EASL™
The Home of Hepatology

DIGITAL LIVER CANCER SUMMIT

ABSTRACT BOOK

5–6 FEBRUARY 2021

www.easl.eu/dlcs2021

Contents

ORAL POSTER ABSTRACT PRESENTATIONS

ORAL POSTER ABSTRACT PRESENTATIONS

O01 Genetic and epigenetic analyses reveal early and distinct alterations in intraductal papillary and tubulopapillary cholangiocarcinogenesis

Benjamin Goeppert[1], Damian Stichel[2], Reka Toth[3], Sarah Fritzsche[1], Moritz Loeffler[1], Anna Melissa Schlitter[4], Olaf Neumann[1], Yassen Assenov[3], Monika Vogel[5], Arianeb Mehrabi[6], Katrin Hoffmann[6], Bruno Köhler[7], Christoph Springfeld[7], Dieter Weichenhan[3], Christoph Plass[3], Irene Esposito[4], Peter Schirmacher[1], Andreas von Deimling[2], Stephanie Roessler[1]

[1]Institute of Pathology, University Hospital Heidelberg, Heidelberg, Germany, *[2]German Cancer Research Center (DKFZ), Clinical Cooperation Unit Neuropathology*, *[3]German Cancer Research Center (DKFZ), Cancer Epigenomics*, *[4]Institute of Pathology, Technical University of Munich*, *[5]Thoraxklinik at University Hospital Heidelberg, Diagnostic and Interventional Radiology*, *[6]Department of General, Visceral and Transplantation Surgery, University Hospital Heidelberg*, *[7]National Center for Tumor Diseases, Department of Medical Oncology*
Email: stephanie.roessler@med.uni-heidelberg.de

Background and Aims:
A detailed understanding of the molecular alterations in different forms of cholangiocarcinogenesis is crucial for a better understanding of cholangiocarcinoma (CCA) and may pave the way to early diagnosis and better treatment options. Two main subtypes of biliary precursor lesions which may lead to invasive CCA are nowadays recognized and described in detail in the current WHO classification of Digestive System Tumours: the microscopic biliary intraepithelial neoplasm (BilIN) and the macroscopically visible intraductal papillary or tubulopapillary neoplasms of the bile duct, IPNB and ITPN, respectively. In this study, we focused on the molecular alterations in IPNB and ITPN to better understand early tumorigenic events and differences between IPNB and ITPN.

Method:
We analyzed a clinicopathologically well-characterized patient cohort (N=54) with high-grade intraductal papillary (IPNB) or tubulopapillary (ITPN) neoplastic precursor lesions of the biliary tract and correlated the results with an independent non-IPNB/ITPN associated CCA cohort (N=294). The triplet sample set of non-neoplastic biliary epithelium, precursor, and invasive CCA was analyzed by next generation sequencing, DNA copy number and genome-wide methylation profiling.

Results:
According to morphological and immunohistochemical analysis of MUC1, MUC2, MUC5AC and CDX2 the cohort consisted of 44 (81.5%) IPNB and 10 (18.5%) ITPN. Anatomically, ITPN were mostly associated with intrahepatic CCA, whereas IPNB co-occurred mostly with distal CCA. IPNB/ITPN were equally associated with small- and large-duct type intrahepatic CCA. Patients with invasive CCA arising from IPNB/ITPN had better prognosis than CCA patients without IPNB/ITPN.
At the molecular level, the most frequently mutated genes in IPNB were TP53, KRAS, SMAD4 and CDKN2A; which are also typically found in extrahepatic CCA, while ITPN had significantly fewer mutations. Most mutations were shared between precursor lesions and corresponding invasive CCA but ROBO2 mutations occurred exclusively in invasive CCA and CTNNB1 mutations were mainly present in precursor lesions. In addition, IPNB and ITPN differed in their DNA methylation profiles and analyses of latent methylation components suggested that IPNB and ITPN may have different cells-of-origin.

Conclusion:
We found that IPNB and ITPN harbor distinct early genetic alterations, IPNB are enriched in mutations typical for extrahepatic CCA, whereas ITPN exhibited few genetic alterations and showed distinct epigenetic profiles, and IPNB/ITPN may represent a distinctive, intermediate form of intra- and extrahepatic cholangiocarcinogenesis.

O02YI CRISPR screening in intrahepatic cholangiocarcinoma

Mollie Wilson[1], Nicholas Younger[2], Luke Boulter[1 3]
[1]MRC Human Genetics Unit., Edinburgh, United Kingdom, *[2]Centre for Inflammation Research, Edinburgh, United Kingdom*, *[3]MRC Human Genetics Unit, Edinburgh, United Kingdom*
Email: luke.boulter@ed.ac.uk

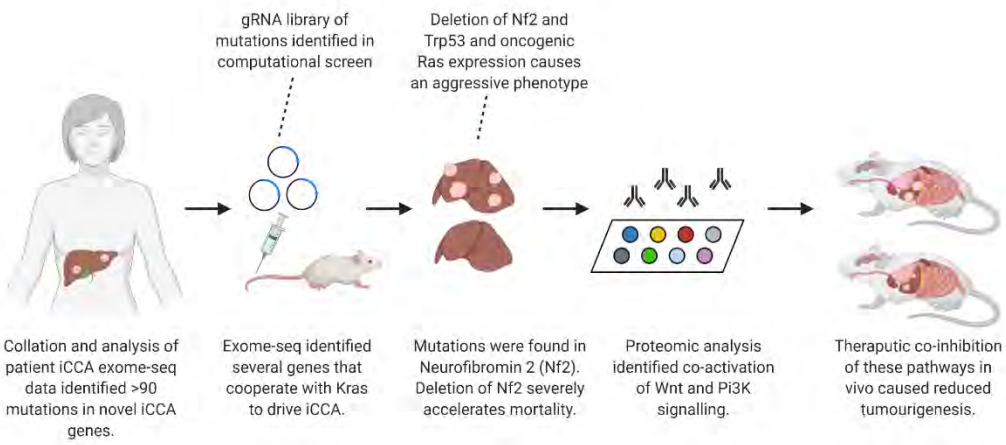
Background and Aims:
Intrahepatic cholangiocarcinoma (iCCA), a malignancy arising from the bile ducts, presents with highly genetically heterogeneous tumours. The lack of understanding surrounding the genetics of this disease has hampered therapeutic approaches. To address the gap in our understanding of the functional genetics of iCCA we developed a computational screen of patient exome sequencing data to comprehensively identify driver mutations in ICC, and found that in addition to known drivers of ICC, a large number of infrequently mutated genes typifies iCCAs. Here, we aimed to address whether these infrequently mutated genes are capable of initiating iCCA formation and determine whether rare iCCA mutations are sufficient to enhance, or modify, iCCA growth.

Method:
A CRISPR-Cas9 library of loss of function mutations in iCCA was generated using data from patients, which was hydrodynamically injected into immune competent mice to delete iCCA-genes, along with expression of oncogenic $Kras^{G12D}$. Whole exome and RNA sequencing as well as proteomic approaches were combined to analyse the resulting tumours, to identify mutations that synergise with oncogenic Kras, define the transcriptional signatures that result from these mutations and uncover novel candidate therapeutic pathways.

Results:
Computational screening of patient exomes identified a list of mutations in iCCA driver genes. Using this approach, we re-identified, canonical mutations in iCCA and >90 mutations in novel genes. To determine which mutations are functional *in vivo*, we performed a CRISPR screen targeting genes found in patient iCCA. Tumours formed that histologically represented human iCCA when the CRISPR-library was co-expressed with $Kras^{G12D}$. Whole exome sequencing of these tumours identified a number of novel CRISPR-induced mutations that interact with $KRas^{G12D}$ to drive iCCA formation and we identified mutations in neurofibromin 2 (Nf2), a cytoskeletal protein whose encoded protein Merlin is at the apex of many important signaling pathways as a gene of interest. Deletion of Nf2 cooperates with $Kras^{G12D}$ to initiate iCCA formation; moreover, Nf2-loss cooperates with Trp53-loss leading to cancer with a significantly accelerated phenotype and increased lethality. Proteomic analysis of tumour tissue from this aggressive model demonstrated that co-activation of Wnt and Pi3K signaling typify these tumours and can be targeted therapeutically to reduce tumour progression.

Conclusion:
Our study demonstrates that Nf2 is a rare driver gene of iCCA that acts in a cooperative manner with oncogenic $Kras^{G12D}$ to accelerate tumourigenesis. Using a combination of *in silico* and *in vivo* modelling holds a great deal of promise in unveiling the contribution of different mutations to iCCA progression and also providing a platform to identify novel therapeutic vulnerabilities.

Figure:


gRNA library of mutations identified in computational screen
Deletion of Nf2 and Trp53 and oncogenic Ras expression causes an aggressive phenotype
Collation and analysis of patient iCCA exome-seq data identified >90 mutations in novel iCCA genes.
Exome-seq identified several genes that cooperate with Kras to drive iCCA.
Mutations were found in Neurofibromin 2 (Nf2). Deletion of Nf2 severely accelerates mortality.
Proteomic analysis identified co-activation of Wnt and Pi3K signalling.
Theraputic co-inhibition of these pathways in vivo caused reduced tumourigenesis.

O03YI Is progression-free survival a robust surrogate endpoint of overall survival in immunotherapy trials of hepatocellular carcinoma?

Giuseppe Cabibbo[1], Ciro Celsa[1], Marco Enea[2], Salvatore Battaglia[3], Giacomo Emanuele Maria Rizzo[1], Anita Busacca[1], Domenica Matranga[2], Massimo Attanasio[3], María Reig[4], Antonio Craxi[1], Calogero Camma[1]

[1]University of Palermo, Section of Gastroenterology and Hepatology, PROMISE Department, Palermo, Italy, *[2]University of Palermo, PROMISE Department, Palermo, Italy*, *[3]University of Palermo, Department of Economics, Business, and Statistics, , Palermo, Italy*, *[4]Barcelona Clinic Liver Cancer, Barcelona, Spain*
Email: calogero.camma@unipa.it

