection rates of CRC and advanced adenoma's were similar. Despite the higher rates of lower GI symptoms, antithrombotics did not lead to more false positive iFOBT. Use of antitrombotics or Aspirin alone does not seem to impact the performance of iFOBT for screening of CRC in the general population.

- G02 -

ANAL PROBLEMS DURING PREGNANCY AND POSTPARTUM : A PROSPECTIVE COHORT STUDY. Y. Dorreman (1), K. Ferdinande (2), K. Roelens (3), S. Weyers (3), W. Ceelen (4), H. Van Vlierberghe (2), D. De Looze (2). [1] Department of Gynecology, Gent, Belgium, Gastroenterology, [2] Department of Hepatology and Gastroenterology, Universiteit Gent, Ghent, Belgium, Gastroenterology, [3] Department of Gynecology, Gent, Belgium, Gynaecology, [4] Department of Digestive Surgery, Gent, Belgium, Digestive surgery.

Introduction : Many pregnant women have anal symptoms during pregnancy and postpartum. The most common proctological problems reported are haemorrhoids, anal fissures and anal incontinence. Literature about this problem is scarce.

Aim : The aim of this study is to determine the prevalence of anal problems and constipation during the second and third trimester of pregnancy, in the immediate postpartum and up to three months after childbirth. We also want to identify the risk factors for the development of anal symptoms.

Methods : This is a prospective cohort study. Women between their 19th and 25th week of pregnancy are included. High-risk pregnancy and non-Dutch speaking are exclusion criteria. Ninety-four women were followed with a symptom questionnaire in the second and third trimester, in the immediate postpartum (within 3 days) and three months postpartum. Descriptive data were obtained from the patient files. A specific proctological diagnosis was presumed on the basis of combined symptoms (rectal bleeding, anal pain and swelling). Constipation was defined by the Rome III criteria. Statistical analysis was performed with SPSS and risk factors were identified using multivariate analysis with binary logistic regression.

Results : Sixty-eight percent of the women developed anal symptoms during the whole study period. Anal symptoms occurred in 50% of the women during pregnancy, in 56,2% in the immediate postpartum and in 62,9% during the three

months postpartum. The most prevalent symptom was anal pain. Constipation was reported by 60,7% during the whole study period. Most prevalent diagnoses were : hemorrhoidal thrombosis (immediate postpartum), hemorrhoidal prolapse (3rd trimester and immediate postpartum) and anal fissure (not episode-related). Anal incontinence was only reported in 2% during the postpartum. Multivariate analysis identified constipation and a history of anal problems as significant risk factors for the development of anal complaints pre- and postpartum.

Conclusions : Two thirds of pregnant women deal with anal symptoms during pregnancy or postpartum, especially hemorrhoidal complications and anal fissure. This high prevalence emphasises the clinical importance of this problem. The most important risk factor is constipation. Therefore, prevention of constipation in pregnant women is recommended.

- G05 -

ENDOSCOPIC SUBMUCOSAL DISSECTION FOR DUODENAL LESIONS : ADVERSE EVENTS AND FOLLOW UP. W. Margos (1), H. Ivekovic (2), L. Shaza (3), C. Yeung (4), H. Piessevaux (5), P. Deprez (5).[1] UCL, Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Gastroenterology, [2] University Hospital Center Zagreb, Zagreb, Croatia, Gastroenterology, [3] Institut Jules Bordet, Brussels, Belgium, Oncologie Digestive et Gastroentérologie, [4] Centre Hospitalier de Jolimont-Lobbes., La Louvière, Belgium, Hépatogastroentérologie, [5] UCL St-Luc, Brussels, Belgium, Hépatogastro-Entérologie.

Introduction : The endoscopic treatment of superficial duodenal lesions can be achieved either by endoscopic mucosal resection (EMR) or endoscopic submucosal dissection (ESD). These two techniques can be combined for the resection of the same lesion (Hybrid Endoscopic Resection, HER). ESD is known to have higher rates of complications (bleeding, perforation) than EMR, to be time consuming and technically challenging. We present results on the adverse events and clinical outcome of ESD/HER compared to EMR in our cohort of patients.

Aim : Evaluation on respective indications, safety, short and long term outcomes of ESD, compared with EMR, among patients with non-ampullary duodenal lesions.

Methods : Single tertiary university center. Retrospective study. Between 2006 and 2016. Procedure was qualified as EMR when submucosal injection and snare excision were used for resection. Procedure was qualified as ESD when any type of endoscopic knife was used. When the resection was achieved with endoscopic knife and a resection loop, the procedure was considered as HER. We divided adverse events in three groups : intra-procedural, early complications (occurring within fifteen days after the procedure) and late complications (occurring after fifteen days). Follow up endoscopy was performed at 3 - 6 months, and then at 12 months in the absence of recurrence. Complete resection rate was defined as the absence of adenomatous tissue in the resection site after resection and first follow up. Recurrence was defined as the presence of adenomatous tissue at the resection site after negative follow up. Results were expressed as medians, and compared with Student's t-test, Pearson's chi-squared test.

Results : Thirty-eight patients underwent ESD/HER procedure out of a total of 111 patients. Histopathological findings in the ESD/HER group showed 7.8% of adenocarcinomas, 15.4% of HGD, and 71.8% of LGD. The median size of specimens was 20 mm. No significant differences were observed between both groups in terms of age, sex, location of the lesions, resection rate or length of hospitalization. There were significant differences in the procedure time (median of 108 min for ESD/HER vs 79 min for EMR) and in intra-procedural complications (44% for ESD/HER vs 23% for EMR, $p=0.01$). Intra-procedural complications occurred in 18 procedures (46%) in the ESD/HER group vs 31 (23%) in the EMR group ($p=0.015$). Intra-procedural events included hemorrhage (ESD/HER 10/39 (25.6%) vs EMR 30/149 (20.1%)) and perforation (ESD/HER 8/39 (20.5%) vs EMR 4/149 (2.7%), $p<0.001$). We observed 15% of early complications (hemorrhage, perforation, pancreatitis) in ESD/HER group vs 9% in the EMR group. There were no-significant difference in early perforation (ESD/HER 3/39 (7.5%) vs EMR 1/149). Only 3 cases of late post-procedural complications were recorded in the EMR group, treated with balloon dilatation. Among all cases of perforation (intra-procedural and early), 50% of them (10/20 cases of ESD/HER) occurred before 2010, and only 5.2% (1/19 cases of ESD/HER) from 2011 until 2016 ($p<0.007$). Complete resection rate was 97.4% in ESD/HER group vs 92.6% in EMR group. Recurrence rate was 9% in ESD/HER group vs 24% in EMR group ($p<0.05$). No mortality was reported.

Conclusions : Standard polypectomy and EMR or piecemeal EMR are preferable treatment for duodenal and small-bowel superficial lesions. ESD may be indicated in specific situations like adherent lesions, en bloc resection in case of suspicion of malignancy and periampullary tumours. In our study, complete resection rate was similar between the two groups, with a significant lower recurrence rate in the ESD/HER group, maybe due to a more "aggressive" approach, but also with a significant higher rate of intra-procedural perforation. In almost all patients, bleeding and perforation were well managed by medical and endoscopic treatment. We didn't find any significant predictive factors, although we believe that extensive fibrosis, redo resection and complex hemostasis may play a role. In expert centers in duodenal EMR, ESD and HER, there was significantly less adverse events with more expertise (5.2 vs 50%).

DIAGNOSTIC YIELD OF CAPSULE ENTEROSCOPY IN PATIENTS WITH IRON DEFICIENCY ANEMIA : RESULTS OF A SINGLE CENTRE RETROSPECTIVE STUDY. A. Bertels (1), J. Nijs (1), S. Vanstraelen (1), L. Van Den Bergh (1), S. Martens (2), H. Verbrugge (3). [1] Sint-Trudo ziekenhuis, Sint-Truiden, Sint-Truiden, Belgium, Gastroenterology, [2] Sint-Trudo ziekenhuis, Sint-Truiden, Sint-Truiden, Belgium, Geriatrics, [3] Sint-Trudo ziekenhuis, Sint-Truiden, Sint-Truiden, Belgium, Internal Medicine.

Introduction : Iron deficiency anemia (IDA) is the most common cause of anemia and is often caused by obscure gastrointestinal bleeding (OGIB). IDA has negative effects on the quality of life and there is a significant impact on morbidity. Capsule enteroscopy (CE) has been used to identify the origin of the OGIB in the small bowel.

Aim : Our study aims to identify the diagnostic yield of CE in OGIB and to clarify if the diagnostic yield is different according the haemoglobin level or the concomitant intake of antithrombotics

Methods : We retrospectively analysed data of 302 patients with IDA who underwent CE in our referral centre. IDA was defined as haemoglobin less than 13 g/dl. All patients underwent a diagnostic gastroscopy and colonoscopy, unable to show abnormalities that could attribute to IDA prior to CE. Patient demographics, lowest haemoglobin level in a 3 month period prior to CE, concomitant antithrombotic therapy and CE findings were evaluated by reviewing medical charts. Diagnostic yield is defined as the number of abnormal endoscopic findings that could be attributed to IDA relative to the total number of examinations.

Results : In total 302 CE studies were performed for IDA. Mean age was 66,6 +/-15,3 years (range 16-93). Lowest haemoglobin level in a 3 months period before CE was 9.1 +/- 2.1 g/dl (range 4.0-12.9). A total of 185 patients showed abnormalities on CE that could attribute to IDA resulting in an overall diagnostic yield of 61%. Findings were angiodysplasia 110 (36%), active bleeding 36 (11.9%), ulceration 30 (9.9%), erosions 21 (7%), neoplasia 4 (1.3%). Capsule retention was present in 1 patient and in 9 patients (3%) the capsule did not reach the caecum. The diagnostic yield of CE in those patients with a haemoglobin level less than 10g/dl (n=199) was 77% versus 26% if the haemoglobin level was above 10 g/dl. In only 1 patient with an active bleeding (n = 36) seen on CE, the haemoglobin level was above 10 g/dl . 27 out of 36 patients with active bleeding were on antithrombotic therapy 52% of the patients were on antithrombotic therapy. The diagnostic yield of CE in those patients on antithrombotic therapy was 70% versus 52% if not taking antithrombotics.

Conclusions : In our study, the overall diagnostic yield of CE in IDA was 61% with angiodysplasia as the most common finding. If the haemoglobin level, prior to CE was less than 10 g/dl, the diagnostic yield is 77% versus 26% if the haemoglobin level was above 10 g/dl. If the patient is taking concomitant antithrombotic therapy the diagnostic yield was 70% versus 52% if not. This suggest that the pre-test probability of positive CE findings is higher with a lower haemoglobin. Also concomitant antithrombotic therapy leads to a higher diagnostic yield. Active bleeding at the moment of CE was also related with low haemoglobin levels and the concomitant intake of antithrombotics.

EFFICACY, SAFETY AND LEARNING CURVE OF ENDOSCOPIC SUBMUCOSAL DISSECTION IN A CONSECUTIVE CASE SERIES BY A SINGLE OPERATOR IN THE WEST. D. De Wulf (1), D. Delooze (2), P. Hindryckx (2), M. Devos (2), F. Baert (3). [1] AZ Delta, Belgium, Gastroenterology, [2] UZ Gent, Gent, Belgium, Gastroenterology, [3] AZ Delta, Roeselare, Belgium, Gastroenterology.

Introduction : Endoscopic Submucosal Dissection (ESD) is a well-established treatment for gastric lesions in the East, and is now increasingly used in the Western world for en bloc endoscopic treatment of large premalignant or early malignant lesions along the GI tract. The procedure is technically challenging and the low number of easily resectable gastric lesions in Europe leads to a steep learning curve.

Aim : The aim of this retrospective study was to evaluate the efficacy, safety and learning curve of a consecutive series of ESD done by a single operator.

Methods : Data on ESD procedures were retrospectively obtained from a single operator in consecutive patients between February 2014 and October 2016. We evaluated the complete (R0) resection rate, the en bloc resection rate, the procedure related events and the rate of remission at follow up. In order to demonstrate a learning curve, the results were evaluated separately for two groups : the first 53 patients in group 1 and the last 53 patients in group 2.

Results : 106 patients with a mean age of 67 yrs (range 19-86) were treated with ESD by a single operator. 106 ESD procedures (79 M, 27 F) were performed in esophagus (n=35), stomach (n=26), rectum (n=40) and colon (n=5). Median size of the resected specimens was 40,5 mm (range 12-90). R0 resection was achieved in 88 patients (83 %), R1 resection in 16 (15 %) and incomplete macroscopic R2 resection in 2 patients (1,9 %). En bloc resection was achieved in 90,5 % (96/106). Global procedure related adverse events occurred in 17/106 patients (16 %). Two major complications (1,9 %) were observed : 1 perforation in the stomach and rectum requiring surgery for closure on the day. The other adverse events were managed endoscopically or conservatively. 4 (3,8 %) small perforations were closed by using endoscopic

clips during the ESD procedure. 4 (3,8 %) delayed bleedings were seen, two of them after restarting Clopidogrel. 3 (2,8 %) inflammatory reactions were managed by IV antibiotics. 3 (2,8 %) esophageal strictures after (almost or) complete circumferential resection were seen and managed with endoscopic dilatation. One stricture developed after adjuvant radiotherapy. Demonstrating a learning curve the R0 resection rate increased from 41/53 (77 %) to 47/53 (88 %) in the 2 consecutive groups. Similar improvements were seen for en bloc resection : 42/53 (81 %) in group 1 and 52/53 (98 %) in group 2. The median procedure time decreased from 109 min in the first group to 60 min in the second group. The two major adverse events were seen at the beginning of the experience curve (second and fourth procedure). There was no significant difference in overall complication rate between the 2 groups : 9/53 (17,0 %) and 7/53 (13,2 %) in the 2 consecutive groups. At median follow up of 16,7 months (range 2-34) the overall recurrence rate (local recurrence and lymph node recurrence) was 6,8 % (6/88) in the R0 treated series. 4 recurrences were seen in group 1, 2 in group 2. All of them could be treated by surgery or chemoradiotherapy or both. According to the ESGE guidelines 3 of the 88 patients with R0 resection underwent additional surgery or chemoradiotherapy for histologically undifferentiated tumours or deep submucosal invasion. Finally two patients with Barrett developed a second primary tumour.

Conclusions : ESD in a Western cohort group is a safe and effective treatment with high en bloc and R0 resection rates and acceptable recurrence rates. There is a considerable learning curve.

- G08 -

ENDOSCOPIC SUBMUCOSAL DISSECTION FOR EARLY ESOPHAGEAL NEOPLASIA. A SINGLE OPERATOR STUDY. D. De Wulf (1), D. Delooze (2), P. Hindryckx (2), M. Devos (2), F. Baert (1). [1] AZ Delta, Roeselare, Belgium, Gastroenterology, [2] UZ Gent, Gent, Belgium, Gastroenterology.

Introduction : Esophageal endoscopic submucosal dissection (ESD) is a well-defined treatment for early esophageal squamous cell cancer (SCC) in the East. In Europe however most early esophageal cancers arise from Barrett's esophagus (BE) with high grade dysplasia and early adenocarcinoma (AC).

Aim : To assess the efficacy, safety and results of ESD for early esophageal tumors in a European population implementing the ESGE guidelines.

Methods : Single operator retrospective cohort study of consecutive patients with an esophageal tumor who underwent ESD between February 2014 and September 2016. A detailed chart review was performed to obtain patient and lesion characteristics, procedural and post-procedural data. Additional treatment after ESD was decided based on the ESGE guidelines. The primary endpoint was the complete (R0) resection rates. Secondary endpoints included the rate of en-bloc resection, procedure related adverse events and recurrence rates at follow-up.

Results : 35 patients (29 M, 6 F) with a median age of 63 yrs (range 19-85) underwent ESD for early esophageal cancer (12 SCC, 21 AC and 2 granular cell tumors (GCT) . The median resected specimen size was 43 mm (range 25-75). A curative resection (R0) was achieved in 76,5 % (26/35). 3 patients had a positive deep margin (2 SCC and 1 AC) and 6 had a positive lateral margin (1 SCC and 5 high grade dysplasia in margin in BE). For those 9 patients further treatment options or follow up were multidisciplinary discussed. No deaths nor major adverse events related to the ESD procedure were observed. There was no delayed bleeding. Minor adverse events included a mediastinal collection (n=1), managed conservative with IV antibiotics and esophageal strictures (n=3), all managed endoscopically. One of the strictures developed after adjuvant radiotherapy. According to the ESGE guidelines : 2 patients with R0 resection received adjuvant treatment : 1 surgery for undifferentiated carcinoma; 1 chemoradiotherapy for deep submucosal (more than 200 micron) invasion . After a median follow-up of 15 months (range 2-32) 8 % (2/24) of the R0 treated patients developed a local recurrence (1 SCC - 1 AC) and 8% (2/24) developed a secondary primary tumor in residual BE. All of them were treated with additional surgery, chemo-radiotherapy or both.

Conclusions : ESD for early esophageal tumors is a safe and effective treatment with high en-bloc and R0 resection rates and acceptable recurrence rates at follow up. ESGE guidelines are used to guide further treatment after ESD. Long term follow-up is awaited to confirm the feasibility of ESD in the West.

- G10 -

COLONOSCOPY QUALITY : IMPLEMENTATION OF COLONOSCOPY QUALITY MONITORING IN A BELGIAN UNIVERSITY HOSPITAL. S. Ouazzani (1), A. Lemmers (1), F. Martinez (2), M. Delhaye (1), M. Arvanitakis (1), O. Le Moine (1), P. Demetter (3), J. Deviere (1), P. Eisendrath (1). [1] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology, [2] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, IT Department, Medico-technical Team, [3] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Department of Pathology.

Introduction : Indicators for colonoscopy quality assessment were developed and promoted during this last decade. However technical and human resources constraints limit local implementation of continuous recording of endoscopic

quality indicators (QI). Automatic system of data extraction and presentation could help endoscopy units in their seek for quality improvement.

Aim : We hereby report our local experience in implementing colonoscopy QI record through an automatic data extraction from two separate databases, and assess the colonoscopy quality at unit and individual levels.

Methods : We locally adapted a company reporting system for colonoscopy by adding in a dedicated tab, selected procedure indicators. Endoscopic QI data from reporting system database (DB) and pathological results from another DB were extracted and merged together in a separated DB. On a regular period basis or on request, key QI are calculated and extracted. It includes adenoma detection rate (ADR), polyp detection rate (PDR), caecal intubation rate (CIR), quality of bowel preparation (using the Boston bowel preparation scale) and type of sedation. During a first period of 6 months starting in January 2016, endoscopists were encouraged to fulfill the dedicated tab on a voluntary basis. In a second period, fulfillment of QI was turned to be mandatory. The completeness of QI recording was evaluated across both periods. Performance measures of all endoscopists were compared to global results of our department and to published targets.

Results : During the first 6 months “free-filling” period, 1935 colonoscopies were performed with a QI tab fully filled in 63.1% of cases. In medical protocols, the CIR for screening colonoscopy was 93.1%, mean Boston bowel preparation score was 7.2 ± 0.66 , with 87% of cases with adequate preparation (Boston score >5), 94.6% of colonoscopies having been performed under propofol sedation. Among QI data, automatically extracted QI (bowel preparation quality, type of sedation) were filled in the specified QI tab in 99.9% and 97.5% respectively; whereas manually filled QI (progression, number of polyps resected and indication) were filled in the specific tab in 79.6%, 76.6% and 76.3% respectively. During this period, the ADR was 32% (range : 0%-61.3%). The PDR, an indicator that does not need a link to the pathology DB, was 37.7% with a mean of 0.94 polyp resected by colonoscopy. During the 4 months “mandatory-filling” period (July-October 2016), 1161 colonoscopies were performed with a QI tab fully filled (both for automatically and manually filled QI) in 100% of cases (the difference with the first period was statistically significant; $p < 0.0001$). The global CIR for screening colonoscopy was 97.9%. Mean Boston bowel preparation score was 7.2 ± 0.76 with 88% of cases with adequate preparation (90% among outpatients and 83.2% among inpatients). Colonoscopies were performed under propofol sedation in 94.9%. During this second period, the global ADR was 32.9% (range : 0%-66.7%). The PDR was 45.8% with a mean of 1.17 polyp removed by colonoscopy.

Conclusions : This study illustrates that quality indicators for colonoscopy assessment in a Belgian tertiary hospital endoscopy unit could be easily implemented with limited human resources by adapting a company reporting system and link it to the pathology department database. Mandatory filling of QI items is the key for system implementation success. Our results were consistent with goals required by international guidelines. This system allows giving feedback to individual endoscopists for self-performance assessment and might be easily adapted in the future following guidelines updates.

- G11 -

ELECTROMAGNETIC-GUIDED PLACEMENT OF A NASOJEJUNAL TUBE IN PATIENTS RECEIVING FECAL MICROBIOTA TRANSPLANTATION. E. Christiaens (1), T. Holvoet (1), J. De Keukelaere (1), T. Van Hulle (1), M. De Vos (1), H. Van Vlierberghe (1), D. De Looze (1). [1] Ghent University Hospital, Ghent, Belgium, Gastroenterology.

Introduction : Accurate placement of jejunal tubes is required for uncomplicated administration of fecal microbiota in fecal microbiota transplantation (FMT) and for enteral feeding in critically ill patients. Placement of nasojejunal tubes typically occurs endoscopically, with or without fluoroscopic guidance or by blind

Introduction. This placement technique requires high skills, has an irradiation burden and it often fails to get the tube in the ideal position. In this study we tested electromagnetic-guided placement of nasojejunal tubes by means of the CORTRAK® feeding tube system (CORPAK Medsystems corp), in which a real-time display shows the relative position of the tube during placement.

Aim : The aim of this study is to evaluate the accuracy, feasibility and patient compliance of electromagnetic-guided placement of nasojejunal tubes.

Methods : As a sub-study of a double blind, placebo controlled randomized clinical trial evaluating fecal microbiota transplantation in refractory irritable bowel syndrome, we evaluated electromagnetic placement of nasojejunal tubes via the CORTRAK® feeding tube system. We analyzed the success rate (defined as placement of the tube nearby or beyond the duodenojejunal flexure), duration of the procedure and the patient compliance measured by a visual analogue scale (VAS) (“0” being not painful and “10” having the worst possible pain).

Results : Between December 2015 and October 2016, a CORTRAK® nasojejunal tube was placed in 26 patients (13 male and 13 female patients). In sixteen of twenty-six patients, the nasojejunal tube was placed nearby or beyond the duodenojejunal flexure, meaning a success rate of 61%. In the remaining 10 patients the tip of the probe was placed in the second or third part of the duodenum. We were able to pass the pylorus in every patient. The median time of placement was 15 minutes (range 3.3 - 40 minutes) and the median distance of placement from the mouth was 105 centimeters (range 90 - 130 centimeters). There was a median VAS score of 3.5 (range 1 - 8).

Conclusions : This study shows that electromagnetic-guided placement of nasojejunal tubes is feasible, can be achieved in a short period of time and occurs with little patient discomfort. The CORTRAK® feeding tube system is a very promising tool that may replace endoscopic guided placement of nasojejunal tubes.

- G12 -

COLD SNARE POLYPECTOMY FOR ADVANCED, FLAT SESSILE LESIONS. L. Van Overbeke (1), M. Ferrante (2), S. Ilegems (2), L. Mortier (2), J. Van Dongen (2). [1] AZ Sint-Maarten, Mechelen, Belgium, Gastroenterology/*Hepatology, [2] AZ Sint-Maarten, Mechelen, Belgium, Gastroenterology.

Introduction : Cold snare polypectomy (CSP) is an accepted technique to remove diminutive or small (6-9 mm) polyps, with minimal risks of complications. There is less risk of immediate bleeding (beside self-limiting), delayed bleeding or perforation. Here we present a series of CSP for advanced flat sessile polyps (> 10 mm).

Aim : To assess the feasibility and safety of CSP for advanced, flat sessile lesions

Methods : This is a retrospective study, conducted in a non-academic centre. From February to October 2016, 25 patients, with sessile polyps (Paris classification 0-IIa, IIb) estimated > 10 mm, underwent CSP. A rim of normal mucosa around the polyp was attempted to be removed. None of the patients took anticoagulants. One patient was taking low dose aspirine, one patient was taking low dose aspirin which was stopped two days before the colonoscopy, one patient stopped clopidogrel 7 days before the procedure and three patients stopped direct acting oral anticoagulants (DOAC) two days before the colonoscopy.

Results : A total of 28 polyps were removed, 19 in the caecum and ascending colon, 7 in the transverse colon, 1 in the descending colon and 1 in the sigmoid. Sixteen were resected in a piecemeal fashion. The mean size of the polyps was 18.6 mm (10 -40 mm). Pathology showed 9 sessile serrated adenomas, 1 traditional serrated adenoma, 1 carcinoma (originated in a traditional serrated adenoma), 8 tubular adenomas, 4 tubulovillous adenoma and 5 hyperplastic polyps. Immediate oozing bleeding was frequent but in all patients rapid, spontaneous haemostasis occurred without need for haemostatic clipping. In one patient a clip was placed in order to restart a DOAC soon after the polypectomy, and in another patient a preventive clip was placed. There were no delayed bleedings nor perforations. Three patients with piecemeal resection, had follow-up colonoscopy after 6 months which showed no residual polyp tissue.

Conclusions : CSP for advanced, flat sessile lesions (0-IIa, IIb) seems feasible and safe. There were no delayed bleedings and no perforations. Immediate bleeding stopped rapidly and spontaneously, which hypothetically can be explained by less endothelial damage, better endothelial function and better haemostasis than with hot snare polypectomy.

- G13 -

CURRENT UTILIZATION AND DIAGNOSTIC YIELD OF RANDOM COLONIC BIOPSIES IN EVALUATION OF CHRONIC DIARRHEA. X. Zhang (1), S. Kane (2), L. Pestana (3). [1] Mayo Clinic, Rochester, United States (the), Internal Medicine, [2] Mayo Clinic, Rochester, United States (the), Gastroenterology, [3] University of California-Davis, Davis, United States (the), Gastroenterology.

Introduction : Chronic diarrhea affects up to 5 percent of the population. Current guidelines recommend performing random biopsies for evaluation of microscopic colitis if macroscopic colonoscopy evaluation is normal. The yield of random biopsies for microscopic colitis has been shown to be low (10-14%).

Aim : As part of a larger quality improvement program, we aimed to study current practice at our tertiary center as to rationale for why random biopsies were not performed when the indication for colonoscopy was “diarrhea”, and determine the yield of those biopsies.

Methods : Retrospective chart review was performed on all outpatient colonoscopies done for the indication “diarrhea” from October 2012 to May 2014. Pertinent patient information including age and sex, clinical variables (duration of symptoms, number of bowel movements, presence of nocturnal bowel movements, anemia and weight loss), and histology were collected.

Results : Six hundred twenty-one colonoscopies were done for the indication “diarrhea.” There were 425 female patients and 196 male patients. Average age was 47 years (range 17 to 93 years). Chronic diarrhea was documented in 513 cases (82%); acute diarrhea was documented in 59 cases (10%). 94 of patients had documented anemia; 172 patients had documented weight loss. Random biopsies were performed in 613 procedures (98.7%) and not collected in 8 colonoscopies (1.3%). Reasons for not pursuing random biopsies included active diverticulitis (1), wrong indication of procedure (2), procedure aborted due to patient instability (2), acute diarrhea that self-resolved by time of colonoscopy (1), and colonoscopy done for fecal microbiota transplant (2). One hundred forty patients yielded abnormal findings (23%), while 474 showed normal histology (77%). Microscopic colitis was found in 73 cases (12%), collagenous in 29 (5%) and lymphocytic colitis in 44 (7%). Other pathological findings included non-specific acute colitis (11), amyloid (2), CMV colitis (1), graft versus host disease (1), and mycobacterium avium-intracellulare infection (1).

Conclusions : Our study demonstrates high instructional compliance with current guidelines to obtain random biopsies in a history of chronic diarrhea. Random biopsies were not obtained only in instances where there was not a valid clinical indication. In addition, the yield of random biopsies at 12% was congruent with previous studies at other institutions. Continued efforts to educate our non-GI colleagues on proper referral for invasive procedures will hopefully lead to higher yields in the future.

- G15 -

ESOPHAGITIS DISSECANS SUPERFICIALIS : A CASE SERIES OF 7 PATIENTS OF A MISDIAGNOSED ENTITY. E. Fiani (1), F. Guisset (1), Q. Fontanges (2), J. Deviere (1), A. Lemmers (1). [1] Erasme Hospital, Brussels, Belgium, (1) Gastroenterology, Hepatopancreatology and Digestive Oncology department, [2] Erasme Hospital, Brussels, Belgium, (2) Pathology Department.

Introduction : Esophagitis dissecans superficialis (EDS) is a rare desquamative disorder of the esophagus, characterized by sloughing of the superficial mucosa. It is a benign entity of uncertain etiology. Most cases of EDS are idiopathic but can be caused by medications, hot beverages, chemical irritants, celiac disease and many skin conditions.

Aim : Knowing that few case series have described this entity, we decided to review all the cases diagnosed in our center to characterize them.

Methods : The pathological institutional database of Erasme University Hospital (Brussels, Belgium) was searched for the diagnosis of EDS. We reviewed retrospectively the clinical and endoscopic findings as well as histological features of all cases of EDS (table 1). During this period of time, 21500 upper gastrointestinal endoscopies have been performed in our institution.

Results : From 2010 to 2016, we identified 7 cases of EDS diagnosed in our institution in this time period. During the same period, 21500 upper gastrointestinal endoscopies were performed (incidence 0.03%). The median age of presentation was 73 years old, with a female predominance (85%). Associated symptoms were variable from weight loss and nausea to epigastric pain, dysphagia and atypical chest pain. The most common co-morbidity found was treated hypertension in 3 patients. There were no skin diseases in any of these patients. Only one patient in our series had an identified potential causal factor (clindamycin), because of the sudden onset of symptoms upon initiating clindamycin for septic arthritis. Endoscopic findings evoked in 2 patients a suspicion of an esophageal tumor; the first one was described as a raised detached lesion of the distal third of the esophagus with suspicion of squamous cell carcinoma (Figure 1) and the second as a suspected tumor of the proximal third of the esophagus (Figure 2). For other patients, EDS was misdiagnosed as unspecific esophagitis in 3, reflux or mycotic esophagitis in 2. Only one patient was suspected to have sloughing esophagitis. Histologic features present in all of those cases were characterized by the presence of a sloughing and necrosis of the superficial layer of the esophageal squamous epithelium with negative anti HSV and anti CMV antibodies, negative periodic acid Schiff stain for fungal infections as well as absence of signs of dysplasia or signs of malignancy. In 2 patients, there was a presence of multiple bacterial colonies on the superficial epithelium. Acute inflammation was reported in 4 of the patients with the presence of eosinophils in the superficial epithelium described in 2 of these patients and of polymorphonuclear leukocytes in 2 other patients (figure 3). An endoscopic follow up 2 months after PPI treatment (with pantoprazole 40 mg once daily) was performed in 2 patients who had an atypical endoscopic presentation with suspicion of a previous suspicion of esophageal neoplasia. A complete healing of the esophageal lesions was observed in these 2 patients (Figure :4).

Conclusions : EDS is a rare benign entity that endoscopists must be aware of in order not to mistake it for other entities such as esophagitis or squamous cell carcinoma. The diagnosis is based on biopsies. The prognosis is good after stopping the causative agent and with PPI treatment.

- G16 -

MULTIMODAL DETECTION OF HELICOBACTER PYLORI INFECTION AND ANTIBIOTIC RESISTANCE IN A MIXED RURAL BELGIAN AREA. P. Bossuyt (1), S. Gils (2), B. Van Den Poel (2), I. Micalessi (2), P. Cuyle (1), S. Carton (1), P. Christiaens (1), V. Moons (1), A. Van Olmen (1), A. Smismans (2), C. Bourgain (3), J. Frans (2). [1] Imelda Hospital, Bonheiden, Belgium, Department of Gastroenterology, [2] Imelda Hospital, Bonheiden, Belgium, Department of Laboratory Medicine, [3] Imelda Hospital, Bonheiden, Belgium, Department of Pathology.

Introduction : Antibiotic resistance in *Helicobacter pylori* (HP) strains is increasing. Currently the resistance pattern guides the therapeutic decision, mainly based on the occurrence clarithromycin resistance. Real world data on HP prevalence and antibiotic resistance in low-urbanized Belgian areas is lacking. Polymerase chain reaction (PCR) is an emerging technique for the detection of HP infection and antibiotic resistance.

Aim : The aim of the study was to evaluate the prevalence of HP infection and clarithromycin resistance with a qualitative real-time PCR.

Methods : Two hundred three consecutive patients presenting for gastroscopy were included. In every patient three sets of biopsies were taken. Biopsy samples were evaluated microscopically with immunohistochemical (IHC) staining (Dako), by PCR (RIDA®GENE *Helicobacter pylori* (R-Biopharm) and culture (in microaerophilic atmosphere on BD *Helicobacter* agar (BD)). Demographic, endoscopic and therapeutic data were collected.

Results : HP infection was detected in 19, 21 and 18 patients by IHC, PCR and culture respectively. Patients with HP positivity on IHC staining had a mean age of 48 years (range 22-88), 53% were males and 32% of these patients had at least one eradication attempt before. Compared to IHC as golden standard, culture and PCR had a sensitivity of 84% and 89% and specificity of 99% and 98% respectively. Clarithromycin resistance, as detected by PCR or culture, was encountered in 6 out of 21 HP positive samples (29%). When considering samples of therapy naïve patients only, 2 out of 15 (13%) tested resistant. In previously eradicated patients, 4 out of 6 came up as resistant (67%) (Fisher's exact test, $p=0.03$). Results on clarithromycin resistance were generally available in less than one day by PCR compared to at least 7 days by culture.

Conclusions : 1/ Prevalence of HP based on detection with PCR in a mixed rural low-urbanized area in Belgium is 10%. 2/ Overall clarithromycin resistance was 29% : 13% in the naïve group versus 67% in patients that were treated before. Standard triple eradication based on a clarithromycin regime remains first choice in treatment naïve patients. 3/ Macrolide resistance detection by PCR is a fast and sensitive technique compared to culture based methods. Further validation is currently ongoing to evaluate whether PCR can replace HP culture for clarithromycin resistance detection as reference method. At this time there is no reimbursement for molecular detection of HP (resistance) in Belgium.

- G17 -

DIFFERENCES AND SIMILARITIES OF GASTROENTEROLOGY TRAINING ACROSS EUROPE : AN INTERNATIONAL SURVEY. G. Laniro (1), M. Maida (1), L. Alrubaiy (2), T. Bokun (3), T. Bruns (4), V. Castro (5), L. China (6), G. Conroy (7), D. Trabulo (8), C. Van Steenkiste (9), R. Voermans (10), J. Burisch (11). [1] Italian Council of Trainees in Gastroenterology, Italy, [2] British Society of Gastroenterology, United Kingdom (the), [3] Youth Section of Croatian Society of Gastroenterology, Croatia, [4] Working Group "Young Gastroenterology" of the German Society for Digestive and Metabolic Diseases, Germany, [5] Swedish Society of Trainees in Gastroenterology, Sweden, [6] British Society of Gastroenterology, Trainees Section, United Kingdom (the), [7] Association of the French residents in hepatogastroenterology, France, [8] Portuguese Society of Trainees in Gastroenterology, Portugal, [9] Jong Vlaamse Vereniging voor Gastro-enterologie, Belgium, Gastroenterologie en Hepatologie, [10] Dutch Society of Trainees in Gastroenterology and Hepatology, Netherlands (the), [11] Trainee Society of the Danish Society of Gastroenterology and Hepatology, Denmark.

Introduction : There is currently no universal European training program in Gastroenterology and Hepatology. The European Board of Gastroenterology and Hepatology (EBGH) have produced guidance¹ regarding expected competences for European Gastroenterology (GI) trainees but it is unclear as to whether these have been incorporated in national curricula. The last evaluation of gastroenterology training across Europe dates back to 2002. 2

Aim : Our aim was to update the picture of gastroenterology training across Europe to identify differences and similarities between current programs.

Methods : We developed a web-based 90-point questionnaire composed of 5 sections to investigate different aspects of gastroenterology training including : institutional rules, clinical activities, endoscopy, ultrasound, academic activities including scientific research, financial/socio-economic/employment issues and pitfalls of training programs. Physicians in their last year of GI training or who had recently finished their training, from 16 European countries (Belgium, Croatia, Denmark, France, Germany, Greece, Italy, Lithuania, Netherlands, Poland, Portugal, Romania, Russia, Serbia, Sweden, UK), were invited to participate in the survey. In 10/16 (62%) countries, physicians were identified through national societies of GI trainees/young gastroenterologists.

Results : A total of 144 physicians answered the survey (last-year trainees 33%, newly graduated gastroenterologists 77%). Overall, major differences in several aspects of training were identified among all evaluated countries, including access to postgraduate training (local or national application) and its duration. Trainees undertake a final exam to complete their training in 11/16 (69%) countries. A minimum number of procedures is required to graduate in 9/16 (56%) countries. Overall European trainees dedicate a median of 12 months of their training period to endoscopy (IQR 6-25) but only a median of 3 months (IQR 0-6) to ultrasound training. The actual workload of trainees is usually higher than that forecast by training programmes and up to 13% of trainees complete their training without the supervision of a mentor. Overall, 70-89% of trainees performed a total number of diagnostic endoscopic investigations that fulfills the requirements of EBGH. However, large differences were found between and within countries, especially for interventional procedures. Only 52% of trainees have access to pancreatobiliary endoscopy during their training. More than 30% of trainees attend few (<10) academic lessons per year. Approximately 48% of trainees do not receive reimbursement for congress-related expenses. Nearly 66% of trainees dedicate ≤ 10 hours/month to scientific research, and 80% of trainees undertake research during their free time. Average monthly salaries range from 1200-5200€ which differs considerably among Countries. In 12/16 (75%) countries trainees are paid for night duties and maternity leave. Only a minority of

trainees perceive that they are very or fully confident regarding activities and topics related to the specialty of gastroenterology and hepatology. Finally, 86% of trainees believe that GI educational programs should be homogenized across Europe.

Conclusions : In this large survey of senior trainees and newly graduated gastroenterologists, considerable differences in several aspects of GI training programs were found both between and within 16 European countries. Nevertheless, there has been some improvement in convergence and adherence to EBGH requirements since 2002.2 Practical training in ultrasound and interventional endoscopy appear to be still insufficient in most countries. In addition there are significant discrepancies between research opportunities and activities, and support for trainees. Such dissimilarities may lead to disparities in quality of training and, consequently, of healthcare across countries. A higher homogenisation of educational programs and training opportunities across Europe is, therefore, strongly desirable.

References :

- 1) The European Board of Gastroenterology and Hepatology Website—<http://www.eubogh.org>
- 2) Bisschops R, Wilmer A, Tack J. A survey on gastroenterology training in Europe. *Gut*. 2002;50 :724-9.

- G18 -

GASTROINTESTINAL BLEEDING UNDER ANTICOAGULATION THERAPY : SYSTEMATIC REVIEW OF THE REBLEEDING RISK, ITS REVERSIBILITY PROFILE AND RISK STRATIFICATION TO SELECT PATIENTS FOR LEFT ATRIAL APPENDAGE OCCLUSION. A. Hadeifi (1), A. Aminian (2), A. Bondue (3), J. Devière (1), A. Lemmers (1). [1] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Brussel, Belgium, Gastroenterology, Hepatopancreatology and Digestive Oncology Department, [2] CHU de Charleroi, Hôpital Marie Curie, Charleroi, Belgium, Cardiology Department, [3] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Brussel, Belgium, Cardiology Department.

Introduction : Percutaneous left atrial appendage occlusion (LAAO) is increasingly recognized as valid alternative therapy to reduce thrombo-embolic risk in patients with non valvular atrial fibrillation (AF) and contraindications for long term oral anticoagulation (OAC) therapy. Patients at high thromboembolic risk with previous gastrointestinal bleeding (GIB) might be at risk of bleeding recurrence in case of resuming anticoagulation. They could be selected for alternative therapies like LAAO. Up to now, there is no scientific consensus for patient selection for LAAO based on recurrent GIB risk.