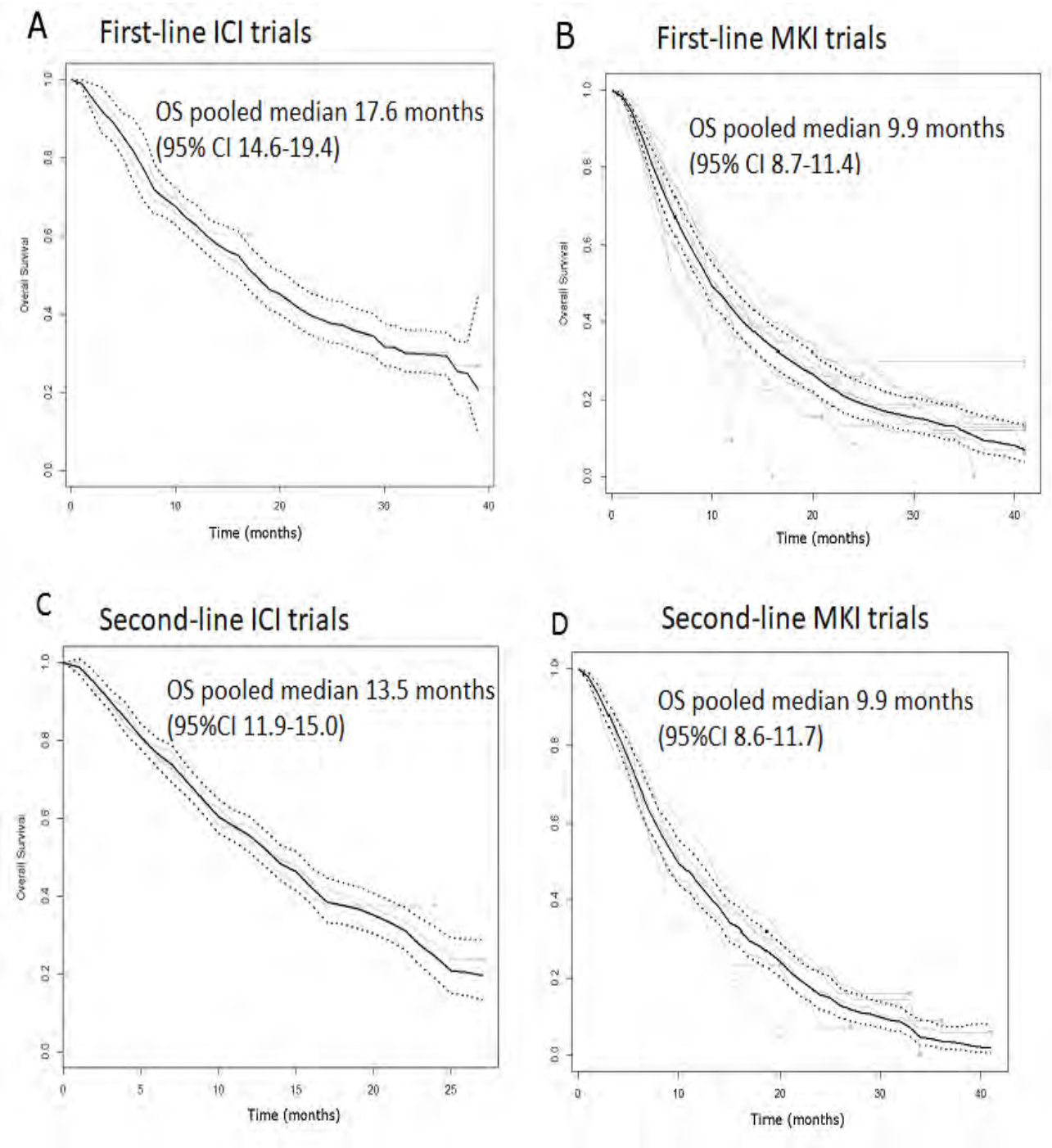
Background and Aims: Radiology-based outcomes, such as progression-free survival (PFS), and objective response rate (ORR), are used as surrogate endpoints in oncology trials. We aimed to assess the surrogacy relationship of PFS with overall survival (OS) in clinical trials of systemic therapies targeting advanced hepatocellular carcinoma (HCC) by novel meta-regression methods.

Method: A search of databases (PubMed, ASCO, ESMO Meeting Libraries, Clinicaltrials.gov) for trials of systemic therapies for advanced HCC reporting both OS and PFS was performed. Individual patient data were extracted from PFS and OS Kaplan-Meier curves. Summary median PFS and OS data were obtained from random-effect model. The surrogate relationships of median PFS, first quartile (Q1), third quartile (Q3), and restricted mean survival time (RMST) for OS were evaluated by the coefficient of determination R^2. Heterogeneity was explored by meta-regression

Results: We identified 49 trials, 11 assessing immune checkpoint inhibitors (ICIs) and 38 multikinase inhibitors (MKIs). In first-line, OS pooled median was 17.6 months (95% Confidence Intervals [CI] 14.6-19.4) for ICIs (Panel A) and 9.9 months (95%CI 8.7-11.4) for MKIs (Panel B); PFS pooled median was 5.5 months (95%CI 2.8-7.7) for ICIs and 4.3 months (95%CI 3.4-5.3) for MKIs. In second-line, OS pooled median was 13.5 months (95%CI 11.9-15.0) for ICIs (Panel C) and 9.9 months (95% CI 8.6-11.7) for MKIs (Panel D); PFS pooled median was 3.5 months (95% CI 2.4-5.2) for ICIs and 3.4 months (95% CI 2.7-4.3) for MKIs. Overall, the correlation between median PFS and median OS was weak (R^2= 0.20. 95% CI -0.02;0.42). Surrogacy robustness varied between treatment classes and PFS endpoints. In ICI-trials only, the correlations between Q1-PFS and Q1-OS and between 12-month PFS-RMST and 12-month OS-RMST were high (R^2=0.89, 95%CI 0.78-0.98, and 0.80, 95%CI 0.63-0.96, respectively). Interaction p-values obtained by meta-regression confirmed the robustness of results.

Conclusion: In trials of systemic therapies for advanced HCC, the surrogate relationship of PFS with OS is highly variable depending on treatment class (ICI or MKI) and evaluation time-point. In ICI-trials, Q1-PFS and 12-month PFS-RMST are robust surrogate endpoints for OS.

Figure: Pooled reconstructed survival curves for overall survival (OS) from clinical trials assessing immune checkpoint inhibitors (ICIs) (First-line: panel A. Second-line: panel C) and multikinase inhibitors (MKI) (First-line: panel B. Second-line: panel D) in advanced hepatocellular carcinoma.



O04 A phase II study of stereotactic body radiotherapy （SBRT） combined with sintilimab in patients with oligometastases of hepatocellular carcinoma (HCC）

Yixing Chen[1], Ping Yang[1], Shi-Suo Du[1], Jian Zhou[2], Cheng Huang[2], Wen-Chao Zhu[1], Yong Hu[1], Yi-Yi Yu[3], Tian-Shu Liu[3], Zhao-Chong Zeng[1]

[1]Zhongshan Hospital, Fudan University, Department of radiation oncology, Shanghai, China, *[2]Zhongshan Hospital, Fudan University, Department of liver surgery , Shanghai, China*, *[3]Zhongshan Hospital, Fudan University, Department of medical oncology, Shanghai, China*

Email:

Background and Aims: SBRT is an emerging treatment option for oligometastatic cancer disease. Preclinical studies have shown the synergistic effects of radiotherapy in combination with immunotherapy. Early clinical data suggest that immunotherapy might augment the local effects of radiotherapy and decrease metastatic recurrence. This study aimed to evaluate the efficacy and safety of SBRT combined with sintilimab (a PD-1 inhibitor) for oligometastases of HCC.

Method: In this single arm, phase II study (NCT03857815), eligibility criteria included oligometastases of HCC (defined as ≤5 metastatic/recurrent lesions), Child-Pugh class A, ECOG PS ≤ 1. Pts received sintilimab 200 mg IV Q3W for up to 12 months or until progressive disease, unacceptable toxicity, or withdrawal. SBRT to all lesions was started at cycle 1 day 1. Primary endpoint was progression free survival (PFS) per RECIST 1.1. Secondary endpoints included toxicity, objective response rate (ORR), disease control rate (DCR) and overall survival (OS).

Results: At data cutoff (Nov 24th, 2020), 18 pts were enrolled with median follow up of 6.8 months (range 0.3-19.0). Median age was 64.5 years (range 37-77), 17 (94.4%) were male, 18 (100%) were HBV+, 4 (22.2%) had extrahepatic metastasis, most pts had prior ≥2 local therapies. SBRT was delivered to a median dose of 54 Gy (range 48-60) in 6 fractions (range 6-10). 15 pts were evaluable for efficacy. ORR was 100% (15/15, 10 CR and 5 PR). Median PFS and OS were not reached. 6-mo PFS rate was 100%. 2 pts has completed treatment of 12 months and 7 pts remain on treatment. Treatment-related adverse events (TRAEs) occurred in 8 (44.4%) of 18 pts. Most common TRAEs were platelet count decreased (16.7%), myositis (16.7%) and ALT/AST increased (11.1%). Grade 3 TRAEs (myositis) occurred in 1 (5.6%) pt. There were no grade 4-5 TRAEs. TRAEs led to treatment discontinuation in 2 (11.1%) pts.

Conclusion: These preliminary data suggest the combination of SBRT and sintilimab has a high response rate and acceptable safety in treatment of oligometastases of HCC. Additional pts and longer follow up are required to further evaluate the efficacy and safety.

Figure: NA

O05 IMbrave150: updated overall survival (OS) data from a global, randomized, open-label Phase III study of atezolizumab (atezo) + bevacizumab (bev) vs sorafenib (sor) in patients (pts) with unresectable hepatocellular carcinoma (HCC)

Richard Finn[1], Shukui Qin[2], Masafumi Ikeda[3], Peter Galle[4], Michel Ducreux[5], Tae-You Kim[6], Ho Yeong Lim[7], Masatoshi Kudo[8], Valeriy Breder[9], Philippe Merle[10], Ahmed Kaseb[11], Daneng Li[12], Wendy Verret[13], Hui Shao[14], Juan Liu[14], Lindong Li[14], Andrew Zhu[15], Ann-Lii Cheng[16]

[1]Jonsson Comprehensive Cancer Center, Geffen School of Medicine at UCLA, Los Angeles, United States, *[2]People's Liberation Army Cancer Center, Nanjing, China*, *[3]National Cancer Center Hospital East, Kashiwa, Japan*, *[4]University Medical Center Mainz, Mainz, Germany*, *[5]Gustave Roussy Cancer Center, Villejuif, France*, *[6]Seoul National University College of Medicine, Seoul, Korea, Rep. of South*, *[7]Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea, Rep. of South*, *[8]Kindai University Faculty of Medicine, Osaka, Japan*, *[9]N.N. Blokhin Russian Cancer Research Center, Moscow, Russian Federation*, *[10]University Hospital La Croix-Rousse, Lyon, France*, *[11]The University of Texas MD Anderson Cancer Center, Houston, United States*, *[12]City of Hope Comprehensive Cancer Center and Beckman Research Institute, Duarte, United States*, *[13]Genentech, Inc., South San Francisco, United States*, *[14]Roche Product Development, Shanghai, China*, *[15]Harvard Medical School, Massachusetts General Hospital Cancer Center, Boston, United States*, *[16]National Taiwan University Cancer Center and National Taiwan University Hospital, Taipei, Taiwan*

Email: rfinn@mednet.ucla.edu

O06YI Identifying effective subtype-specific treatment responses in hepatocellular carcinoma in genetically engineered mouse models

Miryam Müller[1], Stephanie May[1], Lynn Mcgarry[2], Tim Kendall[3], William Clark[1], Ann Hedley[1], Thomas Jamieson[1], Colin Nixon[1], Karen Blyth[1 2], Daniel J Murphy[1 2], Crispin J Miller[1 2], Leo M Carlin[1 2], Owen J Sansom[1 2], Thomas G Bird[1 2 3]

[1]CRUK Beatson Institute, Glasgow, United Kingdom, *[2]University Of Glasgow, Institute of Cancer Sciences, Glasgow, United Kingdom*, *[3]University of Edinburgh, United Kingdom*
Email: m.mueller@beatson.gla.ac.uk

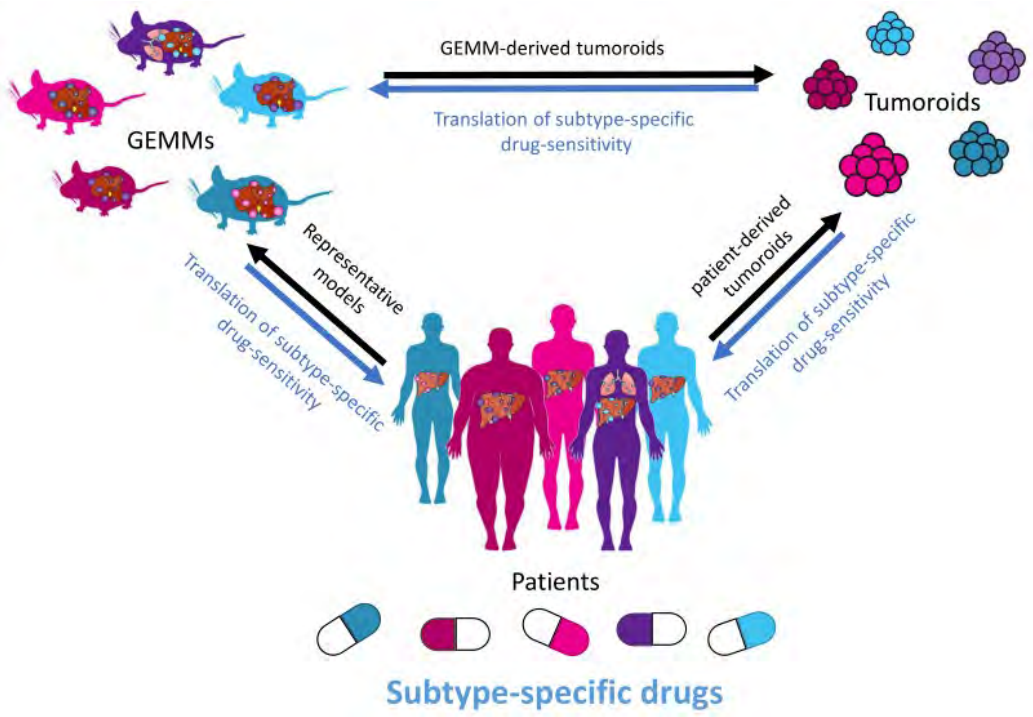
Background and Aims:
Systemic treatment options for advanced hepatocellular carcinoma (HCC) have increased over the last decade but typically have modest efficacy and are generally applied as a one-size-fits-all approach. Refining stratified therapies is hindered by a lack of suitable *in-vivo* models of HCC subtypes. Our aim was to develop a suite of genetically engineered mouse models (GEMMs) representing human subtypes of HCC and to use these to identify and test new therapies.

Method:
We generated the GEMMs combining loxP technology with hepatocyte-specific AAV8-TBG-Cre mediated recombination resulting in clonal HCC outgrowth driven by oncogenes/tumor suppressors identified in human HCC. Over 20 genetic models were disease positioned against human HCC using transcriptomics and histopathological characteristics and cross referenced to classical HCC models. Current standard-of-care therapies were tested for effect on overall survival. Tumoroids were generated from GEMMs and FDA-approved oncology drugs were analyzed for effect on tumoroid growth. Drugs were validated *in vitro* and survival studies were performed in the respective mouse models.

Results:
Targeted hepatocytes expanded from a clonal source to form nodules and ultimately HCC within 3-4 months. The resulting tumors reproduce key features of human disease, including histological patterns, tumor hemorrhage and metastasis. Established tumors respond to first-line tyrosine kinase inhibitors (TKIs; Sorafenib or Lenvatinib) leading to modest but significant survival benefits. We demonstrate lack of efficacy of anti-PD-1 treatment (alone and with TKI) in β-catenin driven tumors, similar to reports in human HCC. Utilizing high-throughput drug screens we identify licensed anticancer drugs, novel in HCC therapy, that markedly increase survival *in vivo,* including in combination with TKIs.

Conclusion:
These models provide a platform for both detailed interrogation of the biological mechanisms driving HCC and improved translational research using a subtype-specific approach to pre-clinical therapeutic identification and testing. We have developed a bank of tumoroids across the range of GEMMs and a pipeline for using these for high-throughput *in vitro* drug screening, followed by subsequent *in vivo* validation. This allows us to rapidly link *in vitro* and *in vivo* efficacy both identifying and testing novel therapies in immunocompetent mice with the ultimate aim of guiding precision medicine trials in human HCC patients.

Figure:


GEMM-derived tumoroids
Translation of subtype-specific drug-sensitivity
GEMMs
Tumoroids
Representative models
Translation of subtype-specific drug-sensitivity
patient-derived tumoroids
Translation of subtype-specific drug-sensitivity
Patients
Subtype-specific drugs

O07 Immunological alterations induced by HBV-TCR T cell immunotherapy associates with treatment response of primary HBV related-HCC.

Anthony Tan[1], Jiehua Jin[2], Jinfang Zhao[2], Fanping Meng[2], Alicia Chua[3], Zi Zong Ho[3], Lu-En Wai[3 4], Sarene Koh[3 4], Antonio Bertoletti[1 4], Fu-sheng Wang[2]

[1]Duke-NUS Medical School, Singapore, Singapore, *[2]The Fifth Medical Center of PLA General Hospital*, *[3]Lion Tcr Pte. Ltd., Singapore, Singapore*, *[4]Singapore Immunology Network*

Email: anthony.tan@duke-nus.edu.sg

Background and Aims:
The application of HBV-TCR T cell immunotherapy in chronic HBV patients with primary HCC have been somewhat apathetic due to the increased risk of on-target off-tumour severe liver inflammatory events. However, such immunological changes after T cell immunotherapy could also indicate a less compromised immune system that can better respond to treatment. Here we longitudinally analysed the immunological, virological, biochemical and radiological alterations in primary HBV-HCC patients receiving HBV-TCR T cell immunotherapy to determine its association with treatment response.

Method:
Patients (n=8) with diffuse non-operable HBV-associated HCC was scheduled to receive escalating doses ($1x10^4$, $1x10^5$, $1x10^6$, $5x10^6$ / kg body weight) of mRNA electroporated HBV-TCR T cells at weekly intervals. Serum and PBMCs were collected at baseline and at regular intervals post T cell infusion to monitor for liver inflammation and the quantity of cytokines and the frequency and activation phenotype of T cells were analysed respectively. Treatment response was evaluated primarily by radiological imaging or by the detection of serum HBV pgRNA as a surrogate.

Results:
2 patients exhibited signs of reversible liver inflammation after receiving the second dose of HBV TCR T cells ($1x10^5$ / kg body weight, ~$6x10^6$ HBV-TCR T cells in total). In one, this is accompanied with the elevation of total bilirubin and clinical symptoms of nausea and jaundice (pt. 1), while the other did not (pt. 2). Interestingly, the peak liver inflammation in pt. 1 coincided with an elevation of activated T cell frequency in the peripheral blood (~20 days post treatment) which was followed by a dramatic destruction of the tumour lesion. While radiological response was not observed in pt. 2, we detected a decline in serum HBV pgRNA levels upon treatment, indicative of a functional HBV-TCR T cell treatment. In two patients with elevations of serum CXCL-9 and CXCL-10, or a detectable increase in activated T cell frequency in the peripheral blood after receiving high doses of HBV-TCR T cells ($5x10^6$ / kg), the tumour load remained stable throughout treatment, while in those with no detectable peripheral blood alterations, tumour load continued to increase.

Conclusion:
Immunological alterations in the peripheral blood induced by T cell immunotherapy is associated with treatment response. This has important implications in the monitoring of primary HBV-HCC patients receiving T cell immunotherapy.

O08YI Non-autonomous induction of endothelial Rela underpins immune-mediated surveillance of premalignant hepatocytes

Kelvin Yin[1 2], Daniel Patten[3], Aaron Lun[1], Martijn Shuijs[1], Adelyne Chan[1], Andrew Young[1], Tim Halim[1], Shishir Shetty[3], Masashi Narita[1], Matthew Hoare[1 4]

[1]Cancer Research UK Cambridge Institute, University of Cambridge, Cambridge, United Kingdom, *[2]The Helmholtz Pioneer Campus, Helmholtz Zentrum München , Neuherberg, Germany*, *[3]Centre for Liver and Gastrointestinal Research, University of Birmingham, Birmingham, United Kingdom*, *[4]Department of Medicine, University of Cambridge, Cambridge, United Kingdom*
Email: chuan-hsin.yin@helmholtz-muenchen.de

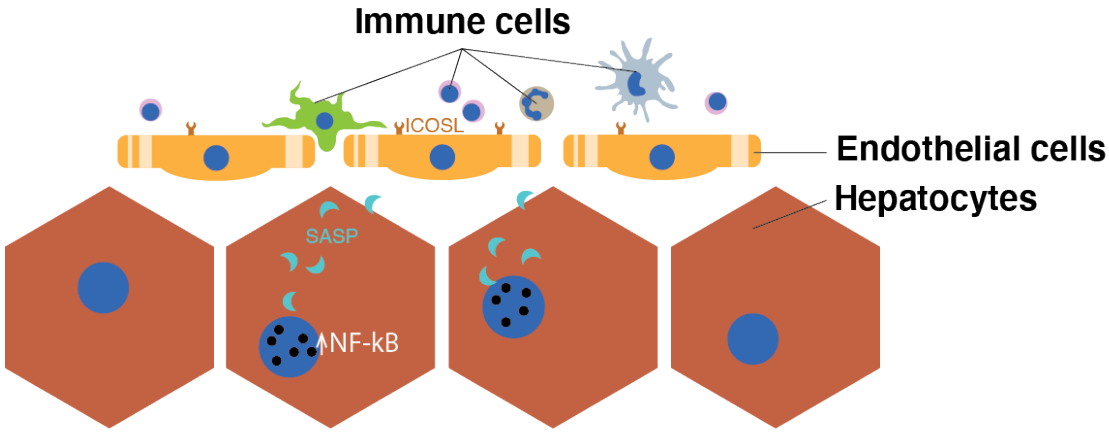
Background and Aims: Oncogene-induced senescence (OIS) is a tumour suppressor mechanism, with profound effects on the microenvironment (ME) through the senescence-associated secretory phenotype (SASP). Previous studies have shown that OIS hepatocytes are progressively cleared by a SASP-driven CD4+ T-lymphocyte-dependent immune response, termed senescence surveillance; failure of this clearance leads to tumorigenesis. The mechanisms underpinning this surveillance of pre-malignant cells are not well understood. We hypothesised that OIS hepatocytes would regulate immunomodulatory endothelial behaviour, controlling immunocyte recruitment and behaviour in the liver ME.