Aim : We aimed to review the literature on gastrointestinal (GI) bleeding recurrence and proposed to define the reversibility profile of each lesion in an organ by organ and lesion by lesion approach to stratify the risk of bleeding individually.

Methods : We systematically collected data from both prospective and retrospective studies from pubmed in order to extract rebleeding risk by etiology. The reversibility profile was defined by type of treatment needed to cure the lesion. Low reversibility (LR) profile was defined as a need for heavy treatment (surgery, radiotherapy, embolisation) to cure the lesion or as diffuse lesions.

Results : The most frequent reported causes of bleeding are peptic gastroduodenal ulcer (60%) for upper GI diverticulosis (40%), colitis (20%) and anorectal diseases (20%) for lower GI and angiodysplasia (23%) for the midgut, these latter being responsible for 5% of all GI bleeding causes. The rate of recurrent GI bleeding under OAC therapy is 5-7% with vitamin K antagonists (VKA), an incidence that might increase with direct OAC. Recurrent bleeding rates for upper GIB, lower GIB and obscure GIB are respectively 5-7%, 16%, and 40,3 %. In the upper GI tract, lesions at high risk of bleeding recurrence are Dieulafoy lesions and angiodysplasia with reported rates up to 40% in some series. In the lower GI tract, lesions at highest risk are diverticular disease, angiodysplasia, colitis and radiation proctitis with bleeding recurrence rates reaching 60%, 20%, 40% and 20% respectively. For the midgut, angiodysplasia (20%) and bleeding of unknown origin (20%) are associated to the highest risk of recurrent bleeding. LR profile lesions with high rebleeding risk are present for diffuse angiodysplasia, colonic diverticulosis and Dieulafoy lesions.

Conclusions : In conclusion, GI lesions at high risk of recurrent bleeding with low reversibility profile are infrequent and include in particular : diffuse angiodysplasia, colonic diverticulosis and Dieulafoy lesions. Patients with AF having those lesions with GIB under anticoagulation might be the best candidates for alternative therapies like LAAO. Larger studies are needed to assess the long-term outcome of patients treated by LAAO for GIB under current oral anticoagulant therapies.

**Belgian Society for Paediatric Gastroenterology, Hepatology and Nutrition
BeSPGHAN**

- H01 -

PROSPECTIVE MONOCENTRIC EVALUATION OF THE RESPONSE TO INITIAL HEPATITIS B VIRUS VACCINATION AND REVACCINATION IN CHILDREN WITH CELIAC DISEASE. T. Roussef (1), S. Vande Velde (2), R. De Bruyne (2), M. Van Winckel (3), P. Schelstraete (4), S. Van Biervliet (5). [1] Ghent University Hospital, Gent, Belgium, Pediatrics, [2] Ghent University Hospital, Gent, Belgium, Pediatric Gastroenterology and nutrition, [3] Ghent University Hospital, Gent, Belgium, Pediatric Gastroenterology and nutrition, [4] Ghent University Hospital, Gent, Belgium, Pediatric pneumology and infectious diseases, [5] Ghent University hospital, Ghent, Belgium, Pediatric gastroenterology and nutrition.

Introduction : Celiac disease (CD) is an autoimmune disease characterized by immune mediated inflammatory damage of the small intestinal mucosa, precipitated by the ingestion of gluten-containing foods. Nonresponse following Hepatitis B virus (HBV) vaccine in a healthy population is 4-10% and can partially be explained by genetic predisposition, especially Human leukocyte antigen (HLA) DQ2 and DQ8 alleles seem to play a primary role. It is known that more than 95% of celiac patients possess these HLA genotypes. Consequently, a lower immunisation rate after HBV vaccine is seen in celiac patients.

Aim : The aim of this study is to prospectively map the responses to HBV vaccine in children with CD. We also investigated if there is a relationship between the patients' responses to HBV vaccination and the dietary compliance.

Methods : At the moment of annual follow-up, we performed a blood analysis and measured the anti-hepatitis B surface antibodies (antiHBs AB) in children with CD followed at the pediatric gastroenterology department of the Ghent university hospital, between 2015 and 2016. Subjects with antiHBs AB <10 IU/L were considered non-responders. Non-responders were advised to take a single intramuscular HBV vaccine booster. Response was checked at the next annual appointment. Compliance to gluten free diet (GFD) and CD activity were monitored as usual, using serum anti-transglutaminase antibody levels (a-TG AB). The results were compared to the 4-10% non-response reported in literature.

Results : 71 children with CD were included of which 24% (n=17) were male. The mean age at diagnosis of CD was 6.1 years (range 1-16 years) and 9.5 years (range 3-17 year) at measurement of antiHBs AB. Of the 31 (43.6%) responders to vaccination, 21 (67.7%) showed low response (10-100 IU/L), 8 (25.8%) intermediate response (100-1000IU/L) and 2 (6.5%) a high response (>1000 IU/L). More than half of the patients were non-responders (40 (56.3%)). Until now, for only 13/40 (32.5%) non-responders, antiHBs AB were available after intramuscular revaccination. Of those 53.8% (n=7) acquired immunity after a single HBV booster. The a-TG AB were still positive in 16/71 (22.5%) CD patients. The a-TG AB ranged from 11-392 U/mL (normal value <7 U/mL). Ten of them were non-responders. Control antiHBs AB titre after booster vaccination was available for 3/10. At control all had normal a-TG AB and 2/3 became responders.

Conclusions : Non-responsiveness to HBV vaccination was more frequently found in children with CD compared to the literature reported non-response. Since more than half of the CD patients have an insufficient response to HBV vaccination this should be checked. A single booster injection was able to induce a response in more than 50% of patients. Furthermore, compliance to the prescribed GFD may possibly improve the immune response to HBV vaccination in children with CD.

- H06 -

IMPACT OF TUBE FEEDING ON PULMONARY FUNCTION IN CHILDREN AND ADULTS WITH CYSTIC FIBROSIS (CF) : A REGISTRY STUDY. D. Libeert (1), S. Wanyama (2), D. Declercq (3), M. Thomas (2), F. De Baets (4), S. Van Biervliet (5). [1] Ghent University Hospital, Gent, Belgium, Pediatrics, [2] Wetenschappelijk instituut volksgezondheid - l'Institut Scientifique de Santé Publique., Brussel, Belgium, Belgian CF registry, [3] Ghent University Hospital, Gent, Belgium, CF Centre dietetics, [4] Ghent University Hospital, Gent, Belgium, Pediatric pulmonology, [5] Ghent University Hospital, Ghent, Belgium, Pediatric Gastroenterology and Nutrition.

Introduction : CF patients often struggle with malnutrition due to pancreatic insufficiency and pulmonary infections. There is a known association between nutritional status and pulmonary function in CF patients. Therefore, tube feeding (TF) is often used to maintain nutritional status and improve pulmonary function. It is often used as last resource when nutritional advice and supplements failed. However, the long-term results on pulmonary function outcomes are not yet clear.

Aim : Evaluation of long-term effect of TF on pulmonary function in CF patients.

Methods : This was a registry based, retrospective longitudinal study using data on all patients included in the Belgian CF registry (BCFR) between 2000 and 2013. The registry is a one-point registration of changes made during the preceding year entered by the CF centers. Cases were defined as patients using tube feeding (TFCF). Index year was the year with the first recording of TF. On index year, cases were compared with 2 control patients matched for age, gender, pancreatic status and genotype class (CoCF) recruited from the BCFR. Data from the year before index, at index en 3

years after index were analysed. A long-term longitudinal evaluation of the trends was done. Results are given as median and interquartile range (IQR).

Results : From the 1482 patients in the registry, 113 TFCF and 223 CoCF were included. The Forced Expiratory Volume in 1 second (FEV1%) of CFTF patients was 51.4 (32.7-73.1) compared to 82.7 (65.6-94.3) in CFCo ($p < 0.0001$) at index. No significant changes were observed when comparing FEV1% from year before start TF, index year and 3 years after start (FEV1% : -1y : 49.1% (33.2-73.2); Index : 51.4% (32.7-73.1); +3yrs : 63.7% (42.3-85.3)). However, when looking at the trend in the years before the index, TFCF patients display a significant decrease in pulmonary function (Δ FEV1% -1.52%/year ($p = 0.0017$)). The rapid decline stopped and pulmonary function stabilised at a lower level after TF. In contrast, CoCF patients showed a stable deterioration (Δ FEV1% -0.48%/year) ($p < 0.0001$) throughout the observation period. TFCF had significantly more hospitalisation days during the index year compared to CoCF (30(10-54); 10(0-15) resp.; $P < 0.0001$). Comparing the year before the index year and 3 years after the index year hospitalisation increases significantly towards the index year ($p = 0.0025$) and decreases again 3 year after the index year ($P < 0.0001$) (14 (3-33); 30 (10-61); 15 (0-45) respectively, (no sign. difference between -1yr and +3 yrs)). However, at every evaluation point TFCF patients were more frequently hospitalised than CoCF. TFCF patients received significantly more intravenous antibiotic (IV AB) treatment at any evaluation point. The IV AB treatment decreased from 1 year before until 3 yrs after index year ($p = 0.0156$) (13 (0-46); 6 (0-24); 0 (0-28) resp. (no sign. diff. between -1yr and index yr)) Although TFCF had were more frequently chronically colonized by Burkholderia cepacia complex (BCC) at the index year ($P < 0.02$), there was no significant difference in specific infections or colonisations (Aspergillus, MRSA, Pseudomonas aeruginosa, BCC) between TFCF and CoCF afterwards. Within the 3-year post-index year, there were significantly more transplantations (19 (16.8%); 6 (2.7%) resp.; $P < 0.0001$) and deaths (10 (8.9%); 4 (1.8%) resp.; $P < 0.007$) in the TFCF compared to CoCF.

Conclusions : The pulmonary function stabilised after the start of tube feeding but did not improve drastically. In parallel, the increase in hospitalisation days and IV antibiotic treatment is reversed. The irreversible pulmonary function loss might imply that we react too late in addressing nutritional deficiencies.

- H07 -

IMPACT OF TUBE FEEDING ON NUTRITIONAL STATUS IN CHILDREN AND ADULTS WITH CYSTIC FIBROSIS (CF) : A REGISTRY STUDY. D. Libeert (1), S. Wanyama (2), D. Declercq (3), M. Thomas (2), F. De Baets (4), S. Van Biervliet (5). [1] Ghent University Hospital, Gent, Belgium, Pediatrics, [2] Wetenschappelijk instituut volksgezondheid - l'Institut Scientifique de Santé Publique, Brussel, Belgium, Belgian CF registry, [3] Ghent University Hospital, Gent, Belgium, CF Centre Dietetics, [4] Ghent University Hospital, Gent, Belgium, Pediatric Pulmonology, [5] Ghent University Hospital, Ghent, Belgium, Pediatric Gastroenterology and Nutrition.

Introduction : CF patients often struggle with malnutrition due to pancreatic insufficiency and pulmonary infections. There is a known association between nutritional status and pulmonary function in CF patients. Therefore, tube feeding (TF) is often used to maintain nutritional status. It is often used as last resource when nutritional advice and supplements failed. However, the long-term results on nutritional status are not yet clear.

Aim : Evaluation of long-term effect of TF on nutritional status in CF patients.

Methods : This was a registry based, retrospective longitudinal study using data on all patients included in the Belgian CF registry (BCFR) between 2000 and 2013. The registry is a one-point registration of changes made during the preceding year entered by the CF centers. Cases were defined as patients using tube feeding (TFCF). Index year was the year with the first recording of TF. On index year, cases were compared with 2 control patients matched for age, gender, pancreatic status and genotype class (CoCF) recruited from the BCFR. Data from the year before index, at index en 3 years after index were analysed. A long-term longitudinal evaluation of the trends was done. Results are given as median and interquartile range (IQR).

Results : From the 1482 patients in the registry, 113 TFCF and 223 CoCF were included in this study. At index, TFCF patients displayed a worse nutritional status than CoCF, which was reflected in a smaller stature (Height z-score : -1.1 (-1.9- -0.5); -0.4 (-1.2- 0.2) resp. ($p < 0.0001$)) and a worse BMI (BMI z score : -1.5 (-2.4- -0.7); -0.4 (-1.1- 0.2) resp. ($p < 0.0001$)). Comparing the BMI z-score 1 year before, at index and 3 years after a significant increase is observed ($p = 0.0012$) (BMI z-score : -1yr : -1.8 (-2.5- -1); index : -1.5 (-2.4- -0.7); +3yrs : -1.1 (-2- 0.3) (no sign. Diff. between -1yr and index)). However, TFCF patients did not normalise their BMI z-score and remained significantly thinner than CoCF patients ($p < 0.0001$). Looking at the BMI trend, TFCF patients displayed a decline in BMI before Introduction of ETF (Δ BMI z-score (SDs) : -0.024 (0.018)). A small non-significant recuperation was observed during index year (Δ BMI z-score : 0.1 (-0.4 - 0.8)) and there was a further significant improvement during the following 3 years, whereas CoCF had a stable BMI evolution. The height z-score for the children remained at a lower level compared to CoCF and did not change over time (H z-score : -1yr : -0.7 (-1.9- -0.4); index : -1.1 (-1.9- -0.5); +3 yrs : -1 (-1.9- -0.4) (n.s.)) At the index year and in the years before, there was no significant difference in the presence of CF related diabetes (CFRD). However, in the 3 years post index, TFCF cases developed more frequently CFRD ($n = 13$ (11.5%), $n = 11$ (4.9%) resp. $P < 0.03$).

Conclusions : Tube feeding restored BMI towards the original curve, which was still lower than the CoCF curve. However, it did not improve growth. In the years after the start of tube feeding, significantly more TFCF patients develop CFRD. Questions remain whether tube feeding speeds up the development of CFRD or that patients with impaired glucose tolerance become more malnourished and this influences the start of tube feeding.

- H08 -

STATE OF HYDRATION AFTER SPORTS IN OBESE CHILDREN BEFORE AND AFTER WEIGHT LOSS. J. Baert (1), S. Van Biervliet (2), J. Vande Walle (3), A. De Guchteneere (4) / [1] Ghent University, Ghent, Belgium, Pediatrics, [2] Ghent University, Ghent, Belgium, Pediatric Gastroenterology and Nutrition, [3] Ghent University, Ghent, Belgium, Pediatric Nephrology, [4] Zeepreventorium, De Haan, Belgium, Pediatric Nephrology.

Introduction : Obese adults have a higher dehydration risk after sports than healthy adults. The latter could not yet be confirmed in obese children.

Aim : The effect of a standardized slimming program on sport-induced dehydration and the renin-aldosterone system (RAA-system) activity in obese children was evaluated in this study.

Methods : Sixty-six obese children (BMI z-score $2,52 \pm 0,32$, aged 15 ± 1 years, blood pressure $135 / 79 (\pm 16 / \pm 9)$ mmHg) following a 1 year residential slimming program were included. Twenty-eight stopped the program prematurely. At the start and the end of the program urine samples for sodium, chloride, potassium, urea, creatinine, protein and osmolality, weight, blood pressure and pulse were collected before and after a Cooper-test.

Results : After 1 year, all clinical parameters in rest decreased significantly (BMI z-score $1,52 \pm 0,43$; blood pressure $121 / 71 (\pm 13 / \pm 10)$ mmHg). In rest the percentage of urinary potassium over the sum of urinary sodium (UNa) and urinary potassium (UK/(UNa+UK) (%)) increased significantly from $40 \% \pm 11$ to $50 \% \pm 11$ at the end of the program. After the Cooper-test only non-obese patients displayed a significant UK/(UNa+UK) (%) increase (49 ± 11 ; 56 ± 12 respectively) ($p < 0,01$) as well as an increase in UNa over urinary creatinine ($0,12 \pm 0,07$; $0,1 \pm 0,05$ respectively) ($p < 0,05$).

Conclusions : There was a significant weight loss after sports at the 2 test periods, associated with significant dehydration. Normalizing the BMI after the program resulted in a significant higher aldosterone-effect (UK/(UNa+UK)), which confirms the re-appearance of a normal functioning RAA-system.

- H09 -

CHARACTERISTICS AT BASELINE OF TUBE FED CYSTIC FIBROSIS (CF) PATIENTS WITH MATCHED CONTROLS : A REGISTRY STUDY. D. Libeert (1), S. Wanyama (2), D. Declercq (3), M. Thomas (2), F. De Baets (4), S. Van Biervliet (5) / [1] Ghent university hospital, Ghent, Belgium, Pediatrics, [2] Wetenschappelijk Instituut Volksgezondheid - l'Institut Scientifique de Santé Publique. Brussel, Belgium, Belgian CF registry, [3] Ghent University Hospital, Ghent, Belgium, CF Centre dietetics, [4] Ghent University Hospital, Ghent, Belgium, Pediatric Pulmonology, [5] Ghent University hospital, Ghent, Belgium, Pediatric Gastroenterology and Nutrition.

Introduction : CF patients often struggle with malnutrition due to pancreatic insufficiency and pulmonary infections. There is a known association between nutritional status and pulmonary function in CF patients. Therefore, tube feeding (TF) is often used to maintain nutritional status. It is often used as last resource when nutritional advice and supplements failed. However, the long-term results are not unequivocal.

Aim : Evaluate the differences at start between patients receiving tube feeding and controls.

Methods : This was a registry based, retrospective longitudinal study using data on all patients included in the Belgian CF registry (BCFR) between 2000 and 2013. The registry is a one-point registration of changes made during the preceding year entered by the CF centers. Cases were defined as patients using tube feeding (TFCF). Index year was the year with the first recording of TF. On index year, cases were compared with 2 control patients matched for age, gender, pancreatic status and genotype class (CoCF) recruited from the BCFR. Results are given as median and interquartile range (IQR).

Results : From the 1482 patients in the registry, 113 received TF during the study period and 223 CoCF were selected. Three included controls dropped out due to death during the follow-up period. Median age at introduction was 10.3 years (1.3-18.4) in TFCF and 9.9 (2.1-18.6) in CFCo. The duration of TF was 2 yrs (1-5 yrs). The use of TF in the CF population markedly decreased over the investigation period. Before 2006, 3.7-5.3% of the CF-population used TF whereas only 1.8-2.2% did afterwards. TFCF patients were diagnosed earlier (0.2 yrs (0-1.2) vs 0.3 yrs (0.1-2) respectively ($P < 0,02$)). Their nutritional status was significantly worse, as one would expect (BMI z-score $-1.3 (-2.4 - -0.2)$ vs. $-0.4 (-0.9 - 0.3)$ resp. ($P < 0,0001$)). They were also significantly shorter (Height z-score $-1.1 (-1.9 - -0.5)$ vs. $-0.4 (-1.2 - 0.4)$ resp. ($P < 0,0001$)). Their pulmonary function was significantly worse (FEV1% : $51.4\% (32.7-73.1)$; $82.7\% (65.6-94.3)$ resp. $P < 0,0001$). However, even the first entry of pulmonary function in the registry was

significantly lower in the later on TFCF patients (FEV1% : 67.6% (47.4- 85.3) vs. 84.4% (68 – 99.7) P<0.0001). They were hospitalised more frequently during the index year (30 days (10-61) vs 1 d (0-12) resp. (P<0.0001)). There was no difference in prevalence of chronic pseudomonas or MRSA infection. However, TFCF were more frequently colonized by Burkholderia Cepacia complex (4 (3.5%) vs. 1 (0.4%) resp. (P<0.05)). No difference in the presence of CF related diabetes was discerned.

Conclusions : At index year patients starting tube feeding had evidently significantly worse nutritional and pulmonary status leading to an increased hospitalization compared to matched controls. Noteworthy is the fact they had already a worse pulmonary function from the first entry in the registry.

- H10 -

THE PREDICTIVE VALUE OF COLON TRANSIT TIME AND ANORECTAL MANOMETRY IN THE APPROACH OF FECAL CONTINENCE IN CHILDREN WITH SPINA BIFIDA. C. Daeze (1), S. Vande Velde (2), A. De Guchteneare (3). [1] Ghent University, Ghent, Belgium, Pediatrics, [2] Ghent University, Ghent, Belgium, Pediatrics, Gastroenterology, [3] Zeepreventorium, De Haan, Belgium, Pediatrics.

Introduction : Only a minority of children with spina bifida (SB) achieve spontaneously fecal continence. Despite adequate bowel management, a substantial part of them still suffers from fecal incontinence, which is associated with major psychological problems. At the present, there is no standard protocol to predict whether these children will achieve continence spontaneously, nor what the designated treatment is for bowel management.

Aim : The aim of the current study is to analyze colon transit time (CTT) and anorectal manometry (ARM) in children with SB as a predictor of spontaneous fecal continence.

Methods : SB patients (2.5-7 years old), followed at the SBRC at the Ghent University Hospital, who underwent a CTT study and/or ARM before starting bowel management, were asked to participate in this retrospective study. A questionnaire about the presence of constipation or fecal incontinence was completed. Constipation was present if ≥ 2 of the Rome III criteria for pediatric functional constipation were fulfilled. Fecal incontinence was defined as fecal loss more than once a month in children >4 years old. Normal values for CTT were based on the results of a study by Vande Velde. [1] Sixteen age and sex-matched controls were selected from this population. Normal values for ARM were based on the results of a study by Kumar et al. [2] Total and segmental CTT was measured using a 6-day method, as described by Abrahamsson. [3] ARM was performed in non-sedated children with a water-perfused latex-free catheter. Ethical approval was obtained (2016/0841).

Results : Twenty-two patients were studied. They all had a CTT study, and seventeen had agreed to ARM. 10/22 patients (45.5%) suffered from constipation, according to Rome III criteria. 5/22 patients (22.7%) became spontaneously continent, 10/22 (45.5%) became pseudo continent with bowel management, and 7/22 (31.8%) remained incontinent. SB patients had a significant longer CTT compared to healthy controls ($p=0,001$), which was mainly due to difference in the left CTT ($p=0,037$) and the rectosigmoidal CTT ($p=0,007$). As expected, constipated SB patients had a significant longer CTT than non-constipated SB patients ($p=0,000$). There was also a significant difference in CTT according to continence status ($p=0,001$). In the group with an abnormal CTT study, 10 patients had undergone ARM, which was abnormal in 6 cases and normal in 4 cases. None of these patients developed continence spontaneously. In case of a normal CTT study (10 patients), 7 had undergone ARM. Four children had a normal resting pressure, they all gained continence spontaneously. The three children with abnormal resting pressure rested incontinent. Comparing the resting pressure in the spontaneous continence group with the incontinent patients, there was no significant difference ($p=0,156$), but this could be due to the small group number of spontaneous continent children.

Conclusions : This study confirms that, in case of a normal CTT and normal resting pressure, spontaneous continence is to be expected. If CTT is abnormal, independent of the result of ARM, there will be need for bowel management in order to develop pseudo-continence. In these cases, ARM is not a designated examination.

References :

1. Velde, S.V., et al., Colon transit time in healthy children and adolescents. *Int J Colorectal Dis*, 2013. 28(12) : p. 1721-4.
2. Kumar, S., et al., Manometric tests of anorectal function in 90 healthy children : a clinical study from Kuwait. *J Pediatr Surg*, 2009. 44(9) : p. 1786-90.
3. Abrahamsson, H., S. Antov, and I. Bosaeus, Gastrointestinal and colonic segmental transit time evaluated by a single abdominal x-ray in healthy subjects and constipated patients. *Scand J Gastroenterol Suppl*, 1988. 152 : p. 72-80.

- H11 -

CLINICAL IMPACT OF INTENSIVE NUTRITIONAL SUPPORT IN PATIENTS TREATED WITH CHEMORADIOTHERAPY FOR LOCALLY ADVANCED ESOPHAGEAL CANCER. D. Persyn (1), K. Plovie (2), M. Abasbassi (3), M. Cool (4), G. Deboever (4), G. Lambrecht (5). [1] KU Leuven, Belgium, Internal Medecine, [2] AZ Damiaan,

Oostende, Belgium, Clinical Nutrition, [3] AZ Damiaan, Oostende, Belgium, Abdominal Surgery, [4] AZ Damiaan, Oostende, Belgium, Gastro-enterology, [5] AZ Damiaan, Oostende, Belgium, Department of Gastroenterology.

Introduction : The prevalence of malnutrition is high in patients with esophageal cancer. The main mechanisms involved in weight loss are a decreased nutrients intake and an alteration in metabolism due to a cytokine-driven inflammatory status. Malnutrition is a risk factor for a poor compliance to chemotherapy and radiation therapy and finally for the oncologic outcome. There is scientific evidence that frequently both conditions exist, but in the advanced stages of disease metabolic alterations play a major role and are responsible for the poor response to nutritional support.

Aim : We retrospectively investigated the effect of intensive nutritional support (INS) on nutritional status in patients treated with chemoradiotherapy (CRT) for locally advanced esophageal cancer in a curative setting.

Methods : During a two-year period 13 patients with locally advanced esophageal cancer were treated pre-operatively with CROSS or Herskovic regimen. They were all screened with NRS-2002 and received INS before surgery. Nutritional support was delivered with oral nutrition support, enteral nutrition (PEG or surgical jejunostomy) and immunonutrition peri-operatively. Weight loss and albumin was recorded before and after completion of CRT. Dose reductions and admissions during CRT or in the pre-operative period were recorded.

Results : The time relaps of the CRT and the surgery was mean 6 weeks. 7 patients had an NRS score of 3 or more. 5 patients had a non-significant drop in albumin level. Mean weight loss was only 4 %. Only one patient received a dose reduction due to haematological toxicity. 1 patient needed to be admitted and treated for pneumonia. 10 patients received enteral nutrition : 6 gastrostomy and 4 surgical jejunostomy. 11 patients received immunonutrition in the peri-operative period.

Conclusions : In a small retrospective study of patients treated intensively with CRT for esophageal cancer before surgery, we managed to have a positive effect on nutritional status with INS. Weight loss was reduced and INS seems to reduce major side effects due to CRT. There is a need for prospective trials to objectivate not only the nutritional parameters but also the impact of INS on the oncological outcome in esophageal cancerpatients in a curative setting.

References :

- Herskovic A, Martz K, Al-sarraf M, et al. Combined chemotherapy and radiotherapy compared with radiotherapy alone in patients with cancer of the esophagus. *N Engl J Med.* 1992;326(24) :1593-8.
- Van hagen P, Hulshof MC, Van lanschot JJ, et al. Preoperative chemoradiotherapy for esophageal or junctional cancer. *N Engl J Med.* 2012;366(22) :2074-84.
- Cong MH, Li SL, Cheng GW, et al. An Interdisciplinary Nutrition Support Team Improves Clinical and Hospitalized Outcomes of Esophageal Cancer Patients with Concurrent Chemoradiotherapy. *Chin Med J.* 2015;128(22) :3003-7.

- H12 -

COST ANALYSIS OF LONG-TERM PARENTERAL NUTRITION FOR BENIGN INDICATIONS. E. Canovai (1), L. Ceulemans (1), G. Peers (2), L. De Pourcq (3), M. Pijpops (3), M. Stas (1), I. Hoffman (4), G. De Hertogh (1), M. Hiele (1), J. Pirenne (1), T. Vanuytsel (1). [1] UZ Leuven, Leuven, Belgium, Leuven Intestinal Failure and Transplantation Center, [2] UZ Leuven, Leuven, Belgium, Financial Department - Medical Administration, [3] UZ Leuven, Leuven, Belgium, Pharmacy Department, [4] UZ Leuven, Leuven, Belgium, Pediatric Department.

Introduction : The primary treatment for long-term intestinal failure is home parenteral nutrition (HPN). It is known that HPN significantly reduces costs compared to in-hospital administered parenteral nutrition. Nevertheless, the total financial burden remains significantly high related to the cost of HPN itself and the continued need for treatment of the underlying disease (UD) and concomitant conditions. It is unknown what the specific costs are for each of these three categories and how the costs evolve over the years.

Aim : To evaluate the cost dynamics of HPN care in a cohort of stable, long-term intestinal failure patients (> 2 years).

Methods : We performed a retrospective analysis of our long-term (>2 years) HPN population for benign indications that commenced between 2000 and 2013. Patients with complicated intestinal failure on the waiting list for intestinal transplantation were excluded from this analysis. Demographics, underlying disease, HPN formulation, admissions, complications and mortality data were recorded. Total costs were calculated from data collected from the hospital pharmacy, financial department of the hospital and home-care companies. These included all the costs of admissions, diagnostics, treatments, outpatient clinics, home-nursing care, medication, HPN material and HPN education. The costs were tabulated per patient and all costs were assigned by cause (HPN related, UD related or unrelated). All data from patients were included until 31/12/2015.

Results : A total of 40 long-term patients, 37 adults (24 females, mean age 58.6 years (range 34-85)) and 3 children (1 female, mean age 11 years (range : 6-16)), were included in the study. The median duration of HPN was 5.3 years (range 2.1-15.1). The main indications for the adults were short bowel (59%) and intestinal dysmotility (32%). All pediatric patients had congenital, extensive small bowel mucosal disease (Tufting enteropathy, Trichohepatoenteric syndrome and Beyler's Disease). A large majority of our patients had at least partial nursing assistance at home (91%). On average, HPN was administered 4.5 days per week (range : 1.5-7), 6461 kcal per week (2270-11290) and 6.1 liters per week (2-

14L). Catheter related blood stream infection rate was low at 0.79/1000 catheter days (Community acquired : 0.61 vs Hospital acquired 0.18). For adults, the median total cost of the first year of HPN was €83 503 (€ 35 364 - € 256 780). In this first year, HPN and associated complications accounted for 69% of costs (€ 57 866, range : €40 433- € 127 960) vs 27% for UD related costs (€ 22 505, range : € 401 - € 194 766) and 4% for unrelated costs (€ 3 133, range : € 1 157 - € 20 794). The total cost dropped by 15% in the second year to € 71 311 (€ 31 955 - € 136 657) and HPN related costs remained the predominant factor (78%). This reduction was mostly due to fewer admissions (year 1 : 2.19 vs year 2 : 1.03 admissions per patient per year) and fewer HPN complications (year 1 : 23 complications in 17 patients, year 2 : 15 complications in 5 patients, year 3 : 3 complications in 3 patients). The costs dropped further over time, as year 5 was 40% cheaper compared to year 1 (€ 58 186 vs 83.503). The cost of HPN itself (bags, ancillary material, education and nursing care) for adults was € 39 031 (€14 131 - € 60 775) in the first year and dropped by 14% in the fifth year. The cost of HPN related complications was € 17 585 (0 - € 86 718) in the first year and accounted for 31% of HPN costs and dropped to 16% in the second year (€ 7 033; € 0 - € 81 452). The costs in pediatric patients were much higher (median first year cost : € 255 467) and more evenly split between UD (51%) and HPN (46%) when compared to adults. This was due to higher cost of underlying disease and higher PN requirements.

Conclusions : In adults, HPN related costs accounted for the majority of the healthcare expenses. The overall costs in children were much higher and UD accounts for a higher percentage of total costs. The costs gradually declined each year due to reduction in complications and admissions. Complications most frequently occurred during the first year after start of HPN and led to significant additional costs.

- H13 -

LONG-TERM RESULTS OF HOME PARENTERAL NUTRITION FOR BENIGN INDICATIONS : A SINGLE CENTER EXPERIENCE. E. Canovai (1), L. Ceulemans (2), M. Laurent (3), L. De Pourq (4), M. Pijpops (4), M. Stas (2), G. De Hertogh (2), M. Hiele (2), J. Pirenne (2), T. Vanuytsel (2). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Leuven Intestinal Failure and Transplantation center, [2] UZ Leuven, Leuven, Belgium, Leuven Intestinal Failure and Transplantation center, [3] UZ Leuven, Leuven, Belgium, Geriatric Department, [4] UZ Leuven, Leuven, Belgium, Pharmacy Department.

Introduction : The primary treatment for intestinal failure still remains home parenteral nutrition (HPN). If intestinal rehabilitation is not achieved within 2 years after the start of HPN, the patient will most likely depend upon lifelong HPN therapy. The aim of this study was to evaluate the incidence and treatment of complications of HPN dependent patients at our tertiary care center.

Aim : To evaluate the results of HPN care in a cohort of long-term intestinal failure patients (> 2 years).

Methods : We performed a retrospective analysis of our long-term, adult HPN population (>2 years) for benign indications that commenced their therapy between 2000 and 2013. Patients were included until 31/12/2015 and analyzed data were : demographics, HPN formulation, underlying disease, complications and survival.

Results : A total of 40 adult patients (26 females, mean age 57 years (range 26-85)) were included in the study. The mean HPN dependency was 6.6 years (range : 2.1-15.1). HPN indications were short bowel syndrome (SBS) in 23 patients (58%), intestinal dysmotility in 13 patients (33%), mechanical obstruction in 3 patients (8%) and extensive small bowel mucosal disease in 1 patient (3%). HPN was administered by tunneled catheters in 82% of cases and implantable ports in 18%. 91% of patients had at least partial nursing assistance at home. On average, HPN was administered 4.4 days per week (range 1.5-7) at 6223 kcal per week (2270- 11970). Catheter related blood stream infection (CRBSI) was the most frequent HPN related complication at a rate of 0.76/1000 catheter days (Community acquired : 0.59 vs Hospital acquired 0.17). A total of 70 CRBSI occurred in 26 patients. Bacterial infections accounted for 87% and fungal infections for 13%. The majority of bacterial infections were caused by Staphylococcus species : S. Epidermidis (43%), S. Aureus (14%) and other staphylococci (8%). In the dysmotility group 85% of patients had at least one CRBSI versus 57% of SBS patients. There were 12 clinically significant occlusions of catheters (0.12/1000 catheter days), of which 8 could be solved conservatively using a urokinase lock while the other 4 needed to be replaced. There were 30 other catheter related problems that required treatment such as ulcerations of subcutaneous ports (n=9), exit site infections (n=5) and line fractures (n=16). 15 patients (38%) initially had an implantable device but switched to tunneled catheters during their HPN care following complications (infections, ulcerations). Intestinal failure associated liver disease (IFALD) developed in 38% of patients (n=15) which was reversed in all but one case by lowering the amount of lipids or changing to third generation lipids. No patients evolved to cirrhosis or liver failure. Eight patients (21%) developed metabolic bone disease with decreased bone mass density on Dual X-ray Energy Absorptiometry. Overall, 5- and 10-year survival were 90.1% and 69.5% respectively. For the SBS group, the 5- and 10-year survival were 84.5% and 68.7%. For the dysmotility group survival was 100% and 83.3% respectively. Eight patients died during follow-up, 3 due to the underlying illness and 5 from unrelated causes. No deaths were related to HPN. Three patients were weaned off HPN completely after 4.4 to 4.6 years of HPN-dependency (2 dysmotility syndrome and 1 with SBS). Two patients (5%; 1 male of 29 and 1 female of 26 years old) underwent isolated intestinal transplantation due to severe HPN related complications (recurrent CRBSI) in both and intractable symptoms of intestinal pseudo-obstruction in the female patient.

Conclusions : In our long-term HPN cohort, the incidence of CRBSI is low but tends to occur more frequently in dysmotility patients. Active screening for IFALD and metabolic bone disease, which were common in this cohort, is warranted to prevent further deterioration. The 5- and 10-year survival rates were in accordance with literature and no patient died because of HPN related problems. The long-term success of HPN relies primarily on a structured, multidisciplinary approach with early detection and prompt treatment of HPN-related complications. Intestinal transplantation was performed in 5% of our long-term HPN population.

SUCCESSFUL DOSE DE-ESCALATION TO ADALIMUMAB 40MG EVERY THREE WEEKS IN PATIENTS WITH CROHN'S DISEASE. S. Van Steenberghe (1), S. Bian (2), V. Ballet (1), S. Vermeire (1), G. Van Assche (1), A. Gils (2), M. Ferrante (3). [1] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology, [2] KU Leuven, Leuven, Belgium, Laboratory for Therapeutic and Diagnostic Antibodies, [3] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology.

Introduction : Although dose escalation is widely used to optimize anti-TNF therapy in case of relapse, less is known about possibilities to de-escalate therapy in patients with Crohn's disease (CD) who are in clinical remission. Dose de-escalation may not only have beneficial economic consequences, it may possibly also decrease adverse events.

Aim : In this retrospective study, the outcome of dose de-escalation to adalimumab (ADM) 40 mg every three weeks (ETW) in patients with CD was studied.

Methods : Out of 703 patients treated with ADM for moderate-to-severe CD in a tertiary referral center, we selected all patients who had received maintenance therapy with ADM 40 mg ETW with serum levels (SL) available before and after dose de-escalation. A sex- and age-matched control group consisted of patients continuing ADM 40mg EOW. ADM SL were measured using RIDASCREEN® monitoring kit (R-biopharm AG). In addition, patient reported outcome (PRO2), C-reactive protein (CRP) and serum albumin were collected. Other baseline variables included disease behavior, disease location, smoking behavior, concomitant therapy, bodyweight, and body mass index. ROC curve analyses were performed to define cut off values for continuous variables. Mann-Whitney U, Wilcoxon Signed Rank test, and Cox regression were performed using SPSS 23.0

Results : We identified 40 patients (11 male, median age 37 years) who dose de-escalated to ADM 40 mg ETW for ADM-related AE (n=1), ADM SL >7µg/mL (n=8), or both (n=31). Most frequently reported AE were skin manifestations (47%), arthralgia (22%) and frequent infectious episodes (22%). Compared to the control population, ADM SL dropped significantly within four months, but without associated clinical or biochemical changes. During a median follow-up of 24 months, 65% of patients maintained clinical response, but 35% needed dose escalation back to ADM 40mg EOW because of clinical relapse (n=8), ADM SL dropping to <4µg/mL (n=2), or both (n=4). CRP <3.5mg/L at dose de-escalation was independently associated with dose escalation free survival [Odds ratio 6.28 (95%CI 1.83-21.59), p=0.004]. We could not define a minimal ADM SL to consider or maintain dose de-escalation. In 53% of 32 patients dose de-escalation was associated with a complete disappearance of AE and this after a median of 4 months (8/15 skin manifestation, 3/7 arthralgia, 5/7 frequent infectious episodes). Study population (n=40) : Median (IQR) ADM serum level : T0 12.0 (9.4-14.4) µg/mL – T1 7.9 (5.8-10.7) µg/mL (p<0.001) Median (IQR) C-reactive protein : T0 1.4 (0.6-3.5) mg/L – T1 1.3 (0.6-5.1) mg/L (p=0.188) Median (IQR) serum albumin : T0 44.0 (42.2-46.9) g/L – T1 43.7 (41.5-47.3) g/L (p=0.533) Median (IQR) PRO2 : T0 0.0 (0.0-6.0) – T1 2.0 (0.0-8.5) (p=0.027) Control group (n=40) : Median (IQR) ADM serum level : T0 9.6 (8.1-11.1) µg/mL – T1 10.1 (8.0-11.7) µg/mL (p=1.000) Median (IQR) C-reactive protein : T0 1.2 (0.4-3.0) mg/L – T1 1.9 (1.0-3.6) mg/L (p=0.020) Median (IQR) serum albumin : T0 44.9 (43.0-47.1) g/L – T1 45.3 (43.3-47.1) g/L (p=0.876) Median (IQR) PRO2 : T0 0.0 (0.0-4.8) – T1 2.0 (0.0-7.0) (p=0.007)

Conclusions : In this retrospective cohort analysis, 65% of patients were able to continue ADM therapy at a dose of 40 mg ETW for a median of 24 months. Furthermore, in half of the patients who experienced ADM related AE at baseline, the AE disappeared completely. Regardless of ADM serum levels, disease remission should be objectively assessed prior to dose de-escalation, since an elevated baseline CRP predicted relapse following de-escalation with subsequent need for increase of ADM dose.