Method: We performed immunofluorescence labelling and flow adhesion assay on *in vitro* models with genetic and/or pharmacological blockade of NF-kB signaling to study the non-autonomous signalling of OIS on liver sinusoidal endothelial cells (LSECs). Hydrodynamic tail vein injection (HDTV) of $NRAS^{G12V}$-containing transposons was used to generate murine hepatocyte OIS in mice models. Immunohistochemistry, flow cytometry, quantitative PCR analyses, mass cytometry and scRNA-sequencing were used to study the *in vivo* ME following hepatocyte OIS.

Results: Using an *in vitro* model of OIS, we have demonstrated a SASP-driven non-autonomous upregulation of NF-kB-target genes in primary human liver sinusoidal endothelial cells (LSECs). This observation was further validated in two independent human endothelial cell lines. Genetic or pharmacological blockade of NF-kB signalling in SASP-primed LSECs abrogated lymphocyte trapping and trans-endothelial migration *in vitro.* Using hydrodynamic tail vein injection (HDTV) of $NRAS^{G12V}$-containing transposons to generate murine hepatocyte OIS, we show hepatocyte OIS non-autonomously upregulates multiple NF-kB-target genes in LSECs, including *Cxcl1* and *Icosl*. Involvement of Icosl-Icos signalling was confirmed by mass cytometry. Furthermore, using an inducible endothelial-specific knock-out of the canonical NF-kB component *Rela* in the context of hepatocyte OIS, we find that endothelial NF-kB signalling is crucial to senescence surveillance. Loss of endothelial *Rela* leads to persistence of OIS hepatocytes and scRNA-Seq shows a complete loss of a *Stat1*-expressing CD4+ lymphocyte subset from the liver ME.

Conclusion: Our findings demonstrate that endothelial cells are a target of non-autonomous signalling from senescent cells *in vitro* and *in vivo.* We have found that immunomodulatory endothelial behaviour, through non-autonomous induction of endothelial NF-kB signalling, forms a crucial part of the immune surveillance of premalignant hepatocytes.

Figure:


Immune cells
ICOSL
Endothelial cells
Hepatocytes
SASP
NF-kB

POSTER ABSTRACT PRESENTATIONS

P001YI Surgery versus Sorafenib in case of BCLC-C hepatocellular carcinoma: an Italian weighted study

Simone Famularo[1 2], Matteo Donadon[2], Federica Cipriani[3], Felice Giuliante[4], Alessandro Ferrero[5], Sarah Molfino[6], Raffaelle Dalla Valle[7], Marco Chiarelli[8], Marcello Maestri[9], Elio Jovine[10], Gianluca Grazi[11], Andrea Ruzzenente[12], Riccardo Memeo[13], Michele Crespi[14], Adelmo Antonucci[15], Davide Bernasconi[16], Fabrizio Romano[1 17], Guido Griseri[18], Luca Aldrighetti[3], Guido Torzilli[2], Franco Trevisani[19]

[1]University of Milan-Bicocca, School of Medicine and Surgery, Monza, Milan, Italy, *[2]Humanitas University, Humanitas Clinical and Research Center, Department of Hepatobiliary and General Surgery, Rozzano, Milan, Italy*, *[3]San Raffaele Hospital, Hepatobiliary Surgery Division,, Milan, Italy*, *[4]Fondazione Policlinico Universitario A. Gemelli, IRCCS, Catholic University of the Sacred Heart, Hepatobiliary Surgery Unit, Rome, Italy*, *[5]Mauriziano Hospital "Umberto I", Department of General and Oncological Surgery, turin, Italy*, *[6]University of Brescia, Department of Clinical and Experimental Sciences, Brescia, Italy*, *[7]University of Parma, Department of Medicine and Surgery, Parma, Italy*, *[8]ASST Lecco, Department of Emergency and Robotic Surgery, Lecco, Italy*, *[9]University of Pavia and Foundation IRCCS Policlinico San Matteo, Unit of General Surgery 1, Pavia, Italy*, *[10]Alma Mater Studiorum, University of Bologna, AOU Sant'Orsola Malpighi, IRCCS at Maggiore Hospital, Bologna, Italy*, *[11]IRCCS - Regina Elena National Cancer Institute, Division of Hepatobiliarypancreatic Unit, Rome, Italy*, *[12]University of Verona, Division of General and Hepatobiliary Surgery, Department of Surgical Sciences, Dentistry, Gynecology and Pediatrics, Verona, Italy*, *[13]Miulli Hospital, Department of Hepato-Pancreatic-Biliary Surgery, Bari, Italy*, *[14] L. Sacco Hospital, Department of Surgery, Milan, Italy*, *[15]Monza Policlinic, Department of Surgery, Monza, Italy*, *[16]University of Milan - Bicocca, Center of Biostatistics for Clinical Epidemiology, School of Medicine and Surgery, Monza, Italy*, *[17]San Gerardo Hospital, Department of Surgery, Hepatobiliary surgery Division, Monza, Italy*, *[18]San Paolo Hospital, HPB Surgical Unit, Savona, Italy*, *[19]Alma Mater Studiorum-University of Bologna, Department of Medical and Surgical Sciences, Semeiotics Unit, Bologna, Italy*
Email: simone.famularo@gmail.com

Surgery versus Sorafenib in case of BCLC-C hepatocellular carcinoma: an Italian weighted study

Background and Aims: The treatment of hepatocellular carcinoma (HCC) in BCLC-C stage is still controversial. The aim of the study was to compare the overall survival (OS) of patients treated with surgery (SURG) vs. sorafenib (SOR) in national cohorts.

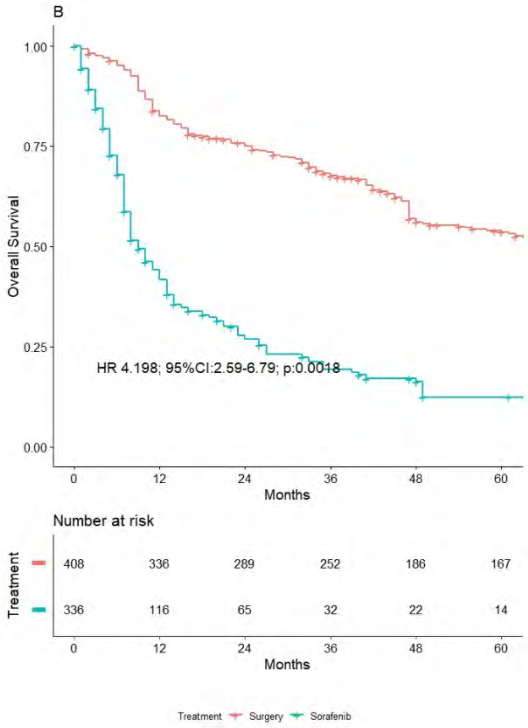
Method: BCLC-C patients without extra-hepatic spread were considered. All patients were at their first tumor treatment. Surgical patients were those from the Hepatocarcinoma-Recurrence-on-the-Liver-Study (HE.RC.O.LE.S.) Group, while Sorafenib patients were those from the Italian-Liver-Cancer (ITA.LI.CA.) Group. Inverse Probability Weighting (IPW) was used to weight the confounders between the two groups. Kaplan-Meier analysis and Cox regression were adopted for survival analysis.

Results: Between 2008 and 2018, 519 patients were eligible, 303 in SURG and 216 in SOR group. ECOG performance status, presence of cirrhosis, steatosis, Child-Pugh(CPT)-score, HBV, HCV, alcohol intake, collateral veins, bilobar disease, localization of the tumor thrombus, alpha-fetoprotein, age and Charlson Comorbidity index were weighted by IPW to create two balanced pseudo-populations (SURG=408 and SOR=335). After IPW, 1-3-5 years OS was 82.5%, 67.6%, 53.6% and 41.7%, 19.3%, 12.4% for SURG and SOR, respectively (p=0.0018). Survival benefits of SURG over SOR were identified in patients ECOG >0 (p<0.001), and segmental portal invasion (p<0.001) only. After adjustment for confounders,

being treated by SOR rather than by SURG was the only independent predictor of mortality (HR=4.266; 95%CI=2.67-6.81; p=0.0014).

Conclusion: In BCLC-C patients surgical resection, when feasible, guarantees an increased survival compared to systemic treatment.

Figure:



P002YI Current Trends of Hepatocellular Carcinoma Characteristics in Greece

Georgios Markakis[1], Andreas Koulouris[2], Maria Tampaki[1], Evangelos Cholongitas[3], George Papatheodoridis[1], Ioannis-Georgios Koskinas[4]

[1]National and Kapodistrian University of Athens, Gastroenterology and Hepatology, Athens, Greece, *[2]University of Crete, Oncology, Heraklion, Greece*, *[3]National and Kapodistrian University of Athens, 1st Dep. of Internal Medicine, Athens, Greece*, *[4]National and Kapodistrian University of Athens, 2nd Dep. of Internal Medicine, Athens, Greece*
Email: markakismd@gmail.com

Background and Aims: Hepatocellular carcinoma (HCC) is the sixth most common cancer and the fourth leading cause of cancer death globally. It is mainly associated with Hepatitis B and C virus (HBV and HCV), alcohol and non-alcoholic fatty liver disease (NAFLD). The aim of this study was to evaluate current HCC characteristics and possible changes during the last decades in Greece.

Method: 300 consecutive patients diagnosed with HCC in two academic hospitals in Athens, Greece from 2000 to 2019, were evaluated for the etiology of liver disease, patient and HCC characteristics and possible changes concerning staging and biological behavior (alpha fetoprotein, aFP) at diagnosis, in two time periods; before 2011 (A) and after 2011 (B). Our findings were also compared with data of Greek patients from previous decades.

Results: Median age of patients with HCC was 64 years, 86% were male and 97% Caucasian. 134 patients (45%) had HBV, 77 (26%) HCV and 89 (30%) non-viral liver disease (nvLD). Median aFP was 78ng/ml and 86% of patients were cirrhotic. At presentation, the largest nodule was <3cm in 18% and >5cm in 50%, while 20% of cases had >3 nodules.

No change was observed among the causes of liver disease between periods A and B. However, there was a trend towards a decreasing proportion of viral etiology, HBV (from 48% to 44%) and HCV (from 30% to 23%), with a relative increased proportion of non-viral liver diseases (from 23% to 33%) (p=.075). Regarding the stage and biological behavior of HCC, patients in period A vs B had less frequently <3 nodules (69% vs 85%, p=.006) and largest nodule <3cm (5% vs 24%, p=.005).

Comparison among different liver diseases yielded a younger median age at HCC diagnosis in HCV vs HBV or nvLD (57 vs 66 or 69 years, p≤.005), higher proportion of men in HBV vs nvLD vs HCV (93% vs 85% vs 75%, p=.002) and a tendency for an increased proportion of cirrhotic patients in HCV vs HBV vs nvLD (93% vs 86% vs 80%, p=.054).

Lastly, compared with data from 1558 Greek HCC patients diagnosed between 1974-2000, there was a decrease in the proportion of HBV (from 59% to 45%) and an increase in the proportion of HCV (13% to 25%) and nvLD (28% to 30%) related HCCs in our current cohort (p<.001).

Conclusion: In Greece, after 2000, there is a decrease in the proportion of HBV and an increase in the proportion of HCV and nvLD related HCC. Over the last two decades, there is a decreasing trend in HBV and HCV and an increasing trend in non-viral induced HCC. Patients with HCV related HCC compared to other liver diseases are younger, less frequently men and tend to be more frequently cirrhotic. After 2011, HCC is being diagnosed at an earlier stage, possibly reflecting an improvement in surveillance strategies.

P003 Patterns of response to atezolizumab (atezo) + bevacizumab (bev) in hepatocellular carcinoma (HCC) from the Phase 1b GO30140 study

Stacey Stein[1], Chih-Hung Hsu[2], Michael Lee[3], Kyung-Hun Lee[4], Baek-Yeol Ryoo[5], Aiwu Ruth He[6], Jessica Spahn[7], Wendy Verret[7], Steve Hack[7], Vincent Gaillard[8], Sven Stanzel[8], Riccardo Lencioni[9]

[1]Yale School of Medicine, New Haven, United States, *[2]National Taiwan University Hospital, Taipei, Taiwan*, *[3]University of Texas MD Anderson Cancer Center, Houston, United States*, *[4]Seoul National University Hospital, Seoul, Korea, Rep. of South*, *[5]Asan Medical Center, University of Ulsan College of Medicine, Seoul, Korea, Rep. of South*, *[6]Georgetown University, Washington, DC, United States*, *[7]Genentech, Inc., South San Francisco, United States*, *[8]F. Hoffmann-La Roche, Basel, Switzerland*, *[9]University of Pisa, Pisa, Italy*
Email: stacey.stein@yale.edu

Background and Aims: Atezo+bev is approved globally for pts with unresectable HCC who have not received prior systemic therapy. Here, we present an analysis of patterns of antitumor response in pts who received atezo+bev in the Phase 1b GO30140 trial (NCT02715531).

Method: In GO30140, pts with unresectable HCC who were systemic treatment–naive and enrolled in the single-arm HCC cohort received atezo 1200 mg IV q3w + bev 15 mg/kg IV q3w until unacceptable toxicity or loss of clinical benefit per investigator. This exploratory analysis investigated patterns of response in these pts, including those with deep (≥65% reduction in the sum of the longest diameters from baseline) and prolonged (≥6 mo) responses, as well as efficacy by response status.

Results: 36% of pts had a response per RECIST 1.1; 62% of responders had a deep response, 65% had a prolonged response and 54% had both a deep and prolonged response (HCC modified [m]RECIST data in table). Pts with deep and/or prolonged responses generally had more favorable prognostic factors vs the overall study population, including lower disease burden and absence of extrahepatic spread and/or macrovascular invasion. Pts achieving any response (CR/PR, regardless of depth or duration), and to a lesser extent SD, showed prolonged OS and PFS vs pts with PD (Table). Also, pts with liver lesions had a numerically higher response rate with atezo+bev than pts with lung lesions (Table).

Conclusion: Deep and/or prolonged responses with atezo+bev seem to be associated with favorable prognostic factors. Pts achieving CR/PR appeared to have the greatest survival benefit. Additional studies are needed to confirm this association.

Table: Response and efficacy by response status

	RECIST 1.1[a] n=104			HCC mRECIST[a] n=104		
Objective response rate, n (%)	37 (36)			41 (39)		
Deep response[b]	23 (62)			30 (73)		
Prolonged response[b]	24 (65)			30 (73)		
Deep and prolonged response[b]	20 (54)			25 (61)		
	CR/PR	**SD**	**PD**	**CR/PR**	**SD**	**PD**
Best response, n (%)	37 (36)	37 (36)	25 (24)	41 (39)	33 (32)	25 (24)
Pts with liver lesions[c]	25 (34)	31 (42)	13 (18)	29 (39)	27 (36)	13 (18)
Pts with lung lesions	9 (26)[d]	9 (26)[d]	17 (49)[d]	10 (28)[e]	9 (25)[e]	17 (47)[e]
Median OS (95% CI), mo	NE	16.1 (13.8-NE)	8.0 (5.1-10.8)	NE	16.1 (11.9-NE)	8.0 (5.1-10.8)
Median PFS (95% CI), mo	NE (13.6-NE)	7.0 (5.4-9.4)	1.9 (1.7-1.9)	NE (13.6-NE)	7.0 (5.3-9.5)	1.9 (1.7-1.9)

Clinical cutoff: Jun 14, 2019.
NE, not estimable.
[a]Per independent review facility. [b]Percentages were calculated based on number of responders.
[c]n=74. [d]n=35. [e]n=36.

P004YI Association between serum magnesium concentration and hepatocellular carcinoma in cirrhotic patients

Simona Parisse[1], Flaminia Ferri[1], Marzia Persichetti[1], Monica Mischitelli[1], Aurelio Abbatecola[1], Michele Di Martino[2], Quirino Lai[3], Sara Carnevale[1], Pierleone Lucatelli[2], Massimo Rossi[3], Mario Bezzi[2], Adriano De Santis[1], Stefano Ginanni Corradini[1]

[1]Sapienza University of Rome, Translational and Precision Medicine, Rome, Italy, *[2]Sapienza University of Rome, Department of Radiological, Oncological and Pathological Sciences, Rome, Italy*, *[3]Sapienza University of Rome, Hepatobiliary and Organ Transplantation Unit, Rome, Italy*
Email: simona.parisse@gmail.com

Background and Aims: Mg (Mg) acts as a cofactor for more than 600 enzymatic reactions and has a central role in many important cellular functions, notably DNA repair, cell metabolism and proliferation. In some tumors, low Mg serum concentration was observed, allowing to hypothesize an increased Mg avidity by neoplastic tissue with a consequent decrease in DNA repair and increase in cell metabolism and proliferation. No data on serum Mg level and hepatocellular carcinoma (HCC) are currently available.

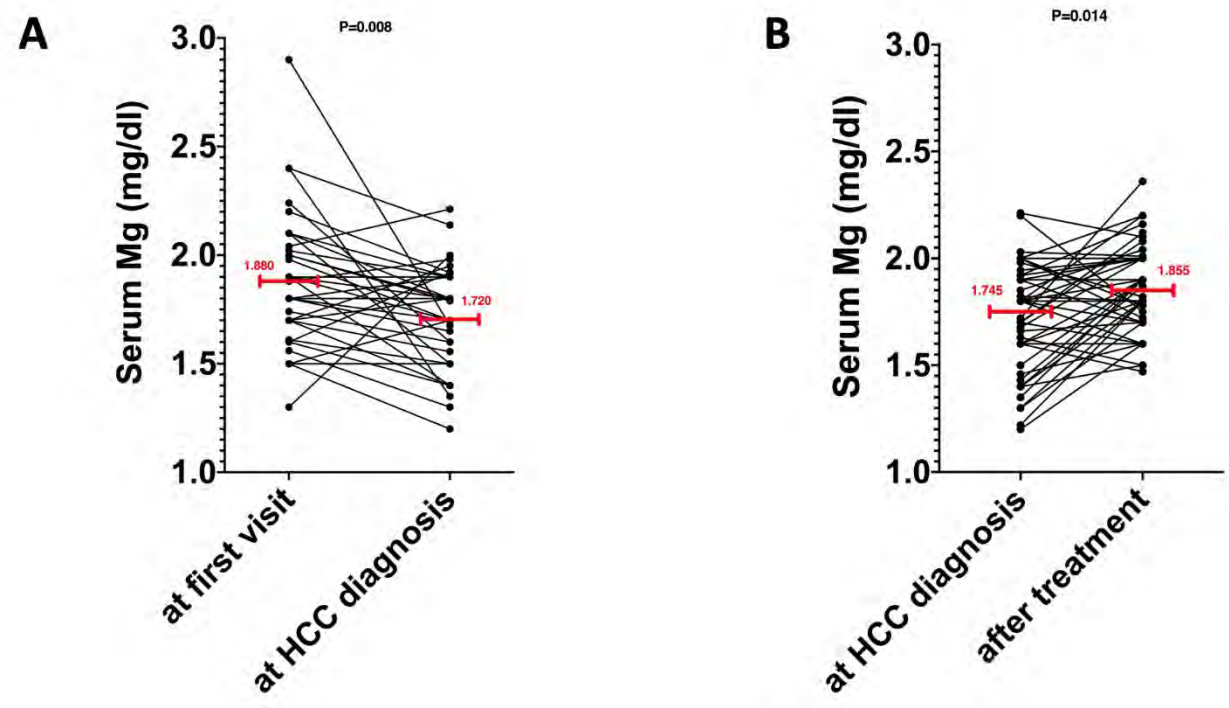
The aims of this study are to compare serum Mg concentrations in cirrhotic patients with and without hepatocellular carcinoma and to analyze, in a subgroup of patients, the differences of serum Mg concentrations before and at the time of HCC diagnosis.

Method: We enrolled 291 patients with an available Mg serum concentration at the beginning of cirrhosis follow-up and/or at HCC diagnosis: 161 patients were cirrhotic without HCC, 90 patients were cirrhotic with an HCC diagnosis at the time of enrollment and 40 patients were enrolled as cirrhotic without HCC and developed HCC during the follow-up time.

Results: Cirrhotic patients with HCC had lower serum Mg concentrations at the time of diagnosis (1.80 95% CI:1.62-1.90) than those without HCC (1.90 95% CI:1.72-2.08, P< 0.001). In the multivariate-adjusted analysis, low serum Mg was independently associated with the presence of HCC, OR = 0.050 (95% C.I. 0.016-0.174, P< 0.001). In a subgroup of 40 patients who developed HCC during the follow-up time, serum Mg concentrations decreased at HCC diagnosis, compared to the corresponding values before diagnosis (1.720 vs 1.880, P=0.008, respectively) and this decrease was correlated with total tumor diameter (r=-0.383, P=0.020). In 54 patients with HCC, we also evaluate the serum Mg at diagnosis and after HCC locoregional treatment, the data analysis underlines an increment of serum Mg after HCC treatment (1.745 vs 1.855, P= 0.014, respectively).

Conclusion: the study results are consistent with the hypothesis that HCC, like other tumors, may be avid for Mg and behave as a Mg trap, disturbing body Mg balance and resulting in lower serum Mg levels.