EARLY FIBROSTENOSIS IN CROHN'S DISEASE IS ASSOCIATED WITH MULTIPLE SUSCEPTIBILITY LOCI ON IMMUNOCHIP ANALYSIS. T. Holvoet (1), P. Bossuyt (2), I. Cleynen (3), I. De Kock (4), P. Hindryckx (1), S. Vermeire (3), D. Laukens (1), M. De Vos (1). [1] Ghent University Hospital, Ghent, Belgium, Gastroenterology, [2] Imelda Hospital, Bonheiden, Belgium, Imelda GI Clinical Research Centre, [3] University Hospitals Leuven, Leuven, Belgium, Gastroenterology, [4] Ghent University Hospital, Ghent, Belgium, Radiology.

Introduction : Fibrostenosis is a common complication of Crohn's disease (CD) occurring in about one third of patients. Although the pathophysiology of intestinal fibrosis is incompletely understood, evidence suggests a genetic contribution. Previous genetic association studies and candidate gene studies with fibrostenotic CD were based on clinical definitions which lack both sensitivity, specificity and have a high inter-observer disagreement. Additionally, the recent genotype-phenotype analysis by the IIBDGC did not consider the time to development of fibrostenotic disease.

Aim : As the genetic risk may be more important in patients with early fibrostenosis, in this study we aimed to identify novel genetic markers by focussing on early fibrostenotic disease.

Methods : In this multicenter, retrospective nested case-control study, performed at the University Hospitals of Ghent and Leuven, computed tomography (CT) and magnetic resonance imaging (MRI) from CD patients obtained between 2002 and 2016 were examined for the presence of ileal fibrostenotic disease. Patients with early fibrostenosis, defined as the presence of bowel thickening with luminal narrowing and prestenotic dilatation on CT/MRI occurring within 5 years following diagnosis of ileal or ileocolic disease (Montreal L1 or L3), and with available Illumina ImmunoChip data were included. The control cohort consisted of inflammatory CD patients, also Montreal L1 or L3, without arguments for fibrostenotic disease after min 10 years follow up. Allelic association was assessed using the PLINK v1.07 software.

Results : In total, 3,024 CT or MRI scans of 2,042 CD patients were screened. 112 patients were selected because of positive arguments for fibrostenosis occurring within 5 years of diagnosis. Of these, ImmunoChip data were available in 60 cases, and 49 (82%) had confirmed stenosis by histopathology. 343 inflammatory CD controls with genotype data were included in the analysis. Of the 156,500 SNPs analysed, only rs35223850 in the MIS18BP1 gene passed genome-wide significance level for association with early fibrostenosis (P -value $< 3.3 \times 10^{-7}$, OR 3.9, 17.5% vs 5% in cases vs controls respectively). The protein encoding MIS18BP1 is known to bind to the SP1 transcription factor, and has been associated with cardiac, liver and kidney fibrosis. Five additional SNPs reached a statistically suggestive significance level of $P < 5 \times 10^{-6}$, including rs116630177 in the IL23R gene, which is of particular interest as this gene has previously been associated with systemic sclerosis.

Conclusions : This carefully phenotyped study reveals an important role for genetic contribution to early development of fibrostenotic complications in CD. Our data suggest a role for MIS18BP1 and the SP1 transcription factor as well as the IL23 pathway in the pathogenesis of early intestinal fibrosis.

- I03 -

THERAPEUTIC MANIPULATION OF THE GUT MICROBIOTA THROUGH DIET TO REDUCE INTESTINAL INFLAMMATION : RESULTS FROM THE FIT TRIAL. J. Sabino (1), S. Vieira-Silva (2), K. Machiels (3), M. Joossens (4), G. Falony (4), M. Ferrante (3), G. Van Assche (3), S. Van Der Merwe (5), C. Matthys (6), J. Raes (4), S. Vermeire (3). [1] KU, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] KU, Leuven, Belgium, Department of Microbiology and Immunology, REGA institute, [3] KU Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [4] KU Leuven, Belgium, Department of Microbiology and Immunology, REGA institute, [5] KU Leuven, Belgium, Department of Hepatology, University Hospitals Leuven, [6] KU Leuven, Belgium, Clinical and Experimental Endocrinology and Department of Endocrinology.

Introduction : The intestinal microbiota is implicated in the pathogenesis of several immune-mediated disorders including inflammatory bowel diseases and has subsequently been the target of different therapeutic interventions, such as faecal microbiota transplantation, pro-, pre- and antibiotics.

Aim : We designed the Food influence on the Intestinal microbioTa (FIT) trial to study the effects of diet on intestinal microbiota changes and inflammation in healthy individuals (part 1) and patients with ulcerative colitis (part 2). We here report the results of the first part of the study.

Methods : The FIT diet consists of a semi-vegetarian diet, high in fiber (> 30 g/day), low in saturated fat and sulphites and exclusion of added sugar, processed foods, carrageenan, and polysorbate-80. Following informed consent, 29 volunteers followed the diet for 1 month and were followed up for 6 months. Faecal calprotectin was measured on fresh faecal samples (Bühlmann ELISA). Dietary compliance was followed with food frequency questionnaires and 3-day food records. 16S rDNA paired-end sequencing targeting the V4 hypervariable region was performed using Illumina MiSeq sequencer. Sequencing depth was downsized to 10000 reads/sample. The RDP classifier was used for taxonomic annotation. Statistical analyses were performed with R.

Results : A significant weight loss was observed after 4 weeks following the FIT diet (t-test, $p < 0.0001$, mean -2.3 Kg, SD -1.5). Strikingly, faecal calprotectin - although within normal ranges in all but 1 individual - significantly decreased after dietary intervention (Wilcoxon test, $p = 0.0008$) and microbial richness significantly increased (OTU observed richness, Wilcoxon test, $p = 0.004$). There was an inverse correlation between the microbial richness at baseline and the magnitude of increase in richness following the diet (Spearman rho -0.51 , $p = 0.0113$). At genus level, Roseburia decreased after the diet, although after multiple testing correction, this was no longer significant. At enterotype level, 27% of individuals which were Bacteroides at baseline shifted towards the Ruminococcus enterotype, 11% of Ruminococcus shifted towards Bacteroides and no shifts were observed in the Prevotella enterotype.

Conclusions : The FIT diet significantly increased intestinal microbial richness in healthy individuals, especially in individuals with low-richness at baseline. The Bacteroides enterotype, frequently associated with dysbiosis, was less resilient to dietary changes. Furthermore, a significant decrease in faecal calprotectin was seen after the diet suggesting additional anti-inflammatory metabolic effects beyond microbial richness and composition. A proof-of-concept study using the FIT diet is currently ongoing in patients with quiescent ulcerative colitis but recent flare, to see if the diet could prevent relapse.

LONG-TERM OUTCOME AND ENDOSCOPIC HEALING RATES FOLLOWING LONG MODIFIED SIDE-TO-SIDE STRICTUREPLASTIES. J. Van Stappen (1), A. De Buck Van Overstraeten (2), M. Ferrante (1), D. Vanbeckevoort (3), G. Van Assche (1), S. Vermeire (1). [1] University Hospitals Leuven, Leuven, Belgium, Gastroenterology, [2] University Hospitals Leuven, Leuven, Belgium, Abdominal surgery, [3] University Hospitals Leuven, Leuven, Belgium, Radiology.

Introduction : A long modified side-to-side isoperistaltic strictureplasty (SSIS) is an option in patients undergoing surgery for extensive stricturing Crohn's disease (CD) to avoid extensive small-bowel resections.

Aim : The aim of this study was to assess the endoscopic healing rates six months following SSIS, and to analyse the long-term outcome of these patients including need for re-intervention and/or re-introducing medical therapy.

Methods : The electronic medical records of all 40 patients (16 men and 24 women; median age 39 years; range 16-73 years) who underwent a long modified SSIS between 2010 and 2015 at our tertiary referral centre, were reviewed. In all patients, SSIS was performed because of extensive stenotic CD (>20 cm) of the (neo-) terminal ileum. Each patient received the same standardized follow up (FU) with clinical and endoscopic evaluation after median time of 6 months (interquartile range, IQR, 6-8 months). We also analysed disease recurrence during follow up necessitating medical or surgical re-intervention.

Results : Median FU period was 33 months (IQR, 15-47 months). Only 10 patients (25%) continued medical treatment immediately after surgery because of remaining disease activity in the colon or systemic disease activity with a high risk of clinical relapse and 30 patients received no medical treatment for the first 6 months until ileocolonoscopy. At month 6, 24/40 patients (60%) showed important mucosal improvement of the strictureplasty side, with increasing healing observed from distal to more proximal. At the end of FU, the cumulative clinical relapse rate was 62.5% (25 patients), median time to relapse was 13 months (IQR 6.7-16.5 months). Two patients necessitated endoscopic balloon dilatation of the most proximal anastomosis side of the SSIS for symptoms related to subobstruction. Only 2 patients (5%) so far needed surgical re-intervention; one patient developed recurrent stenosis at the inlet of the SSIS, another patient needed revision due to adhesions. No resection of any strictureplasty was required. Of the 25 patients with clinical relapse, 18 patients (72%) were started on anti-TNF antibodies or vedolizumab, 4 patients received budesonide and 2 patients azathioprine. At the last FU, 27/40 patients (67.5%) patients had durable response including 10 patients in clinical remission (no treatment). 13 patients failed medical therapy and changed treatment and/or 2 received surgery.

Conclusions : The long modified SSIS is a safe procedure with good long-term outcome. Postoperative ileocolonoscopy after six months showed a remarkable tendency for mucosal healing. The exact mechanism needs further investigation. With a median follow up of 2.5 years, surgical reintervention rates were very low and two thirds of patients showed durable response or were in remission on (very often) previously-failed treatments. Keywords Side-to-side isoperistaltic strictureplasty, Crohn's disease, mucosal healing.

METAGENOMICS AND METABOLOMICS OF PATIENTS WITH INFLAMMATORY BOWEL DISEASE AND THEIR UNAFFECTED RELATIVES. M. Vancamelbeke (1), J. Sabino (2), L. Deroover (2), G. Vandermeulen (2), A. Luypaerts (2), M. Ferrante (2), G. Falony (3), S. Vieira-Silva (3), K. Verbeke (2), J. Raes (3), I. Cleynen (4), S. Vermeire (2). [1] KU, Leuven, Belgium, Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders, [2] KU, Leuven, Belgium, Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders, [3] KU, Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Molecular Bacteriology, [4] KU, Leuven, Belgium, Department of Human Genetics, Laboratory for Complex Genetics.

Introduction : Dysbiosis, intestinal barrier dysfunction and metabolic alterations of the gut microbiota have been implicated in the pathogenesis of inflammatory bowel disease.

Aim : We studied the faecal microbiome and metabolome, as well as intestinal permeability of multiple-affected families with Crohn's disease (CD) or ulcerative colitis (UC) to investigate which factors are associated with disease.

Methods : Faecal and urine samples were obtained from 84 individuals of 19 families (37 CD, 11 UC and 36 unaffected first-degree relatives (FDR)). Faecal microbial profiling was done using 16S rDNA paired-end sequencing (Illumina MiSeq). Sequencing depth was downsized to 10,000 reads/sample. Taxonomic annotation was performed with the RDP classifier. Faecal volatile organic metabolites were measured using GC-MS. Metabolite data were relatively quantified to an internal standard, and subject-specific compounds were discarded. Metabolite profiles were clustered by PLS-DA (Unscrambler). Small intestinal permeability (IP) was measured using a 2-hour lactulose-mannitol urine test. Statistical analyses were conducted in R with multiple testing correction (Benjamini-Hochberg).

Results : Microbial richness and composition were significantly different in patients with CD compared to UC and FDR ($p < 0.05$), whereas these comparisons were not significant for UC versus FDR. Vector fitting confirmed diagnosis as

the main driver of the variability in microbial composition ($p < 0.001$), followed by family ID ($p = 0.02$). The genera discriminating CD and FDR included 16 known and new genera, such as *Faecalibacterium*, *Ruminococcus* and *Gemmiger* (corrected $p < 0.05$). Analysis of the metabolites also showed separate clusters for CD and FDR, while samples from UC patients partially overlapped with both groups. In contrast to the microbiota results, family did not significantly drive the metabolic profiles. The chemical classes associated with FDR were short- and medium-chain fatty acids, while samples from CD patients were associated with esters. Comparison of individual metabolites identified eight compounds with significantly different levels for CD versus FDR (corrected $p < 0.05$). Among these, acetic acid and butyric acid are known for their anti-inflammatory properties and beneficial effect on gut barrier function. A subset of CD patients (30%) had increased small IP values, but this trait was not associated with any of the individual metabolites, nor bacterial genera.

Conclusions : Significantly different metagenomic and metabolomic profiles were observed between CD patients and healthy individuals with a shared familial background. *Faecalibacterium*, *Ruminococcus* and *Gemmiger* genera, amongst others, drive the phenotype of CD, as do esters and lower levels of short-chain fatty acids.

- I07 -

DEFECTS IN ER STRESS AND AUTOPHAGY GENES TRANSLATE INTO INCREASED FUNCTIONAL ER STRESS LEVELS IN PATIENTS WITH INFLAMMATORY BOWEL DISEASE. W. Vanhove (1), K. Nys (2), I. Arijis (3), I. Cleynen (4), M. De Bruyn (5), H. Korf (6), M. Ferrante (7), G. Van Assche (7), S. Vermeire (7). [1] KU Leuven, Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, [2] KU Leuven, Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, [3] Hasselt University, KU Leuven, Belgium, Faculty of Medicine and Life Sciences, Hasselt University & Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, KU Leuven, [4] KU Leuven, Belgium, Laboratory for Complex Genetics, Department of Human Genetics, [5] KU Leuven, Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine & Laboratory of Immunobiology, Department of Microbiology and Immunology, Rega Institute for Medical Research, [6] KU Leuven, , Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine & Hepatology, Department of Clinical and Experimental Medicine , [7] University Hospital Leuven, KU Leuven, Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Department of Clinical and Experimental Medicine, KU Leuven & Department of Gastroenterology and Hepatology, University Hospitals Leuven.

Introduction : The crucial role of the intestinal epithelium in inflammatory bowel disease (IBD) is underscored by (genetic) association of epithelial homeostasis pathways such as bacterial sensing, autophagy and ER stress signaling. Reducing epithelial ER stress has therefore gained attention as a novel therapeutic approach. Nevertheless, molecular tools for patient stratification and therapeutic decision making are lacking.

Aim : We hypothesized that treatment selection could be improved by better patient stratification and molecular characterization. Therefore, this study investigated whether ER stress profiles could be quantified in patient-derived (ex vivo) intestinal epithelial cell (IEC) cultures.

Methods : IBD patients ($n = 35$) undergoing endoscopic evaluation were selected and stratified based on the number of IBD-associated ER stress risk alleles in *XBP1* (rs35873774) and *ORMDL3* (rs2872507). In addition, autophagy risk alleles in *ATG16L1* (rs2241880), *IRGM* (rs10065172 and rs4958847), *MTMR3* (rs2412973), *LRRK2* (rs11175593) and *ULK1* (rs12303764) were also investigated since autophagy is a compensatory ER stress resolving mechanism. For this second analysis, patients were grouped into genetic risk quartiles based on the combined ER stress and autophagy risk allele (RA) distribution (Q1 : ≤ 4 RA, Q2 : 5 RA, Q3 : 6 RA and Q4 : ≥ 7 RA). As described previously, we were able to culture IECs derived from mucosal biopsies. These IEC cultures were subjected to ER stress using thapsigargin (Tg, $0.4 \mu\text{M}$) and the ER stress response was measured in cell lysates with a binding immunoglobulin protein (BiP)-ELISA. Statistical analyses were performed with Mann-Whitney U tests ($\alpha = 0.05$).

Results : Median [IQR] Tg-mediated BiP-induction (vs. untreated) read-outs were 2.67 [1.01-6.07], 1.87 [1.50-3.16], 1.70 [1.32-2.41] and 4.48 [3.76-4.64] in IECs from patients carrying 0 ($n = 4$), 1 ($n = 17$), 2 ($n = 11$) or 3 ($n = 3$) ER stress risk alleles, respectively. Patients with 3 ER-stress-related risk alleles had significantly more epithelial ER stress (BiP) induction rates when compared to patients with 1 or 2 risk alleles ($p = 0.026$ and 0.043 , respectively). When risk alleles in autophagy genes were added, median [IQR] Tg-mediated BiP-induction read-outs were 1.34 [1.08-1.91], 2.16 [1.68-4.05], 3.60 [1.39-4.48] and 2.41 [1.61-3.27] in IECs from patients belonging to Q1 to Q4, respectively. Patients in Q2 ($n = 10$), Q3 ($n = 7$) and Q4 ($n = 10$) had significantly higher ER stress induction rates when compared to Q1 ($n = 8$) ($p = 0.034$, 0.040 and 0.034 , respectively).

Conclusions : IBD patients with an increased genetic risk for ER stress and autophagy have increased ER stress induction rates as measured in patient-derived IECs. These patients would benefit most from ER stress reducing therapies such as tauroursodeoxycholic acid (TUDCA), which has already shown to reduce inflammation in murine IBD models. We thus present a novel tool for molecular characterization of IBD patients for which pilot studies should be considered.

INCREASED BASELINE TNF-DRIVEN PATHWAYS OBSERVED IN PATIENTS WITH CROHN'S DISEASE NOT RESPONDING TO INFLIXIMAB. B. Verstockt (1), I. Arijts (1), M. De Bruyn (1), S. Verstockt (2), G. Van Assche (1), C. Breynaert (3), S. Vermeire (1), M. Ferrante (1). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), KU Leuven, Leuven, Belgium, Department of Clinical and Experimental Medicine, [2] Laboratory of Complex Genetics, KU Leuven, Leuven, Belgium, Department of Human Genetics, [3] Laboratory of Clinical Immunology, KU Leuven, Belgium, Department of Microbiology and Immunology.

Introduction : Anti-TNF therapy (infliximab, IFX) is effective for treating Crohn's Disease (CD) but 15-25% of patients fail to respond. Pathophysiological understanding of primary response (R) and non-response (NR) to IFX might help to predict who will benefit most from it. Additionally, it may highlight other potential therapeutic targets in non-responders.

Aim : Identifying pathophysiological mechanisms and predictive markers for primary (non-) response to IFX in CD patients.

Methods : Inflamed colonic mucosal biopsies from 17 CD patients (11 R and 6 NR, median age 31.8 years) before first IFX infusion were studied. Total RNA was analysed for whole genome expression via Affymetrix Human Genome U133 Plus 2.0 Arrays, followed by a Weighted Gene Co-expression Network Analysis. A false discovery rate <0.1 was considered biologically significant. Gene set enrichment and upstream regulation analyses were performed with Ingenuity Pathway Analysis. Mann-Whitney U-test or Fisher's exact test were used, when appropriate.

Results : Network analysis identified 70 gene clusters of which 4 (including 2179 probe sets) were correlated with (N) R to IFX. Consensus clustering using these identified probe sets perfectly discriminated R from NR. Although disease activity and CRP were not significantly different between R and NR at baseline, pathway analysis showed increased (a) granulocyte adhesion and diapedesis, TREM-1 signalling, IL-6 signalling, inhibition of matrix metalloproteases and NF- κ B signalling at baseline in NR. Upstream regulation analysis identified TNF and TGF β 1 as the strongest upstream regulators. Also TREM-1 was identified as a potential upstream regulator. Interestingly, the previously identified top 5 differentially expressed genes between IFX R and NR are regulated by TNF and/or TGF β 1 and TREM-1. Colonic mRNA levels of TNF, TGF β 1 and TREM-1 showed a significantly higher expression in IFX NR vs R. Finally, we hypothesized that NR with increased TNF-driven pathways at baseline may need more TNF-blockade. We therefore retrospectively reviewed the need for dose escalation within the first year after IFX induction and found that 50.0% of NR received dose escalation, all successfully leading to R to IFX.

Conclusions : At baseline several inflammatory pathways differ between IFX R and NR. TNF was the strongest predicted upstream regulator and colonic TNF mRNA levels were higher in IFX NR, suggesting that local cytokine production is (partially) driving these upregulated pathways. These patients may benefit from a higher dose of anti-TNF to neutralise gut inflammation. Additionally, therapy directed against TREM1, a triggering receptor expressed on myeloid cells, may also be a potential treatment strategy in these patients.

MOLECULAR PROFILING OF EARLY CROHN'S DISEASE REVEALS A PROMINENT ROLE FOR WNT5A. S. Verstockt (1), J. Van Der Goten (2), M. Vancamelbeke (2), B. Verstockt (2), L. Van Lommel (3), F. Schuit (3), P. Rutgeerts (2), M. Ferrante (2), S. Vermeire (2), I. Arijts (2), I. Cleynen (1) / [1] KU, Leuven, Belgium, Department of Human Genetics, Laboratory for Complex Genetics, [2] KU, Leuven, Belgium, Department of Clinical and Experimental Medicine, Translational Research Centre for Gastrointestinal Disorders, [3] KU, Leuven, Belgium, Department of Cellular and Molecular Medicine, Gene Expression Unit.

Introduction : Crohn's disease (CD) is characterized by a chronic inflammation of the gut, progressing to stricturing and/or penetrating complications in most patients. Effective intervention before the onset of bowel damage, and thus in the early phase of the disease, will be required to optimize patient outcomes.

Aim : We aimed to define the molecular landscape of early CD by using the unique post-operative recurrence (POR) model.

Methods : Ileal mucosal biopsies were obtained during colonoscopy from (1) 25 patients with early recurrence CD (Rutgeerts' score i2b, i3 or i4) within 18 months after ileo-colonic resection with ileo-colonic anastomosis (= POR CD); (2) 19 CD patients within 18 months after diagnosis (= new CD); and (3) 14 active CD patients >3 year after diagnosis and/or >3 year after ileo-colonic anastomosis (= late CD). Ileal biopsies from 12 controls were included as comparison. Total RNA was used to study mRNA and microRNA (miRNA) expression via Affymetrix Human Gene 1.0 ST and Affymetrix miRNA 2.0 arrays, respectively. A false discovery rate (FDR) <5% and >2-fold change (mRNA) or >1.5-fold change (miRNA) were considered biologically significant. Gene and miRNA expression profiles were integrated using the Ingenuity microRNA Target Filter.

Results : When comparing POR, new and late CD with controls, we observed respectively 353, 608 and 614 significantly differentially expressed gene probe sets. Comparative analyses of the miRNA expression profiles in POR, new and

late CD versus controls identified respectively 13, 5 and 1 significantly differential signal(s). Integration of dysregulated genes and miRNAs in POR CD found 64 miRNA-mRNA pairs with negative correlation in expression profiles, five of which experimentally supported in literature : hsa-let-7g-5p is known to target PRDM1 and PTGS2, hsa-miR-30d-5p targets SLC7A11 and WNT5A, and hsa-miR-196a-5p targets ANXA1. To be sure that POR i2b/i3/i4 represents a true baseline model for early disease, we looked at gene expression in ileal biopsies from 3 CD patients with uninflamed post-operative ileum (i0), and 6 CD patients with POR i1. Comparison of i0, and i1 vs. controls identified respectively 1, and 123 significantly differentially expressed gene probe sets. WNT5A was the only dysregulated gene in i0, and showed an increased expression with an increasing Rutgeerts' score ($p < 0.0001$), which was validated with qRT-PCR.

Conclusions : We showed an important mRNA dysregulation in new/late CD, while dysregulated miRNA expression was more pronounced in POR CD. WNT5A, a non-canonical Wnt ligand, seems to have a key role throughout, being the only dysregulated gene in i0 CD patients, showing an increased expression with increasing Rutgeerts' score, and being targeted by one of the dysregulated miRNAs. WNT5A is known to be involved in reparative inflammation.

- 111 -

NAFAMOSTAT MESILATE, A SERINE PROTEASE INHIBITOR, AMELIORATES CHRONIC COLITIS VIA SUPPRESSION OF T-BET. H. VAN SPAENDONK (1), S. FRANQUE (2), J. DE MAN (1), B. DE WINTER (1) / [1] University of Antwerp, Antwerp, Belgium, Laboratory of Experimental Medicine and Pediatrics, Division of Gastroenterology, [2] Antwerp University Hospital, Edegem, Belgium, Gastroenterology and Hepatology.

Introduction : The gastrointestinal tract is constantly exposed to high levels of endogenous and exogenous proteases. Their proteolytic activity is tightly regulated by endogenous antiproteases, since excessive proteolysis can cause tissue damage. Increasing evidence suggests that a protease/antiprotease dysbalance might play a role in gastrointestinal diseases such as inflammatory bowel disease (IBD), making protease inhibition a potential therapeutic intervention.

Aim : We aimed to investigate the effect of a serine protease inhibitor, nafamostat mesilate, on chronic colitis in a murine transfer model.

Methods : Colitis was induced in immunodeficient SCID mice by the adoptive transfer of CD4⁺CD25⁻CD62L⁺ T-cells, isolated from BALB/c mice; controls were injected with PBS. Animals were treated twice a day with vehicle or nafamostat mesilate (5 mg/kg, i.p.) starting from week 2. The following groups were included : control mice treated with vehicle (CONTROL; n=8) or nafamostat mesilate (CONTROL+NFM; n=8) and colitis mice treated with vehicle (COLITIS; n=8) or nafamostat mesilate (COLITIS+NFM; n=8). Every 2 weeks, colonic inflammation was assessed by clinical outcomes (body weight, stool consistency, mobility and piloerection) and an endoscopic scoring system. After sacrifice at week 4, colonic inflammation was assessed by macroscopy and cytometric bead array (CBA) for FN- γ and IL-6. Messenger RNA of transcription factors that regulate T helper (Th) cell differentiation such as T-bet (Th1) and protease-activated receptors (PAR) were quantified using RT-qPCR technique. Data are represented as mean \pm SEM.

Results : Chronic colitis resulted in a significant increase in colonic inflammation. In the COLITIS group the body weight dropped to 88.1 \pm 1.9 % of the initial body weight at week 4 whereas CONTROL mice gained weight over this time course (103.2 \pm 1.0%). The clinical disease and colonoscopic colitis score significantly increased to a maximum of 7.1 \pm 0.3 and 8.3 \pm 0.5 respectively at week 4 in the COLITIS group (vs 0.0+0.0 in CONTROL). Also the macroscopic score was increased in the COLITIS group versus CONTROL with a respective score of 8.8 \pm 0.4 versus 0.0 \pm 0.0. Nafamostat mesilate was able to significantly reduce these inflammatory signs, resulting in an ameliorated body weight (95.0 \pm 2.8%), an improved clinical (5.1 \pm 0.6), colonoscopic (6.7 \pm 0.6) and macroscopic score (7.0 \pm 0.8) Quantification of colonic cytokines by CBA confirmed these findings with a significant upregulation of IFN- γ and IL-6 in the COLITIS group versus CONTROL, respectively 202.1 \pm 43.2 vs 0.7 \pm 0.3 and 133.6 \pm 48.5 vs 1.9 \pm 0.9 pg/ml. Treatment with nafamostat mesilate significantly decreased these levels to respectively 104.0 \pm 20.4 and 35.3 \pm 6.5 in the COLITIS+NFM group. RT-qPCR experiments showed an upregulation of the relative mRNA expression of T-bet and PAR-4 in COLITIS (versus CONTROL), respectively 3.5 \pm 0.4 and 2.3 \pm 1.1, while other markers remained unchanged. Treatment with nafamostat mesilate significantly lowered the increased mRNA expressions (respectively 2.3 \pm 0.3 and 0.5 \pm 0.1 in COLITIS+NFM group versus CONTROL).

Conclusions : Our results show that treatment with a serine protease inhibitor ameliorates the course of experimental colitis. The beneficial effect of nafamostat on the Th1 transcription factor T-bet and major effector cytokine IFN- γ make us hypothesize that the Th1 T-cell subset plays a pivotal role in the observed anti-inflammatory effect of nafamostat mesilate. We additionally hypothesize that nafamostat mesilate acts through PAR-4 signaling on the crosstalk between innate and adaptive immunity to induce the switch in CD4⁺ T-cell differentiation.

MUCOSAL RECOLONIZATION AFTER ILEOCECAL RESECTION DIFFERS IN CROHN'S DISEASE PATIENTS DEVELOPING POSTOPERATIVE RECURRENCE. K. Machiels (1), M. Pozuelo Del Río (2), J. Sabino (3), A. Santiago (2), D. Campos (2), A. Wolthuis (4), A. De Buck Van Overstraeten (4), A. D'hoore (4), G. Van Assche (3), M. Ferrante (3), P. Rutgeerts (3), G. De Hertogh (5), S. Vermeire (3), C. Manichanh (2). [1] University Hospitals Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] Vall d'Hebron Research Institute, Barcelona, Spain, Department of Gastroenterology, [3] University Hospitals Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [4] University Hospitals Leuven, Leuven, Belgium, Department of abdominal surgery, [5] University Hospitals Leuven, Leuven, Belgium, Department of Pathology.

Introduction : Mucosa-associated bacteria are believed to play a more prominent role in the pathogenesis of Crohn's disease (CD) as they are in closer contact to the gut immune system.

Aim : Our aim was to study temporal changes of the microbiota in CD patients undergoing ileocecal resection and to identify the predictive value of recurrence-related microbiota.

Methods : A total of 204 samples from CD patients undergoing ileocecal resection with ileocolonic anastomosis were prospectively collected : biopsies were taken at the time of surgery from the resected intestine (histologically inflamed (N=63) and non-inflamed ileum (N=56)) and from the neoterminal ileum (N=85) during postoperative endoscopy at month 6. Postoperative endoscopic recurrence (POR) was defined by a Rutgeerts score ≥ 2 . The microbiota was evaluated by 16S rDNA sequencing using an Illumina MiSeq platform. Calculation of alpha and beta diversity and statistical analysis were performed in QIIME.

Results : At the time of surgery, the inflamed mucosa had a lower abundance of Actinomyces (FDR=0.05) compared to the non-inflamed mucosa. Six months after resection, alpha diversity increased significantly compared to baseline samples in patients with recurrence ($p=0.011$) but not in patients without recurrence. An enrichment in Lachnospiraceae was observed in all patients at month 6 after surgery when compared to baseline samples (FDR<0.001). In recurrence patients, also Fusobacteriaceae (FDR=0.002) and Halomonadaceae (FDR=0.07) increased significantly after surgery when compared to baseline. Patients without recurrence on the other hand showed a decrease of Peptostreptococcaceae (FDR=0.08). At month 6, patients experiencing POR had a higher abundance of taxa belonging to Negativicutes (FDR=0.04) and Fusobacteria (FDR=0.04) compared to the remission patients. A small subset of CD patients was on antibiotics at the time of surgery. Alpha diversity of the inflamed and non-inflamed mucosal microbiota was significantly reduced in antibiotics users (N=6 and 7; $p=0.004$ and 0.009 respectively). A strong impact was seen on many taxa including a reduction of Clostridia, Bacteroidaceae and increase of Flavobacteria and Bacilli.

Conclusions : At the time of resection, differences in microbiome composition between inflamed and non-inflamed mucosa are limited. The impact of ileocecal resection on the mucosal microbiome is defined by a general increase of Lachnospiraceae. Recolonization after ileocecal resection more specifically in patients developing recurrence differs from patients without recurrence by an increase of members belonging to Fusobacteriaceae and Halomonadaceae families. Postoperatively, the increased levels of Fusobacteria and Negativicutes (Veillonellaceae), which previously have been associated with inflammation in pediatric patients with new onset of CD, may be involved in the development of early POR.

PERIOPERATIVE USE OF VEDOLIZUMAB IS NOT ASSOCIATED WITH SHORT-TERM POSTOPERATIVE INFECTIOUS COMPLICATIONS IN PATIENTS WITH ULCERATIVE COLITIS UNDERGOING (PROCTO) COLEC-TOMY WITH ILEAL POUCH-ANAL ANASTOMOSIS. M. FERRANTE (1), N. SCHILS (1), A. DE BUCK VAN OVERSTRAETEN (2), S. VERMEIRE (1), G. VAN ASSCHE (1), A. WOLTHUIS (2), A. D'HOORE (2). [1] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology, [2] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Abdominal Surgery.

Introduction : Vedolizumab, a bowel focused anti-adhesion molecule, is effectively used in patients with Crohn's disease (CD) and ulcerative colitis (UC). Preoperative use of vedolizumab has recently been associated with increased risk of short-term postoperative infectious complications.

Aim : We assessed this risk in a single-center cohort of patients with UC undergoing (procto)colectomy with ileal pouch-anal anastomosis (IPAA).

Methods : A chart review was performed in all patient undergoing (procto)colectomy with IPAA between September 2006 (initiation of vedolizumab in clinical trials) and September 2016. Patients receiving an investigational medical product besides vedolizumab within 14 weeks of (procto)colectomy or receiving a permanent ileostomy were excluded. Short-term postoperative infectious complications were evaluated within 30 days after (procto)colectomy and included pouch related complications, surgical site infections, and infections outside the surgical site. The comprehensive complication index (CCI) was calculated based on all complications reported within 30 days of (procto)colectomy.

Results : We identified 170 patients undergoing (procto)colectomy (46% female, median age 38 years, median disease duration 6 years). Thirty-four patients (20%) received vedolizumab within 14 weeks, 60 (35%) received anti-TNF within 8 weeks, 32 (19%) received a moderate-to-high dose (≥ 20 mg/day) of prednisone, and 71 (42%) received no therapy at time of (procto)colectomy. Surgery was laparoscopy-assisted in 131 patients (77%). Pouch construction was performed at first stage in 47 patients (28%), more frequent in patients with dysplasia/cancer (85% vs. 13%, $p < 0.001$), and less frequent in patients under vedolizumab (9% vs. 32%, $p = 0.005$), anti-TNF (15% vs. 35%, $p = 0.006$), or steroids (0% vs. 34%, $p < 0.001$). In multivariate analysis, the only risk factor for short-term postoperative infectious and overall complications was the construction of the pouch at first stage [Odds ratio 2.40 (95%CI 1.18-4.90), $p = 0.016$ and 3.11 (1.52-6.40), $p = 0.002$, respectively]. No significant difference could be observed between different treatment categories and development of short-term postoperative complications (anastomotic leakage, pouch related complications, surgical site infectious complications, any infectious complication, any non-infectious complication, or overall complications). Also the CCI and postoperative hospitalization stay were comparable between each treatment category, and only elevated in patients undergoing pouch construction at first stage [20.9 (0.0-30.8) vs. 0.0 (0.0-20.9), $p = 0.001$, and 11 (9-17) vs. 7 (5-10) days, $p < 0.001$, respectively].

Conclusions : In this large single-center cohort of patient with UC undergoing IPAA surgery, perioperative use of vedolizumab was not associated with short-term postoperative (infectious) complications. However, in patients under biological therapy or moderate-to-high dose of steroids pouch construction should be postponed to a second stage of surgery.

- 116 -

ADALIMUMAB DOSE ESCALATION AND DE-ESCALATION IN UC : INCIDENCE AND PREDICTORS OF SUCCESS : A REAL LIFE BELGIAN COHORT STUDY. S. Van De Vondel (1), F. Baert (2), C. Reenaers (3), P. Bossuyt (4), P. Hindryckx (5), S. Vanden Branden (6), L. Amininejad (7), P. Dewint (8), W. Van Moerkercke (9), J. Rahier (10), M. Ferrante (1). [1] University Hospitals Leuven, Leuven, Belgium, Leuven, Belgium, Dept of Gastroenterology and Hepatology, [2] AZ Delta, Roeselare, Belgium, Dept of Gastroenterology, [3] University Hospital CHU of Liège, Liège, Belgium, Liège, Belgium, Dept of Gastroenterology, [4] Imelda Hospital, Bonheiden, Belgium, Bonheiden, Belgium, Dept of Gastroenterology, [5] Ghent University Hospital, Ghent, Belgium, Gent, Belgium, Dept of Gastroenterology, [6] Onze-Lieve-Vrouw Hospital, Aalst, Belgium, Aalst, Belgium, Dept of Gastroenterology, [7] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Brussel, Belgium, Dept of Gastroenterology, Hepatopancreatology and Digestive Oncology, [8] AZ Maria Middelaers, Ghent, Belgium, Gent, Belgium, Dept of Gastroenterology, [9] AZ Groeninge, Kortrijk, Belgium, Kortrijk, Belgium, Dept of Gastroenterology, [10] CHU Mont-Godinne, Yvoir, Belgium, Dept of Gastroenterology.

Introduction : Adalimumab (ADM) is efficacious in inducing and maintaining remission in ulcerative colitis (UC). In randomized trials dose escalation from 40 mg every other week (EOW) to 40 mg every week (EW) was needed in 20-25% within 1 year. Real life data show higher escalation rates and attempts for dose de-escalation in UC have not been studied.

Aim : This study aimed to assess the need for, the outcome of, and predictors of dose escalation and de-escalation in a large cohort of UC patients (pts) treated with ADM.

Methods : Consecutive pts from 10 Belgian centers that initiated ADM treatment for active UC before September 1st 2015 were included in this cohort study. We performed a detailed retrospective chart review including pts demographics, disease characteristics, previous and concomitant medication including prior infliximab (IFX) use and the reason for discontinuation. Primary clinical benefit was based on physician global assessment (PGA) and absence of rectal bleeding at week 10. Similarly, success of dose-escalation was defined based on a positive PGA and absence of rectal bleeding on two consecutive visits at least 3 months apart. Success of dose de-escalation was defined as persistent ADM use at a dose of 40mg EOW for ≥ 6 months after dose de-escalation. Cox regression and backward Wald multivariate analysis were used to identify variables associated with a) a primary clinical benefit of ADM, b) success of dose escalation and c) success of dose de-escalation.

Results : We included 231 pts [67% male, median (IQR) age at diagnosis 30.6 (22.9-44.8) years, median disease duration at start of ADM 5.5 (2.6-11.8) years, 63% previously exposed to infliximab (IFX)]. A primary clinical benefit was achieved in 101 pts (44%) and was less frequent in pts previously failing IFX [37% vs. 50%, OR 0.51 (95%CI 0.28-0.95), $p = 0.035$]. One hundred and sixty four pts (71%) needed ADM discontinuation (N=56) or dose escalation (N=129) after a median of 2.8 (1.7-5.1) months. Disease duration ≥ 5 years [0.58 (0.43-0.80), $p = 0.001$]; IFX stop for primary non response or loss of response [1.41 (1.03-1.93), $p = 0.032$] and primary clinical benefit [0.30 (0.21-0.42), $p < 0.001$] were independently associated with ADM discontinuation or dose escalation. Dose escalation was successful in 77/129 (60%). Only primary response independently predicted successful dose escalation [3.08 (1.46-6.49), $p = 0.003$]. Dose de-escalation was attempted in 71% (55/77) and was successful in 80% (43/54), but no predictive markers could be identified. After a median of 40.7 (20.0-71.6) months, 35 (15%) needed colectomy (Fig 2), which was predicted independently by PSC [6.70 (2.06-21.81), $p = 0.002$], Mayo 3 baseline endoscopic activity [2.53 (1.01-6.38), $p = 0.049$], and absence of primary clinical benefit [2.94 (1.12-6.25), $p = 0.026$].

Conclusions : Early ADM dose escalation is needed in the majority of UC patients and has a success rate of 60%, which is predicted by a primary response to the treatment. Most patients can be successfully de-escalated at a later timepoint.

- I17 -

CORRELATION OF DURABILITY OF RESPONSE, SERUM TROUGH CONCENTRATIONS AND OUTCOME PARAMETERS : LONG-TERM FOLLOW-UP OF THE TROUGH CONCENTRATION ADAPTED INFLIXIMAB TREATMENT (TAXIT) TRIAL. L. Pouillon (1), M. Ferrante (1), G. Van Assche (1), P. Rutgeerts (1), M. Noman (1), N. Vande Casteele (2), A. Gils (3), S. Vermeire (1). [1] University Hospitals Leuven, Leuven, Belgium, Gastroenterology and Hepatology, [2] University of California San Diego, CA, United States (the), Medicine, [3] University of Leuven, Leuven, Belgium, Pharmaceutical Sciences.