Figure:
Single values and mean of Mg concentration at the beginning of follow-up for cirrhosis and at HCC diagnosis (A) in a subgroup of 40 patients and at HCC diagnosis and after HCC treatment (B) in a subgroup of 54 patients



P005YI The best potential treatment for recurrent hepatocellular carcinoma after surgery: a machine learning predictive model for treatment allocation based on an Italian multicentric database

Simone Famularo[1 2], Matteo Donadon[2], Federica Cipriani[3], Federico Fazio[4], Francesco Ardito[5], Francesca Carissimi[1], Maurizio Iaria[6], Pasquale Perri[7], Simone Conci[8], Nicholas Pontarolo[9], Quirino Lai[10], Giuliano La Barba[11], Stefan Patauner[12], Sarah Molfino[13], Paola Germani[14], Alberto Manzoni[15], Enrico Pinotti[16], Matteo Zanello[17], Luca Fumagalli[18], Cecilia Ferrari[19], Maurizio Romano[20], Antonella Del Vecchio[21], Davide Paolo Bernasconi[22], Maria Grazie Valsecchi[22], Adelmo Antonucci[23], Riccardo Memeo[21], Giacomo Zanus[20], Guido Griseri[19], Marco Chiarelli[18], Elio Jovine[17], Mauro Zago[16], Giuseppe Zimmitti[15], Paola Tarchi[14], Gianluca Baiocchi[13], Antonio Frena[12], Giorgio Ercolani[11], Massimo Rossi[10], Marcello Maestri[9], Andrea Ruzzenente[8], Gianluca Grazi[7], Raffaelle Dalla Valle[6], Fabrizio Romano[1], Felice Giuliante[5], Alessandro Ferrero[4], Luca Aldrighetti[3], Guido Torzilli[2]

[1]Building U8 - University of Milan Bicocca, School of Medicine and Surgery, Monza, Italy, *[2]Humanitas Research Hospital, Department of Hepatobiliary and General Surgery, Rozzano, Italy*, *[3]San Raffaele Hospital, Hepatobiliary Surgery Division, Milano, Italy*, *[4]Mauriziano Umberto I Hospital, Department of General and Oncological Surgery, Torino, Italy*, *[5]Agostino Gemelli University Policlinic, Hepatobiliary Surgery Unit,, Roma, Italy*, *[6]University of Parma, Department of Medicine and Surgery, Parma, Italy*, *[7]IRCCS - Regina Elena National Cancer Institute, Division of Hepatobiliarypancreatic Unit, Roma, Italy*, *[8]University of Verona, Division of General and Hepatobiliary Surgery, Department of Surgical Sciences, Dentistry, Gynecology and Pediatrics, Verona, Italy*, *[9]Fondazione I.R.C.C.S. Policlinico San Matteo, Unit of General Surgery 1, Pavia, Italy*, *[10]Policlinico Umberto I, Hepato-Biliary Surgery and Organ Transplantation Unit, Roma, Italy*, *[11]Ospedale Morgagni - Pierantoni, General and Oncologic Surgery, Forlì, Italy*, *[12]Ospedale di Bolzano, Department of General and Pediatric Surgery, Bolzano, Italy*, *[13]University of Brescia, Department of Clinical and Experimental Sciences, Brescia, Italy*, *[14]University Hospital of Trieste, Trieste, Surgical Clinics, Trieste, Italy*, *[15]Fondazione Poliambulanza Istituto Ospedaliero, Department of General Surgery, Brescia, Italy*, *[16]Policlinico San Pietro, Department of Surgery, Ponte San Pietro, Italy*, *[17]University of Bologna, AOU Sant'Orsola Malpighi, IRCCS at Maggiore Hospital,, Alma Mater Studiorum, Bologna, Italy*, *[18]Hospital Alessandro Manzoni Lecco, Department of Emergency and Robotic Surgery, Lecco, Italy*, *[19]St. Paul's Hospital, HPB Surgical Unit,, Savona, Italy*, *[20]University of Padua, Department of Surgical, Oncological and Gastroenterological Science (DISCOG), Padova, Italy*, *[21]Regional General Hospital F. Miulli, Department of Hepato-Pancreatic-Biliary Surgery, Acquaviva delle Fonti, Italy*, *[22]Building U1 - University of Milan - Bicocca, Bicocca Bioinformatics Biostatistics and Bioimaging Centre - B4, School of Medicine and Surgery, Milano, Italy*, *[23]Monza Policlinic, Department of Surgery, Monza, Italy*
Email: simone.famularo@gmail.com

The best potential treatment for recurrent hepatocellular carcinoma after surgery: a machine learning predictive model for treatment allocation based on an Italian multicentric database.

Background and Aims: How to select effective treatments for patients with hepatocellular carcinoma recurrence (rHCC) after surgery is still unclear. The aim was to develop a machine-learning predictive model of survival after recurrence (SAR) under curative treatments (CT), meaning redo-surgery or thermoablation versus Sorafenib (SOR) or chemoembolization (TACE).

Method: 23 Italian centers participating to HE.RC.O.LE.S. register shared their data on rHCC. Treatment effect modifiers (TEM) were selected and a model for SAR was fitted. This was used to estimate the potential outcome after CT, SOR or TACE. After best potential treatment (BPT) establishment, a comparison was made with the received treatment. A tree-diagram to choice the BPT was created.

Results: Between 2008 and 2019, 701rHCC were enrolled. Patients submitted to CT were 293 (41.8%), while 188 (26.8%) underwent SOR and 220 (31.4%) TACE. At Cox regression, being treated by SOR (HR 3.678; 95%CI:2.664-5.078; p<0.001) and by TACE (HR 1.995; 95%CI:1.431-2.782; p<0.001); recurrent nodules>1 (HR 2.115; 95%CI:1.618-2.765; p<0.001) and size>5cm (HR 2.438; 95%CI:1.773-3.352; p<0.001); bilobar recurrence (HR 1.826; 95%CI:1.406-2.373; p<0.001); concomitant extra-hepatic spread (HR 2.782; 95%CI:2.052-3.772; p<0.001) and time to recurrence (HR1.793; 95%CI-1.440; 2.232,p<0.001) were identified as TEMs and then used to build the model. The AUC was 78.3% for mortality risk at 3-years, and 79.4% at 5-years after recurrence. The BPT was CT for 583 patients (84.4%), SOR for 48 (5.7%) and TACE for 70 (9.9%). Those factors were employed to build 16 risk-profiles to develop a patient-tailored algorithm for the BPT allocation.

Conclusion: Curative treatments were the best potential treatments.When surgery or thermoablation are feasible, they should have hierarchical priority in rHCC treatment regardless of the tumor features.

Figure:

P006YI Cholangiocarcinoma progression depends on the uptake and metabolization of extracellular lipids

Mikel Ruiz de Gauna[1], Francesca Biancaniello[2 3], Pedro Miguel Rodrigues[2], Francisco Gonzalez-Romero[1], Ibone Labiano[2], Igor Aurrekoetxea[1], Ainhoa Lapitz[2], Ane Nieva-Zuluaga[1], Asier Benito-Vicente[4 5], Maider Apodaka-Biguri[1], María Jesús Perugorria[2], Diego Saenz de Urturi[1], Beatriz Gómez Santos[1], Helen Carr-Ugarte[1], Xabier Buque[1], Igotz Delgado[1], Luis Bujanda[2], Cesar Augusto Martín[4 5], Jesus Maria Banales[2], Patricia Aspichueta[1 6]

[1]University of the Basque Country UPV/EHU, Faculty of Medicine and Nursing, Department of Phisiology, 48940 Leioa, Spain, *[2]Department of Liver and Gastrointestinal Diseases, Biodonostia Health Research Institute, Donostia University Hospital, University of the Basque Country UPV/EHU, CIBERehd, Ikerbasque, San Sebastian, 20014 San Sebastian, Spain*, *[3]Department of Translational and Precision Medicine, "Sapienza" University of Rome, Rome, Italy*, *[4]Department of Molecular Biophysics, Biofisika Institute (University of Basque Country and Consejo Superior de Investigaciones Científicas (UPV/EHU, CSIC)), 48940 Leioa, Spain*, *[5]Department of Biochemistry and Molecular Biology, University of the Basque Country (UPV/EHU), 48940 Leioa, Spain*, *[6]Biocruces Bizkaia Health Research Institute, Cruces University Hospital, 48903 Barakaldo, Spain*
Email: mikelrgm@hotmail.com

Background and Aims: Metabolic reprogramming is a major hallmark of cancer and a potential source of new targets for treatment. Little is known about the dysregulation of lipid metabolism in cholangiocarcinoma (CCA), a group of highly heterogeneous biliary malignancies characterized by dismal prognosis due to the lack of effective treatments and high chemoresistance features. Current evidence suggests that *de novo* fatty acid (FA) synthesis, upregulated in hepatocellular carcinoma, is downregulated in CCA, pointing towards exogenous lipids as the main source of FA for these tumor cells. Here, we aimed to investigate the dysregulation of lipid metabolism in CCA human cells and the role in tumor proliferation and progression.

Method: Enrichment analysis of the proteome of 5 different human CCA cell lines and 5 primary cultures of normal human cholangiocytes (NHCs) was performed by full proteomic mass spectrometry. CCA cell proliferation and migration *in vitro*, as well as their tumorigenic capacity in a xenograft mouse model were analysed in all cell lines. FA uptake and metabolic fate were measured through the incorporation of ^{3}H-oleate. Cellular uptake of very-low-density (VLDLs), low-density (LDLs) and high-density lipoproteins (HDLs) was studied after lipoprotein fluorescent labelling. Furthermore, ^{14}C-palmitate oxidation rate was quantified and concentration of different lipid species was measured. For inhibition of the FA oxidation (FAO) rate etomoxir was used.

Results: The most dysregulated proteins in CCA cells are linked to energetic metabolism; of note, 12.6% of these proteins are involved in the metabolism of lipids and lipoproteins. The EGI1 CCA cell line showed the highest proliferative and migration capacity compared to the other CCA cell lines in both *in vitro* and *in vivo*. Further metabolic studies in high (EGI1) *vs* low (HUCCT1) proliferative CCA cells, as well as in a primary culture of NHCs (NHC3) used as control, showed that that both CCA cell lines incorporated more oleic acid than NHC3. EGI1 also showed greater uptake of VLDLs and HDLs than NHC. Additionally, the esterification of extracellular FAs into triglycerides (TGs) was upregulated in both CCA cell lines when compared to NHC, sustaining a greater storage of TGs in the tumor cells. The FAO rate was also found specifically upregulated in EGI1, and consequently, pharmacological inhibition of FAO induced a more pronounced inhibition of their proliferation features compared to HUCCT1.

Conclusion: Different classifications of CCAs have been previously proposed based on their transcriptomic profiles, including a “proliferation-like” subtype. We here show that highly proliferative human CCA cells relies on lipid and lipoprotein uptake to fuel the FA catabolism, suggesting that inhibition of FAO and/or uptake of lipoproteins and FAs could be a potential strategy for treatment of “proliferation-type” CCAs.

Figure:

P007 Sequential Treatment of Sorafenib-Regorafenib versus Sorafenib-Physician's Choice: A Propensity Score Matched Analysis

Andrea Casadei Gardini[1], Yeonghak Bang[2], Sara Lonardi[3], Caterina Vivaldi[4], Ilario Giovanni Rapposelli[5], Mario Scartozzi[6], Changhoon Yoo[2]

[1]San Raffaele hospital, oncology, *[2]Asan Medical Center, Korea, Rep. of South*, *[3]Institute Oncology Veneto, Padova, Italy*, *[4]University of Pisa, Pisa, Italy*, *[5]Oncological Institute Romagnolo, Forlì, Italy*, *[6]Università degli Studi di Cagliari, Cagliari, Italy*
Email: casadeigardini@gmail.com

Background and Aims:
Regorafenib has been shown to improve clinical outcomes compared to placebo,becoming the standard second-line therapy for sorafenib-progressed and -tolerated HCC patients,but an evaluation of its use in a real-life setting has not been previously published.

Method:
This is a multinational, multicentre, retrospective study from Italy and Korea.A propensity score model was developed to control the results for baseline variable imbalances between the arm treated with sorafenib and regorafenib (S-R) and the arm treated with sorafenib and physician choice (S-P). Survival analysis was conducted on the matched population.

Results:
After the application of propensity score matching, we analysed 99 patients in the arm treated with S-R and 99 patients in the arm treated with S-P. For the S-R group, the median OS was 22.2 months (95% CI: 17.1–27.4), compared to 17.9 months(95% CI: 15.1–50.0) for the S-P group (Fig 1A). The results of the univariate analysis showed a 31% reduction of death risk for patients treated with S-R(p=0.0382) compared to patients treated with S-P.

From second line, for patient treated with regorafenib the median OS was 10.1 months (95% CI: 8.0–30.0) compared to 9.0 months (95% CI: 6.1–30.0) for the patient group treated with physician choice (Fig 1B). The results from the univariate analysis showed a 35% reduction of death risk for patients treated with regorafenib (95% CI: 0.46–0.93; p=0.0193) compared to patients treated with physician choice.

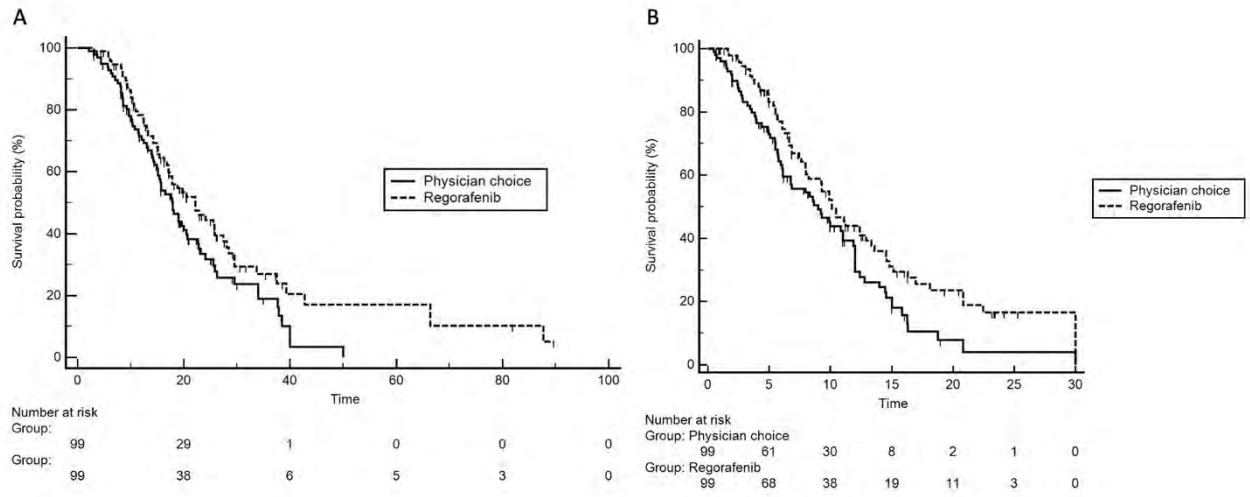
The forest plot highlighted that the sequence sorafenib and regorafenib conferred a better OS compared to sorafenib and physician choice in patients with age > 65 years (HR 0.53; p=0.0307) , male (HR 0.64; p=0.0279) (Supplementary Fig 2B), BCLC-B (HR 0.33; p=0.0230) (Supplementary Fig 2C), HCV-positive patients (HR 0.48; p=0.0068), HBV-positive patients (HR 0.34; p=0.0085) (Supplementary Fig 2E), extrahepatic spread (HR 0.50; p=0.0130) and NLR <3 (HR 0.49; p=0.0125). Conversely, patients with NLR>3 had a better OS in patients treated with sorafenib and physician choice (Ref NLR<3; HR 1.72; p=0.048) compared to patients treated with sorafenib and regorafenib.

Interaction tests highlighted the predictive role of AFP, NLR and extrahepatic spread.

Conclusion:

This study provides more proof of the superiority of the S-R treatment over the S-P treatment approach in advanced HCC patients from a real-life setting.

Figure:


A
Survival probability (%)
100
80
60
40
20
0
0
20
40
60
80
100
Time
Physician choice
Regorafenib
Number at risk
Group:
99 29 1 0 0 0
Group:
99 38 6 5 3 0
B
Survival probability (%)
100
80
60
40
20
0
0
5
10
15
20
25
30
Time
Physician choice
Regorafenib
Number at risk
Group: Physician choice
99 61 30 8 2 1 0
Group: Regorafenib
99 68 38 19 11 3 0

P008 Study on Regorafenib Combined with Transcatheter Arterial Chemoembolization in the Treatment of Advanced Hepatocellular Carcinoma after First Line Targeted Therapy

Long Jin[1], Haochen Wang[1], Yanjing Han[1], Shasha Cao[1], Guang Chen[1], Jianan Yu[1], Tianhao Su[1], chao yin[1], Zhiyuan Zhang[1], Jian Wei[1], Guowen Xiao[1], Yuefeng Hu[1], Zeran Yang[1], Zhenfeng Li[1]

[1]Beijing Friendship Hospital, Department of Interventional Radiology, Beijing , China
Email: longerg@hotmail.com

Background and Aims: The purpose of this study is to study the efficacy and safety of regorafenib combined with TACE in the treatment of advanced HCC patients after first-line targeted treatment.

Method: From October 2019 to September 2020, patients with unresectable HCC who had previously received targeted therapy were included in the study. Inclusion criteria: 1. Once received at least one targeted therapy of Sorafenib, Rivatinib or Apatinib, and the disease progressed; 2. Patients with BCLC B / C HCC; 3. Child Pugh A / B liver function; 4. ECOG score 0-1; 5. At least one measurable lesion was evaluated. The patients were treated with routine TACE (lipiodol + epirubicin + gelatin sponge). After one week of operation, oral administration of Regorafenib (80-160mg, once a day) was started for 3 weeks and stopped for 1 week. Objective response rate (ORR), disease control rate (DCR), progression free survival rate (PFSR) at 3 and 6 months and safety were evaluated according to modified-RECIST.

Results: A total of 19 patients were treated with Regorafenib after the failure of first-line targeted therapy during this period. Two patients were excluded because the ECOG score were 2 points. 1 patient lost the follow-up one week after taking the drug. 16 patients were included in the study. By October 2020, the longest follow-up time for all patients was 12 months. Tumor progression occurred in 3 patients during the treatment. The 3-month progression free survival rate (PFSR) was 92.7%. The 6-month progression free survival rate (PFSR) was 81.3%. After treatment, 1 patient was CR, 9 patients were PR, 3 patients were SD and 3 patients were PD. ORR was 62.5%, DCR was 81.3%. The most common grade 3 / 4 adverse reaction was hypertension (grade 3 in 2 cases and grade 4 in 1 case). No treatment-related death occurred.

Conclusion: For patients with advanced hepatocellular carcinoma treated after first-line targeted therapy, Regorafenib combined with TACE has achieved higher ORR and DCR, and only 3 patients have grade 3 / 4 adverse reactions. The 3 and 6 month progression free survival rate (PFSR) was 92.7%. and 81.3% respectively. Therefore, Regorafenib combined with TACE is effective and safe in the treatment of advanced hepatocellular carcinoma.

Table 1. Baseline Characteristic

Parameter	Regorafenib+TACE
Median age	56.8
Sex	
Male	14(87.5%)
Female	2(12.5%)
BCLC	
B	4(25.0%)
C	12(75.0%)
Child-pugh	
A	14(87.5%)
B	2(12.5%)
AFP	
＜400	12（75.0%）
≥400	4(25.0%)
HBV	
Y	12(75.0%)
N	4(25.0%)
Vascular invasion	
Y	5(31.3%)
N	11(68.7%)
Extrahepatic sites	
Y	6(37.5%)
N	10(62.5%)
First line drugs	
Sorafenib	11(68.7%)
Rivatinib	1(6.3%)

Apatinib	4(25.0%)

Table 2 Efficacy Outcomes

Parameter	Regorafenib+TACE
CR	1
PR	9
SD	3
PD	3
ORR	62.50%
DCR	81.30%

Table 3 Adverse Events with Any Grades

	Any grade	＞3
Hand-foot Reaction	8	0
Weight decreased	3	0
Astriction	2	0
Proteinuria	1	0
Weak	2	0
Hypertention	0	3
Diarrhea	1	0
Hoarseness	2	0

P009YI Hepatocellular carcinoma prognostic scores: filling the gap?

Michele Campigotto[1], Mauro Giuffrè[1], Anna Colombo[1], Alessia Visintin[1], Alessandro Aversano[1], Martina Budel[1], Anna Piccin[1], Carolina Aliai Micol Cavalli[1], Riccardo Sigon[1], Flora Masutti[2], Cristiana Abazia[2], Fabio Tinè[2], Saveria Lory Croce'[1 2 3]

[1]Università degli Studi di Trieste, Dipartimento Clinico Universitario di Scienze Mediche, Chirurgiche e della Salute, Trieste, Italy, *[2]Azienda Sanitaria Universitaria Giuliano Isontina, Clinica Patologie del Fegato, Trieste, Italy*, *[3]Fondazione Italiana Fegato, Basovizza, Italy*
Email: campigottomichele@gmail.com

Background and Aims:
Hepatocellular carcinoma is the most common primitive liver malignancy with steadily incidence and death rates. Among unmet clinical needs proper staging, prognosis evaluation and treatment allocation are of main concern. Despite Barcelona Clinic Liver Cancer (BCLC) staging system is the most widely used according to European and American guidelines, it showed several limits, especially the strictness in treatment recommendation and the inclusion of a heterogeneous populations in stages B and C. To overcome these limitations several other staging systems have developed but many of them are not validated for clinical use. We thus aimed to evaluate prognostic ability of fifteen staging systems in a cohort of local patients in north- east of Italy.

Method:
We retrospectively evaluated a cohort of 140 patients of our Liver Clinic Unit with radiological or histological diagnoses of hepatocellular carcinoma between 2006 and 2017. Follow- up ended in 2018. Clinical and biochemical features were analyzed together with treatment allocation (i.e. curative or palliative treatment) according to commonly used BCLC staging system. Patients were than classified according to many different prognostic systems: ITA.LI.CA., BCLC, CLIP, JIS, HKLC, Tokyo score, Okuda, GRETCH, NIACE, MESH, ALBI (and derived), HAP, STATE, SNACOR, NSP. Pearson's Chi-square test and Mann-Whitney U test were used to compare discrete and continuous variables among groups, respectively. Overall survival was defined as the time from the date of diagnosis of HCC to the date of death or data censoring. Log- rank test was used to compare differences in survival and Kaplan- Meyer curves employed to estimate median survival.

Results:
Using the ITA.LI.CA prognostic system median survival was 57.9 months for stages 0-1, 43 months for stages 2-3, 21.7 months for stages 4-5 and 10, 4 months for stage > 5. Using the BCLC staging system median survival was > 81, 1 months for stage 0, 44, 9 months for stage A, 21, 3 months for stage B and 3, 1 months for stage C. The Kaplan-Meier curves are shown in Figure 1. The best prognostic performance was achieved by the ITA.LI.CA score ($P < 0.001$), followed by HKLC, GRETCH, BCLC and CLIP ($P = 0.001$); the other score systems showed less accuracy.