Introduction : The Trough Concentration Adapted Infliximab Treatment (TAXIT) randomized controlled trial showed that targeting patients' infliximab trough concentrations in a 3-7 µg/mL window resulted in a more efficient use of the drug in patients with inflammatory bowel disease (IBD). However, following dose optimization, continued concentration-based dosing was not superior to clinically-based dosing for achieving a co-primary endpoint of clinical and biological remission after 1 year.

Aim : The aim of this study was to evaluate the long-term outcome of all 226 patients who completed the TAXIT maintenance phase. Durability of response to infliximab was correlated with serum trough concentrations and important quality of care outcome parameters, including need for IBD-related hospitalization, need for abdominal surgery and steroid use.

Methods : This was a retrospective analysis.

Results : With a median follow-up of 41 months after the completion of the TAXIT trial, 167/215 (78%) patients were still on continued treatment with infliximab, and 48/215 (22%) patients needed to stop (11 patients were lost to follow-up). Among the 48 patients who discontinued infliximab, 10/27 (37%) randomized previously to the clinically-based dosing arm did so within 1 year, compared to 2/21 (10%) patients randomized to the concentration-based dosing arm ($p < 0.05$). Among the 167 patients who continued infliximab, the dosing scheme was intensified in 56 patients and de-intensified in 27 patients, compared to the end of the TAXIT maintenance phase. Median trough concentrations of infliximab at the end of follow-up were 4.73 µg/mL (IQR=3.3-6.42). Five patients developed immunogenicity within 1 year after TAXIT and all had been randomized to the clinically-based dosing arm. In patients continuing on infliximab, the rates of IBD-related hospitalization (16/167 patients or 9.6%), abdominal surgery (4/167 patients or 2.4 %) and steroid use (6/167 patients or 3.6%) during the entire follow-up period were very low and significantly better than in patients who had to discontinue infliximab ($p < 0.001$).

Conclusions : In this long-term follow-up of the TAXIT trial, infliximab discontinuation occurred earlier in patients treated in the clinically-based dosing arm than in patients treated in the concentration-based dosing arm. Targeting infliximab trough concentrations to a therapeutic window led to a highly durable treatment response, and was associated with very good outcomes including very low (<5%) surgical rates and steroid use.

- I18 -

RECENT ANTI-TNF EXPOSURE PREDICTS LOWER VEDOLIZUMAB TROUGH CONCENTRATIONS IN PATIENTS WITH CROHN DISEASE. A. Gils (1), E. Dreesen (1), G. Compennolle (1), M. Peeters (1), E. Brouwers (1), S. Tops (1), V. Ballet (2), M. Noman (2), M. Ferrante (2), G. Van Assche (2), S. Vermeire (2). [1] Katholieke Universiteit Leuven, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences, [2] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Vedolizumab (VDZ) specifically inhibits the $\alpha 4\beta 7$ integrin interaction with mucosal addressin cell adhesion molecule (MAdCAM)-1 and has been approved for the treatment of patients with moderate to severe Crohn's disease (CD) and ulcerative colitis (UC). We have previously shown that anti-tumor necrosis factor-alpha (anti-TNF) treatment influences MAdCAM-1 expression in gut biopsies.

Aim : We studied the impact of recent anti-TNF exposure on the VDZ trough concentrations (TC) and response.

Methods : From 75 patients (46 CD, 29 UC) starting IV VDZ therapy in a tertiary referral center, VDZ and anti-TNF serum concentrations (median [IQR]) were measured at trough during VDZ induction (w2 and w6) and early maintenance therapy (w10 only for patients with CD, w14 and w22) using in-house developed ELISAs. Clinical response was evaluated by clinical symptoms and physician global assessment. Biological response (CRP decrease $\geq 50\%$ from baseline) and remission (CRP ≤ 5 mg/L) were assessed at w6 and w22 in patients with CD who had a baseline CRP > 5 mg/L ($n = 25$). All patients underwent endoscopy at baseline. Twenty-eight patients with CD underwent endoscopy after w22 to assess mucosal healing and all patients with UC received sigmoidoscopy at w10 (mucosal healing was defined as a Mayo endoscopic sub-score of 0 or 1).

Results : Clinical response was achieved in 46% (21/46) and 66% (19/29) of the patients with CD and UC. Only in patients with UC, a significant exposure-response correlation was found between VDZ TC up to w22 and clinical response (data not shown, $p < .0001$). Biological response and remission at w6 were achieved in 48% (12/25) and 28% (7/25) of the patients with CD. Patients with biological response and remission had significantly higher VDZ TC at w6 ($p = .002$ and $p = .0007$, resp.). At w22, 48% (12/25) and 32% (8/25) of the patients with CD were in biological response and remission. Patients in biological remission had significantly higher TC at w6, w10 and w22 (data not shown, $p = .02$, $p = .04$ and $p = .01$). Mucosal healing was achieved in 18% (5/28) of the patients with CD and in 66% (19/29) of the patients with UC. Patients with UC with mucosal healing had significantly higher VDZ TC up to w22 compared to patients with no healing ($p = .02$ and $p = .006$, $p = .03$ and $p = .04$ for w2, w6, w14 and w22, resp.). Patients with CD with mucosal healing had significantly higher VDZ TC at w6 and w10 ($p = .006$, $p = .03$, resp.). Most patients (93%, 70/75) were previously exposed to anti-TNF. Of these, 30 (43%) had still detectable anti-TNF concentrations at the first VDZ infusion. Patients with CD who were recently exposed to anti-TNF (<16 weeks before the start of VDZ therapy, $n = 38$) had significantly lower VDZ TC at all time points, compared to patients with no recent anti-TNF exposure (w2 : 22.7 $\mu\text{g/mL}$ [19.3 – 31.6], $n = 18$ vs. 31.6 $\mu\text{g/mL}$ [23.7 – 38.1], $n = 28$, $p = .05$; w6 : 16.8 $\mu\text{g/mL}$ [10.7 – 23.2], $n = 18$ vs. 28.5 $\mu\text{g/mL}$ [20.6 – 38.8], $n = 28$, $p = .006$; w10 : 16.4 $\mu\text{g/mL}$ [9.6 – 24.4], $n = 18$ vs. 27.6 $\mu\text{g/mL}$ [16.5 – 42.2], $n = 26$, $p = .03$; w14 : 19.5 $\mu\text{g/mL}$ [11.6 – 22.8], $n = 16$ vs. 26.7 $\mu\text{g/mL}$ [17.7 – 40.8], $n = 28$, $p = .005$; w22 : 6.8 $\mu\text{g/mL}$ [3.2 – 11.2], $n = 16$ vs. 15.5 $\mu\text{g/mL}$ [9.9 – 22.3], $n = 27$, $p = .005$). We observed numerical though non-significant lower response rates in patients with recent anti-TNF exposure (data not shown, $p > .1$).

Conclusions : A clear exposure-response correlation was observed as early as w2 and up to w22, with significant impact of higher VDZ TC on meaningful outcomes such as clinical response, biological response and remission and endoscopic healing. The inverse association between recent anti-TNF exposure and VDZ TC in patients with CD is intriguing and might be explained by a residual effect of anti-TNF treatment on MAdCAM-1 expression.

- 119 -

NATURAL HISTORY OF DYSPLASIA AND COLORECTAL CANCER IN INFLAMMATORY BOWEL DISEASE IN BELGIUM TERTIARY CARE CENTERS. A. Cremer (1), C. Liefferinckx (2), M. De Vos (3), J. Rahier (4), F. Baert (5), T. Moreels (6), E. Macken (6), E. Louis (7), J. Devière (2), A. Van Gossum (2), S. Vermeire (8), D. Franchimont (2). [1] Erasme University Hospital, Anderlecht, Belgium, Gastroenterology, [2] Erasme University Hospital, Brussels, Belgium, Gastroenterology, [3] University Hospital Gent, Gent, Belgium, Gastroenterology, [4] CHU Mont-Godinne, Yvoir, Belgium, Gastroenterology, [5] AZ Delta, Roeselare, Belgium, Gastroenterology, [6] Antwerp University Hospital, Edegem, Belgium, Gastroenterology, [7] University Hospital Liege, Liege, Belgium, Gastroenterology, [8] University Hospitals Leuven, Leuven, Belgium, Gastroenterology.

Introduction : Inflammatory bowel disease (IBD) patients are at increased risk of developing dysplasia and colorectal cancer, namely colitis-associated colorectal cancer (CAC). Several risk factors have been identified. Ulcerative colitis (UC) and Crohn's disease (CD) patients are recommended to undergo screening and surveillance colonoscopy according to National and International recommendations. However, large study populations are needed to understand the natural history of dysplasia and CAC and improve their management in IBD patients.

Aim : The aim of our study was to evaluate the natural history of low-grade dysplasia (LGD), high-grade dysplasia (HGD) and CAC in IBD patients in Belgium tertiary IBD centers.

Methods : This is a national long-term follow-up retrospective study of IBD patients who had confirmed dysplasia and/or CAC from 1990-2016 in Belgium tertiary referral regional and academic centers within the Belgian Inflammatory Bowel Disease Research and Development (BIRD) group. Clinical, endoscopic and pathologic data were retrieved and reviewed through retrospective electronic chart review. All biopsies and surgical specimen were reviewed by a second independent expert IBD pathologist to confirm the diagnosis of dysplasia and/or CAC.

Results : 195 IBD patients (105 (54%) CD, 83 (43%) UC, and 7 (3%) unclassified IBD) with in total 466 lesions (dysplasia or CAC) were identified. From these 466 lesions, a total of 391 were LGD (346 raised, 45 flat lesions), 40 were HGD (35 raised, 5 flat lesions), and 35 were CAC. Median age at IBD diagnosis was 42 years old (IQR : 29-57). Median disease duration at diagnosis of first diagnosed lesion (index lesion), either dysplasia or CAC, was 8 years (IQR : 1- 17). From the 195 affected patients, 161 (83%) had only dysplasia (either LGD or HGD), while 34 (17%) had CAC (26 CD and 8 UC; 20 men and 14 women) during IBD follow-up. Median disease duration was significantly longer in patients with CAC compared to those with dysplasia (13 (IQR : 4-27) vs 7 (IQR : 1-16) years; $p = 0.03$). Findings on surgery and follow-up colonoscopies based on index lesion are provided in table 1. Overall 11 (7.6%) out of 146 patients with firstly diagnosed LGD have progressed to more advanced neoplasia (6 HGD and 5 CAC) after a median follow-up of 43 months (IQR : 16-79). 27/34 (79%) IBD patients with CAC were diagnosed with CAC without evidence of prior dysplasia, while 7/34 (21%) (2 with prior HGD and 5 with prior LGD) developed CAC secondarily after a median follow-up of 43 months (IQR : 13-76). Among them, 4 (57%) had early stage CAC, and 3 (43%) had advanced stage (stages II-IV) at diagnosis, while in patients without prior history of dysplasia, 10 (37%) had early stage at diagnosis. CAC was diagnosed during colonoscopy in 26 patients (3 before IBD diagnosis, 5 at the time of IBD diagnosis and 18 during IBD

follow-up) and on surgical pathological specimen in 8 known IBD patients where surgery was initially performed either for pre-existing dysplasia (n=2), or for IBD therapeutic management (n=6). Median age at CAC diagnosis was 57 years old (IQR : 44-65). Among the patients diagnosed with dysplasia (n=168), 14 (8.3%) (9 HGD, 5 LGD) underwent partial or total colectomy with dysplasia as indication and a CAC was diagnosed on the surgical specimen in only 2 patients, while the 12 other patients had lower or same lesion grade.

Conclusions : This is one of the largest cohorts of IBD patients with dysplasia and CAC never described so far. The rate of progression of LGD to advanced neoplasia (HGD and CAC) remains low with the limitation of a retrospective study. CAC diagnosis is mostly done during colonoscopy with no prior history of dysplasia. CAC was found incidentally at surgery for indications of dysplasia and refractory disease.

- I21 -

ANTI-INFLIXIMAB ANTIBODY CONCENTRATIONS GUIDE THERAPEUTIC DECISION-MAKING IN PATIENTS WITH CROHN'S DISEASE LOSING CLINICAL RESPONSE. E. Dreesen (1), T. Van Stappen (1), V. Ballet (2), S. Tops (1), G. Compennolle (1), G. Van Assche (2), M. Ferrante (2), S. Vermeire (2), A. Gils (1). [1] Katholieke Universiteit Leuven, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences, [2] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Loss of clinical response (LOR) to infliximab (IFX) maintenance therapy in patients with Crohn's disease (CD) may necessitate treatment intensification.

Aim : We explored the pharmacokinetics and effectiveness of infusion interval shortening (IS) and dose doubling (DD) and whether IFX and antidrug antibody (ADA) trough concentrations (TC) can guide therapeutic decision-making.

Methods : A retrospective cohort study was conducted, including 93 patients with CD who received a double dose IFX (10 mg/kg body weight) and/or a next infusion after a shortened interval following LOR during maintenance therapy. IFX TC and ADA were measured at consecutive time points just before (at T0) and after (at T+1) the treatment intensification. ADA were quantified using an in-house developed drug tolerant assay. We compared the short-term evolution of IFX exposure, immunogenicity, clinical response (evaluated by clinical symptoms and physician global assessment) and biological response and remission (based on CRP) during DD, IS and combined DD+IS.

Results : Overall, treatment intensification significantly increased the IFX TC from 1.2 $\mu\text{g/mL}$ [0.3 – 3.6] to 3.6 $\mu\text{g/mL}$ [0.5 – 10.2] (93 paired samples, $p < 0.0001$). An ADA concentration below 481 ng/mL eq. predicted a therapeutic post-treatment TC (3 $\mu\text{g/mL}$, 100% specificity, 51% sensitivity, AUROC 0.83, $p < 0.0001$). When ADA were undetectable, all treatment intensification interventions significantly increased the serum IFX TC and DD+IS resulted in a larger TC increase compared to DD alone (IS : 3.1 $\mu\text{g/mL}$ [0.7 – 5.7] to 5.4 $\mu\text{g/mL}$ [1.9 – 15.6], $n=17$, $p=0.0002$; DD : 3.4 $\mu\text{g/mL}$ [1.3 – 6.0] to 5.5 $\mu\text{g/mL}$ [2.9 – 11.8], $n=25$ $p < 0.0001$; DD+IS : 1.7 $\mu\text{g/mL}$ [1.2 – 2.2] to 10.7 $\mu\text{g/mL}$ [8.3 – 13.7], $n=7$, $p=0.02$). When ADA were detectable but below the 481 ng/mL eq. cut-off, only DD significantly increased the serum IFX TC (IS : 0.7 $\mu\text{g/mL}$ [0.4 – 1.1] to 1.2 $\mu\text{g/mL}$ [0.4 – 2.3], $n=9$, $p=0.1$; DD : 1.5 $\mu\text{g/mL}$ [0.4 – 3.7] to 7.2 $\mu\text{g/mL}$ [3.3 – 13.5], $n=12$, $p=0.002$; DD+IS : 0.3 $\mu\text{g/mL}$ [0.3 – 0.7] to 5.2 $\mu\text{g/mL}$ [2.8 – 5.9], $n=5$, $p=0.06$). When ADA were above the 481 ng/mL eq. cut-off, neither treatment intensification intervention was effective for increasing the serum IFX TC (IS : 0.3 $\mu\text{g/mL}$ [0.3 – 0.3] to 0.3 $\mu\text{g/mL}$ [0.3 – 0.4], $n=8$, $p=0.3$; DD : 0.3 $\mu\text{g/mL}$ [0.3 – 0.3] to 0.3 $\mu\text{g/mL}$ [0.3 – 0.3], $n=8$, $p=1.0$; DD+IS : 0.3 $\mu\text{g/mL}$ [0.3 – 0.3] to 0.3 $\mu\text{g/mL}$ [0.3 – 0.3], $n=1$). A significant TC increase was associated with clinical response in patients undergoing IS, only when they had no detectable ADA (from 2.3 $\mu\text{g/mL}$ [0.7 – 4.4] to 4.9 $\mu\text{g/mL}$ [1.7 – 12.1], $n=15$, $p=0.009$). Even when ADA were detectable but below the 481 ng/mL eq. cut-off, a significant TC increase was associated with clinical and biological response and remission in patients undergoing DD (clinical response : from 2.0 $\mu\text{g/mL}$ [1.0 – 6.2] to 9.2 $\mu\text{g/mL}$ [4.4 – 13.5], $n=8$, $p=0.008$; biological response : from 1.5 $\mu\text{g/mL}$ [0.4 – 3.1] to 6.0 $\mu\text{g/mL}$ [3.3 – 13.5], $n=8$, $p=0.02$; biological remission : from 1.7 $\mu\text{g/mL}$ [0.3 – 3.4] to 4.0 $\mu\text{g/mL}$ [3.2 – 13.7], $n=7$, $p=0.03$).

Conclusions : Dose doubling is more effective than interval shortening for restoring therapeutic TC and clinical and biological response and remission in patients with low ADA titers. When ADA titers are high, neither treatment intensification strategy is effective.

- I22 -

FILGOTINIB, A SELECTIVE JAK1 INHIBITOR, INDUCES CLINICAL REMISSION IN PATIENTS WITH MODERATE-TO-SEVERE CROHN'S DISEASE : RESULTS FROM THE PHASE 2 FITZROY STUDY. S. Vermeire (1), C. Arnaud (2), V. Muls (3), M. De Vos (4), A. Vijverman (5), O. De Wit (6), T. Van Kaem (7), L. Meuleners (7), C. Tasset (7), A. Van Der Aa (7), P. Harrison (7). [1] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology, [2] Clinic Saint-Joseph, Liege, Belgium, Department of Gastroenterology, [3] St. Pierre University Hospital Center, Brussel, Belgium, Department of Gastroenterology, [4] University Hospital Ghent, Ghent, Belgium,

Department of Gastroenterology, [5] Citadelle Hospital, Liege, Belgium, Department of Gastroenterology, [6] University Hospital Saint-Luc, , Belgium, Department of Gastroenterology, [7] Galapagos NV, Mechelen, Belgium, Department of Development.

Introduction : Filgotinib is an oral, selective Janus kinase 1 (JAK1) inhibitor, which has demonstrated efficacy in patients with rheumatoid arthritis.

Aim : This Phase 2 study was designed to evaluate the efficacy and safety of filgotinib in patients with active Crohn's disease.

Methods : Between January 2014 and July 2015, 311 patients were screened in 9 countries across Europe, including Belgium. In 6 clinical sites, 32 Belgian Crohn's patients were screened of which 20 were eligible, mainly anti-TNF non-responders. In total, 174 patients with moderate-to-severe Crohn's disease [CDAI : 220 to 450, endoscopic evidence of active disease, centrally read] were randomized to receive 200mg filgotinib (FIL) or placebo (PBO) QD (3 : 1) for 10 weeks. Thereafter, patients continued to receive filgotinib (200mg or 100mg QD) or placebo for another 10 weeks. Anti-TNF-naïve (73/174, 42%) as well as anti-TNF non-responders (101/174, 58%) were included. Immunosuppressants had to be discontinued, but stable steroids were maintained until Week 10. Key efficacy and safety data from the 10-week induction period are presented, including the primary endpoint of clinical remission (CDAI < 150) at Week 10.

Results : Baseline characteristics were comparable in the filgotinib-treated and placebo groups, including mean disease duration (8.3 y), mean CDAI score (293), mean SES-CD endoscopic score (14.6), mean CRP (15.6 mg/L, 41% > 10mg/L) and oral corticosteroids use (51%, mean daily dose 20.8 mg/day). Filgotinib induced clinical remission in 47% of the patients, compared to 23% in the placebo arm ($p=0.0077$) after 10 weeks. Significantly more patients in the filgotinib arm showed a clinical response (FIL :59%, PBO :41%, $p=0.0453$) and improved quality of life (PRO2 mean change from baseline FIL :-21.9; PBO :-15.6; $p=0.0321$; IBDQ changes from baseline FIL : 33.8, PBO : 17.6 ; $p=0.0046$) than in the placebo arm. Effect of filgotinib on IBDQ was evident in all IBDQ subcomponents (mean change from baseline): bowel symptoms (FIL :10.0; PBO :5.6; $p=0.0040$), systemic symptoms (FIL :5.7; PBO :2.9; $p=0.0044$), emotional status (FIL :12.1; PBO :6.1; $p=0.0094$), and social functioning (FIL :6.2; PBO :2.9; $p=0.0202$). Numerically more patients on filgotinib had their CRP normalized (FIL :27%, PBO :14%) and showed an improvement of at least 50% in SES-CD endoscopy score (F :25%, P :14%), with this difference being more pronounced in local endoscopy readings (F :39%, P :23%). Histopathology overall total score on biopsies taken after 10 weeks of treatment was significantly more decreased in the filgotinib group compared to placebo (F :-3.5, P : -0.6; $p=0.0359$). In general, filgotinib was safe and well tolerated in this patient population. Similar incidences in early discontinuations, SAEs and AEs including infections were observed, with the majority of the SAEs related to worsening of Crohn's disease. An increase in mean haemoglobin concentration was observed, without difference between filgotinib and placebo. No clinically significant changes from baseline in mean neutrophil counts or liver function tests were observed.

Conclusions : Filgotinib is the first JAK inhibitor showing efficacy in moderate-to-severe Crohn's disease patients, as demonstrated by induction of clinical remission and response, and improved quality of life. The efficacy and safety data of filgotinib suggest a favourable risk/benefit profile, showing its potential as an oral treatment with a novel mechanism of action for the treatment of IBD. Currently, a global Phase III program (DIVERSITY) is ongoing with filgotinib to confirm the Phase II FITZROY results in Crohn' patients. In addition, filgotinib is also being assessed in patients with ulcerative colitis (Phase IIB/III SELECTION study).

- 123 -

IBD NURSES AS INTEGRAL PART OF A MULTIDISCIPLINARY IBD TEAM : PROSPECTIVE STUDY ON VIEW ON PATIENT OUTCOMES. S. Coenen (1), E. Weyts (1), P. Geens (1), S. Vermeire (1), M. Ferrante (1), M. Noman (1), V. Ballet (1), K. Vanhaecht (2), G. Van Assche (1). [1] UZ Leuven, Leuven, Belgium, IBD, [2] UZ Leuven, Leuven, Belgium, Quality.

Introduction : Inflammatory bowel diseases (IBD) are chronic gastrointestinal conditions with great impact on patient's social and professional life. Information, education and empowerment will help to optimize and ameliorate disease outcomes. In this process IBD nurses play a key role. They support the patient to better understand the risks and benefits of the disease management plan and facilitate prompt recognition of symptoms. In this way, IBD nurses can accomplish better compliance, more early intervention during flares and as a consequence, improve patient outcomes.

Aim : We prospectively investigated the effect of IBD nurses on improvement of quality of patient's care.

Methods : In Sep 2016, a second IBD nurse joined the multidisciplinary IBD team in our tertiary referral center. In order to standardize the assessments and measurements, all contacts (phone, e-mail and personal contacts) were prospectively collected and in detail by using a standard record. Patient characteristics, type of contact and interventions performed by the IBD nurse were categorized in the record and outcomes were reported.

Results : During Sep and Oct 2016, 703 patient contacts were recorded by the two IBD-nurses (43% male, median age 35 years, 77% Crohn's disease; 65% of patients on biologicals). The vast majority of nurse-patient contacts were phone calls (64%) and a minority involved personal contacts (28%) and e-mails (9%). Most of the contacts of the IBD nurse were assigned to providing disease information and support (24%), to the planning of procedures and consultations

(21%), to administration (12%) and to the follow-up on medication-related matters (11%). In addition 11% of the patients contacted the IBD nurses for flare management, 9% for psychosocial support, 7% for the start of new therapy and 6% for education on therapy. Most of the interventions performed by the nurses involved comforting patients (22%), calling patients for follow-up (19%), for blood or stool sampling (15%) and planning an urgent outpatient visit (9%). Besides, in house made information brochures were provided during 5% of the contacts, medication was initiated for 5% of the patients and 4% of the patients were referred to the general practitioner. By planning 75 urgent outpatient visits, the IBD nurses intervened earlier during a flare and as a result 17 emergency room visits could be avoided in this 2-month period only. Another 92 outpatient appointments could be avoided through counseling by phone and for 10 patients education and follow-up on therapy resulted in better compliance.

Conclusions : Standardized measurement of outcomes is a 'hot topic' in today's clinical practice. Prospective and standardized reporting of each nurse-patient contact allows us to measure important patient outcomes. In this way variability in reporting can be reduced, and the care we provide to our patients can rapidly be monitored and improved.

- I25 -

PERIOPERATIVE USE OF VEDOLIZUMAB SEEMS NOT ASSOCIATED WITH SHORT-TERM POSTOPERATIVE INFECTIOUS COMPLICATIONS IN PATIENTS WITH CROHN'S DISEASE UNDERGOING RIGHT HEMICOLECTOMY WITH ILEOCOLONIC ANASTOMOSIS. N. Schils (1), A. De Buck Van Overstraeten (2), S. Vermeire (1), G. Van Assche (1), A. Wolthuis (2), A. D'hoore (2), M. Ferrante (3). [1] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology, [2] University Hospitals Leuven, Leuven, Belgium, Department of Abdominal Surgery, [3] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Vedolizumab, a bowel focused anti-adhesion molecule, is effectively used in patients with Crohn's disease (CD). Preoperative use of vedolizumab was recently associated with increased risk of short-term postoperative infectious complications.

Aim : We assessed the risk of short-term postoperative infectious complications linked to vedolizumab in a single-center cohort of patients with CD undergoing right hemicolectomy with ileocolonic anastomosis (RHC).

Methods : A chart review was performed in all patient referred for RHC between September 2006 and September 2016 to identify patients receiving vedolizumab within 14 weeks of RHC. Next, age- and sex-matched control patients were identified who received anti-TNF within 8 weeks of RHC, a moderate-to-high dose of steroids at RHC, or no therapy at RHC. Short-term postoperative infectious complications were evaluated within 30 days and included anastomotic leakage, surgical site and other infections. The comprehensive complication index (CCI) was calculated based on all complications reported within 30 days of RHC.

Results : We identified 12 patients receiving vedolizumab within 14 weeks of RHC (75% female, median age 31 years, median disease duration 12 years, previous RHC 33%). Surgery was laparoscopy-assisted in 9 patients (75%). Compared to the vedolizumab group, control groups did not differ significantly in demographics, disease behavior, and surgical characteristics, except for patients in the moderate-to-high steroid dose group who had a shorter disease duration and more often used concomitant immunosuppressive therapy at RHC. No significant difference could be observed between the vedolizumab group and both the steroids and no therapy group regarding postoperative complications. Although patients in the selected anti-TNF group more often experienced non-infectious complications including prolonged ileus, venous thrombosis and urinary retention [67% vs. 8%, Odds ratio 22.22 (95% CI 2.05-250.00), p=0.009], a similar infectious complication rate [58% vs. 50%, 1.40 (0.28-6.99), p=0.682] and a similar low CCI [10.5 (8.7-28.8) vs. 4.4 (0.0-22.2), p=0.128] was observed. The postoperative hospitalization stay tended to be higher in this selected anti-TNF group [9 (6-10) vs. 6 (4-10) days in the vedolizumab group, p=0.078]. Both CCI and postoperative hospitalization stay were not significantly different between the other treatment categories.

Conclusions : In this small matched case-control study with patient undergoing RHC for refractory CD, we could not observe an increased risk of short-term postoperative (infectious) complications in patients receiving vedolizumab in the preoperative setting. On the contrary, compared to anti-TNF therapy a decreased risk of postoperative non-infectious complications could be observed. Larger multi-center data sets are required to provide more evidence of a save use of vedolizumab in the preoperative setting.

- I26 -

EXAMINATION OF THE EFFECTS OF ANTIBIOTICS ON THE RELEASE OF CAMPYLOBACTER CONCISUS ZONULA OCCLUDENS TOXIN. H. Chung (1), A. Tay (2), S. Octavia (1), J. Chen (1), F. Liu (1), R. Ma (1), R. Lan (1), S. Riordan (3), M. Grimm (4), L. Zhang (1). [1] University of New South Wales, Sydney, Australia, School of Biotechnology and Biomolecular Sciences, [2] University of Western Australia, Perth, Australia, School of Pathology and

Laboratory Medicine, [3] Prince of Wales Hospital, Sydney, Australia, Gastrointestinal and Liver Unit, [4] University of New South Wales, Sydney, Australia, St George and Sutherland Clinical School.

Introduction : *Campylobacter concisus* is a Gram-negative oral bacterium that is associated with human inflammatory bowel disease (IBD) due to its significantly higher prevalence in the intestinal tract of patients with IBD. Zonula occludens toxin (Zot) is a *C. concisus* virulence factor. Previous studies showed that *C. concisus* Zot toxin damaged intestinal epithelial barrier and induced production of TNF- α in both intestinal epithelial cells and macrophages.

Aim : In this study we examined whether antibiotics including those used in the treatment of IBD affect the release of Zot toxin.

Methods : *C. concisus* strain 13826, a zot-positive *C. concisus* strain, was used in this study. The bacterium was incubated with various concentrations of 5 different antibiotics at 37°C, rotating at 160rpm. Following a 24-hour incubation period, the supernatants were collected and concentrated using Amicon Ultra 10K columns. Cell were collected, exposed to a threefold freeze-thaw method with liquid nitrogen and passed through a 26-gauge needle 10 times to produce whole cell lysates. The presence of *C. concisus* Zot toxin in the supernatants and whole cell lysates were determined by Western-blot using a commercially produced Zot-specific antibody. Colony forming units of *C. concisus* was also determined.

Results : Zot was not detected after incubation with all antibiotics.

Conclusions : Antibiotics examined in this study inhibited the growth of *C. concisus* but did not induce the release of *C. concisus* Zot toxin.

- I27 -

URSODEOXYCHOLIC ACID AND ITS TAURINE/GLYCINE CONJUGATED SPECIES REDUCE COLITOGENIC DYSBIOSIS AND EQUALLY SUPPRESS EXPERIMENTAL COLITIS IN MICE. L. Van Den Bossche (1), P. Hindryckx (1), L. Devisscher (1), S. Devriese (1), S. Van Welden (1), T. Holvoet (1), R. Vilchez-Vargas (2), M. Vital (3), D. Pieper (3), J. Vanden Bussche (4), L. Vanhaecke (4), T. Van De Wiele (2), M. De Vos (1), D. Laukens (1). [1] Ghent University, Ghent, Belgium, Department of Gastroenterology, [2] Ghent University, Ghent, Belgium, Center for Microbial Ecology and Technology, [3] Helmholtz Centre for Infection Research (HZI), , Germany, Department of Medical Microbiology, [4] Ghent University, Ghent, Belgium, Department of Veterinary Public Health and Food Safety.

Introduction : The promising results with secondary bile acids in experimental colitis suggest that they may represent an attractive and safe class of drugs for the treatment of inflammatory bowel diseases (IBD). However, the exact mechanism by which bile acid therapy confers protection from colitogenesis is currently unknown.

Aim : Since the gut microbiota plays a crucial role in the pathogenesis of IBD, and exogenous bile acid administration may affect the community structure of the microbiota, we examined the impact of the secondary bile acid ursodeoxycholic acid (UDCA) and its taurine/glycine conjugates on the fecal microbial community structure during experimental colitis.

Methods : Acute colitis was induced in mice by administration of 4% dextran sodium sulfate to the drinking water for 7 days. Mice were treated with 500 mg/kg/d UDCA, tauroursodeoxycholic acid (TUDCA), glyoursodeoxycholic acid (GUDCA), or placebo by oral gavage. At day 9 of colitis, fecal microbiota profiles were determined through 16S rRNA Illumina MiSeq sequencing and mice were sacrificed at day 10 to assess the severity of inflammation. Ultra-high performance liquid chromatography and high resolution mass spectrometry were performed on fecal samples to analyze the extent of biotransformation of orally administered UDCA, TUDCA and GUDCA.

Results : Daily administration of UDCA, TUDCA and GUDCA equally lowered the severity of colitis, as evidenced by reduced body weight loss, colonic shortening and expression of inflammatory cytokines. Illumina sequencing demonstrated that bile acid therapy during colitis did not restore fecal bacterial richness and diversity but normalized the colitis-associated increased ratio of Firmicutes to Bacteroidetes. Interestingly, administration of bile acids prevented the loss of *Clostridium* cluster XIVa and increased the abundance of *Akkermansia muciniphila*, bacterial species known to be particularly decreased in IBD patients. Orally administered UDCA, TUDCA and GUDCA were extensively metabolized in vivo, resulting in a similar fecal bile acid composition.

Conclusions : We conclude that UDCA, which is an FDA-approved drug for cholestatic liver disorders, could be an attractive treatment option to reduce dysbiosis and improve inflammation in human IBD.

- I28 -

IDENTIFICATION OF PROTEINS DISCRIMINATING INFLAMMATION INDUCED DYSPLASIA FROM INFLAMMATION IN ULCERATIVE COLITIS BY LASER CAPTURE MICRODISSECTION AND LABEL FREE PROTEOMICS – A PILOT STUDY. A. Merli (1), F. Quesada Calvo (1), C. Massot (1), N. Bletard (2), N. Smargiasso (3), D. Baiwir (4), G. Mazzucchelli (3), M. De Pauw-Gillet (5), M. Malaise (6), E. De Pauw (7), P. Delvenne (2),

M. Meuwis (1), E. Louis (1). [1] ULg and CHU de Liège, Liège, Belgium, Translational Gastroenterology, GIGA-R and Hepato-Gastroenterology and Digestive Oncology unit, [2] CHU de Liège, Liège, Belgium, Department of Preclinical and Biomedical Sciences - Laboratory of Experimental Pathology, [3] Université de Liège, Liège, Belgium, Department of Chemistry - Mass Spectrometry Laboratory, [4] Université de Liège, Liège, Belgium, GIGA-Technology platforms : Proteomics Platform, [5] Université de Liège, Liège, Belgium, Department of Preclinical and Biomedical Sciences - Mammalian Cell Culture Laboratory, [6] CHU de Liège, Liège, Belgium, Department of Clinical Sciences - Rheumatology, [7] Université de Liège, Liège, Belgium, Department of Chemistry - Mass Spectrometry Laboratory.

Introduction : Chronic colonic inflammation in ulcerative colitis (UC) may induce dysplasia, which can itself progress and transform into neoplasia. Diagnosis of dysplasia in UC remains difficult particularly when tissue inflammation is present.

Aim : The aim of this retrospective pilot study was to highlight proteins specifically associated with inflammation induced dysplasia in UC.

Methods : We performed a pilot experiment on 15 Formalin-Fixed, Paraffin-Embedded (FFPE) samples isolated from 5 cases of UC patients with an Adenoma-Like Mass (ALM). We compared the proteomes of the ALM, the inflammatory (I) and the normal (NL) tissues of each patient. We performed Laser Capture Microdissection (LCM) in order to collect only epithelial cells, avoiding inflammatory infiltrating ones. Label free proteomic analysis using a 2D-nanoUPLC coupled with a hybrid Quadrupole-Orbitrap was applied, as well as differential analysis on the paired samples. Immunohistochemistry (IHC) characterisation of one of the selected proteins of interest was used for validation.

Results : Out of 985 quantified proteins, 7 were found significantly more abundant in ALM compared to I tissues, with 6 being only detected in ALM using proteomics. One of these is Solute Carrier Family 12 member 2 (SLC12A2), also known as Na-K-2Cl co-transporter 1 (NKCC1), a protein involved in ionic balance, in T-cell migration promotion and in some features involved in cancer development like proliferation, migration or invasion. IHC results obtained were in correlation with proteomic results and showed that SLC12A2 was more abundant in ALM tissue than in I and NL tissues, with a signal clearly delimiting the dysplastic region from the surrounding inflammatory tissue.

Conclusions : This pilot experiment shows a different proteomic profile in inflammation-associated dysplasia and simple inflammation. This should be replicated using other types of dysplasia in IBD (DALM and flat dysplasia). SLC12A2 could be a potential biomarker of inflammation-associated dysplasia.

- I29 -

TROUGH LEVELS (TLS) AT INDUCTION : IMPACT ON LONG TERM RESPONSE WHEN RE-INITIATING INFLIXIMAB. C. Liefferinckx (1), C. Minsart (1), J. Toubeau (2), A. Cremer (3), L. Amininejad (3), E. Quertinmont (1), J. Devière (3), A. Gils (4), A. Van Gossum (3), D. Franchimont (3). [1] Université Libre de Bruxelles Faculté de Médecine, Brussels, Belgium, Laboratory of Experimental Gastroenterology, [2] Université de Mons, 700, Belgium, Department of Electrical Engineering, [3] Hôpital Erasme, Brussels, Belgium, Department of Gastroenterology, [4] KU, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences

Introduction : Infliximab (IFX) is indicated for the treatment of inflammatory bowel disease (IBD) (ulcerative colitis (UC) or Crohn disease (CD)). Nevertheless, a significant proportion of patients will experience a loss of response (LOR) to IFX over time which may require despite optimization a switch to another anti-TNF or to swap out to another biotherapy. We have recently reported that week 2 and 6 IFX through levels (TLs) can be predictive of treatment failure and long term response. Only one study has shown that week 14 TLs can be predictive of long term response on re-initiation of IFX therapy.

Aim : Our objective is to analyze the early impact of previous anti-TNF exposition on the pharmacokinetics in order to predict LOR and need of switch during maintenance treatment.

Methods : 269 IBD patients (194 CD and 75 UC) have been treated with IFX on follow-up (median + range), (EC approved). 2331 samples were prospectively collected and measured retrospectively by ELISA in parallel with clinical data. 91 samples, with TL measured $< 1\mu\text{g/ml}$, were analyzed for IFX ATI using drug-sensitive bridging ELISA. Statistical analysis used Kolmogorov-Smirnov test after determination of non-normality of compared distributions thanks to χ^2 goodness-of-fit test.

Results : At follow-up, patients were subdivided in three groups : long-term responders, patients who had LOR but responded to optimization or patients who had LOR but did not respond to optimization and were switched to another biotherapy. Each group was subdivided according to naïve or previous treatment with anti-TNF (Infliximab or Adalimumab) status. During induction (week 2 and 6 combined), in the LOR switched group, median IFX TL was significantly lower in previously exposed patients (10/28) than in naïve patients (18/28) ($0.92\mu\text{g/ml}$ [0.12-4.4 $\mu\text{g/ml}$] VS $6.6\mu\text{g/ml}$ [0.15-19.93 $\mu\text{g/ml}$], $p=0.044$) (Figure 1A). Inversely, there was no statistical difference between median TL in the LOR optimized group between naïve (36/60) and previously exposed patients (24/60) ($9.38\mu\text{g/ml}$ [0.17-14.91 $\mu\text{g/ml}$] VS $11.82\mu\text{g/ml}$ [0.17-14.91 $\mu\text{g/ml}$], $p=0.52$) as well as in naïve (50/64) and previously exposed (14/64) Long-term responders ($9.57\mu\text{g/ml}$ [1.44-11.97 $\mu\text{g/ml}$] vs $11.91\mu\text{g/ml}$ [0.12-19.93 $\mu\text{g/ml}$], $p=0.92$). Overall, among

the previously exposed patients, the LOR switched group had a lower median IFX TL (0.92µg/ml [0.12-4.40µg/ml]) compared to the Long-term responders (9.57 µg/ml [0.44-11.97µg/ml], p= 0.015) and LOR optimized group (11,82 µg/ml [0.23-12.09µg/ml], p=0.005) (Figure 2). The percentage of ATI occurrence was statistically lower in the Long-term responders (5.7% (n= 2/35)) than in the LOR optimized (37.5% (n= 12/32), p=0.002) and LOR switched groups (40% (n=10/25), p=0.002). Interestingly, among the LOR switched group, the percentage of ATI occurrence was similar in patients whether naïve or previously exposed to anti-TNF (38,8%, n= 7/18 VS 42,9%, n= 3/7, p=0.86) (Figure 1B). The same observation was found in the LOR optimized group (25%, n= 3/12 VS 45%, n= 9/20, p=0.45). No comparison was possible within the Long-term responders group due to the low presence of ATI.

Conclusions : In the group of patients who do not respond to any optimization strategy (LOR switched group), patients previously exposed to anti-TNF seem to have lower IFX TLs at induction than naïve patients. This may not be related to immunogenicity as the presence of ATI was similar in patients whether naïve or previously exposed to anti-TNF.

- I30 -

EFFICACY OF VEDOLIZUMAB FOR INDUCTION OF CLINICAL RESPONSE AND REMISSION IN PATIENTS WITH MODERATE TO SEVERE INFLAMMATORY BOWEL DISEASE WHO FAILED AT LEAST TWO TNF ANTAGONISTS. M. De Vos (1), B. Dhooghe (1), S. Vermeire (2), E. Louis (3), M. Fazia (4), A. Elewaut (5), P. Bossuyt (6), F. Baert (7), C. Reenaers (8), M. Van Gossum (9), E. Macken (10), M. Ferrante (2), P. Hindryckx (1), S. Dewit (11), T. Holvoet (1), D. Franchimont (12). [1] Ghent University Hospital, Ghent, Belgium, Gastroenterology, [2] University Hospitals Leuven, Leuven, Belgium, Gastroenterology, [3] Centre Hospitalier Universitaire de Liège, Liège, Belgium, Gastroenterology, [4] UZ Brussel, Jette, Belgium, Gastroenterology, [5] AZ Groeninge, Kortrijk, Belgium, Gastroenterology, [6] Imelda GI clinical research, Imelda general hospital, Bonheiden, Belgium, Gastroenterology, [7] AZ Delta, Roeselare, Belgium, Gastroenterology, [8] Centre Hospitalier Universitaire de Liège Domaine Universitaire du Sart Tilman, Liège, Belgium, Gastroenterology, [9] UZ, Brussels, Belgium, Gastroenterology, [10] UZ Antwerpen, Antwerp, Belgium, Gastroenterology, [11] Mariaziekenhuis Noord-Limburg, Overpelt, Belgium, Gastroenterology, [12] Erasme Hospital, Brussels, Belgium, Gastroenterology.

Introduction : Vedolizumab is a recently available, monoclonal antibody targeting $\alpha 4\beta 7$ integrin for the treatment of ulcerative colitis (UC) and Crohn's disease (CD).

Aim : To evaluate the efficacy of vedolizumab induction therapy in refractory UC and CD patients in real life.

Methods : A cohort of 149 moderately to severely active UC and CD patients that failed or showed intolerance to at least 2 TNF antagonists received vedolizumab in a medical need program (April-September 2015) in 37 Belgian centers. Rates of clinical response and remission were retrospectively evaluated at week 10 for UC and week 14 for CD using the physician's assessment (for response only) and established disease activity scores.

Results : Ninety-five patients ((35 UC,60 CD) had sufficient data for analysis. For UC patients, clinical response was observed in 77% based on physician's assessment and 67% based on the Mayo score. The corresponding percentages for CD patients were 78% and 64%. Clinical remission rates based on activity scores (Mayo score ≤ 2 , respectively CDAI ≤ 150 or Harvey Bradshaw index ≤ 4) were 8.6% and 40% for UC and CD, respectively. No safety concerns were reported.

Conclusions : Approximately 70% of adult refractory UC and CD patients achieved a clinical response after 10 to 14 weeks of vedolizumab treatment in this real-life cohort.

- I31 -

USABILITY OF A HOME-BASED TEST FOR THE MEASUREMENT OF FECAL CALPROTECTIN IN IBD PATIENTS. C. Bello (1), A. Roseth (2), J. Guardiola (3), C. Renaers (1), C. Van Kemseke (1), E. Louis (4). [1] CHU de Liège, Liège, Belgium, Gastroenterology, [2] Lovisenberg Diakonale Hospital Oslo, Oslo, Norway, Gastroenterology, [3] Hospital Universitari de Bellvitge-IDIBELL, Barcelona, Spain, Gastroenterology, [4] CHU Liege, Liège, Belgium, Gastroenterology.

Introduction : Fecal calprotectin (FC) correlates well with mucosal healing and risk of relapse in inflammatory Bowel Disease (IBD). An obstacle for a broader use of FC measurement in routine practice is the need to bring stool samples at the hospital or the send them by mail.

Aim : The aim of our work was to test the usability of a home-based test for the measurement of FC in IBD patients.

Methods : IBD patients in clinical remission or mild disease activity and declaring motivation to perform home-based FC measurements were prospectively recruited in three IBD centres in Oslo, Barcelona and Liège. They received a standardized training. They were instructed to collect and extract stools and to measure FC with a dedicated tool and smartphone application, 5 times at two weeks intervals over a 8 weeks period. The included patients had to fill in a

usability questionnaire made of simple questions and Linkert scales at the first and the last FC measurement. Two global scores were calculated integrating the different aspects of usability : the System Usability Scale (SUS : 0-100) and the Global Usability Score (GUS : 0-85). FC was also centrally measured by ELISA.

Results : 58 patients were recruited, including 18 ulcerative colitis (UC) and 40 Crohn's disease (CD), 30 females. Median (IQR) age was 35 yrs (27-40), median (IQR) HBI in CD was 0 (0-4), median (IQR) Clinical Mayo in UC was 0 (0-1). Over the 58 included patients, 42 performed at least one FC measurement and 27 performed all the FC requested measurements. The median (IQR) GUS (0-85) at the first and last use were 74 (69-80) and 77 (68-83), respectively; the median (IQR) SUS (0-100) at the first and last use were 85 (78-90) and 81 (70-88), respectively. Adherence to the planned measurements and usability of the tool were higher in females and in less severe disease. The inter-class correlation coefficient between home-based and centrally measured FC was 0.85.

Conclusions : Around three quarters of the patients who declared themselves motivated to use home-based test of FC measurement actually did it, but only half of them fully adhered to the planned measurements. Usability scores for the home-based test were high. There was a very good correlation with the centrally measured FC by ELISA.

- I32 -

CLINICAL RELEVANCE OF DETECTING ANTI-INFLIXIMAB ANTIBODIES WITH A DRUG-TOLERANT ASSAY : POST-HOC ANALYSIS OF THE TAXIT TRIAL. T. Van Stappen (1), N. Vande Castele (1), G. Van Assche (2), M. Ferrante (2), S. Vermeire (2), A. Gils (1). [1] KU Leuven, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences, [2] University Hospitals Leuven, Leuven, Belgium, Department of Clinical and Experimental Medicine.

Introduction : Anti-drug antibodies (ADA) may develop in up to 51% of patients treated with infliximab maintenance therapy and are associated with infusion reactions and impaired response. Drug-sensitive assays do not detect ADA in the presence of drug and underestimate ADA formation. Drug-tolerant assays have therefore been developed and markedly increased the detection of ADA, although their clinical relevance remains to be shown.

Aim : Our goal was to evaluate the clinical relevance of anti-drug antibodies (ADA) measured using a drug-tolerant assay in comparison to a drug-sensitive assay in a post-hoc analysis of the Trough Concentration Adapted Infliximab Treatment (TAXIT) randomized controlled trial.

Methods : Patients who presented with an infliximab trough concentration (TC) $<3 \mu\text{g/mL}$ at screening (n=76) underwent dose escalation to achieve therapeutic TCs between $3-7 \mu\text{g/mL}$ prior to randomization. Serum samples were reanalyzed at screening, after optimization and one year after randomization using a drug-tolerant ADA assay.(1)

Results : Using a drug-tolerant assay, the immunogenicity detection rate increased from 21% (drug-sensitive assay) to 63% at screening, from 0% to 51% after optimization and from 3% to 42% one year after randomization. ADA concentration (median [interquartile range, IQR]) in ADA+ patients grouped into quartiles according to ADA concentration at screening, decreased throughout the study : from 220 [116-737] ng/mL eq at screening to 112 [78-180] ng/mL eq after optimization and to 95 [0-166] ng/mL eq at the end of the study. Patients in ADA Q4 required a higher cumulative infliximab dose (2390 [880-2998] mg) to achieve target TCs compared to ADA negative patients (450 [365-680] mg, $p<0.001$) and patients in ADA Q1/Q2 (425 [344-863]/483 [398-719], $P<0.001$). However, all but one patient belonging to ADA Q4 were also ADA positive using a drug-sensitive assay. Once dose optimized, patients have similar rates of clinical, biological and endoscopic remission after one year of treatment, regardless of ADA status in a drug-tolerant assay.

Conclusions : Upon dose intensification, low concentration ADA, not detectable using a drug-sensitive assay, disappear in more than half of the patients over time and are clinically non-relevant. In contrast, high concentration ADA which are typically also detected in a drug-sensitive assay, persist over time and necessitate a higher cumulative dose and drug cost. In the latter group, proactive drug switching may be more cost-efficient. References : (1)Van Stappen T, Bollen L, Vande Castele N et al. Rapid test for infliximab drug concentration allows immediate dose adaptation. Clin. Transl Gastroenterol 2016; in press.

- I33 -

INTESTINAL ORGANOID DERIVED FROM INFLAMED TISSUES REACH TRANSCRIPTION LEVELS COMPARABLE TO NON-INFLAMED TISSUES AND HEALTHY CONTROLS. M. NOBEN (1), N. HENDRIKS (2), S. VERMEIRE (3), G. VAN ASSCHE (3), C. VERFAILLIE (4), M. FERRANTE (3) / [1] KU Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] KU Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [3] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology, [4] KU Leuven, Belgium, Stem Cell Institute Leuven.

Introduction : The intestinal epithelium is the first line of contact between the host and microbiota, and other luminal particles which may have a pathogenic role in patients with inflammatory bowel disease (IBD). Previously we showed that culturing organoids from healthy controls (HC) and patients with Crohn's disease (CD) or ulcerative colitis (UC) works with equal efficiency, and that the transcriptional profiles are largely comparable (LGR5, MUC2, among others).

Aim : Here we investigated how the inflammatory burden affects cultured organoids.

Methods : Biopsies were derived from both the inflamed and non-inflamed mucosa of patients with IBD, including 5 patients with UC (3 males, median disease duration 4.6 years), and 7 patients with colonic Crohn's disease (CDc, 4 males, median disease duration 10.1 years), and 3 female HC. Next, crypts were isolated and colon organoids were derived following previous described methods (Sato et al., *Gastroenterology*, 2011). RNA was isolated both from the original biopsies, as well as from organoids at the end of passage 1 of culture (14 days after isolation, kept in expansion medium with Wnt3a, EGF, Noggin, Rspo1, B27, nicotinamide, p38-inhibitor, A83-01). We used RTqPCR to assess expression levels of the following genes : LGR5, CXCL8 (Interleukin-8), CXCL3, IL1 β , IFN γ , and TNF α . Ct values were normalized on the geometric mean of 3 reference genes (RPS14, HPRT1, and B2M). DeltaCt values were used for statistical analysis for : IBD vs HC for biopsy and organoid data and inflamed vs non-inflamed in the IBD groups.

Results : The intestinal stem cell marker LGR5 was equally expressed in the different groups and was enriched in organoids compared to biopsies (HC vs UC vs CD). A significant decreased in expression of TNF α was observed in organoids compared to biopsies for the CDc groups (in organoids derived from normal as well as inflamed tissue), while expression levels of TNF α in organoids were equal between the groups. CXCL8 and CXCL3 were upregulated in organoids compared to biopsies (regardless of the inflammatory status at the site of biopsy). mRNA levels of IL1 β and IFN γ were less expressed in organoids compared to primary biopsies, suggesting that removal from the inflammatory milieu leads to loss of the inflammatory phenotype.

Conclusions : Organoids derived from inflamed tissue have equivalent transcriptional profiles to those from non-inflamed tissue from the same patient, or from healthy controls. We identified that CXCL8 and CXCL3 transcripts are induced upon culture in all samples tested, suggesting that these markers might be less suitable for investigating a response in inflammation. Expression of other genes was not seen following culture and these may hence be more suitable for measuring inflammation in organoids. Most importantly, we could not detect significant differences in gene expression of a number of inflammatory genes in cultured organoids derived from inflamed and non-inflamed tissue of HC and IBD patients, indicating that removal from the in vivo inflamed environment results in loss of the inflammatory phenotype.

- I34 -

SEVERITY OF INFLAMMATORY BOWEL DISEASE IN IMMIGRANTS FROM MOROCCAN AND CAUCASIAN ORIGIN. L. De Ville De Goyet (1), T. Moreels (1), M. Denis (1), B. De Vroey (1), N. De Suray (1), C. Mouradides (1), A. Taha (1), C. De Bouvere (2), H. Piessevaux (1), O. Dewit (1). [1] Cliniques Universitaires St Luc, Brussels, Belgium, Hépatogastroentérologie, [2] Cliniques Universitaires Saint-Luc, Belgium, Hépatogastro-Entérologie.

Introduction : Moroccans are a growing minority in Belgium since the 1960s. Few studies have examined the characteristics of inflammatory bowel disease (IBD) in this population.

Aim : The aim of our study was to compare IBD characteristics between immigrants from Moroccan and Caucasian origin.

Methods : We performed a retrospective chart review of all first and second generation immigrants with Crohn's disease (CD) or Ulcerative colitis (UC) followed in our IBD Center from 2010 to 2016. Disease characteristics (Montreal classification), clinical variables and treatment were extracted to define disease severity and prognosis.

Results : A total of 83 Moroccans (57 CD / 26 UC, 53 men) and 72 Caucasian immigrants (42 CD / 30 UC, 37 men) patients were analyzed. For CD patients, penetrating disease (26/57 vs 11/42), perineal localization (24/57 vs 12/42) were more frequent in Moroccan. Although there were no significant differences in medication prescription between the two groups (immunosuppressive :IS (49/57 vs 32/42), biologics (32/57 vs 23/42), there was a higher number of hospitalizations (37/57 vs 15/42) and surgeries (33/57 vs 19/42) in the Moroccan population especially with the most severe disease who needed a definitive stoma (5/57 vs 0/42). For UC patients (26 Moroccans, 30 Caucasians), no differences in localization, medications or need for hospitalization were observed. Colectomy was performed in 4 Moroccans and 3 Caucasians.

Conclusions : We report a higher proportion of CD in Moroccan compared to Caucasian immigrants in our active IBD cohort. Moreover, Moroccan CD patients tend to have more severe disease, characterized by more penetrating disease, more perineal disease leading to higher rates of CD-related surgery and definitive stoma. Further research is required to confirm these observations and determine if these findings reflect genetic or environmental differences.

APPLICATION OF DRIED BLOOD SPOTS FOR PHARMACOKINETIC PROFILING OF GOLIMUMAB-TREATED PATIENTS WITH ULCERATIVE COLITIS. I. Detrez (1), V. Ballet(2), G. Van Assche (2), S. Vermeire (2), M. Ferrante (2), A. Gils (1) / [1] KU Leuven, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences, [2] UZ Leuven, Leuven, Belgium, Department of Clinical and Experimental medicine.

Introduction : Preventing loss of response to golimumab, an anti-tumour necrosis factor (TNF) biologic, is a challenge for clinicians treating patients with ulcerative colitis (UC). Although drug serum concentrations and anti-drug antibody serum concentrations have suggested an association between golimumab exposure and clinical outcome, detailed information on absorption, distribution and elimination of the drug in a real-life cohort of patients is lacking. Dry blood spot (DBS) sampling involves a finger prick to apply whole blood to a sampling paper after which drug is extracted.

Aim : We wanted to study if golimumab efficacy can be increased by exploring its full pharmacokinetic (PK) profile in UC patients by intensive sampling via DBS.

Methods : First, DBS were obtained through spotting of 45 μ L of golimumab (0.2-20 μ g/mL) or anti-golimumab antibody (20-200 ng/mL) spiked in whole citrated blood, to a filter paper. After punching, DBS were extracted and DBS extracts were analysed on both the MA-GOM171D8/MA-GOM159B8-HRP ELISA and the MA-GOM159B8 bridging ELISA. Extraction efficacy, accuracy, imprecision, sensitivity and robustness were determined as well as the impact of anti-golimumab antibodies on the detection of golimumab (and vice versa). Second, DBS were obtained by spotting blood obtained through a finger prick of eight golimumab-treated patients with UC (six females, mean (SD) age of 43 (16) years and median [IQR] duration under golimumab of 100 [72-114] weeks) of whom serum was taken simultaneously by venepuncture, allowing the calculation of a real-life conversion factor between golimumab serum concentration and DBS extract.

Results : The selected extraction condition yielded an average extraction efficiency of 54% and 53% for golimumab and anti-golimumab determination, respectively. Overall-assay accuracy and imprecision were between 80-120% and <15%, respectively, for each concentration analysed. Storing the sampling papers at room temperature for one month or the extracts at -20°C for three months did not impair DBS recovery. The presence of golimumab hampered the detection of anti-golimumab and vice versa. A real-life conversion factor of 3.8 ± 0.3 (n = 6) from DBS to serum was calculated. The blood volume per spot did not influence the results if it had at least a diameter of six mm, which was not the case in two out of eight patients and is the main drawback of this method.

Conclusions : The described DBS method is robust and can be used as a patient friendly and inexpensive method to perform rich sampling in patients treated with biologic agents. Proper patient education on how to sample is essential and will result in an accurate determination of exposure to golimumab in patients with UC. The method will be applied in a prospective cohort of ten patients with UC by collecting 20-40 DBS per patient over time.

INVESTING IN WORKABILITY OF PATIENTS WITH IBD : RESULTS OF A PILOT PROJECT ACTIV84WORK (ACTIVATE FOR WORK). S. Coenen (1), M. Schmickler (2), D. Staes (3), F. Wieme (3), T. Swinnen (4), X. Dekeuleener (5), T. Van Hooland (6), K. Van Den Cruys (7), S. Vermeire (1). [1] UZ Leuven, Leuven, Belgium, IBD, [2] Mensura, Belgium, [3] CCV, Belgium, [4] SD Worx, Belgium, [5] Proximus, Belgium, [6] Abbvie, Waver, Belgium, [7] Novellas healthcare, Belgium.

Introduction : Inflammatory bowel diseases (IBD) are chronic gastrointestinal conditions mainly affecting young people of active working age (15 to 40). Many of the symptoms of these diseases (frequent diarrhea, urgency, incontinence and/or fatigue) often make it difficult to actively participate in the workplace and commute to and from work especially during periods of flare. Activ84worK was a pilot project to stimulate professional activity and reduce absenteeism in IBD patients by providing patients with more flexible working conditions including teleworking.

Aim : The aim of Activ84worK was to improve both the well-being of the patient as well as his/her employer, and to contribute to a reduction in absenteeism in society by focusing on the workability of people with IBD.

Methods : Activ84worK was a collaboration between Abbvie, Mensura, Proximus, SD Worx and University Hospitals Leuven (UZ Leuven) with the support of the patient association "Crohn- en Colitis Ulcerosa Vereniging (CCV vzw) in Flanders, Belgium". Since April 2015, IBD patients were recruited by CCV and the gastroenterology department of UZ Leuven. Patients who showed interest, were contacted by Novellas healthcare (specialized in international healthcare recruitment and patient support programs) for screening and follow-up in the program. Informed consent was signed and both the employee and employer were followed for a duration of 6 months. The first and last contacts were face-to-face meetings, with employee and employer, and 5 intermediate contacts with the employee were done by phone or email. The project was evaluated from three perspectives : the benefit of teleworking for the employer, the employee, and the effect on society at large by measuring absenteeism.

Results : Between April 2015 and October 2016, 71 patients showed interest in the Activ84worK program, 19 were eligible to participate (because teleworking was allowed by the employer) and 14 completed the program (29% male, 29% private companies). Over the period of 6 months, all patients expressed their enthusiasm for tailored and flexible working conditions thanks to the option of teleworking. The case studies, based on interviews conducted with participating employees, indicated that removing work-related stress factors (such as not having a toilet nearby, or not being able to take a rest when needed) resulted in employees feeling much more at ease. Concretely, this led to fewer days of sick leave for most patients, a higher degree of workability and focus of employees, and a decrease in costs of absenteeism for employers and society. The pilot project was seen as a very positive experience by both parties. In addition, more openness was created between the employee and the employer, the taboo on the disease was lifted, and this had an overall positive impact on the work-life balance of patients.

Conclusions : This pilot project showed that teleworking and flexible working hours improved labor participation of patients with IBD. The results of this project are now used to inspire policy-makers and employers to gain maximum support for the chronically ill eager to work. We feel this initiative should be extended to a larger cohort of patients and should also be tested in other chronic diseases.

- 137-

MET DELETION IN MRP8+ NEUTROPHILS IS PROTECTIVE DURING DDS-INDUCED COLITIS VIA TH17 PATHWAY. M. Stakenborg (1), E. Meroni (1), G. Goverse (1), M. Di Matteo (2), M. Mazzone (2), G. Matteoli (3). [1] KU, Leuven, Belgium, TARGID, [2] KU, Leuven, Belgium, Department of Oncology, [3] KU Leuven, Belgium, TARGID.

Introduction : Neutrophils are essential to maintain intestinal homeostasis as they provide a first line of defense against invading pathogens. During inflammation, neutrophils also aid in the recruitment of other immune cells and facilitate the immune response in the gut. However, during chronic inflammatory conditions, such as Inflammatory Bowel Disease (IBD), excessive neutrophil accumulation can lead to tissue damage, delayed tissue repair and loss of homeostasis.

Aim : Thus, in the current study, we aim to identify the role and function of neutrophils recruited during intestinal inflammation and their contribution in the pathogenesis of colitis.

Methods : To block neutrophil chemoattraction and cytotoxicity in response to its ligand hepatocyte growth factor, we used the neutrophil-specific Mrp8-Cre line backcrossed with Metfl/fl. Acute colitis was induced in MRP8Cre/WT METfl/fl (KO) mice and MRP8WT/WT METfl/fl littermate controls (WT) by 2,5% dextran sodium sulfate (DSS) in drinking water for 5 days. Disease progression was assessed via a standardized disease activity index (DAI) including body weight loss, stool consistency and blood in the faeces. During chronic colitis, mice were subjected to 3 cycles of 2.5% DSS for 5 days followed by 2 weeks of drinking water. Colonic immune cells were assessed by flow cytometry. Data are expressed as mean \pm SEM; t-test was performed; $p < 0.05$ is considered significant.

Results : During the third cycle of chronic DSS colitis, KO mice displayed a decreased DAI ($p < 0.01$) and body weight loss ($p < 0.05$) compared to WT mice. Moreover, flow cytometric analysis revealed a reduced amount of ROS+ neutrophils (WT; $3.17 \pm 0.84 \times 10^5$, KO; $0.69 \pm 0.26 \times 10^5$, $p < 0.05$), eosinophils (WT; $3.00 \pm 0.62 \times 10^5$, KO; $0.80 \pm 0.20 \times 10^5$, $p < 0.05$), and macrophages (WT; $9.98 \pm 0.59 \times 10^5$, KO; $6.46 \pm 0.73 \times 10^5$, $p < 0.05$), implying a protective effect of MET deletion in MRP8+ neutrophils during chronic intestinal inflammation. To further elucidate the observed phenotype, we performed acute DSS colitis. In line, KO mice subjected to acute DSS colitis showed an improved disease course with reduced body weight loss and DAI and a comparable decrease in the amount of neutrophils, eosinophils and macrophages. Moreover, the percentage of FoxP3+ T regulatory cells was increased in KO mice compared to their WT counterparts (WT; $29.27 \pm 2.14\%$, KO; $41.05 \pm 2.11\%$, $p < 0.05$), pointing towards a return to homeostasis in the KO colon. Strikingly, analysis of CD3+ CD4+ T cells showed a predominant decrease of the percentage IL17A+ Th17 (WT; $26.87 \pm 1.85\%$, KO; $14.19 \pm 2.11\%$, $p < 0.01$) and IL17A+ IFN γ + Th1-like Th17 (WT; $10.62 \pm 0.50\%$, KO; $4.70 \pm 1.51\%$, $p < 0.01$) in KO mice compared to WT mice, while no differences were observed in the percentage of IFN γ + Th1 cells (WT; $12.03 \pm 0.61\%$, KO; $12.03 \pm 3.00\%$, ns).

Conclusions : In the present study, we showed that MET is required for neutrophil chemoattraction and cytotoxicity during colitis. MET deletion in neutrophils seems to be essential to limit inflammation in the lamina propria. In addition, MET deletion in neutrophils was associated with a specific reduction of Th17 cells. Further understanding the mechanisms underlying neutrophil function during colitis will aid in the development of novel therapeutic strategies to treat IBD patients.

PROFILE STUDY : PROSPECTIVE EVALUATION OF STEP UP THERAPY IN PATIENTS WITH EARLY UC IN BELGIUM. P. Bossuyt (1), F. Baert (2), J. Coenegrachts (3), M. De Vos (4), O. Dewit (5), M. Ferrante (6), F. Fontaine (7), F. Mana (8), J. Vandervoort (9), T. Moreels (10). [1] Imelda Hospital, Bonheiden, Belgium, Department of gastroenterology, [2] AZ Delta, Roeselare, Belgium, Department of gastroenterology, [3] Jessa Hospital, Hasselt, Belgium, Department of gastroenterology, [4] University Hospital Ghent, Ghent, Belgium, Department of gastroenterology, [5] Cliniques Universitaires Saint-Luc, Belgium, Department of Gastroenterology, [6] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of Gastroenterology, [7] CHC, Liège, Belgium, Department of Gastroenterology, [8] UZ Brussel, Jette, Belgium, Department of gastroenterology, [9] Onze-Lieve-Vrouw Ziekenhuis, Aalst, Belgium, Department of gastroenterology, [10] Antwerp University Hospital, Edegem, Belgium, Department of Gastroenterology.

Introduction : The natural history of ulcerative colitis (UC) is unpredictable. The current approach is gradual step-up (SU) therapy in the majority of patients. Data on the need for and factors influencing SU therapy beyond 5ASA or steroids are understudied.

Aim : To describe the first year SU therapy in patients with early UC failing on 5-ASA or steroids.

Methods : In this prospective multicentre observational trial patients with UC failing on 5-ASA and/or steroids were followed for 12 months. Patient characteristics, demographics, medical therapy, biomarkers, therapy adherence and quality of life were evaluated at every out-patient visit.

Results : A total of 103 patients (54% male, median age 40 years, median disease duration 17 months) were included. Only 2% were active smokers, while 51% were ex-smokers. Of the 103 patients 34%, 24% and 42% were 5-ASA-refractory, cortico-dependent and cortico-refractory respectively. After 1 year of follow up 81% of patients had mild or inactive UC based on the Mayo score. Sixty percent of patients had been treated with immunomodulators and 30% with biological therapy. Eighteen percent used combination therapy, representing only 54% of patients on anti-TNF therapy. The median time to initiation of immunomodulators and anti-TNF was 1 day and 55 days respectively, with a quicker initiation of anti-TNF treatment in cortico-dependent (34 days; 95% CI : 0-148) and cortico-refractory (57 days; 95% CI : 2-181) patients as compared to 5-ASA-refractory patients (97 days; 95% CI : 17-262). In total, 24/43 (56%) cortico-refractory patients started anti-TNF treatment. This was a significantly higher number compared to 4/25 (16%) of the cortico-dependent group ($p=0.002$) and 7/35 (20%) of the 5-ASA-refractory group ($p=0.002$). Biomarkers (CRP and platelet count) and clinical scores were numerically higher at initiation of anti-TNF therapy compared to immunomodulators. Whereas the use of faecal calprotectin was negligible (7%) in therapeutic decision making. Two patients underwent colectomy. Based on the results of the MMAS-8 questionnaire, patients with severe disease at baseline presented a lower median MMAS-8 score throughout the study period and thus were less adherent to therapy.

Conclusions : In patients with early UC a step up approach leads to good clinical outcomes at 1 year. Immunomodulators are initiated very early in patient flaring on 5-ASA or steroids, and up to 30% will be on anti-TNF treatment within 1 year, with cortico-refractory patients having the highest risk. Surprisingly, combination therapy is not used very often in daily clinical practice. The gradual SU and the acceleration of the therapy are based on sanguine biomarkers and clinical scores, not on faecal calprotectin levels.

DISEASE COURSE AND OPERATIVE RISK AFTER DIAGNOSIS OF ILEAL PENETRATING CROHN'S DISEASE : A COHORT STUDY. P. Bossuyt (1), C. Debeuckelaere (1), M. Ferrante (2), A. De Buck Van Overstraeten (3), T. Billiet (1), D. Vanbeckevoort (4), A. Wolthuis (3), A. D'hoore (3), G. Van Assche (1), S. Vermeire (1). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of gastroenterology, [2] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of gastroenterology, [3] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of abdominal surgery, [4] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of Radiology.

Introduction : Although penetrating complications are common in Crohn's disease (CD), little is known about the disease course and operative risk after diagnosis of small bowel penetrating CD complications.

Aim : To study the disease course and need for surgery in patients presenting with penetrating ileal CD.

Methods : In this cohort study, all cross-sectional imaging exams (CT and/or MRI) performed between 2006 and 2014 in patients with CD in a tertiary referral centre were reviewed for the presence of ileal penetrating complications (defined as abscesses, phlegmons or fistula). Demographic, clinical, biochemical, radiological and endoscopic factors were retrospectively assessed in these patients as well as the need for surgery (intestinal resections and/or strictureplasties) and postoperative complications.

Results : In total, we identified 1803 cross-sectional imaging exams in 957 CD patients. In 113 patients penetrating ileal CD complications were identified. The vast majority of these patients were sent to surgery (86%) over time. The median time to surgery was 1 month. Based on univariate analysis, the presence of abscesses ($p=0.003$) and increased C-reactive protein $>22\text{mg/L}$ (based on ROC curve analysis with AUC of 0.723) at documentation of the penetrating complication

($p=0.015$), were significantly associated with subsequent surgery. The post-operative course was complicated in 14% of patients. Surgery within one month after first documentation of penetrating disease ($p=0.004$) and previous CD related surgery ($p=0.01$) were significantly associated with postoperative complications. The presence of prestenotic dilation on imaging resulted in less postoperative complications ($p=0.02$). Previous therapy (corticosteroids, immunomodulators, anti-TNF alpha therapy) had no impact on the complication rate.

Conclusions : The vast majority of patients with penetrating ileal CD were sent to surgery over time. Abscesses and high inflammatory burden are the most important factors driving the multidisciplinary decision for surgery. Early surgery within one month after documentation of the penetrating CD manifestation was more likely to be associated with a complicated post-operative course, especially anastomotic leakage.

- I40 -

THE MODIFIED POSTOPERATIVE ENDOSCOPIC RECURRENCE SCORE FOR CROHN'S DISEASE : DOES IT REALLY MAKE A DIFFERENCE IN PREDICTING CLINICAL RECURRENCE? P. Rivière (1), S. Vermeire (2), G. Van Assche (2), P. Rutgeerts (2), A. De Buck Van Overstraeten (3), A. D'hoore (3), M. Ferrante (2). [1] Hôpital Haut-Lévêque, Bordeaux, France, Gastroentérologie et Nutrition, [2] UZ Leuven Gasthuisberg, Leuven, Belgium, Gastroenterology, IBD, [3] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of Abdominal Surgery.

Introduction : The endoscopic Rutgeerts' score is widely used to guide post-operative management of patients with Crohn's disease (CD). It is unclear whether all lesions from the i2 category should be considered clinically relevant. The modified Rutgeerts' score differentiates lesions at the anastomosis with or without < 5 isolated neo-terminal ileal erosions (i2a) from presence of ≥ 5 isolated neo-terminal ileal erosions with or without anastomotic lesions (i2b), but its predictive value has not been validated.

Aim : We investigated if clinical relapse (CR) and need for endoscopic/surgical intervention (ESI) differ between i2a and i2b endoscopic recurrence (ER).

Methods : This was a retrospective, single-center study including all consecutive patients with an i2 ER observed 6-12 months after right hemicolectomy with ileo-colonic anastomosis. The modified Rutgeerts' score was attributed based on the available endoscopic report and on the images captured during endoscopy. CR was defined as the occurrence of CD related symptoms along with biological, endoscopic (i3-i4) and/or radiologic signs of disease activity. ESI was defined as the need for balloon dilatation or stricturoplasty at site of the anastomosis, or new right hemicolectomy. Kaplan-Meier curves were plotted for time from index endoscopy to CR and ESI.

Results : The study population consisted of 94 patients [43 males, median age at index endoscopy 37 years], operated between December 2000 and December 2013. At index endoscopy, 53 patients (56%) had an i2a ER, and 41 (44%) an i2b ER. At endoscopy, the two groups were not different regarding disease characteristics and post-operative prophylactic therapy. Medical treatment was optimized or initiated according to index colonoscopy in 8 (15%) patients with i2a and 20 (49%) with i2b ER (Odds ratio (OR) 5.2 (95%CI 2.0-14.6), $p<0.001$). During a median (IQR) follow-up of 78 (37-109) months, CR and ESI were observed in 47 (50%) and 21 (22%) patients, respectively. As shown in Figures 1 and 2, the modified i2a and i2b scores were not predictive of CR and ESI (Log Rank $p=0.37$ and $p=0.10$, respectively). Median time to clinical relapse was 86.4 and 44.5 months in i2a and i2b patients, respectively. Cumulative probability of clinical relapse at 3 years was 31.8% (95%CI 17%-44%) and 46.3% (95%CI 28.7%-59.6%) in i2a and i2b patients, respectively. Also after exclusion of patients with immediate post-endoscopy treatment optimization, the modified i2a and i2b scores were not predictive of CR and ESI (Log Rank $p=0.73$ and $p=0.34$, respectively). A previous ileocolonic resection (OR 2.0 (95%CI 1.1-3.9), $p=0.04$) was associated with CR; immediate post-operative prophylactic therapy by anti-TNF was protective against CR ($p=0.03$). Post-operative prophylactic therapy by thiopurine was protective against ESI ($p=0.02$).

Conclusions : In this cohort, no difference was observed in terms of clinical relapse and need for endoscopic or surgical intervention between i2a and i2b ER after a right hemicolectomy with ileocolonic anastomosis in CD patients. Further study is needed to confirm these results and evaluate the outcome of Rutgeerts' score i2 patients.

- I41 -

PURSUIT-SC LONG TERM EXTENSION. P. Gibson (1), W. Reinisch (2), W. J Sandborn (3), P. Rutgeerts (4). [1] Alfred Hospital, Melbourne, VIC, Australia, Australia, Gastroenterology, [2] Universitätsklinik für Innere Medizin III, Vienna, Austria, Austria, Gastroenterology, [3] University of California San Diego, La Jolla, CA, USA, United States (the), Gastroenterology, [4] University Hospital, Gasthuisberg, Leuven, Belgium, Belgium, Gastroenterology.

Introduction : As maintenance therapy for UC is required long term, the current study examined the efficacy and safety through 4 years of maintenance golimumab among golimumab induction responders who were randomized to receive

golimumab during the maintenance study through week 52 and continued to receive golimumab during the long-term extension.

Aim : To evaluate safety & efficacy through 4 years (i.e. Wk228) of SC Golimumab maintenance in pts with moderately-severely active UC.

Methods : Among 1354 patients randomized and treated in induction, 1228 were enrolled into maintenance. During PURSUIT-Maintenance, GLM induction responders (464) were randomized to SC PBO, GLM 50mg, or GLM 100mg q4wks thru wk52 while 129 PBO induction responders continued on PBO; 635 non-responders to PBO or GLM induction received GLM100mg q4wks. Pts completing treatment thru wk52 & evaluation at wk54 were eligible to enter a study extension (LTE) for an additional 3y. Pts entered LTE at same GLM dose they received at the end of main study. During LTE, PBO- or GLM 50mg pts could cross over to GLM 100mg q4wks upon worsening of UC. GLM Pharmacokinetics (PK) and immunogenicity were evaluated during the LTE. Efficacy analyses are based on pts randomized to GLM at wk0 of maintenance who continued GLM during LTE. Safety analyses are based on all pts treated with GLM at any time from wk0 of induction thru wk228.

Results : 672 pts entered LTE : 666 were treated (96 PBO, 93 GLM50mg, 477 GLM100mg). The majority of study agent discontinuations were in PBO, 83.3% vs 37.2% GLM combined. PBO pts at study unblinding were discontinued ; majority of GLM pts discontinued due to AE and unsatisfactory therapeutic effect. PK analysis showed that serum GLM concentrations were dose proportional and maintained over time in pts receiving GLM50 mg or GLM100mg q4w in the LTE. The number of patients positive for antibodies to GLM was numerically higher in the 50mg vs100mg dose regimen (4.4% versus 3.7%). Efficacy analyses are based on observed data. At wk216 99.3% of pts receiving GLM had PGA of 0/1; 77.6% had PGA of 0; 76.1% had an IBDQ score >170. The proportion of pts not receiving corticosteroids (CS) thru week 216 was maintained; 92.5% were CS-free at wk216. AEs/100 pt years of follow-up thru wk 54 and 228 are presented (Table). Through Wk228, rates of AEs of special interest (e.g. demyelination, TB) and malignancy remained comparable to wk54. There were 4 NMSC [3-GLM 100mg & 1-GLM 50mg], 2 lymphoma [both GLM 100mg], & 7 solid tumors [5-GLM 100mg, 2 -GLM 50mg]). There were 9 deaths overall : 2 (previously reported) cardiac & respiratory insufficiency-[PBO], biventricular heart dysfunction-[GLM 50 mg] & 7 additional deaths : MI, gall bladder cancer, advanced rectal cancer, adenocarcinoma of the colon, sepsis, accidental nitrous oxide overdose, & massive aspiration post-colectomy (1 patient was not treated during the study extension, remaining GLM 100mg;).

Conclusions : These data continue to support a positive benefit/risk profile for GLM in the treatment of moderate-severe UC. GLM treatment for up to 4y maintained clinical benefit with no new safety signals observed; safety profile was similar to that observed through wk54.

- I42 -

ANTIBODIES TOWARDS VEDOLIZUMAB APPEAR FROM WEEK 2 ONWARDS AND DISAPPEAR UPON TREATMENT. S. Bian (1), H. Tang (1), M. Peeters (1), G. Compennolle (1), E. Dreesen (1), G. Van Assche (2), M. Ferrante (2), S. Vermeire (2), A. Gils (1). [1] KU, Leuven, Belgium, Department of Pharmaceutical and Pharmacological Sciences, [2] University Hospitals Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : Vedolizumab (VDZ), a monoclonal antibody (MA) that specifically binds to $\alpha 4\beta 7$ integrin, has demonstrated efficacy in patients with moderate-to-severe Crohn's disease (CD) and ulcerative colitis (UC). VDZ trough concentrations (TC) are typically >30 $\mu\text{g/ml}$ during induction and >10 $\mu\text{g/ml}$ during maintenance therapy, making it challenging for immunogenicity assays to detect anti-VDZ antibodies (AVA). The GEMINI trials reported AVA at ≥ 2 consecutive time points in 0.4% and 1% of patients with CD and UC, respectively.

Aim : To develop and validate a drug resistant AVA assay using a combination of VDZ/AVA complex precipitation and acid dissociation; to evaluate the incidence and the time course of AVA in 75 VDZ-treated patients (46 CD, 29 UC)

Methods : We developed a drug-resistant AVA assay in which AVA are complexed with an excess of VDZ followed by VDZ/AVA complex precipitation, acidification of the precipitate, coating and detection of released AVA using biotinylated VDZ. MA-VDZ19C11, a MA towards VDZ, was validated as calibrator. Drug resistance was examined by determination of the recovery of MA-VDZ19C11 in the presence of 3 different concentrations of VDZ. The cut-off was determined using 20 VDZ naïve patients. Cross reactivity with serum from 2 anti-infliximab antibody and 2 anti-adalimumab antibody positive patients, as well as with serum containing high concentration of rheumatoid factor (150 U/mL) was determined. The assay was subsequently applied to serum samples from 75 VDZ-treated patients (46 CD, 29 UC) taken at trough during induction (w6) and maintenance (w22). VDZ TC, AVA and CRP were determined in all available sera of patients positive for AVA at w6/22.

Results : MA-VDZ19C11 yielded dose-response curve ranging from 25–1600 ng/mL in 1/125 diluted serum allowing detection of AVA concentrations up to 200 $\mu\text{g/mL}$ equivalents. Spiking 1, 10 and 100 $\mu\text{g/mL}$ of VDZ to 40 $\mu\text{g/mL}$ MA-VDZ19C11 yielded recoveries of $89 \pm 7\%$ (mean \pm SD, n = 3), $90 \pm 11\%$ and $82 \pm 5\%$, respectively, confirming the complete drug resistance of the AVA assay. Cut-off for quantification was determined to be 1.1 $\mu\text{g/mL}$ MA-VDZ19C11 equivalents and none of the sera tested revealed cross-reactivity. Among the 75 VDZ-treated patients, 1 patient (1.3%) had AVA antibodies at w6 determined by a classical drug-sensitive bridging assay whereas 4 patients (5.3%) were AVA

positive on ≥ 2 time points using the drug-resistant AVA assay. AVA antibodies appeared from w2 onwards but disappeared over time. None of the 4 ADA-positive patients required VDZ intensification.

Conclusions : Using a drug-resistant AVA assay, AVA are detected in 5.3% of patients. Antibodies appear from w2 onwards and disappear upon time indicating their transient character.

- I43 -

SERUM PROTEOMIC ANALYSIS DEFINES NOVEL CIRCULATING INFLAMMATORY MARKERS FOR CROHN'S DISEASE AND RESPONSE TO ANTI-TNF THERAPY. H. Korf (1), J. Sabino (1), V. Ballet (2), M. De Bruyn (1), G. Van Assche (2), M. Ferrante (2), S. Vermeire (2). [1] KU Leuven, Belgium, Translational Research in Gastrointestinal Disorders (TARGID), Clinical and Experimental Medicine, [2] UZ Leuven Gasthuisberg, Leuven, Belgium, Department of Gastroenterology and Hepatology.

Introduction : A major challenge in understanding and managing inflammatory bowel disease (IBD) is the tremendous heterogeneity of the disease. Most therapeutic strategies are nevertheless aiming at reducing proinflammatory cytokines such as tumor necrosis factor (TNF). Anti-TNF agents are effective for IBD, but clinical remission and mucosal healing are observed in only 30-50% of patients.

Aim : We aim towards more tailored diagnostic and/or management practices by investigating biomarkers of disease activity and predictors of response to therapy.

Methods : Using the Proximity Extension Assay (PEA) technology, proteomic analysis was performed on paired serum samples from 18 patients with Crohn's disease (CD) before (week 0) and after (week 14) initiation of anti-TNF therapy (infliximab and adalimumab), as well as 20 healthy controls. A total of 90 parameters were analyzed with the Proseek Inflammation Panel (OLINK). Patients featuring endoscopic response (evaluated within a median of 41 days after the last sampling date) were grouped as responders. Wilcoxon rank-sum and Mann-Whitney tests (GraphPad Prism) were used and $p < 0.05$ were considered significant.

Results : A biomarker signature was observed for active CD with increased baseline levels of IL-8 ($p=0.006$), MCP3 ($p=0.012$), MCP4 ($p=0.048$), CXCL1 ($p=0.012$), TNFSF14 ($p=0.025$), CASP-8 ($p=0.01$), EN-RAGE ($p=0.02$), PDL-1 ($p=0.003$), TGFbeta1 ($p=0.031$), IL10RB ($p=0.038$), and decreased levels of 4E-BP1 ($p=0.034$) and FGF19 ($p=0.03$) compared to healthy controls. Three markers (IL-8 ($p=0.005$), MCP3 ($p=0.032$) and EN-RAGE ($p=0.042$)) decreased significantly in patients responding to therapy. Decreased levels of additional markers following response to anti-TNF therapy was observed (VEGFA ($p=0.009$), CDCP ($p=0.032$), IL-6 ($p=0.011$), MCP1 ($p=0.027$), IL17A ($p=0.024$), OSM ($p=0.014$), TGFA ($p=0.042$), CCL11 ($p=0.019$) and CCL3 ($p=0.032$). Finally, patients responding to treatment showed higher levels of IL-8 ($p=0.037$), SCF ($p=0.033$) and DNER ($p=0.026$) compared to their non-responder counterparts.

Conclusions : Utilizing the novel PEA technology, we identified a panel of inflammatory markers associated with CD activity. A combination of markers including IL-8, MCP3 and EN-RAGE was associated with response to therapy. Our results show the potential for serum proteomics to identify response following anti-TNF therapy, but validation in an independent cohort is required.

- I44 -

MATRIX METALLOPROTEINASE/MMP-9 DEFICIENCY DOES NOT INFLUENCE CHANGES IN GUT MICROBIOTA IN A MODEL OF ACUTE DEXTRAN SODIUM SULPHATE/DSS-INDUCED COLITIS. M. De Bruyn (1), J. Sabino (2), D. Vandeputte (3), S. Vermeire (4), J. Raes (3), G. Opdenakker (5). [1] Rega Institute for Medical Research, KU Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Immunobiology & Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] KU Leuven, Belgium, Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [3] Rega Institute for Medical Research, KU Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Molecular Bacteriology, [4] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology & Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [5] Rega Institute for Medical Research, KU Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Immunobiology.

Introduction : Commensal microbiota help to educate the immune system in the periphery and a number of involved immune cells have recently been characterized. However, specific molecular determinants in these processes are not known and, reciprocally, little information exists about single host determinants that alter the microbiome. Matrix metalloproteinase (MMP)-9 deficiency has previously been linked to alterations in gut microbiota composition in a model of infectious colitis (Rodrigues DM et al., BMC Microbiol. 2012 Jun 13;12 :105).

Aim : To investigate whether MMP-9 deficiency influences changes in gut microbiota in a model of inflammatory colitis induced by dextran sodium sulphate (DSS).

Methods : Acute colitis was induced in MMP-9 knockout (KO) mice (n=10) and their wild-type (WT) littermates (n=10) via oral administration of 3% DSS for 7 days followed by 2 days of regular drinking water. Control mice (10 WT and 10 MMP-9 KO) received normal drinking water throughout the experiment. Both genotypes were raised under identical environmental conditions for more than 15 years and were co-housed during the experiment according to phenotype (control vs DSS). Fecal samples were collected at time of sacrifice and immediately frozen at -80°C. Illumina MiSeq sequencer was used for 16S rDNA paired-end sequencing targeting the V4 hypervariable region. Sequencing depth was downsized to 10000 reads/sample. Taxonomic annotation was performed with Ribosomal Database Project. PICRUSt was used for metagenome prediction and analyzed with STAMP software (version 2.1.3). R software (version 3.3.0) was used for statistical analysis with multiple testing correction (Bonferroni).

Results : No significant differences in clinical or histopathological parameters were found between both genotypes (WT and MMP-9 KO) after induction of acute colitis. Observed microbial richness (genus level, t-test) and microbiota composition (Bray-Curtis dissimilarities, adonis) were not significantly influenced by genotype. In contrast, weight loss, disease activity index, cage and phenotype (control vs DSS) did significantly influence the intestinal microbiota composition (envfit, $r^2 > 0.7$, $p = 0.001$). The genera *Bacteroides* and *Alistipes* explained most of the variability in microbiota composition between genotype in the control group, whereas this was the case for the genera *Bacteroides* and *Allobaculum* in the DSS group (Constrained Principal Coordinate Analyses, capscale). After multivariate analysis (MaAsLin, $p < 0.05$), however, cage was identified as the sole driver of microbiota composition variability. Functional profiling indicated that both genotype and phenotype influenced the metagenome (PICRUSt). However, after multiple testing correction, only phenotype remained significantly associated with changes in metagenomic profile. The top metagenomic pathways influenced by DSS included ion coupled transporters and fructose and mannose metabolism.

Conclusions : Changes in gut microbiota composition were mainly driven by DSS and were not significantly altered by MMP-9 gene knockout.

- I45 -

RESPONSE TO THIOPURINES IS INDEPENDENT OF ATG16L1 GENOTYPE. B. Verstockt (1), A. Matthias (2), K. Strongili (2), I. Cleynen (3), M. Parkes (2). [1] Translational Research Center for Gastrointestinal Disorders (TARGID), KU Leuven, Leuven, Belgium, Department of Clinical and Experimental Medicine, [2] University of Cambridge and Addenbrooke's Hospital, United Kingdom (the), Gastroenterology Unit, [3] Laboratory of Complex Genetics, KU Leuven, Leuven, Belgium, Department of Human Genetics.

Introduction : Thiopurines, like azathioprine (AZA) and 6-mercaptopurine (6-MP), are effective in maintaining remission in Crohn's disease (CD). Due to potential adverse effects and the increased availability of biological therapies, their role might be questioned.

Aim : We aimed to identify clinical characteristics associated with an increased response to thiopurines. Additionally we wanted to validate the recently discovered ATG16L1 T300A single nucleotide polymorphism (SNP), associated with response to thiopurines.

Methods : Medical records of 230 British CD patients were retrospectively assessed. Response to thiopurines was defined as continued usage up to chart assessment or termination because of prolonged disease remission on thiopurine monotherapy; non-response as requirement for significant additional therapy (>1 course corticosteroid / year or addition of biologics) after 4 months on thiopurine. Patients who stopped thiopurines because of intolerance or immediately started combo therapy were excluded from genetic association analysis. Genotyping data (rs2241880, T300A) were available for 128 patients (UK IBDGC). Association analysis was performed via PLINK (chi-square test). A p-value <0.05 was considered significant.

Results : Most (87.0%) of the 230 included patients (111 men, median age at diagnosis 21 years) were administered AZA, with 23.9% ever receiving 6-MP. 24.8% of all patients had to stop thiopurines due to side effects (6 leukopenia, 5 abnormal LFT's, 6 pancreatitis, 1 lymphoma and 39 patients - intolerance unknown). A response rate of 57.8% was observed in patients who tolerated therapy. No difference in response rates was noticed depending on either disease location, disease behaviour, sex or smoking status. Conversely, absence of perianal disease was significantly associated with response to thiopurines (OR=2.8, $p = 0.003$). The ATG16L1 minor allele, A, was not represented more often in responders compared to non-responders (minor allele frequency 41.8% vs 43.0% respectively, $p = 0.85$). Additionally we could not identify a significantly higher proportion of AA homozygotes in thiopurine responders (14/71 vs 9/34, $p = 0.63$), as identified in a Dutch cohort.

Conclusions : Although thiopurines have to be stopped due to intolerance in approximately one quarter of patients, they can still maintain clinical remission in an important subset. Apart from the absence of perianal disease, we could not identify any clinical parameters which might help to stratify patients towards thiopurine monotherapy. Similarly, we could not validate the previously reported predictive value of ATG16L1 genotyping.

SERUM MARKER PANEL FOR EARLY DETECTION OF ENDOSCOPIC HEALING WITH INFLIXIMAB IN PATIENTS WITH ULCERATIVE COLITIS. M. De Bruyn (1), R. Ringold (2), M. Ferrante (3), G. Van Assche (4), G. Opdenakker (5), A. Dukler (2), S. Vermeire (4). [1] Rega Institute for Medical Research, KU Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Immunobiology & Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] Kepler Diagnostics, Inc., United States (the), Kepler Diagnostic, Inc., [3] University hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology & Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [4] University Hospitals Leuven, KU Leuven, Leuven, Belgium, Department of Gastroenterology and Hepatology & Department of Clinical and Experimental Medicine, Translational Research Center for Gastrointestinal Disorders (TARGID), [5] Rega Institute for Medical Research, KU Leuven, Belgium, Department of Microbiology and Immunology, Laboratory of Immunobiology.

Introduction : The need for surrogate markers to detect endoscopic healing in inflammatory bowel disease (IBD) is imminent. Previously, neutrophil gelatinase B-associated lipocalin and matrix metalloproteinase-9 (NGAL-MMP-9) complex was found to be superior to CRP for detection of endoscopic healing with infliximab (IFX) in patients with ulcerative colitis (UC) (de Bruyn M, *Inflamm Bowel Dis*. 2014). The combination of NGAL-MMP-9 with CRP and neutrophils increased the sensitivity and specificity. Cathelicidin LL-37 is an antimicrobial protein found in lysosomes of neutrophils and plays a role in innate immune defense. Chitinase 3-like 1 (CHI3L1) is secreted by neutrophils and is a growth factor for vascular endothelial cells and fibroblasts. Both markers were previously associated with IBD (Koon HW, *Gastroenterology* 2011 and Buisson A, *Aliment Pharmacol Ther* 2016).

Aim : To study if LL-37 and/or CHI3L1 could improve detection of endoscopic healing with IFX in UC patients.

Methods : Serum samples were obtained from 145 UC patients (41% female, median [interquartile range, IQR] age 41.3 [30.8-51.9] years) who underwent endoscopy following IFX initiation and from 75 healthy individuals who served as controls (56% female, median [IQR] age 33.6 [29.2-51.8] years). Endoscopic healing with IFX was defined as a Mayo endoscopic subscore of 0 or 1. CRP, NGAL-MMP-9 and neutrophils were previously determined, and LL-37 and CHI3L1 were measured with ELISA (Hycult Biotech and R&D systems, respectively). For all markers, optimal cut-offs were determined with ROC analysis and binary variables were entered in a logistic regression model to generate the Ulcerative Colitis Response Index (UCRI). Non-parametric statistical tests were performed and p-values <0.05 were considered significant.

Results : Median (IQR) time to serum sampling after start of IFX was 8.2 (6.0-14.0) weeks. 83 patients (57%) had endo-scopic healing, whereas 62 patients (43%) did not have endoscopic healing. Median [IQR] LL-37 levels (ng/ml) were significantly lower in healers (24.3 [16.1-41.4]) compared to non-healers (37.3 [24.0-53.8], p=0.002), but remained elevated compared to controls (16.7 [10.2-27.1]; p<0.001). Median [IQR] CHI3L1 levels (ng/ml) were significantly lower in healers (20.9 [14.3-34.4]) compared to both non-healers (30.0 [22.7-53.9], p<0.001) and controls (31.9 [19.6-48.6], p=0.003). UCRI consisted of a combination of CRP (Odds ratio [95% confidence interval] 3.3 [1.4-7.5]), CHI3L1 (3.1 [1.3-7.7]), neutrophils (4.9 [2.1-11.2]) and LL-37 (2.5 [1.0-6.4]). The area under the curve (AUC) of UCRI was 0.83 and quartile (Q)1 (0.0-2.6) was able to discriminate healing with 54% sensitivity, 92% specificity, 60% negative predictive value (NPV) and 90% positive predictive value (PPV), whereas Q4 (7.2-9.8) was able to discriminate non-healing with 37% sensitivity, 95% specificity, 67% NPV and 85% PPV. Finally, UCRI could detect endoscopic healing as early as 3 weeks after IFX initiation (Hazard ratio [95% CI] 4.1 [2.6-6.5]).

Conclusions : In the search for surrogate markers of endoscopic healing, UCRI (CRP, CHI3L1, neutrophils and LL-37) was shown to accurately identify UC patients who fail to achieve healing with IFX and may help in early decision making to switch treatment.

IMPACT OF ILEOCECAL RESECTION IN CROHN'S DISEASE PATIENTS ON FECAL MICROBIOTA. K. Machiels (1), V. Pascal (2), J. Sabino (3), A. Santiago (2), D. Campos (2), A. Wolthuis (4), A. De Buck Van Overstraeten (4), A. D'hoore (4), G. Van Assche (3), M. Ferrante (3), P. Rutgeerts (3), C. Manichanh (2), S. Vermeire (3). [1] University Hospitals Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [2] Vall d'Hebron Research Institute, Barcelona, Spain, Department of Gastroenterology, [3] University Hospitals Leuven, Leuven, Belgium, Translational Research Center for Gastrointestinal Disorders (TARGID), [4] University Hospitals Leuven, Leuven, Belgium, Department of Abdominal Surgery.

Introduction : Dysbiosis of the intestinal microbiota is implicated in Crohn's disease (CD) and may play an important role in triggering postoperative disease recurrence (POR).

Aim : We hypothesized that the fecal microbial recolonization process after ileocecal resection differs between patients developing recurrence and patients remaining in remission, and further aimed to identify other factors influencing the microbial composition.

Methods : Fecal samples from 54 CD patients undergoing ileocecal resection with ileocolonic anastomosis were prospectively collected before surgery and at month 1, 3 and 6 after surgery. POR - defined by a modified Rutgeerts score ≥ 2 on endoscopy was assessed at month 6. The microbiota was evaluated by 16S rDNA sequencing using an Illumina MiSeq platform. Calculation of alpha and beta diversity and statistical analysis were performed in QIIME.

Results : Patients developing early POR (N=23) harbored more Coriobacteriaceae, Corynebacteriaceae and Micrococcaceae in their faecal samples before surgery, than patients without recurrence (N=31) ($p < 0.04$). During the first 3 months postoperatively, no significant taxonomic differences were observed between both patient groups. At month 6, recurrence patients had a higher relative abundance of Fusobacteria (FDR=0.09). The impact of resection on the fecal microbiome was shown by an increase of Negativicutes (FDR=0.02) and reduction of Bifidobacteriales (FDR=0.04) in all CD patients whereas recurrence patients additionally were marked by an increase of Fusobacteria (FDR=0.03) and decrease of Faecalibacterium ($p=0.04$). Smoking (N=16) was not associated with early POR in this cohort, but smoking did impact on the fecal microbiota. Alpha diversity was significantly reduced in active smokers at baseline ($p=0.028$), month 3 ($p=0.016$) and month 6 ($p=0.023$) after surgery. In general, smokers were characterized by an enrichment of Veillonellaceae (FDR=0.09) and reduction of the two most abundant families from the order Clostridiales, namely Ruminococcaceae (FDR=0.005) and Lachnospiraceae (FDR=0.03). Within these families, the relative abundance of essential types of butyrate and other short-chain fatty acids-producing bacteria such as Faecalibacterium, Roseburia, Dorea, Coprococcus, Blautia and Ruminococcus were depleted.

Conclusions : Ileocecal resection has an impact on the fecal microbiota composition which mostly affects members of Negativicutes and Bifidobacteriales. The microbial differences between patients developing recurrence and patients remaining in remission are minor during the first 3 months whereas early recurrence at month 6 was mainly associated with an enrichment of Fusobacteria. Although smoking was not associated with early POR, it did show a significant impact on the microbial composition which might have potential implications at later stages of the disease.

BELGIAN GROUP FOR DIGESTIVE ONCOLOGY (BGDO)

- 001 -

YPT0N+ : THE OUTCASTS IN PATHOLOGICAL COMPLETE TUMOR RESPONSE AFTER NEOADJUVANT CHEMORADIATION FOR ESOPHAGEAL CANCER. HOW DO THEY FARE? G. Vervloet (1), L. Depypere (1), J. Moons (1), H. Van Veer (1), W. Coosemans (1), P. Naftoux (1). [1] University Hospitals Leuven, Leuven, Belgium, Thoracic Surgery.

Introduction : Little is known about the prognostic significance of residual nodal disease in otherwise complete pathologic responders (ypT0N+) after neoadjuvant chemoradiation (nCRT) for esophageal cancer. Current staging systems and treatment guidelines do not provide prognostic information or management recommendations for ypT0N+ patients and often a postsurgical follow-up policy is adapted.

Aim : The purpose of this study is to analyze the long-term outcomes of esophageal cancer patients with a pathologic response characterized as ypT0N+ following nCRT and esophagectomy.

Methods : From our prospectively build single institution database, 466 consecutive esophageal cancer patients undergoing esophagectomy after nCRT between 1996 and 2016 were collected. ypT0N+ responders were identified and were compared to patients with pathological complete response (ypT0N0) and to pathological non-complete responders (both ypT+N0 and ypT+N+).

Results : Out of 466 patients, 149 (32.0%) ypT0N0, 31 (6.7%) ypT0N+, 141 (30.3%) ypT+N0 and 145 (31.1%) ypT+N+ patients were identified. Clinical staging before nCRT was comparable between all groups. Likewise, no statistical difference ($p=0.109$) was found in the number of resected lymph nodes across the groups with a mean of 24.8 nodes in ypT0N0; 30.0 nodes in ypT0N+; 24.8 nodes in ypT+N0 and 26.4 nodes in ypT+N+ patients. Median overall survival (OS) was worse in ypT0N+ (21.7 months) and ypT+N+ (16.8 months) patients compared to ypT0N0 (55.2 months) and ypT+N0 patients (42.0 months). Stratification according to histology showed a significantly ($p<0.0001$) higher complete pathological response on primary tumor of 62.5% in 184 squamous cell carcinomas (SCC) compared to 23.0% in 282 adenocarcinomas (ADC). The proportion of patients with residual nodal disease in complete responders on primary tumor, being 22% in ADC and 15% in SCC, showed no statistical difference ($p=0.25$). In ADC, locoregional recurrence in ypT0N+ patients (43%) was comparable to ypT+N+ (31%) and more common compared to ypT0N0 (7%) and ypT+N0 (10%) patients, which is reflected in median overall survival rates of 20.6, 17.5, 53.0 and 36.6 months for the respective groups. Median overall survival in ADC is significantly determined by the number of positive lymph nodes, being 21.7 months for pN1 and 2.7 months for pN2/3 ($p=0.005$) in ypT0N+ and 33.7 months for pN1 and 16.2 months for pN2/3 ($p=0.031$) in ypT+N+. In SCC, locoregional recurrences were found in 17% of ypT0N+, 33% of ypT+N+, 11% of ypT0N0 and 22% in ypT+N0 patients and median overall survival was 26.6, 15.6, 55.2 and 43.8 months respectively. In SCC ypN+ the number of affected lymph nodes showed no statistically significant difference on overall survival.

Conclusions : Residual nodal disease in esophageal cancer patients with complete response in the primary tumor following nCRT has a poor prognosis and behaves similar to non-complete responders with positive lymph nodes. However stratification by histology shows that this is especially true in ADC but seems determined by the number of involved lymph nodes. These findings open the debate on how to treat these ypT0N+ patients with adjuvant treatment after surgery.

- 002 -

PRIMARY TUMOR SIDEDNESS HAS AN IMPACT ON PROGNOSIS AND TREATMENT OUTCOME : RESULTS FROM THREE RANDOMIZED STUDIES OF PANITUMUMAB PLUS CHEMOTHERAPY VERSUS CHEMOTHERAPY (PLUS BEVACIZUMAB) IN 1ST AND 2ND LINE RAS/BRAF WT MCRC. N. Boeckx (1), A. Toler (2), K. Op De Beeck (1), G. Kafatos (3), V. Deschoolmeester (4), C. Rolfo (5), K. Lowe (6), G. Van Camp (7), G. Demonty (8), M. Peeters (5). [1] Universiteit Antwerpen, Antwerpen, Belgium, Center for Oncological Research (CORE) / Center of Medical Genetics, [2] Amgen, Inc, Thousand Oaks, CA, United States (the), Center for Observational Research, [3] Amgen Ltd, Uxbridge, United Kingdom (the), Center for Observational Research, [4] Universiteit Antwerpen / Antwerp University Hospital, Antwerpen, Belgium, Center for Oncological Research (CORE) / Department of Pathology, [5] Universiteit Antwerpen / Antwerp University Hospital, Antwerpen, Belgium, Center for Oncological Research (CORE) / Department of Oncology, [6] Amgen, Inc, Seattle, WA, United States (the), Center for Observational Research, [7] Universiteit Antwerpen, Antwerpen, Belgium, Center of Medical Genetics, [8] Amgen Inc, Brussels, Belgium, Regional Medical Development.

Introduction : Previous communications have reported that the location of the primary tumor has a prognostic impact in metastatic colorectal cancer (mCRC). Other studies comparing biological agents in patients with KRAS or RAS WT mCRC revealed that the primary tumor location might also influence the efficacy of these agents. It has been proposed that tumor sidedness is a surrogate for the different biological characteristics of the primary tumor.

Aim : In this analysis, we aim to evaluate the association between tumor sidedness and anticancer treatment efficacy in mCRC patients with a RAS/BRAF WT primary tumor.

Methods : Data from three randomized Amgen-sponsored clinical trials were analyzed for treatment outcomes in relation to primary tumor sidedness. Our analysis includes a first-line phase 3 study (PRIME; NCT00364013, panitumumab plus FOLFOX vs FOLFOX alone), a first-line phase 2 study (PEAK; NCT00819780, panitumumab plus mFOLFOX6 vs bevacizumab plus mFOLFOX6) and a second-line phase 3 study (181; NCT00339183, panitumumab plus FOLFIRI vs FOLFIRI alone). Only RAS/BRAF WT patients were analyzed in order to have a biomarker refined patient population. Information on tumor sidedness (left/right colon) was obtained from the free-text surgery descriptions and from the original pathology reports. Primary tumors located in the caecum to transverse colon were categorized as right-sided tumors and those located from the splenic flexure to the rectum were categorized as left-sided tumors.

Results : We were able to determine the tumor location in more than 80% of the included patients. Between 80% and 85% of these patients were categorized as patients with left-sided CRC. The number of patients with right-sided tumors in the different treatment arms was relatively small (R/L panitumumab arm vs comparator arm : PRIME : 26/156 vs 32/148, PEAK : 13/52 vs 13/53, 181 : 22/143 vs 26/144). In patients with left-sided tumors, better PFS and OS outcomes were observed in the panitumumab arm compared to the comparator arm (PRIME - OS : 32.5 vs 23.6 months, HR=0.67; PFS : 12.9 vs 9.3 months, HR=0.69 / PEAK - OS : 43.4 vs 32.0 months, HR=0.77; PFS : 14.6 vs 11.5 months, HR=0.67 / 181 - OS : 20.1 vs 16.9 months, HR=0.97; PFS : 8.0 vs 6.6 months, HR=0.89). The response rate (complete response plus partial response) was also higher in the panitumumab arm compared to the comparator arm in patients with left-sided mCRC (PRIME : 70.3 vs 54.8%; PEAK : 63.5 vs 58.5%; 181 : 50.7 vs 13.5%). In right-sided CRC, the small number of patients does not allow drawing definitive conclusions, but a lack of efficacy of panitumumab was not confirmed, contradicting previous reports on the impact of tumor sidedness (PRIME - OS : 22.5 vs 21.5 months, HR=0.94; PFS : 8.9 vs 7.3 months, HR=0.71; RR : 52.0 vs 41.2% / PEAK - OS : 22.5 vs 23.3 months, HR=0.63; PFS : 10.3 vs 12.6 months, HR=0.88; RR : 69.2 vs 46.2% / 181 - OS : 11.9 vs 10.9 months, HR=0.84; PFS : 6.8 vs 3.7 months, HR=0.62; RR : 19.0 vs 3.8%). However, it was clear that patients with right-sided tumors did worse for all parameters compared to those with left-sided tumors in all treatment arms which confirms the prognostic effect of tumor location.

Conclusions : The results of these retrospective analyses in a homogenous RAS/BRAF WT subpopulation confirm that tumor sidedness has a prognostic impact in mCRC as patients with right-sided mCRC were associated with poor prognosis regardless of treatment received. Moreover, the addition of panitumumab provides a benefit over chemotherapy with or without bevacizumab in left-sided tumors. No final conclusions can be drawn on the optimal treatment in patients with right-sided primary tumors due to the low number of patients in this subgroup.

- 003 -

INTERLEUKIN 15-STIMULATED NATURAL KILLER CELLS CAN KILL BOTH PANCREATIC CANCER AND STELLATE CELLS. J. Van Audenaerde (1), J. De Waele (1), E. Marcq (1), E. Lion (2), J. Van Den Bergh (2), R. Jesenofsky (3), A. Masamune (4), G. Roeyen (5), P. Pauwels (6), F. Lardon (1), M. Peeters (7), E. Smits (1). [1] University of Antwerp, Antwerp, Belgium, Center for Oncological Research, [2] University of Antwerp, Antwerp, Belgium, Laboratory of Experimental Hematology, [3] University of Heidelberg, Heidelberg, Germany, Department of Medicine II, [4] Tohoku University Graduate School of Medicine, Sendai, Japan, Division of Gastroenterology, [5] Antwerp University Hospital, Edegem, Belgium, Department of Hepatobiliary, Endocrine and Transplantation Surgery, [6] Antwerp University Hospital, Edegem, Belgium, Department of Pathology, [7] Antwerp University Hospital, Edegem, Belgium, Department of Oncology.

Introduction : Pancreatic ductal adenocarcinoma (PDAC) is the 4th leading cause of cancer-related death in Western countries with a 5-year survival rate below 5%. Within the tumour microenvironment, a strong desmoplastic reaction occurs that is orchestrated by activated pancreatic stellate cells (PSC), resulting in a functional and mechanical shield. Tackling this stromal shield is needed to overcome treatment resistance.

Aim : Since conventional therapies have limited effects, we investigated innate immunotherapy for PDAC, more specifically the possibility to kill pancreatic cancer cells (PCC) as well as PSC with interleukin (IL-15 activated natural killer (NK) cells.

Methods : Peripheral blood NK cells were purified from healthy controls and stimulated overnight with recombinant human IL-15. The effect of IL-15 stimulation on the surface expression of several NK cell receptors was evaluated. NK cell-mediated cytotoxicity against three PCC lines and three PSC lines was measured using a 4h flow cytometric annexin V/propidium iodide assay. The impact of cell-to-cell contact and soluble mechanisms on NK cell-mediated killing were assessed using a transwell system and neutralizing antibodies, respectively. Subsequently, the cytotoxic potential of NK cells towards PSC, both isolated from PDAC patients, was tested in an autologous setting. Finally, we charted the expression of several NK cell ligands on PSC using flow cytometry.

Results : IL-15-activated NK cells have the capacity to significantly kill PCC and PSC cell lines (9-35% and 20-50%, respectively), as compared to resting peripheral blood NK cells, in a contact-dependent manner. NK cell-mediated killing was confirmed in the autologous setting in 4 out of 5 patients. IL-15 induces significant upregulation of TIM-3 and

NKG2D. Our experiments investigating the mechanism of IL-15-activated NK cell cytotoxicity point towards involvement of these two receptors. Furthermore, we observed expression of PD-L1, PD-L2, MICA/B, ULBPs and Galectin-9 on PSC of PDAC patients.

Conclusions : We are the first to demonstrate that both human PCC and PSC can be killed in vitro by IL-15-stimulated NK cells. Our results in the autologous setting stresses the therapeutic potential of IL-15 in the treatment of pancreatic cancer and reveals possible future targets to tackle remaining PSC.

- 004 -

CHARACTERIZATION OF THE IMMUNE MICROENVIRONMENT AND RELATION TO PREOPERATIVE TREATMENT OF SYNCHRONOUS RESECTION OF PRIMARY TUMOR AND LIVER COLORECTAL CANCER METASTASES. M. Van Den Eynde (1), B. Mlecnik (2), G. Bindea (3), J. Machiels (4), A. Jouret-Mourin (5), P. Baldin (6), A. Kartheuser (7), D. Leonard (8), C. Remue (7), J. Gigot (8), C. Hubert (9), Y. Humblet (4), N. Haicheur (10), F. Marliot (10), F. Pagès (10), J. Galon (3). [1] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Digestive Oncology, [2] Centre de Recherche des Cordeliers, Paris, France, Integrative cancer Immunology, [3] Centre de Recherche des Cordeliers, Paris, France, Integrative cancer immunology, [4] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Oncology, [5] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Pathology, [6] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Pathology, [7] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Digestive Surgery, [8] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Digestive Surgery, [9] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Digestive Surgery, [10] Hôpital Européen Georges Pompidou, Paris, France, Immunology.

Introduction : Previous reports suggest that the adaptive Th1 immune response observed in resected primary colorectal tumor (PCT) and liver colorectal metastases (LCM) is a recognized prognostic factor and is often associated with a tumor response to preoperative treatment.

Aim : We investigated, in a subgroup of patients synchronously resected for PCT and LCM, if this immune microenvironment could be different in PCT compared to LCM and the relation to the preoperative treatment administered

Methods : From a cohort of 161 patients undergoing curative LCM resection, 29 patients with synchronous resection of LCM (n = 102) and PCT after different preoperative treatment were analyzed. The density of immune cells (CD3, CD8, CD45RO, CD20, FoxP3) was quantified with a dedicated image analysis software on whole-slide imaging in the core (CT) and invasive margin (IM) of all synchronous LCM and PCT. Comparisons were made using the Wilcoxon-Mann-Whitney test. A CD3/CD8 Immunoscore (I) was calculated and ranged from 0 (I0), when low densities of both cell types are found in the CT and IM of the tumor, to 4 (I4) when high densities for both markers are found in both regions. Distribution of (I) was analyzed and compared using the Fisher's exact test.

Results : Global analysis of immune cell density in LCM and corresponding PCT showed no significant correlation. Compared to PCT, LCM were more frequently associated with a high T-cells infiltration in CT and IM ($p < 0.001$). Conversely, high CD20 and FoxP3 density were higher in the CT of PCT ($p < 0.005$). PCT had low proportion of I3-4 (13,8%) compared to the least (41,4%, $p=0,04$), the mean of all (62,1%, $p=0,0003$) and the most infiltrated LCM per patient (65,5%, $p=0,0001$). A significant difference for immune microenvironment was observed in LCM and PCT after preoperative treatment. Anti-EGFR treatment was significantly associated with an increase of T-cells in CT of LCM but not in PCT ($p < 0.001$).

Conclusions : PCT had different immune microenvironment and was significantly associated with a worse immunoscore compared to the synchronously resected LCM. Preoperative treatment had different impact on synchronous PCT and LCM immune microenvironment. Anti-EGFR treatment increases T cells densities in the CT of the LCM but not in the PCT, possibly suggesting a specific treatment effect in this tumor region.

- 005 -

PROGNOSTIC RELEVANCE OF PANCREATIC NEUROENDOCRINE TUMORS GRADING ON EUS-FNA. L. Boutsen (1), I. Borbath (2), A. Van Maanen (3), A. Mourin (4), B. Weynand (5). [1] Université catholique de Louvain, Belgium, Faculté de médecine, [2] Cliniques Universitaires Saint-Luc, Belgium, Hépatogastro-entérologie et Oncologie digestive, [3] Cliniques Universitaires Saint-Luc, Belgium, Centre du Cancer Institut Roi Albert II, [4] Cliniques Universitaires Saint-Luc, Belgium, Anatomopathologie, [5] UZ Leuven Gasthuisberg, Leuven, Belgium, Pathology.

Introduction : In the WHO 2010 classification, resection specimens of pancreatic neuroendocrine tumors (pNETs) are graded using the Ki67-labeling index (Ki67-LI) (G1 : Ki67-LI < 2% ; G2 : 3-20%, G3 : > 20%) . These past few years, endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) has become an important diagnostic tool of pNETs by collecting cytological samples. Although many studies have considered the diagnostic accuracy of EUS-FNA, only few

have dealt with grading of pNETs in EUS-FNA. This study is an extension of a previously published paper from our team, that assessed prognostic value of Ki67-LI on EUS-FNA in 46 pNETs (33 surgically resected).

Aim : To compare Ki67-LI on cytology with the ones obtained on surgical specimens. Analysis regarding influence of tumor size and number of counted cells in FNA grading will secondly be addressed, along with overall survival (OS) and progression free survival (PFS) estimates of all patients based on cytological grade.

Methods : Between 1996 and 2013, 102 pNETs from 101 patients (57 required surgery) were retrospectively included in this multicentered study. All of them underwent EUS-FNA (22 or 25-gauge needle) at the time of diagnosis. Cytological Ki67-LI was evaluated on FNA material of the 102 tumors (200 cell count). In a subgroup of 29 FNA specimens, more than 2000 cells were counted (14 patients underwent surgery). For patients who underwent surgery, Ki67-LI of resected tumor was assessed (more than 2000 counted cells) and compared with Ki67-LI of the corresponding FNA specimen. All patients were followed-up until June 2016.

Results : Cytological grade was consistent with histological grade in 39/57 cases hence a concordance rate of 72% when using a 5% cut-off between G1 and G2 tumors. Concerning Ki67-LI absolute values, correlation remained significant : $r=0.443$, $p=0.001$; and raised to $r=0.824$, $p<0.001$ when only cases with more than 2000 counted cells). Mean tumor size was significantly smaller when cytological and histological grading was consistent ($p=0.023$, 5% cut-off). Thirty-eight of 101 patients died during a median follow-up of 70.5 months. The median OS of the entire population is 235.30 months. OS is significantly different between tumor grade based on cytological Ki67-LI (log rank test, $p<0.001$) with a 5% cut-off (G1 : 235.30 months, G2 : 36.35 months and G3 : 10.95 months; HR vs. G1 : 3.78 and 12.55). The median PFS is significantly greater (log rank test, $p<0.001$) for patients with a G1 tumor than for those with a G2 (39.80 months) or a G3 (10.07 months) tumor (HR vs. G1 : 2.61 and 14.70).

Conclusions : The current results indicate that pNETs cytological grading is accurate when tumor size is < 2 cm and more tumor cells are counted on FNA. Discrepancies are seen among G2 tumors that are often considered G1 on FNA material due to tumor heterogeneity. Nevertheless EUS-FNA is a valuable tool to distinguish patients with a good (G1) or a poor (G3) prognosis, in terms of both OS and PFS. Our results show that EUS-FNA is helpful to clinicians by providing important prognostic information leading to adequate therapeutic decisions.

- 006 -

RETROSPECTIVE STUDY ON THE INCIDENCE OF CHEMOTHERAPY SIDE EFFECTS AND EVALUATION OF THE HURRIA SCORE AND G8 CGA AS PREDICTIVE TOOLS FOR TOXICITY IN OLDER PATIENTS WITH GASTROINTESTINAL CANCER. M. De Man (1), S. Laurent (1), K. Geboes (1). [1] UZ Gent, Gent, Belgium, Digestive Oncology and Gastroenterology.

Introduction : Cancer is a disease that mainly affects older people and age is considered as a risk factor for chemotherapy toxicity. G8 is a validated comprehensive geriatric assessment (CGA) screening and has a proven prognostic value for functional decline and survival. The Hurria score (H score) is a validated predictive risk stratification schema for chemotherapy toxicity in older patients.

Aim : The aim of the study is to evaluate the incidence of side effects of chemotherapy treatment in older patients with gastrointestinal tumours and to evaluate G8 and Hurria score as predictive tools.

Methods : This retrospective study based on prospectively collected data evaluates patient ≥ 70 years diagnosed with gastrointestinal cancer and who started a new treatment line with chemotherapy (adjuvant, first or further line) between May 2015 and November 2016. Drug dosing was at the discretion of the treating physician. Data on patient, tumour and treatment were collected at baseline, including age, Eastern Cooperative Oncology Group (ECOG) score, weight, length, haematocrit, renal function, activities of daily live (ADL), instrumental activities of daily live (IADL), tumour location and chemotherapy dose are collected at baseline. G8 and H score were performed. Chemotherapy related grade 3/4 side effects (CTCAE 4.03), dose modifications and unexpected hospital admission were recorded during the first treatment period defined as the time between start and first evaluation. Incidence of side effects, number of patients (pts) and events (ev), were compared in patients with high risk (Hh) versus low (Hl) or intermediate risk (Hi) according to H score and in patients with $G8 < 14$ versus patients with $G8 \geq 14$. Chi square test and Student t-test were used to compare respectively the number of patients and events in the risk groups.

Results : 64 pts (M42/F22) were included, median age was 76 (range 70-90) y. 22 pts (34%) had 31 (48%) grade 3/4 side effects, 25 pts (39%) 29 (45%) dose reductions and 25 pts (39%) 32 (50%) treatment delay. 9 pts (14%) needed unplanned hospital admission. Most frequent side effects were neutropenia : 15 pts (23%), 20 ev and gastrointestinal 9 pts (14%) 12 ev. Hurria score was significantly correlated with incidence of grade 3/4 side effects, both for pts Hl/i 14/52 versus Hh 8/12 pts ($p=0.01$) as for events Hl/i 18 versus Hh 13 ev ($p=0.03$). G8 score was not significantly correlated with grade 3/4 side effects for patients : $G8 < 14$ 12/37 versus $G8 \geq 14$ 10/27 pts ($p=0.70$) nor for events : $G8 < 14$: 16 versus $G8 \geq 14$ 15 ev ($p=0.45$). There was no significant correlation between neither Hurria nor G8 score and treatment delay, reduction or unplanned hospital admissions neither for number of patients nor events : delay Hl/I 19/52 versus Hh 6/12 pts ($p=0.39$), Hl/i 24 versus Hh 8 ev ($p=0.41$), $G8 < 14$ 14/37 versus $G8 \geq 14$ 10/27 pts ($p=0.95$), $G8 < 14$ 18 versus $G8 \geq 14$ 12 ev ($p=0.80$), reduction : Hl/I 20/52 versus Hh 5/12 pts ($p=0.84$), Hl/i 22 versus Hh 7 ev ($p=0.52$), G

8 <14 16/37 versus G 8 ≥ 14 9/27 pts (p=0.40), G 8 < 14 :20 versus G 8 ≥ 14 9 ev (p=0.16). Hospital admission : HI/I 7 versus Hh 2 (p=0.77), G8<14 :4 versus G8 ≥ 14 :5 (p=0.38).

Conclusions : During chemotherapeutic treatment of older patients with gastrointestinal cancer the incidence of side effects, dose adjustments and unexpected hospital admissions is high. The Hurria predictive score can define a group of patients with higher risk of grade 3/4 side effects. Performing this score can guide clinicians in treatment decisions in older patients and can be a basis for further research on optimizing treatment strategies in the elderly.

- O07 -

PRESSURIZED INTRAPERITONEAL AEROSOL CHEMOTHERAPY (PIPAC), A NEW SURGICAL TECHNIQUE FOR THE TREATMENT OF UNRESECTABLE PERITONEAL CARCINOMATOSIS. W. Willaert (1), S. Cosyns (1), L. Van De Sande (1), P. Pattyn (1), W. Ceelen (1). [1] Ghent University Hospital, Gent, Belgium, Gastro-Intestinal Surgery.

Introduction : Patients with unresectable peritoneal carcinomatosis treated with systemic chemotherapeutics have a bad prognosis. Pressurized intraperitoneal aerosol chemotherapy (PIPAC) can be a valuable adjunct. PIPAC is a minimally invasive and repeatable technique to deliver chemotherapeutic drugs into the peritoneal cavity.

Aim : We report about the practical organization and implementation of this technique, its indications and its impact on patients' early postoperative recovery and disease.

Methods : Every practical step until the first procedure was retrospectively reviewed together with the indications to perform this technique, the details of the procedures in each patient and the early postoperative recovery and survival.

Results : To perform PIPAC, a certificate is needed. Working with vaporized chemotherapeutics in the operation room is potentially dangerous. Therefore, an extensive checklist was made; two simulation procedures were performed; and several meetings with nurses, the department of security (for preventive actions and preparation in case of aerosol leakage in the operation room), pharmacists (for suited preparation and administration of chemotherapeutics) ; anesthesiologists (for patient monitoring outside the operation room and early postoperative follow-up) and a specialist of pressure injectors were necessary. No increased platinum concentrations were detected in the air during the first two PIPACs. Fifty-three PIPACs have been performed in 26 patients during 14 months (1 PIPAC (N=9); 2 PIPACs (N=8) ; 3 PIPACs (N=8); 4 PIPACs (N=1)). The primary disease was cancer of colorectal (N=7), gastric (N=6), ovarian (N=3), cholangio (N=1), esophageal (N=3), mesothelial (N=1), breast (N=1), small intestinal (N=1), LAMN (N=1), MANEC (N=1) and CUP (N=1) origin. Twenty-three patients had a history of systemic chemotherapy and six patients previously underwent cytoreductive surgery combined with intraperitoneal chemotherapy. The mean time between diagnosis of peritoneal carcinomatosis and the first PIPAC was 9.8 (mean) [1-56] months. All PIPAC procedures were uneventful, except one iatrogenic small intestinal perforation that was successfully sutured. Two cases were not performed due to portal hypertension-induced-bleeding and malignant adhesions. Postoperative recovery was uneventful except for one patient, who developed a toxic inflammation of the abdominal wall, which was successfully treated with antibiotics. The main reasons for not performing multiple PIPACs were disease progression (N=4) and because patients were too weak (N=5). PIPAC was usually performed in day clinic and was combined with systemic chemotherapy in 12 patients. The latter therapy can be reinstituted one week after PIPAC. Eleven patients died due to disease progression, of whom two underwent three PIPACs. Of those who underwent restaging after termination of two or three PIPACs, stable disease was observed in five patients and disease progression was found in six cases.

Conclusions : PIPAC is a new surgical procedure for the treatment of unresectable peritoneal carcinomatosis. This technique is safe for the surgical team under controlled circumstances. Its practical implementation requires extensive teamwork. The impact of this chemotherapeutic procedure on patients' postoperative recovery is limited. It is advisable to combine PIPAC with systemic chemotherapy and to start with PIPAC as soon as possible when peritoneal carcinomatosis has been diagnosed. In the near future, we will start with experimental and clinical studies on PIPAC.

- O12 -

PROTEOMIC DIFFERENTIAL DISTRIBUTION OF 53BP1 PROTEIN IN SERRATED AND CONVENTIONAL ADENOMAS VALIDATED BY HISTOLOGICAL CHARACTERISATION. F. Quesada Calvo (1), M. Merli (1), C. Massot (2), D. Baiwir (3), G. Mazzucchelli (4), N. Smargiasso (4), M. Gillet (5), N. Bletard (6), J. Somja (6), C. Coimbra Marques (7), M. Malaise (8), M. Polus (9), E. De Pauw (10), P. Delvenne (6), M. Meuwis (11), E. Louis (2). [1] University of Liege, Liège, Belgium, Translational Gastroenterology GIGA-R, [2] CHU de Liège, Liège, Belgium, Hepato-Gastroenterology and Digestive Oncology Department, CHU of Liège and GIGA-R, [3] University of Liege, Liège, Belgium, Proteomic facility, GIGA, [4] University of Liege, Liège, Belgium, Laboratory of Mass Spectrometry, Chemistry Dep., [5] University of Liege, Liège, Belgium, Histology and Cytology, [6] CHU Liege, Liège, Belgium, Pathological anatomy and cytology, [7] CHU de Liège, Liège, Belgium, Chirurgie Abdominale, Sénologique, Endocrine

et de Transplantation, [8] CHU de Liège, Liège, Belgium, Rheumatology, [9] CHU de Liège, Liège, Belgium, Hepato-Gastroenterology and Digestive Oncology Department, CHU of Liège, [10] University of Liege, Liège, Belgium, Laboratory of Mass Spectrometry, Chemistry, [11] CHU Liege, Liège, Belgium, Hepato-Gastroenterology and Digestive Oncology Department, CHU of Liège and GIGA-R.

Introduction : Sessile serrated adenoma/polyp (SSA/p) is a precancerous lesion, mostly located in the right side of the colon (cecum, ascending and transverse colon). The difficulty is to visualize this lesion during colonoscopy because of its subtle appearance.

Aim : Our aim was to generate data that might help SSA/p diagnosis and understanding the development of SSA/p.

Methods : We compared proteomes of serrated polyps (SSA/p) and conventional adenomas using residual human formalin fixed paraffin embedded (FFPE) samples. FFPE-FASP method was applied on samples before label free proteomic analysis. Immunohistochemistry (IHC) characterisation of one candidate marker was performed for tissue validation on an independent set of samples including : conventional adenomas (low and high-grade dysplasia), serrated polyps (hyperplastic polyps, SSA/p and traditional serrated adenoma) and finally normal colon (taken at the margin of colorectal cancer (CRC) or of diverticular disease).

Results : Proteomics provided 765 proteins (out of 5992 proteins identified) significantly discriminating conventional adenomas from serrated lesions. We selected 53BP1 (Tumor suppressor p53-binding protein 1) among these for IHC validation, because of its tumor suppressor gene function and role as a mediator of DNA damage checkpoint. 53BP1 appeared significantly up-regulated in proteomes of low and high grade adenomas compared to these of normal tissue and SSA/p. 53BP1 IHC signal was located in the nucleus and the percentage of positive nucleus decreased in serrated polyps, especially in crypts and in the border epithelium, confirming part of the proteomic results.

Conclusions : This study highlights potential marker proteins, including 53BP1 from which IHC signal was strongly decreased in some serrated polyps. The loss of 53BP1 has been associated with tumour progression and poor prognosis, while little is currently known about its involvement in precancerous CRC lesions. Early 53BP1 decrease of expression in the nucleus and therefore possible loss of function in some epithelial cells could reflect important changes occurring during dysplasia to neoplasia progression in serrated lesions.

- O13 -

CYTOREDUCTIVE SURGERY AND HYPERTHERMIC INTRAPERITONEAL CHEMOTHERAPY FOR PERITONEAL CARCINOMATOSIS OF COLORECTAL ORIGIN: RESULTS OF THE COMBINATION OF A CLOSED ABDOMEN TECHNIQUE COMBINED WITH OXALIPLATIN. S. Barbois (1), D. Leonard (1), E. Cotte (2), S. Van Den Broeck (1), C. Remue (1), R. Bachmann (1), N. Abbes Orabi (1), F. Lois (3), P. Forget (3), M. De Kock (3), P. Laterre (4), X. Wittebole (4), Y. Humblet (5), M. Van Den Eynde (6), E. Danse (7), A. Dragean (7), A. Jouret-Mourin (8), G. Leclercq (9), L. Delcarte (1), A. Souchet (1), A. Kartheuser (1). [1] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, , Belgium, Colorectal Surgery Unit, [2] CHU Lyon Sud, Lyon, France, Department of Digestive, Thoracic and Endocrine Surgery, [3] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Anesthesiology, [4] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Intensive Care Medicine, [5] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Medical Oncology, [6] UCL, Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Department of Medical Oncology, [7] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Radiology, [8] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Pathology, [9] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Pharmacy.

Introduction : Peritoneal carcinomatosis (PC) of colorectal origin has long been considered a terminal stage of the disease. Cytoreductive surgery (CS) combined with intraperitoneal hyperthermic chemotherapy (HIPEC) has gradually become treatment option of choice for the PC offering favorable oncological results.

Aim : The aim of this work was to analyse our results after Oxaliplatin HIPEC according to a closed abdomen technique.

Methods : From 2007 to 2015, consecutive patients undergoing CS and HIPEC to treat CP of colorectal origin were included. Demographic, surgical and pathological data was collected. Survival analysis was performed according to Kaplan Meyer.

Results : 64 patients underwent 71 CS with Oxaliplatin HIPEC according to a closed abdomen technique. There were 28 men and 36 women with a median age of 58 years (18-77). Eighteen patients (25.3%) had synchronous resectable liver metastases. The average level of carcinoembryonic antigen was 2,7UI/ml. All procedures were classified CCR-0. The median peritoneal carcinomatosis index (PCI) reached 6 (0-30). Fifteen cases (21.1%) had a Clavien-Dindo >2 complication. Postoperative mortality was 1.4%. Median follow-up was 26 months. Thirty-one patients (43.7%) received neoadjuvant chemotherapy, and 47 (66.2%) adjuvant chemotherapy. Eleven patients (17.2%) presented an isolated recurrence of CP and 32 patients (50%) relapsed as distant metastases. The overall (OS) and disease-free (DFS) 5-years survival were 44.5% and 19.1%, respectively. PCI < 7 lead to improved DFS (p=0.009).

Conclusions : HIPEC with Oxaliplatin using a closed abdomen technique for treating CP of colorectal origin offers acceptable oncological results. Recurrence rate remains significant, mainly due to distant metastasis. PCI < 7 significantly improves DFS.

- O14 -

GOOD COMPLIANCE TO FAST-TRACK PROGRAM IMPROVES OUTCOME AFTER COLORECTAL SURGERY.

A. Hartmann (1), C. Trefois (1), D. Leonard (1), C. Remue (1), R. Bachmann (1), N. Abbes Orabi (1), F. Lois (2), P. Forget (2), A. Kartheuser (1). [1] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Colorectal Surgery Unit, [2] Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Belgium, Department of Anesthesiology.

Introduction : Fast Track (FT) is a multimodal perioperative approach which aims to optimize physiologic response to the surgical stress and consequently improve postoperative outcome. Currently, our FT protocol consists of a total of 19 pre-, per- and postoperative measures including thorough pre-operative patient education, the absence of pre-operative fasting, the use of minimally invasive surgery, fluid restriction during the anesthesia, the use of non-morphine based analgesia, the absence of abdominal drains, of naso-gastric tube and of bladder catheter, early enteral nutrition and early mobilisation of the patient.

Aim : Our goal was to evaluate the compliance to protocol i.e. the number of FT items implemented for each patient and to analyse the effect of compliance to the FT protocol on postoperative outcome and post-operative hospital stay (POHS). We also aimed to identify isolated FT measures able to influence outcome.

Methods : This retrospective study involves a cohort of consecutive patients who underwent colorectal surgery within a FT protocol between 2007 and 2013. Beside basic demographics, adherence to protocol, postoperative complications and postoperative hospital stay (POHS) were recorded. Both univariate and multivariate analysis were performed to determine the predictive value of the FT protocol compliance and of specific FT items on surgical outcome and POHS.

Results : There were 157 men and 127 women with a mean age of 58 years. Compliance to the FT protocol reached a median of 18 out of 20 items. The median hospital stay was 3 days (2 – 49). Overall complications rate was 34.9% and 7.4% when Dindo-Clavien classification > 2 was considered. Risk factors of postoperative complications were male sex (p=0.038), laparotomy (p=0.015), neoplasia (p=0.0069) and number of comorbidities (p = 0.00877). Higher compliance to the FT protocol reduces the complication rate (p=0.00004), severity of complication (p=0.002) and POHS (p=<0.00001). We have not been able to identify any specific isolated FT measure able to influence post-operative outcome.

Conclusions : Greater adherence to FT protocol decreases postoperative complications and POHS. Our data supports a holistic effect of the FT protocol rather than specific isolated measures to improve patient's postoperative outcome.

- O15 -

CENTRAL VENOUS PRESSURE DROP AFTER HYPOVOLEMIC PHLEBOTOMY IS A STRONG INDEPENDENT PREDICTOR OF INTRAOPERATIVE BLOOD LOSS DURING LIVER RESECTION.

A. Ryckx (1), C. Christiaens (2), F. Vansteenkiste (1), P. Steelant (3), G. Sergeant (4), M. Clarysse (1), H. Pottel (5), I. Parmentier (6), M. D'hondt (1). [1] AZ Groeninge, Kortrijk, Belgium, Department of Digestive and Hepatobiliary/Pancreatic Surgery, [2] AZ Groeninge, Kortrijk, Belgium, Department of Nephrology, [3] AZ Groeninge, Kortrijk, Belgium, Department of Anesthesiology, [4] Virga Jesse Hospital, Hasselt, Belgium, Department of Abdominal and Hepatobiliary Surgery, [5] KU, Leuven, Belgium, Interdisciplinary research center, KULeuven campus Kortrijk, [6] AZ Groeninge, Kortrijk, Belgium, Department of Oncology and Statistics.

Introduction : Perioperative red blood cell transfusion (RBCT) is associated with earlier recurrence and higher cancer-related mortality after liver resection of hepatocellular carcinoma, intrahepatic cholangiocarcinoma and colorectal liver metastases. By reducing CVP, pressure in the central and hepatic veins and intravascular wall pressure in the sinusoids drops significantly, leading to lower estimated blood loss (EBL). Intraoperative hypovolemic phlebotomy (HP) has been suggested to reduce central venous pressure (CVP) prior to hepatectomy.

Aim : Our goal was to analyse the safety and feasibility of HP and the predictive value of CVP drop after HP on intraoperative blood loss and postoperative renal function.

Methods : A retrospective review of a prospective database of 100 consecutive patients (M/F : 43/57 , mean (range) age : 65 (23-89) years) undergoing liver resection with HP was performed. There were no exclusions based on preoperative ASA-score or hemoglobin/serum creatinin levels. Hypovolemic phlebotomy was performed before the start of parenchymal division with a volume of whole blood corresponding to approximately 0.7% of the patient's weight. After HP, there was no replacement of autologous blood by i.v. fluids to keep patients in a hypovolemic state. The collected

blood was reinfused after parenchymal transection at a rate of 50ml/min. Primary outcome variable was estimated blood loss (EBL), secondary outcome was postoperative serum creatinin (Scr). A multivariate linear regression analysis was performed to identify predictors of intraoperative blood loss. P-value <0.05 was considered statistically significant.

Results : In all patients the target weight of HP was achieved, there were no clotting or stasis issues during withdrawal, storage or reinfusion. Median (range) CVP prior to blood salvage was 8 (4 - 30) mm Hg. Median volume of hypovolemic phlebotomy was 400 (200 - 1000) ml. After HP, CVP decreased to a median of 3 (-2 - 16) mm Hg, resulting in a median CVP drop of 5.5 (2 - 14) mm Hg. Median estimated EBL during liver resection was 165 (0 - 800) ml. Median preoperative serum creatinin (Scr) was 0.82 g/dl (0.5-1.74) and postoperative Scr was 0.74 g/dl (0.44-1.68) . CVP drop was associated with EBL (P<0.001). There was no significant impact of CVP drop on postoperative Scr. There was a transfusion rate of 2%. A univariate analysis showed a significant correlation of gender, pre-operative hemoglobin, non-anatomical resection and use of a Pringle maneuver to increased blood loss. Type of surgery (laparoscopic, open or conversion) had no impact on EBL. In a multivariate analysis CVP drop and wedge/non-anatomical/metastasectomy were predictors of lower EBL. In contrast posterosuperior resection, use of straight bipolar device (Ligasure) and duration of surgery were significantly related to higher blood loss.

Conclusions : CVP drop after HP is a strong independent predictor of EBL during liver resection. The technique of HP is safe and feasible with strict adherence to anesthetic protocol and has no negative impact on renal function. We advocate the routine use of HP to reduce perioperative blood loss and transfusion rates in liver surgery. Measurement of CVP drop as a predictive tool can help surgeons to decide if a laparoscopic approach is safe for resection of lesions in proximity to the vena cava, hepatocaval confluence or hepatic veins.

- O16 -

RADIOEMBOLIZATION USING YTTRIUM 90 MICROSPHERES FOR UNRESECTABLE LIVER METASTASES : A SINGLE CENTRE EXPERIENCE. Y. Gokburun (1), F. Zech (2), R. Lhommel (3), Y. Humblet (4), I. Borbath (5), M. Demorcy (6), P. Goffette (7), M. Van Den Eynde (8). [1] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Hepatogastroenterology, [2] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Internal medicine, [3] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Nuclear medicine, [4] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Oncology, [5] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Hepatogastroenterology, [6] cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Institut Roi Albert II, [7] cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Interventional radiology, [8] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Digestive Oncology.

Introduction : Yttrium 90-loaded microspheres transarterial radioembolization is a selective internal radiotherapy (SIRT) directed against liver tumors. Many studies have demonstrated its efficacy for hepatocellular carcinoma (HCC), liver metastases of neuroendocrine tumors (NET) and colorectal (CRC) liver metastases.

Aim : Our purpose was to evaluate in our centre the efficacy and safety of SIRT for liver-dominant or exclusive tumors known to be less vascularized (patients with HCC and NET were here not analyzed).

Methods : Between 2011 and 2015, patients with liver dominant or exclusive metastases candidate for SIRT were prospectively registered in clinical database at the Cliniques universitaires St-Luc. All these patients underwent a pre-therapeutic workup (liver angiography + 99mTc-MAA scintigraphy) to prepare the hepatic vascularization and exclude treatment contraindication (99mTc-MAA extrahepatic uptake). SIRT was performed 1 to 2 weeks after the workup for eligible patients. Disease response was evaluated by FDG-PET-scan (6 and 12 weeks). Median overall (OS) and progression-free survival (PFS) were estimated using an accelerated life failure log-logistic model and compared by the Smirnov and weighted Gehan logrank tests. Other comparisons were made using the Wilcoxon-Mann-Whitney, Smirnov and Chi-squared tests.

Results : 76 patients (CRC=46, cholangiocellular=7, ocular melanoma=13 and others cancers types=10) were evaluated. 36 patients (47.4%) underwent SIRT and 40 patients (52.6%) were not treated (control group). Except for 99m Tc-MAA liver uptake, the 2 groups (SIRT vs Control) were not significantly different. mOS for SIRT patients (vs control) was significantly higher in the whole cohort (13,5 vs 6,8 months, p=0.0055) and CRC cohort (12,3 vs 5,5 months, p=0,007). Liver metastases burden >50%, FDG-PET progression after SIRT and progression in the treated liver were associated with a worse OS (p<0,05). No significant PFS difference was observed between groups. Disease control rate after SIRT was 51.6% and 32,3% at 6 and 12 weeks. None disease characteristics (including 99m Tc-MAA liver uptake) were significantly associated with tumor response. Serious adverse event occurs in 2,6% (n=2 : 1 SIRT, 1 control).

Conclusions : As compared to the control group, SIRT was safe and was associated with an OS benefit for patients with liver dominant or exclusive malignancies.

A PROSPECTIVE STUDY ON THE IMPACT OF A NET SPECIFIC MULTIDISCIPLINARY TUMOR BOARD ON INDIVIDUAL TREATMENT PLANS. W. Lybaert (1), T. Vandamme (2), M. Simoens (3), I. Dero (4), W. Demey (5), T. Rondou (6), M. Ulenaers (7), P. Abrams (8), C. Mattelaer (9), A. Driessen (10), B. Op De Beeck (11), O. D'archambeau (11), G. Roeyen (12), L. De Backer (2), M. Peeters (2) - On Behalf Of The Steering Committee And All Net-Specialists In "Netwerk" (13) / [1] AZ Nikolaas, Sint-Niklaas, Belgium, Oncology Department, [2] UZA, Edegem, Belgium, Oncology Department, [3] ZNA Antwerpen, , Belgium, Gastroenterology Department, [4] GZA, Wilrijk, Belgium, Gastroenterology Department, [5] AZ Klina, , Belgium, Oncology Department, [6] Sint-Jozef Kliniek Bornem, Bornem, Belgium, Gastroenterology Department, [7] AZ Heilige Familie Rumst, Rumst, Belgium, Gastroenterology Department, [8] GZA, Wilrijk, Belgium, Endocrinology Department, [9] ZNA Antwerpen, Belgium, Pathology Department, [10] UZA, Edegem, Belgium, Pathology Department, [11] UZA, Edegem, Belgium, Radiology Department, [12] UZA, Edegem, Belgium, Hepatobiliary Surgery Department, [13] NETwerk, Edegem, Belgium, Oncology Department.

Introduction : Multidisciplinary care is ideal in the management of patients with cancer and is associated with improvements in diagnosis, treatment planning, survival, patient satisfaction and clinician satisfaction. Traditionally, multidisciplinary tumor boards (MDTs) focus on treating common cancers. Whereas for rare cancers the need for thorough MDTs remains a current challenge to face because knowledge on rare cancers is often confined to certain practicing physicians or only centers where patients are seen in high volume. To cope with this, a multi-institutional health network (nine hospitals) was set up in the region of Antwerp-Waasland in Belgium for the multidisciplinary care of gastroenteropancreatic neuroendocrine tumor (GEP-NET) patients, called "NETwerk". In this scenario, local multidisciplinary teams refer all GEP-NET patients to be discussed among NET specialists in "NETwerk" experienced in diagnostics and treatment. This would introduce the end of local substandard treatment.

Aim : The objective of this interim analysis is to determine the efficacy of the NET specific MDTs within "NETwerk" in altering the treatment plan of NET patients.

Methods : For each GEP-NET patient from nine different hospitals, a NET specific MDT discussion takes place (bi-weekly). This requires input from different specialisms including, medical oncology, endocrinology, gastroenterology, surgery, pathology, nuclear medicine, diagnostic and interventional radiology teams, represented by NET specialists. The objective is to develop an integrated individualized NET management plan. A prospective study about NET specific MDTs concerning 146 consecutive NET patients of "NETwerk" was performed. As each NET patient was presented, record was made of the local treatment plan and compared with the treatment plan after discussion within "NETwerk". Treatment plans were categorized in : surgery, chemotherapy, radiotherapy, SIRT, PRRT, TACE, SSA and angiogenesis/mTOR inhibitor. In case additional examinations are required, this is indicated as "further diagnostics" and in case no further treatment is planned, results are recorded as "cured", "follow-up" and "palliative".

Results : The total number of NET cases enrolled in "NETwerk" and discussed at a NET MDT is 146. Among the 146 discussed GEP-NET cases, 58 were previously diagnosed, 70 were newly diagnosed and 18 GEP-NET patients were follow-up cases within "NETwerk". In 30% of the cases (n=44), the NET specific MDT proposed another treatment plan as compared to the local MDT decision. For 18 out of these 44 NET cases, the local MDT proposed a follow-up plan whereas the NET specific MDT required a change to : chemotherapy (n=1), surgery (n=3), SSA (n=2), further diagnostic (n=10) and cured (n=2). For 102 NET cases, both treatment plans were the same with the majority categorized as follow-up (n=34) and SSA (n=20).

Conclusions : "NETwerk" addresses the needs of both new and follow-up NET patients. A remarkable matter is that the further exploration of diagnostics and staging is often premised by the NET specialists while locally a clinical follow-up was proposed. Further diagnostics can include DOTANOC scan or other additional imaging, extra blood parameters, echo-endoscopy, endocrinological screening. This creates the opportunity to more sensitive and specific management of NETs on an individual base. The establishment of "NETwerk" is clearly associated with registration of medical data of a substantial population of NET patients. The collection of 146 records in 7 months creates perspectives for clinical trials.

Introduction OF EARLY INTEGRATION OF PALLIATIVE CARE IN DIGESTIVE ONCOLOGY : IS IT A CHALLENGE? G. Vanbutsele (1), K. Pardon (1), S. Van Belle (2), V. Cocquyt (2), M. De Laat (3), V. Surmont (4), K. Ecclou (1), K. Geboes (5), L. Deliens (1). [1] Vrije Universiteit Brussel & Ghent University, Brussels, Belgium, End-of-Life Care Research Group, [2] Ghent University Hospital, Ghent, Belgium, Department of Medical Oncology, [3] Ghent University Hospital, Ghent, Belgium, Palliative Care Team, [4] Ghent University Hospital, Ghent, Belgium, Department of Respiratory Medicine/Thoracic Oncology, [5] Ghent University Hospital, Ghent, Belgium, Department of Gastroenterology, Division of Digestive Oncology.

Introduction : Evidence suggests that early integration of palliative care (PC) in standard care improves patients' quality of life. In daily practice, referral to PC still occurs late in the disease trajectory for patients with digestive cancer. An im-

portant barrier to such early integration is the misconception of patients that PC is only provided at the end of life. Consequently, clinicians are hesitant about early integration of PC, they fear it will alarm patients and will decrease hope.

Aim : We wanted to know how often and for which reasons digestive oncologists did not introduce early integration of PC and examined why patients with advanced digestive cancer declined possible early integration of PC.

Methods : Systematic registration (1) of reasons of digestive oncologists for not introducing a randomized controlled trial of early integration of PC in the Ghent University Hospital to eligible patients with advanced cancer with a one year survival estimate and (2) of reasons of non-participation by patients.

Results : During the inclusion period (April 2013-March 2016), 142 patients with advanced digestive cancer were found to be eligible. The trial was introduced to 92 (65%) of patients with advanced digestive cancer and in total 57 (62%) patients were included. Thirty-two patients (35%) declined participation, most frequent reasons for non-participation were : not interested (n=18; 56%) and perceiving the term “palliative care” as threatening (n=9; 28%). The oncologists’ reasons for not introducing the trial to 38 (27%) patients were not clearly specified, for 12 patients (8%) they deemed that the patient couldn’t handle an early
Introduction of PC.

Conclusions : Digestive oncologists were able to introduce early integration of PC to two third of eligible patients and deemed that a minority of patients wouldn’t be able to handle such an early

Introduction. Thirty-two percent of patients refused participation, 28% because they specifically perceived the term ‘palliative care’ as threatening. This shows that PC is still not entirely understood by cancer patients with advanced digestive cancer. Efforts are needed to further clarify the scope of palliative care to this population.

BELGIAN PANCREATIC CLUB (BPC)

- P02 -

SINGLE VERSUS MULTIPLE PANCREATIC STENTS IN CHRONIC PANCREATITIS : A RETROSPECTIVE CLINICAL STUDY. C. Musala (1), M. Arvanitakis (2), J. Deviere (2), M. Delhay (2). [1] ULB Hôpital Erasme, Brussels, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology, [2] ULB Hôpital Erasme, Brussels, Belgium, Department of Gastroenterology, Hepatopancreatology and Digestive Oncology.

Introduction : Endoscopic ductal drainage by pancreatic stents (PS) in chronic pancreatitis (CP) has been shown to improve pain.

Aim : This study aims to compare the patients outcome after the placement of single pancreatic stent versus multiple stents.

Methods : Between 2004-2012, among 286 patients with CP treated by PS (single or multiple), 199 do not meet inclusion criteria (because of biliary stricture, pancreatic pseudocyst / surgery, loss of follow-up, diagnostic error). Stent(s) were removed after stricture resolution, if the patient was pain free (Izbicki pain score (IS) <10/100). Patients were followed-up until the last visit, surgery or death. Data were analyzed by intention-to-treat (ITT) and per-protocols analysis (PPA). The primary end points were to evaluate the rate of stent removal and the improvement in the IS at the end of follow-up (FU). Secondary end points included duration of stenting, number of procedures, need for restenting, and endocrine/ exocrine insufficiencies.

Results : 87 patients (63men; median age 51years) including both randomized (single vs multiple PS, n=24) and retrospectively analyzed patients (n=63), with mean baseline IS at 41/100, were included in this study. 51 patients received initially a single stent (group 1) and 36 patients \geq 2PS (group 2). Baseline characteristics were similar in both groups. During the study period, 37 patients changed of groups : 31(61%) passed from single to multiple stents and 6 (17%) from multiple to single stent. Migration was observed in 12 patients. Stent removal was possible in 72/87 patients (83%) without significant difference between the 2 groups in ITT (42/51, 82.4% vs 30/36, 83.3%, p=0.57), nor in PPA (19/26, 73% vs 53/61, 75%, p=0.25). The number of endoscopic procedures was 3 (1-11) and restenting was required after stent removal or migration in 28/84 patients (33%) without difference according to stenting group. Median FU after removal was 54 months (1-127) with no significant difference between both groups. Surgery was required in 10 patients. Mortality was recorded in 4 patients. A significant difference was observed between the 2 groups regarding the median stenting duration (16 months for group 1 vs 23 months for group 2, p=0.03). For all alive patients in whom stent(s) were removed/ migrated and had not been operated on (n= 68), we observed a significant improvement of IS : from 41(7-100)/ 100 to 10(0-82)/ 100 at the end of FU (p<0.001). Endo/exocrine insufficiencies, at the end of FU, were comparable in both groups (43%/ 38%).

Conclusions : Endoscopic therapy with stenting in CP patients is efficient regarding pain resolution, even after the stent(s) removal. A large proportion of patients changed from single to multiple stents because of pain. However, this study did not identify any benefit from multiple PS as compared to single PS.

- P03 -

MULTIDISCIPLINARY APPROACH DURING LONG-TERM FOLLOW-UP IS ESSENTIAL AFTER SURGERY FOR CHRONIC PANCREATITIS TO ACHIEVE BEST PATIENT OUTCOME. A. Craeynest (1), F. Ververken (1), A. Vanlander (2), F. Berrevoet (3). [1] Ghent University, Ghent, Belgium, Algemene en Hepatobiliaire Heelkunde, [2] Ghent University Hospital, Ghent, Belgium, Algemene en Hepatobiliaire Heelkunde, [3] University Hospital Ghent, Ghent, Belgium, Algemene & Hepatobiliaire Heelkunde.

Introduction : Adequate treatment for chronic pancreatitis includes a wide variety of options being conservative management, endoscopic and surgical treatment. In recent years the role of surgery for these patients increased with several publications showing better outcome than after endoscopic treatment in selected patients.

Aim : The objective of the study was to evaluate longterm patient outcome after surgical treatment of chronic pancreatitis

Methods : Retrospective assessment of the patients surgically treated for chronic pancreatitis from 2005 till 2013 was performed. The aim of this study was to evaluate the indication for surgery, the previous treatment modalities used and the long-term follow-up, considering complications, quality of life, pain coping and this in relation with the abstinence of alcohol and tobacco. Several questionnaires were used including the EORTC QLQ – PAN28, the PCCL, the McGill and EQ-5D.

Results : In total 45 patients were surgically treated with chronic pancreatitis as the main indication. Only 19 patients agreed on participation and a free outpatient visit. Seven patients died during the follow-up period. Six patients had a drainage procedure, while 13 had a classical or pyloruspreserving pancreaticoduodenectomy. Mean age was 50 years with a range in follow-up from 2 till 10 years. The overall outcome considering QoL was 69% with no difference between drainage and resection procedures. As soon as a complete alcohol abstinence was achieved patients had a signifi-

cantly better EQ-5D score than alcoholic patients. As only 2 patients stopped smoking completely, no correlation could be detected between QoL and tobacco use after surgery. Using the PCCL questionnaire for coping, surgery showed that the mean score for internal coping was 3.16 (out of 6-scale), but most patients still have some degree of pain.

Conclusions : Patient outcomes after surgical treatment for chronic pancreatitis are acceptable but longterm multidisciplinary guidance and follow-up are essential to improve quality of life and coping.

- P04 -

PANCREATIC EXOCRINE INSUFFICIENCY AFTER PANCREATICODUODENECTOMY IS MORE PREVALENT WITH PANCREATICOGASTROSTOMY THAN WITH PANCREATICO-JEJUNOSTOMY. A RETROSPECTIVE MULTICENTRE OBSERVATIONAL COHORT STUDY. G. Roeyen (1), M. Jansen (1), L. Ruysinck (2), T. Chapelle (1), A. Vanlander (2), B. Bracke (1), V. Hartman (1), D. Ysebaert (1), F. Berrevoet (2). [1] Universitair Ziekenhuis Antwerpen, Antwerpen, Belgium, Hepatobiliary, Endocrine and Transplantation Surgery, [2] Universitair Ziekenhuis Gent, Gent, Belgium, General, Hepatobiliary and Liver Transplantation Surgery.

Introduction : Recently, pancreaticogastrostomy (PG) has attracted renewed interest as a reconstruction technique after pancreaticoduodenectomy (PD), as it may imply a lower risk of clinical pancreatic fistula than reconstruction by pancreaticojejunostomy (PJ).

Aim : We hypothesise that PEI (Pancreatic Exocrine Insufficiency) is more common during clinical follow-up after PG than it is after PJ.

Methods : This study compares the prevalence of PEI in patients undergoing PD for malignancy with reconstruction by PG versus reconstruction by PJ. PEI during the first year of follow-up was defined as the intake of pancreatic enzyme replacement therapy (PERT) within one year postoperatively and/or an abnormal exocrine function test.

Results : A total of 186 patients, having undergone surgery at two university hospitals, were included in the study. PEI during the first year postoperatively was present in 75.0% of the patients with PG, compared to 45.7% with PJ ($p<0.001$). Intake of PERT within one year after surgery was found to be more prevalent in the PG group, i.e. 75.8% versus 38.5% ($p<0.001$). There was a trend towards more disturbed exocrine function tests after PG ($p=0.061$).

Conclusions : PEI is more common with PG reconstruction than with PJ reconstruction after pancreaticoduodenectomy for malignancy.

- P05 -

AUTOIMMUNE PANCREATITIS IN CHILDREN : WORKING GUIDELINES FOR DIAGNOSIS AND MANAGEMENT. I. Scheers (1), J. Palermo (2), S. Freedman (3), M. Wilschanski (4), U. Shah (5), M. Abu-El-Haija (2), B. Barth (6), D. Fishman (7), C. Garipey (8), M. Giefer (9), M. Heyman (10), R. Himes (7), S. Husain (11), T. Lin (2), Q. Liu (12), M. Lowe (11), M. Mascarenhas (13), V. Morinville (14), C. Ooi (15), E. Perito (10), D. Piccoli (13), J. Pohl (16), S. Schwarzenberg (17), D. Troendle (6), S. Werlin (18), B. Zimmerman (19), A. Uc (19), T. Gonska (20). [1] Cliniques Universitaires Saint-Luc, Belgium, Pediatric gastroenterology, hepatology and nutrition, [2] Cincinnati Children's Hospital Medical Center, Cincinnati/United States of America, United States (the), [3] Harvard Medical School, Beth Israel Deaconess Medical Center, Boston/United States of America, Boston, United States (the), [4] Hadassah Hebrew University Hospital, Jerusalem/Israel, Jerusalem, Israel, [5] Harvard Medical School, Massachusetts General Hospital for Children, Boston/United States of America, Boston, United States (the), [6] University of Texas Southwestern Medical School, Dallas/United States of America, Dallas, United States (the), [7] Baylor College of Medicine, Houston/United States of America, Houston, United States (the), [8] Nationwide Children's hospital, Columbus/United States of America, Columbus, United States (the), [9] Seattle Children's Hospital, Seattle/United States of America, Seattle, United States (the), [10] University of California at San Francisco, San Francisco/United States of America, San Francisco, United States (the), [11] Children's Hospital of Pittsburgh of UPMC, Pittsburgh/United States of America, Pittsburgh, United States (the), [12] Keck School of Medicine, University of Southern California, Children's Hospital Los Angeles, Los Angeles/United States of America, Los Angeles, United States (the), [13] The Children's Hospital of Philadelphia, Philadelphia/United States of America, Philadelphia, United States (the), [14] Montreal Children's Hospital, McGill University, Montreal/Canada, Montreal, Canada, [15] Discipline of Paediatrics, School of Women's and Children's Health, Medicine, University of New South Wales and Sydney Children's Hospital Randwick, Sydney/Australia, Sydney, Australia, [16] University of Utah, Salt Lake City/United States of America, Salt lake City, United States (the), [17] University of Minnesota Masonic Children's Hospital, Minneapolis/United States of America, Minneapolis, United States (the), [18] Medical College of Wisconsin, Milwaukee/United States of America, Milwaukee, United States (the), [19] University of Iowa Carver College of Medicine, Iowa City/United States of America, Iowa City, United States (the), [20] Hospital for Sick Children, Toronto/Canada, Toronto, United States.

Introduction : Autoimmune pancreatitis (AIP) is an increasingly recognized disease entity, but data in children are limited. Pediatric gastroenterologists relied on adult AIP guidelines but disease presentation and outcome of AIP in children might differ from the adult experience.

Aim : We aim to develop a working definition and diagnostic approach for AIP in children.

Methods : Clinical data, imaging, histology, and treatment modalities were collected using 2 different approaches : (1) a systematic literature search and (2) children with an AIP diagnosis from the largest multicenter study of chronic pancreatitis in children (INSPPIRE) and from Cliniques St-Luc (CUSL). We then sought expert opinion from pediatric pancreatologists.

Results : We identified 44 AIP cases, 26 from literature review, 14 from the INSPPIRE and 4 from CUSL cohort. The median age at diagnosis was 13.2 years. Abdominal pain (39/44, 87%) and/or obstructive jaundice (20/44, 45%) were the most reported symptoms at diagnosis. Elevated IgG4 levels was seen in only 8/38 (21%). Cross-sectional imaging was abnormal in all children mainly showing hypointense global or focal gland enlargement (35/43, 81%), irregularity of the main pancreatic duct (29/43, 67%) and common bile duct stricture (25/43, 58%). Lymphoplasmacytic inflammation, pancreas fibrosis and ductal granulocyte infiltration were the main histologic findings (18/25, 72%). Children with AIP had a prompt clinical response to steroids. Complications of AIP included impaired exocrine (4/25, 16%) and/or endocrine (3/27, 11%) function.

Conclusions : AIP in children is a distinct subtype of pancreatitis. Based on these observations, we established working guidelines to help identification and management of children with AIP and pave the way for future studies.

- P06 -

SUPPURATIVE CHOLANGITIS IN CHRONIC PANCREATITIS : REPORT OF A RARE STONE RELATED COMPLICATION. Y. Mourabit (1), M. Palazzo (2), M. Delhaye (1), M. Arvanitaki (1), O. Lheureux (3), J. Deviere (1), A. Lemmers (1). [1] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Brussel, Belgium, Gastroenterology, [2] Hôpital Beaujon, Clichy, France, Gastroenterology, [3] Erasme Hospital, Université Libre de Bruxelles, Brussels, Belgium, Brussel, Belgium, Intensive Care Unit.

We report the case of a 66-year-old woman with a history of chronic calcifying pancreatitis and former cholecystectomy, who presented with pancreatic severe abdominal pain, jaundice and pyrexia. Initial work up showed an inflammatory syndrome (CRP level 340 mg/L), hyperbilirubinemia (7mg/dl) and on abdominal CT-Scan a pancreatic calculus in the papilla with dilatation of the common bile duct, dilation of hepatic bile ducts and dilation of the main pancreatic duct. The patient rapidly went into septic shock and was admitted to the ICU under vasoactive drugs and broad spectrum antibiotics. An endoscopic retrograde cholangiopancreatography was performed. The latter showed a forced papilla with a white pancreatic stone impacted in the papilla. A large endoscopic biliary sphincterotomy was performed with release of pus. There was no common bile duct stone nor biliary stricture. Bile duct clearance was optimal after biliary sphincterotomy. A pancreatic stone was still visualized in the pancreatic sphincter after biliary sphincterotomy. Therefore, an endoscopic pancreatic sphincterotomy was performed followed by balloon extraction of the impacted stone and other main pancreatic duct fragments. Finally a naso-biliary catheter with flush and a pancreatic stent were left in place. Clinical evolution was excellent with resolution of shock, sepsis, pain and jaundice under antibiotics. The final diagnosis was a "pancreatic cholangitis", a biliary obstruction responsible of suppurative cholangitis due to a pancreatic stone impaction in the papilla in the setting of chronic calcifying pancreatitis. Biliary obstruction is not an unusual complication of chronic pancreatitis, mainly caused by inflammatory or fibrotic strictures of the distal bile duct, carcinoma of head of pancreas or less commonly by compression from pseudocysts. However obstruction due to pancreatic calculi causing ampullary obstruction and leading to obstructive jaundice is extremely rare, with only 14 cases described in the literature. Even scarcer are the five cases of cholangitis due to obstructive pancreatic calculi. As in our case, all reported cases were successfully managed with endoscopic treatment. Available iconography : abdominal CT 2 years before, abdominal CT at admission, endoscopic view of the papilla during ERCP, radioscopy view of ERCP.

- P07 -

HOW TO PLACE A SELF-EXPANDABLE METAL STENT (SEMS) INTO THE AFFERENT LIMB AFTER A ROUX-EN-Y WHIPPLE RESECTION USING THE SINGLE-BALLOON ENTEROSCOPE? A. Taha (1), I. Borbath (1), P. Deprez (1), T. Moreels (1). [1] Cliniques Universitaires Saint-Luc, Belgium, Hépatogastro-Entérologie.

Self-expanding metal stents (SEMS) can be used as a palliative treatment for malignant gastrointestinal obstruction. However, their through-the-scope insertion beyond Treitz' angulus remains challenging, because the SEMS device does not fit into the working channel of the single-balloon enteroscope. We present a method to overcome the mismatch between the SEMS deployment device and the single-balloon enteroscope working channel. A 76-year-old woman with a history of Roux-en-Y Whipple resection for pancreatic head cancer, was referred to our institution because of obstructive jaundice. She first underwent percutaneous transhepatic biliary drainage and cholangiography confirming the presence

of a suspect obstruction of the afferent limb 30 cm distally from the hepaticojejunostomy. Next, we performed a peroral single-balloon enteroscopy under fluoroscopic control to reach the malignant obstruction in the afferent limb. Endoscopic biopsies confirmed pancreatic adenocarcinoma recurrence. A 0.035 inch guidewire was inserted into the stricture. The extend of the tumor stenosis was defined using contrast dye injection under fluoroscopic control. The guidewire was left in place in the afferent limb beyond the malignant stricture. Next, the enteroscope was removed leaving the overtube in place with the balloon inflated in the afferent limb to maintain a stable position of the overtube. Then an uncovered self-expandable metal stent device (Wallflex 22x60mm, Boston Scientific) was inserted over the guidewire through the overtube until it passed through the stenosis and was deployed under fluoroscopic control. SEMS insertion resulted in relief of obstruction of the afferent limb. The patient was discharged two days later with normal oral diet tolerance, and the percutaneous biliary drain was removed with progressive improvement of liver function tests. Chemotherapy (gemcitabine) was initiated. Three months later, the patient presented with a recurrence of cholangitis suggestive of tumoral invasion of the uncovered metal stent in the afferent limb, which was confirmed during a second single-balloon enteroscopy. We used the same single-balloon overtube-assisted technique to insert a new partially covered SEMS (ComVi Enteral Colonic Stent 22x100 mm, Taewoong) inside the first stent. The procedure resulted in a quick relief of enteral in-stent obstruction, and the patient was discharged two days later and continued chemotherapy. She finally died 13 months after the first enteral stent insertion due to malignant disease progression. This case emphasizes the usefulness, safety and technical advantages of the single-balloon overtube-assisted technique for the placement of self-expandable metal stents in the small intestine in case of malignant stenosis, especially in cases of Roux-en-Y altered anatomy where conventional endoscopes cannot reach.

- P08 -

MULTIVISCERAL TRANSPLANTATION FOR PORTOMESENERIC THROMBOSIS AND UNRESECTABLE NEUROENDOCRINE TUMOR. E. Canovai (1), L. Ceulemans (2), N. Duchateau (2), G. De Hertogh (2), W. Laleman (3), M. Hiele (2), M. Sainz-Bariga (2), D. Monbaliu (2), I. Jochmans (2), T. Vanuytsel (2). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Abdominal Transplantation, [2] UZ Leuven, Leuven, Belgium, Leuven Intestinal Failure and Transplantation center, [3] UZ Leuven, Leuven, Belgium, Hepatology Department.

Introduction : Multivisceral transplantation (MVTx), defined as the en bloc transplantation of the abdominal viscera (stomach, duodenum, pancreas, liver and small bowel), is mostly performed for complicated diffuse and complete portomesenteric thrombosis. Experience is limited and the procedure still in its relative infancy, but the number of cases is likely to rise with the advent of new indications.

Case description : In August 2011, a 20-year-old male was referred to our department with diffuse and complete portomesenteric thrombosis. Imaging showed extensive hepatosplenomegaly with diffuse perfusion defects. Additional endoscopic ultrasound showed degenerative changes in the pancreas with no signs suggesting inflammation or malignancy. In December 2013, despite medical treatment, the patient's condition severely deteriorated with cholestasis, ascites requiring regular percutaneous drainage, variceal bleeding and intestinal failure. The only therapeutic option left was MVTx. In March 2014, MVTx was performed. First, the celiac trunk and the superior mesenteric artery were embolized to reduce perioperative bleeding, followed by en bloc resection of native stomach, duodenum, pancreas, liver and bowel, and finally en bloc transplantation of the corresponding organs. The patient recovered well, became nutritionally independent and was discharged at day 66 post-transplant. He became nutritionally independent 2 months post-transplant. Unexpectedly, the resection specimen showed an underlying grade III (Ki67 of 20.3%) pancreatic tail neuro-endocrine carcinoma with direct invasion of the spleen and diffuse liver metastasis with diffuse portomesenteric tumor thrombosis. The resection margins were negative. There was no nodal involvement and further imaging showed no residual tumor activity. Everolimus –by virtue of its anti-tumoral properties- was associated to classical tacrolimus and steroids-based immunosuppression. No adjuvant therapy was given. 2.5 years post-transplant, regular MRI scan and serum chromogranin levels have remained normal and the patient is leading a normal life.

Conclusion : This case confirms that MVTx is an effective treatment for portomesenteric thrombosis. In addition, it demonstrates that MVTx could be considered in selected cases of extensive and otherwise unresectable intra-abdominal tumors. In the latter, MVTx – complete splanchnic exenteration and replacement- allows the greatest chance for complete tumoral clearance. In addition, arterial embolization immediately pre-transplant could induce tumor necrosis and prevent perioperative vascular seeding.

- P09 -

SPLenic NEUROENDOCRINE TUMOR METASTASIS OR SIMPLE SPLenic CYST? D. Persyn (1), F. Van Der Cruyssen (2), K. Hertveldt (3), E. D'hondt (4), G. Deboever (5), M. Cool (5), G. Lambrecht (5). [1] UZ Leuven Gasthuisberg, Leuven, Belgium, Internal Medicine, [2] KU Leuven, Leuven, Belgium, Third Year Master Student Faculty of

Medicine, [3] AZ Damiaan, Oostende, Belgium, Pathology, [4] AZ Damiaan, Oostende, Belgium, Nuclear Medicine, [5] AZ Damiaan, Oostende, Belgium, Gastroenterology.

We present a case of a 54-year-old caucasian male with a growing cystic like lesion in the lower splenic pole. In October 2014 he underwent a middle segmental pancreatic resection for a non-functioning pancreatic neuroendocrine tumor. Lab tests showed an elevated plasma level of chromogranin A (198ng/mL, reference range < 40-170 U_g/L), normal CA 19.9 (< 0.6U/mL, reference range 0-37U/mL) and three separate measurements of 24 h urinary 5-Hydroxyindoleacetic Acid (5-HIAA) excretions were normal (respectively 4mg/24h; 3.2mg/24h and 2.8mg/24h, cutoff level < 9 mg/24 h). Pathology report showed the presence of a well-differentiated neuroendocrine tumor (pT2N0M0) of uncertain behavior. There was a low mitotic count (< 2 per 10 HPF) and a Ki-67 index of 2-5%. He received no adjuvant therapy. In February 2016, fifteen months after the initial diagnosis and surgery, a growing cystic like lesion in the lower splenic pole was identified on follow up CT-imaging. The lesion was 21mm in June 2015 and was now measuring 35mm diagonally, MRI confirmed a T2-hyperintense lesion without internal enhancement or evidence of other lesions. Plasma level of chromogranin A was stable : in 07-2015 147 ng/mL and in 02-2016 155 ng/mL. A somatostatin receptor scintigraphy with ¹¹¹In-pentetreotide showed profound radiotracer accumulation in the spleen. A multidisciplinary decision was made to perform a splenectomy. The final pathology report showed the presence of a benign mesothelial cyst. The differential diagnosis of a splenic cystic lesion is broad and includes : pseudocysts, true cysts (epithelial, transitional or mesothelial), inflammatory cysts (parasitical), benign neoplasm (lymphangioma) and malignant neoplasm (cystic metastasis). There are multiple potential causes for a false-positive interpretation of an octreotide scan. Accumulation of the tracer in the nasopharynx and pulmonary hilar areas may be seen with respiratory infections. The tracer can also accumulate in the lungs following radiation of bleomycin therapy. Recent surgical and colostomy sites are known to accumulate tracer. Tracer capitation can also be seen in normal organs (thyroid, liver, spleen, bowel ...) and in non-neoplastic disorders (autoimmune diseases, granulomatous diseases). Several studies have shown that positron emission tomography (PET) using gallium-68 (⁶⁸Ga-DOTATOC-PET/CT) appears to have a higher sensitivity and specificity in the diagnosis of small lesion. A recent study in 2015 confirmed that ⁶⁸Ga-DOTATOC-PET/CT was superior to ¹¹¹In-pentetreotide SPECT for the detection of NET metastases (especially localized in the skeleton and liver).

Conclusions : The differential diagnosis of a splenic cystic lesion is broad. Somatostatin receptor scintigraphy and positron emission tomography using gallium 68 are functional imaging techniques that are based on somatostatin receptor overexpression in neuroendocrine tumors. Recent studies are showing more evidence for the superiority of an ⁶⁸Ga-DOTATOC-PET/CT to the ¹¹¹In-pentetreotide SPECT for the detection of NET metastases. The uptake of radionuclide tracer can also produce a false-positive result. This has to be taken into account to avoid wrong decisions.

Reference : Van Binnebeek S, Vanbilloen B, Baete K, et al. Comparison of diagnostic accuracy of (¹¹¹In)-pentetreotide SPECT and (⁶⁸Ga)-DOTATOC PET/CT : A lesion-by-lesion analysis in patients with metastatic neuroendocrine tumours. *Eur Radiol.* 2016;26(3) :900-9.

RADIOLOGY, PATHOLOGY AND NUCLEAR MEDICINE

- R02 -

HETEROTOPIC GASTRIC MUCOSA IN THE GALLBLADDER : A CASE REPORT. D. STRYBOL (1), P. TERRY (2), G. DE HERTOGH (1) / [1] University Hospitals Leuven, Leuven, Belgium, Pathology, [2] University Hospitals Leuven, Leuven, Belgium, Radiology.

We report a case of an unexpected lesion found in the gallbladder of a 42-year-old woman. She was recently diagnosed with a breast carcinoma. Ultrasonography of the liver, performed as a staging investigation, showed a 2.3 cm sessile polypoid mass in the gallbladder. An additional CT scan confirmed this finding and showed no other lesions. Radiologically the lesion was suspected for gallbladder carcinoma. Transserosal growth could not be excluded. A laparoscopic cholecystectomy with local lymph node resection was performed. Macroscopically we found a soft, red, raised 3 cm large mass in the body of the gallbladder. Histological examination showed fundic-type gastric mucosa with several cystic spaces corresponding to dilated foveolae. A few foci of heterotopic pancreatic tissue were also present. There was no intestinal metaplasia nor dysplasia. The surrounding mucosa of the gallbladder showed extensive cholesterosis, but no cholecystitis or signs of cholelithiasis.

- R03 -

(PERI)ANAL BASAL CELL CARCINOMA : A RARE ENTITY AND DIAGNOSTIC CHALLENGE ON BIOPSY. E. Lecoutere (1), J. Van Dorpe (1), D. De Looze (2), M. De Man (3), P. Pattyn (4), T. Boterberg (5), A. Hoorens (1). [1] UZ Gent, Gent, Belgium, Pathology, [2] UZ Gent, Gent, Belgium, Gastroenterology, [3] UZ Gent, Gent, Belgium, Gastroenterology - Digestive Oncology, [4] UZ Gent, Gent, Belgium, Gastrointestinal Surgery, [5] UZ Gent, Gent, Belgium, Radiotherapy.

Introduction : A 73-year-old man was referred to our hospital for suspicion of recurrence of a carcinoma of the anal region. Two years earlier he was diagnosed with a (peri)anal squamous cell carcinoma.

Aim : A biopsy performed at that time reported a carcinoma compatible with either squamous cell carcinoma with focal basaloid differentiation or basaloid squamous cell carcinoma. In situ hybridisation for HPV low risk (6 and 11) genotypes and HPV high risk (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58 and 66) genotypes was negative.

Methods : The patient was treated with radiotherapy (50 Gy) in combination with 5-fluorouracil/mitomycin. His medical history included transurethral resection of the prostate for benign prostatic hyperplasia and colonic polyps (hyperplastic polyps as well as adenomatous polyps with low grade dysplasia).

Results : Two years after the radiochemotherapy the patient complained of anal pain. Biopsy of the recurrent lesion showed prominent retraction artifact and was suggestive of basal cell carcinoma. Staging gave no arguments for metastatic disease (rcT2N0M0). Radical abdominoperineal resection was performed and confirmed the diagnosis of basal cell carcinoma. The tumour was located at the anal margin with deep invasion into the anal canal. There was no in situ carcinoma or dysplasia of the overlying squamous epithelium. The tumour predominantly displayed features of nodular basal cell carcinoma with a focal adenoid pattern. There was invasion of the underlying connective tissue, up to the surrounding striated muscle layer. Resection margins were free of tumour, there was no lymphovascular or perineural invasion and no lymph node involvement (0/10). As it was a tumour of the anal margin with invasion of the anal canal, the tumour was staged rpT3N0 according to staging criteria for skin carcinomas.

Conclusions : Perianal basal cell carcinomas are extremely rare as these tumours usually occur in sun-exposed areas of the body. Possible causes include prior radiation therapy of the pelvic region, scarring, burning, chronic inflammatory skin diseases and systemic immunosuppressive therapy. This tumour can be difficult to distinguish from squamous cell carcinoma, especially on biopsy. Histological features such as presence of retraction artifact, HPV in situ hybridisation and immunohistochemistry including EpCAM, Bcl-2, p16 and SOX2, as well as a high rate of suspicion can help in making the differential diagnosis.

- R04 -

PSEUDO-SIGNET RING CELLS IN ENDOSCOPIC GASTRIC BIOPSIES : CLINICAL AND PATHOLOGICAL FEATURES OF 3 CASES. H. Dano (1), A. De Potter (2), S. Rogge (3), A. Hayani-Klalfaoui (4), K. Remery (5), K. Dhaene (6), A. Jouret-Mourin (1), L. Libbrecht (7). [1] Cliniques Universitaires Saint-Luc, Belgium, Pathology, [2] AZ Sint-Lucas Gent, Gent, Belgium, Pathology, [3] AZ Sint Lucas, Gent, Belgium, Gastroenterology, [4] Centre hospitalier régional de Mons, Belgium, Gastroenterology, [5] AZ Oudenaarde, Oudenaarde, Belgium, Gastroenterology, [6] ASZ,

Aalst, Belgium, Pathology, [7] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Pathology.

Introduction : A few case reports, mainly based on gastrectomy specimens, have described benign gastric pseudo-signet ring cells mimicking signet ring cell carcinoma.

Aim : Because of the potential diagnostic pitfall and the resulting clinical implications, we aimed to describe our experience with this entity.

Methods : Pathology databases were searched to identify possible cases of pseudo-signet ring cells and data was correlated with clinical findings and follow-up

Results : We identified 3 female patients with ages of 32, 37 and 59 years showing focal clusters of pseudo-signet ring cells in the upper part of the lamina propria in a background of chronic gastritis, which was HP-positive in 2 patients. Pseudo-signet ring cells were E-cadherin positive and rarely Ki-67 positive and reticulin staining showed that they were surrounded by a glandular basement membrane. At the time of biopsy, clinical suspicion of gastric cancer was present in 1 of 3 patients under the form of a suspect ulcer. Re-biopsy of the stomach was performed in 2 patients and these showed no atypical cells and further follow-up showed no evidence of gastric cancer.

Conclusions : When atypical cells suspicious for signet ring cell carcinoma are found in a gastric biopsy, the possibility of pseudo-signet ring cells has to be seriously considered when these cells are non-proliferative and present in a focal and superficial localization within glandular structures. This entity occurs in the background of gastritis and repeat biopsy appears sufficient to confirm the benign diagnosis.

- R06 -

EPSTEIN-BARR VIRUS-ASSOCIATED GASTRIC CARCINOMA UNDIFFERENTIATED NON-SMALL GASTRIC CANCER WITH PDL-1 EXPRESSION. A CASE REPORT. S. Sirimsi (1), A. Driessen (1), S. Declercq (2), M. Peeters (3), K. Traest (4), P. Pauwels (1). [1] University Hospital Antwerp, Antwerp, Belgium, Department of Pathology, [2] ZNA Middelheim, Antwerpen, Belgium, Department of Pathology, [3] University Hospital Antwerp, Antwerp, Belgium, Department of Oncology, [4] ZNA Jan Palfijn, Merksem, Belgium, Department of Pathology.

Epstein-Barr virus-associated gastric carcinoma (EBVaGC) is one of the four histological subtypes of gastric carcinoma. Epstein-Barr virus (EBV) is present within the malignant cells in 9% of all gastric adenocarcinomas and the presence of EBV is significantly more frequent in males and in cardiac tumors of the stomach. EBV genomes are preferably detected in gastric carcinomas using in situ hybridization (ISH) for EBV-encoded small RNAs (EBER). We report on a 53-year-old man diagnosed with undifferentiated non-small cell gastric carcinoma, detected on endoscopic gastric biopsies in 2016. The CT-scan showed lung metastasis. Epstein-Barr virus (EBV) was detected in nearly all tumor cells with in situ hybridization for EBV-encoded small RNAs (EBER). At least 70% of the tumor cells showed membranous expression for programmed death-ligand 1 (PD-L1). The correlation between EBV positivity and high expression levels of PDL1 has been documented recently. No surgical operation was performed, since the laparoscopic examination showed growth of the tumor through the gastric wall. The patient was treated with chemotherapy, however with only partial response. Due to the overexpression of PD-L1 immunotherapy has to be considered. Conclusion : Epstein-Barr virus-associated gastric carcinomas are now targets for immunotherapy, making EBV-testing important in cases of metastatic gastric cancer.

- R07 -

AN UNUSUAL POLYP AT THE ANORECTAL TRANSITION. G. Broeckx (1), F. Lockefeer (2), P. Potvin (3), H. Bontinck (4), A. Driessen (1). [1] University Hospital Antwerp, Antwerp, Belgium, Department of Pathology, [2] St Jozefkliniek, Bornem, Belgium, Department of Pathology, [3] St Jozefkliniek, Bornem, Belgium, Department of Gastroenterology, [4] St Jozefkliniek, Bornem, Belgium, Department of Surgery.

An 87-years-old male presented with rectal blood loss and soiling to the gastroenterology unit. Rectal examination revealed a large internal hemorrhoidal mass. Because of its size transanal resection of the mass was performed. Gross examination of the specimen showed a polypoid lesion (size 2 cm). Microscopically the tumour is composed of nests of moderately sized cells with some signet cell appearance. Numerous mitotic figures were observed in the tumour. Because of its resemblance to a goblet cell carcinoid immunohistochemistry was performed. The tumour expressed several neuroendocrine markers, such as synaptophysin and CD56 and weakly chromogranin. Besides these markers the nuclei of the tumour cells were also positive for CDX2. The proliferation marker Ki67 was very high, around 80%. Morphologically the tumour showed features of an adenocarcinoma with mucus-differentiation and neuro-endocrine differentiation. Therefore, a diagnosis of a mixed adeno-neuroendocrine tumour (MANEC) was made. The tumour was a pT2 tumour, invading the muscularis propria of the intestine. According to the WHO a mixed adeno-neuroendocrine tumour or MANEC is arbitrarily defined as a composite or collision tumour with areas of an adenocarcinoma or squamous cell

carcinoma intermingled with or separated from a neuroendocrine carcinoma/tumour, each composing at least 30 % of the neoplasm. Both components should be malignant. Our case is however a special type of MANEC, namely the amphicrine type, in which there is an expression of the neuro-endocrine markers in mucus-differentiated cells. In the same tumour cell we observed a dual expression of the neuro-endocrine markers and CDX2, a nuclear transcription factor for intestinal differentiation. The amphicrine type is an extremely rare type of MANEC, of which only case-reports have been published. It has been observed in different localisations such as stomach, pancreas and rarely in liver, gallbladder and small bowel. In the colon, most MANECs are composite tumours, characterized by a very poor prognosis. To our knowledge, the amphicrine type of MANEC has never been described at the anorectal region. The dual expression of neuroendocrine and intestinal markers, combined with the mucus differentiation in the same cell supports the hypothesis that this tumour originates from a pluripotent stem cell.

- R08 -

POSSIBLE LINK BETWEEN LONGSTANDING EOSINOPHILIC OESOPHAGITIS AND THE DEVELOPMENT OF OESOPHAGEAL CANCER. A. Taha (1), C. Mouradides (1), A. Mourin (2), M. Nollevaux (3), J. Martinet (4), A. Frère (5), J. Coche (6), P. Deprez (7). [1] Cliniques Universitaires St Luc, Brussels, Belgium, Gastroenterology Department, [2] Cliniques Universitaires St Luc, Brussels, Belgium, Pathology department, [3] CHU ULC Namur and Laboratory of Gastroenterology, Istitut de Recherche Expérimentale et Clinique, UCL, Brussels, Brussels, Belgium, Pathology department, [4] CHU ULC Namur and Laboratory of Gastroenterology, Istitut de Recherche Expérimentale et Clinique, UCL, Brussels, Brussels, Belgium, Gastroenterology Department, [5] CHR Citadelle, Liège, Belgium, Gastroenterology Department, [6] Clinique St-Pierre, Ottignies, Belgium, Gastroenterology Department, [7] Cliniques Universitaires Saint-Luc, , Belgium, Gastroenterology Department.

Eosinophilic oesophagitis is a clinico-pathological immune-mediated disease, with eosinophilic infiltration of >15 per high-power field, leading to a complex inflammatory cascade resulting in esophageal wall remodeling and fibrosis. The disease was first described in the 1990s, with an increasing prevalence in Western countries, affecting mostly children and adults below 50 years. Pathological characteristics are eosinophilic infiltration of the esophageal mucosa, increased basal cell hyperplasia, with subsequent subepithelial fibrosis and muscle hypertrophy. Limited but effective therapeutic strategies are available and long term maintenance treatment is required to avoid relapse. The natural history of the disease is not well understood, and there is no reported increased risk of malignancy. However, this case series presents 4 patients with malignant esophageal lesions in the setting of eosinophilic oesophagitis. This case series highlights the possible, but previously unknown, link between longstanding eosinophilic oesophagitis and the development of esophageal cancer. Further research is necessary to better understand the pathogenesis and long term history of eosinophilic oesophagitis and the relationship with malignancy.

Case 1 : A 63-year-old non-smoking lady was diagnosed with eosinophilic oesophagitis in 2008. She had a history of long-standing dysphagia and endoscopic features of ringed oesophagus. Histology showed evidence of eosinophilic infiltration of >15 eosinophils per high-power field (eos/hpf). She was treated with topical steroids with significant symptomatic improvement. During her endoscopic follow up, a villous lesion in the mid-oesophagus was found with histological evidence of high grade epidermoid cancer with persistent eosinophilic infiltration.

Case 2 : A 70-year-old man underwent gastroscopy for dysphagia in 2002, showing an oesophageal ulcer and oedema, with eosinophilic infiltration > 15 eos/hpf. He was started on PPI treatment. As a result of persistent symptoms, a new endoscopic evaluation was performed which revealed the presence of Barrett's esophagus, with no signs of dysplasia. In 2010 high grade dysplasia developing in previously described Barrett's esophagus was discovered. This was treated by radiofrequency ablation (RFA). In 2012, he developed high grade intraepithelial neoplasia in the mid-oesophagus which was subsequently treated with endoscopic submucosal dissection. In 2015, a second high grade intraepithelial neoplasia was diagnosed. **Case 3** An 84-year-old man was treated for well differentiated adenocarcinoma of the lower oesophagus in the setting of Barrett's esophagus by endoscopic submucosal resection in 2015. Endoscopic follow up showed residual high grade dysplasia that was treated with RFA. Two months later an oesophageal ulcer at the site of the previous RFA treatment showed eosinophilic oesophagitis (>15 eos/hpf) and areas of intraepithelial neoplasia. **Case 4** An 82-year-old man known to have eosinophilic oesophagitis needed multiple endoscopic dilatations because of a lower esophageal stenosis. Follow-up gastroscopy showed an epidermoid carcinoma developing in the mid-oesophagus, treated with endoscopic submucosal dissection. Endoscopic characteristics were oesophageal wall oedema, furrows and rings, with biopsies confirming the diagnosis of eosinophilic oesophagitis with of >15 eos/hpf, without residual neoplasia.

- R11 -

CD70-POSITIVE COLORECTAL CANCER ASSOCIATED FIBROBLASTS : PROGNOSTIC MARKER AND THERAPEUTIC TARGET. J. Jacobs (1), V. Deschoolmeester (2), K. Zwaenepoel (3), C. Hermans (2), C. Rolfo (4),

M. Peeters (5), F. Lardon (2), V. Siozopoulou (6), E. Smits (2), P. Pauwels (7). [1] University of Antwerp, Antwerp, Belgium, center for oncological research (CORE), [2] University of Antwerp, Antwerp, Belgium, Center for oncological research (CORE), [3] Antwerp University HOSPITAL, Edegem, Belgium, Pathology, [4] Antwerp University Hospital, Edegem, Belgium, Phase 1-Early Clinical Trials Unit, [5] Antwerp University Hospital, Edegem, Belgium, Oncology, [6] Antwerp University Hospital, Edegem, Belgium, Pathology, [7] University of Antwerp, Antwerp, Belgium, Pathology.

Introduction : Several studies have reported that tumor progression and invasiveness are determined not only by the malignant cancer cells themselves but also by the surrounding tumor microenvironment, including cancer-associated fibroblasts (CAFs). On the other hand, a total depletion of CAFs marks more aggressive tumor, indicating that different CAF subpopulations have opposing tumor-promoting or tumor-inhibitory roles. Unfortunately, markers to identify these different subsets of CAFs are lacking. It has been described that tumors hijack the immune checkpoint molecule CD70 to facilitate immune evasion by increasing the amount of suppressive regulatory T cells (Tregs). Nevertheless, the expression patterns of CD70 in colorectal cancer (CRC) have never been described before.

Aim : Due to the lack of specific markers to target CAFs with tumor-promoting properties and the limited clinical successes of immune checkpoint blockade in CRC, we have examined the expression pattern of CD70 in CRC with a particular focus on CAFs.

Methods : The expression of CD70 was analyzed by immunohistochemistry on 51 CRC specimens and linked with clinicopathological parameters. In addition, an association of CD70 with Tregs (CD4+FOXP3+) was explored. Finally, a primary CAF cell line was used to study the effect of CD70 on the tumor microenvironment in vitro.

Results : We revealed expression of CD70, not just on the malignant cells but on the majority of CAFs in invasive CRC specimens. Thereby, CD70-expression was significantly correlated with negative clinicopathological parameters including liver metastasis (P=0.007), differentiation (P=0.053) and advanced stage (P=0.001). In addition, CD70-positive CAFs proved to be an independent prognostic marker for inferior overall survival and progression-free survival. We have also detected a significant association between Treg infiltration and CD70-expressing CAFs (P=0.012). In vitro data on the effects of CD70 on CRC behavior are currently being analyzed.

Conclusions : We have identified a new targetable CAF subpopulation, marked by the expression of CD70 and equipped with strong tumor-promoting properties. Thereby, we have found evidence of a potential cross talk between CD70+ CAFs and Treg, paving the way towards immune escape of the tumor. We believe that targeting CD70 holds great potential in CRC, especially in light of the limited immunotherapeutic options available in colorectal cancer.

- R12 -

T-CELL INFILTRATION ASSESSED IN PRETHERAPEUTIC BIOPSIES OF PATIENTS WITH LOCALLY ADVANCED RECTAL ADENOCARCINOMA (LARC) IS ASSOCIATED WITH TUMOR RESPONSE AND RELAPSE AFTER CHEMORADIOTHERAPY (CRT) AND RECTAL SURGERY. M. Van Den Eynde (1), A. Kirilovsky (2), C. Elsissy (2), F. Marliot (2), H. Nacilla (2), C. Dragean (3), E. Danse (4), M. Denis (5), A. Kartheuser (6), C. Remue (7), D. Leonard (8), R. Bachman (7), P. Baldin (9), Y. Humblet (10), A. Jouret-Mourin (9), F. Pagès (2). [1] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Digestive Oncology, [2] Hôpital Européen Georges Pompidou, Paris, France, Immunology, [3] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Radiology, [4] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Radiology, [5] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Gastroenterology, [6] cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Digestive surgery, [7] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Digestive Surgery, [8] Cliniques Universitaires Saint Luc, Woluwe-Saint-Lambert, Belgium, Digestive Surgery, [9] Cliniques universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Pathology, [10] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Oncology.

Introduction : Pre-operative CRT followed by total mesorectal excision (TME) is nowadays the standard of care for patient with LARC (cT3-T4N0 or cTxN+). Currently, pathologic complete response occurs in +/- 15% after CRT. Colorectal cancer T-cell infiltration is a strong prognostic factor for survival after primary tumor resection.

Aim : Our aim was to determine whether T-cell infiltration in pretherapeutic tumor biopsy (PTB) could be predictive of tumor response and relapse after CRT + TME

Methods : Between 1999 and 2012, patients with LARC who underwent CRT + TME and with available clinical follow-up and PTB (with sufficient tumor cells density) were identified at the Cliniques universitaires St-Luc. The density of CD3 (T cells) and CD8 (cytotoxic) was quantified on immunostained PTB slides and analyzed with a dedicated image analysis software on whole-slide imaging. Comparisons were made using the Wilcoxon-Mann-Whitney test. Cumulative disease-free survival (DFS) was performed using the Kaplan-Meier estimator and compared by log-rank tests. Cox regression we used for uni- and multi-variate analysis. P value of less than 0.05 was considered statistically significant.

Results : 154 patients (sex ratio M/F 1.8; mean age 65 years-old; upper (20%), mid (29%) and low rectum (51%), synchronous metastases (11%)) were analyzed. High CD3 and CD8 PTB densities were significantly associated with a

higher pathological response (Dworak 3-4) and lower ypTNM stage after CRT +TME ($p < 0,05$). Higher CD3 and CD8 PTB densities were associated with higher patient DFS (CD3 : HR=3,36, $p=0,007$; CD8 : HR=2,62, $p=0,04$). These results were confirmed in uni and multivariate analysis. CD3 and CD8 PTB densities added to pathological response (ypTNM/Dworak) but also clinical response (cTNM) after CRT + TME increases significantly the accuracy prediction of tumor relapse.

Conclusions : Pretherapeutic T-cell infiltration of LARC is predictive of tumor response and relapse after CRT +TME. This biomarker could be helpful for patient treatment decision. It must be validated in larger patient cohorts.

- R13 -

CORRELATION OF TUMORAL NUCLEI PERCENTAGE WITH RAS MUTANT ALLELE FREQUENCY IN COLORECTAL ADENOCARCINOMA : A QUALITY ASSESSMENT TOOL OF NEXT GENERATION SEQUENCING ANALYSIS SHOWING LIMITED INTRA-TUMORAL HETEROGENEITY FOR RAS MUTATION STATUS. P. Baldin (1), A. Dekairelle (2), H. Dano (1), L. Libbrecht (3), A. Jouret-Mourin (1). [1] Cliniques Universitaires Saint-Luc, Belgium, Pathology, [2] Cliniques Universitaires Saint-Luc, Belgium, Génétique, [3] Cliniques Universitaires Saint-Luc, Woluwe-Saint-Lambert, Belgium, Pathology.

Introduction : Some studies have suggested that visual estimation for the assessment of percentage of tumoral nuclei in a sample in the setting of molecular analysis is not adequate (Smits et al. *Mod Pathol* 27 :168 and Viray et al. *Arch Pathol Lab Med* 137 :1545). However, Haley et al. (*Mod Pathol* 28 :1390) contradicted this by showing that tumoral nuclei percentage does correlate with mutant allele frequency, implying that visual estimation can be accurate and that colorectal adenocarcinomas show limited heterogeneity for RAS mutations.

Aim : To investigate this issue further using our own data.

Methods : NGS analysis performed on colorectal carcinoma at the molecular diagnostic platform of the Saint-Luc university clinics between January and October 2016 were evaluated for positivity rates of different RAS mutations. Tumoral nuclei percentage was assessed on a HE-stained slide in the region used for NGS by one pathologist (PB) and these data were correlated with the mutant allele frequency.

Results : Of 71 cases analysed, mutation of KRAS codon 12, KRAS codon 13, other KRAS mutations and NRAS mutations were seen in 30%, 7%, 9% and 4%, respectively, which is in accordance with the positivity rate on large patient sets in the literature (cfr. Boley et al. *BMC cancer* 16 :825), confirming the overall quality of our NGS analysis for RAS in colorectal carcinoma. As expected, mutations were mutually exclusive. Furthermore, there was a significant correlation between tumoral nuclei percentage visually assessed by a pathologist and the mutant allele frequency ($p=0.0037$ and correlation coefficient of 0.425, according to Pearson correlation test). This is a further confirmation of the good quality of our NGS analysis, including visual scoring by the pathologists. The significant correlation also implies that intra-tumoral heterogeneity for RAS mutations in colorectal adenocarcinoma is rather limited

Conclusions : Our results show that correlation of tumoral nuclei percentage with mutant allele frequency is a good additional quality assessment tool for NGS analysis of RAS in colorectal adenocarcinoma. Furthermore, we found evidence supporting the hypothesis that RAS mutation status shows little intra-tumoral heterogeneity.

- R14 -

RIGHT ILIAC FOSSA PAIN : ROLE OF IMAGING AT THE EMERGENCY DEPARTMENT. N. De Vos (1), F. Vanhoenacker (2). [1] Ghent University, Ghent, Belgium, Department of Radiology and nuclear medicine, [2] AZ Sint-Maarten, Mechelen, Belgium, Department of Radiology.

Introduction : Appendicitis is the most common abdominal condition requiring emergency surgery. The exact role of imaging in the work-up of patients presenting at the emergency department with right lower abdominal pain remains unsettled.

Aim : To determine which factors influence the use of abdominal ultrasound (US) and computed tomography (CT) in the work-up of these patients

Methods : Data from patients, presenting at the emergency department with right lower abdominal pain between July 2015 and July 2016, were retrospectively reviewed. For each patient, presentation during the night shift, Alvarado score, use of US, use of CT, and final diagnosis were determined. Pearson Chi-Square test was used to determine whether patient age under 18, presentation during night shift or Alvarado score influenced the use of medical imaging.

Results : 76 patients were selected (43 females, 33 males). Mean age was 36.7 years (range 5-88). 14 patients were minor (under 18 years old). Imaging (US and/or CT) was performed in 82.9% of patients. Imaging was less frequently performed if the Alvarado score was lower than 5 ($p < 0.05$). Ultrasound was more frequently performed in minor patients ($p < 0.05$) and CT was more frequently performed in patients over 18 years old ($p < 0.05$). Although imaging

was more frequently performed during the night shift, this difference was not statistically significant ($p = 0.083$).

Conclusions : Alvarado score and patient age have a statistically significant influence on the use of medical imaging in the work-up of patients presenting at the emergency department with right lower abdominal pain. Presentation during night shift also influences the use of medical imaging, although not statistically significant.

- R15 -

SAFETY AND EFFICACY OF TRANSARTERIAL RADIOEMBOLIZATION FOLLOWING CHEMOEMBOLIZATION WITH DRUG ELUTING BEADS FOR HEPATOCELLULAR CARCINOMA. E. Dresen (1), E. Klompenhouwer (1), C. Verslype (2), V. Vandecaveye (1), G. Maleux (1). [1] UZ Leuven, Leuven, Belgium, Department of Radiology, [2] UZ Leuven, Leuven, Belgium, Department of Hepatology.

Introduction : Transarterial chemoembolization (TACE) is the most widely used locoregional treatment for intermediate stage hepatocellular carcinoma (HCC). Transarterial radioembolization (TARE) is an emerging interventional treatment that could be complementary or an alternative to TACE.

Aim : To determine the safety and efficacy of TARE in patients who have previously undergone transarterial chemoembolization with drug-eluting beads (DEB-TACE) for HCC.

Methods : We retrospectively identified 30 patients who received one or more sessions of DEB-TACE prior to TARE for HCC in the period 2007-2016. There were 15 patients in Barcelona Clinical Liver Cancer Stage stage B (50%) and 15 (50%) in stage C. Adverse events grade > 3 were graded according to Common Terminology Criteria for Adverse events. Response on MRI was determined by mRECIST and ADCratio. Survival was determined since the first TACE and since the TARE procedure.

Results : Patients had a mean of 1.7 TACE procedures (range 1-4) prior to TARE. The indication to switch to TARE was progressive disease (63.3%) and stable disease (33.3%) despite DEB-TACE, or new portal vein thrombosis (3.3%). Grade 3-4 adverse events following TARE included : fatigue (20%), bilirubin increase (10%), cholecystitis (3.3%) and a gastric ulcer (3.3%). Radiologic response rates based on mRECIST and ADCratio were 43% and 64%, respectively. Radiological progression rate was 36% for both mRECIST and ADCratio. Three patients (10%) were downstaged within the Milan criteria and received a liver transplantation. Of the 19 patients who died during the follow-up period, the mean overall survival since first TACE was 24 months (range 4-45), the mean overall survival since TARE was 15 months (range 1-27).

Conclusions : TARE following DEB-TACE is a safe and efficient treatment strategy in patients with HCC, with the potential to downstage to liver transplantation.