Conclusion:
Our evaluation showed ITA.LI.CA. as the most effective staging system in the local population. Moreover, it does not propose a rigid therapeutic algorithm opposing to BCLC system, since numerous variables can influence choice. Its reproducibility and applicability seem also good.

Figure:

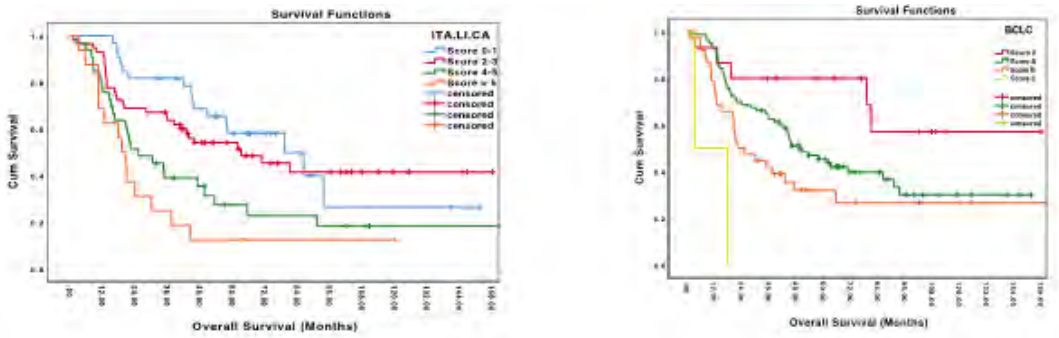


Figure 1: Kaplan- Meier curves for ITA.LI.CA prognostic score (on the left; p<0.001) and BCLC prognostic score (on the right; p=0.001).

P010 A real-world evaluation of baseline characteristics, treatment patterns, and survival outcomes in patients with advanced hepatocellular carcinoma and elevated baseline alpha-fetoprotein levels receiving second-line therapy in the United States

Allicia C. Girvan[1], Yongmei Chen[1], Zhanglin Cui[1], Elena Gugel[1], Kelly Rae KIrsch[1], Kristin M. Sheffield[1]

[1]Eli Lilly and Company Corporate Center, Indianapolis, United States
Email: girvan_allicia_c@lilly.com

Background and Aims: Elevated serum alpha-fetoprotein levels (AFP) of ≥ 400 ng/mL predict poor outcomes in patients with advanced hepatocellular carcinoma (HCC). This real-world study evaluated patient demographics and clinical characteristics, treatment patterns, and survival outcomes by baseline serum AFP levels measured at or close to second-line systemic therapy (2L) initiation in patients with advanced HCC in the United States (US).

Method: This retrospective observational study used electronic health records from the ConcertAI database, comprising de-identified patient data from outpatient oncology practices in the US. Adult patients with advanced HCC who initiated 2L between April 2017 and January 2020 were identified from this database. The AFP cohort was categorized by baseline serum AFP levels into < 400 ng/mL and ≥ 400 ng/mL AFP groups. Demographics, clinical characteristics, and treatment patterns were summarized using descriptive statistics. Overall survival (OS) from initiation of 2L to death was estimated by the Kaplan-Meier method for overall study cohort and by AFP group.

Results: Of the 586 patients with advanced HCC identified in the ConcertAI database, only 106 (18%) had data on baseline serum AFP. Median age of the AFP cohort was 64 years; 90% were males. Approximately 51% (n = 54/106) had Eastern Cooperative Oncology Group (ECOG) performance status 0/1 and 59% (n = 63/106) had albumin-bilirubin (ALBI) score 2. Of the 59 patients with documented HCC Staging data at baseline, 88% (n = 52/59) had HCC Stage IIIB–IV. Overall, the most frequently prescribed agents in 2L were nivolumab (n = 29/106, 27%), sorafenib (n = 9/106, 9%), and lenvatinib (n = 5/106, 5%). Approximately 45% (n = 48/106) had ≥ 400 ng/mL AFP and 55% (n = 58/106) had < 400 ng/mL AFP. The mean (SD) age was 63 (6.7) years for the ≥ 400 ng/mL AFP group and 66 (10.2) years for the < 400 ng/mL AFP group. Approximately 69% (33/48) patients with ≥ 400 ng/mL AFP received active therapy in 2L; 31% (15/48) were on supportive care only. For patients with < 400 ng/mL AFP, 55% (32/58) received active therapy in 2L; 45% (26/58) were on supportive care only. Median unadjusted OS of patients with ≥ 400 ng/mL AFP was 5.2 months (95% confidence interval [CI]: 3.2–10.0) and that of patients with < 400 ng/mL AFP was 8.0 months (95% CI: 6.3–16.2).

Conclusion: This study provides real-world evidence of poor survival prognosis in terms of OS for patients with advanced HCC and elevated baseline serum AFP levels. Despite evidence for AFP as a useful prognostic biomarker in HCC, our study found low rates of testing for AFP in outpatient oncology practices in the US.

P011YI Increased platelet aggregation in patients with cirrhosis and hepatocellular carcinoma: a new potential therapeutic target?

Alberto Zanetto[1 2], Elena Campello[3], Sarah Shalaby[1 2], Martina Gambato[2], Alessandro Vitale[4], Umberto Cillo[4], Fabio Farinati[1], Francesco Paolo Russo[1 2], Patrizia Burra[1 2], Paolo Simioni[3], Marco Senzolo[1 2]

[1]Gastroenterology, Department of Surgery, Oncology, and Gastroenterology, Padova University Hospital, Padova, Italy, Italy, *[2]Multivisceral Transplant Unit, Department of Surgery, Oncology, and Gastroenterology, Padova University Hospital, Padova, Italy, Italy*, *[3]Thrombotic and Hemorrhagic Diseases Unit, General Internal Medicine, Padova University Hospital, Padova, Italy, Italy*, *[4]Hepatobiliary Surgery and Liver Transplantation Center, Department of Surgery, Oncology, and Gastroenterology, Padova University Hospital, Padova, Italy, Italy*
Email: alberto.zanetto@yahoo.it

Background and Aims: Hyper-functional platelets are increasingly recognized as important players in cancer progression and metastasis and have been proposed as potential therapeutic target in multiple types of cancers. Whether this could be considered in patients with cirrhosis and hepatocellular carcinoma (HCC) is currently unknown as platelet function in these patients has not yet been investigated. Therefore, we evaluated platelet function in patients with cirrhosis and HCC.

Method: Patients with cirrhosis with and without HCC were prospectively recruited over a 6 months period. Platelet aggregation, a marker of platelet function, was assessed by impedance whole blood aggregometry with adenosine diphosphate (ADP), arachidonic acid (ASPI), and thrombin receptor agonist peptide (TRAP) stimulation.

Results: One-hundred patients with cirrhosis were recruited (50 with and 50 without HCC). As shown in the table, severity of cirrhosis and platelet count were comparable between the groups. Patients with HCC demonstrated higher ADP-, ASPI-, and TRAP- induced platelet-aggregation, all indicative of platelet hyper-function. Remarkably, HCC-driven platelet hyper-function was confirmed after adjusting the analysis for severity of cirrhosis and thrombocytopenia.

Conclusion: In patients with cirrhosis, HCC is associated with a significantly increased platelet aggregation. Further studies are required to evaluate whether inhibition of hyper-functional platelets can mitigate HCC-related morbidity and mortality in patients with cirrhosis.

Figure:

	HCC (n=50)	No HCC (n=50)	
Age, years	65 (58-69)	61 (55-71)	
Male gender, %	80	66	
Child class A/B/C, %	46/36/18	58/26/16	
MELD	11 (8-16)	10 (8-14)	
Platelet count*, x10^9/L	90 (64-124)	110 (67-140)	
Alpha-fetoprotein, ng/mL	9 (4-47)	3 (2-4)	
History of previous treatment(s) for HCC, %	45	-	
Multinodular, %	68	-	
Total tumor volume, cm^3	9 (5-16)	-	
BCLC staging 0/A/B/C/D, %	10/19/57/8/6	-	
			p value
Platelet aggregation, AUC			
ADP	45 (35-68)	28 (18-43)	<0.001
ASPI	47 (29-62)	28 (18-45)	<0.001
TRAP	85 (66-121)	75 (52-94)	0.01

Median values reported with 25th and 75th percentile values in parenthesis.
Abbreviations: MELD: Model for End-Stage Liver Disease; HCC: hepatocellular carcinoma; BCLC: Barcelona Clinic Liver Cancer; AUC: area under curve.
*When patients with and without HCC were matched according to Child class, those with HCC demonstrated a significantly higher platelet count in Child A and B but not in Child C class.

P012 Comparison of Sorafenib and Lenvatinib as first line therapy in Unresectable Hepatocarcinoma: a multi-center propensity score matching analysis

Margherita Rimini[1], Toshifumi Tada[2], Sara Lonardi[3], Takashi Kumada[4], Caterina Vivaldi[5], Atsushi Hiraoka[6], Kojiro Michitaka[6], Masanori Atsukawa[7], Masashi Hirooka[8], Kunihiko Tsuji[9], Toru Ishikawa[10], Koichi Takaguchi[11], Kazuya Kariyama[12], Ei Itobayashi[13], Kazuto Tajiri[14], Noritomo Shimada[15], Hiroshi Shibata[16], Hironori Ochi[17], Satoshi Yasuda[18], Stefano Cascinu[19], Andrea Casadei-Gardini[19]

[1]Policlinico of Modena, Department of Oncology, Modena, Italy, *[2]Japanese Red Cross Society Himeji Hospital, Department of Internal Medicine, Himeji, Japan*, *[3]Veneto Institute of Oncology, IOV-IRCCS, Department of Oncology, Padua, Italy*, *[4]Gifu Kyoritsu University, Faculty of Nursing, Ogaki, Japan*, *[5]University of Pisa, Department of Translational Research and of New Surgical and Medical Technologies, Pisa, Italy*, *[6]Ehime Prefectural Central Hospital, Gastroenterology Center, Matsuyama, Japan*, *[7]Nippon Medical School, Division of Gastroenterology and Hepatology, Departmente of Internal Medicine, Tokyo, Japan*, *[8]Ehime University Graduate School of Medicine, Department of Gastroenterology and Metabology, Matsuyama, Japan*, *[9]Teine Keijinkai Hospital, Center of Gasteoenterology, Sapporo, Japan*, *[10]Seiseikai Niigata Hospital, Department of Gastroenterology, Niigata, Japan*, *[11]Kagawa Prefectural Central Hospital, Department of Hepatology, Takamatsu, Japan*, *[12]Okayama City Hospital, Department of Gastroenterology, Okayama, Japan*, *[13]Asahi General Hospital, Department of Gastroenterology, Asahi, Japan*, *[14]Toyama University Hospital, Department of Gastroenterology, Toyama, Japan*, *[15]Otakanomori Hospital, Division of Gastroenterology and Hepatology, Kashiwa, Japan*, *[16]Tokushima Prefectural Central Hospital, Department of Gastroenterology, Tokushima, Japan*, *[17]Matsuyama Red Cross Hospital, Hepato-biliary Center, Matsuyama, Japan*, *[18]Ogaki Municipal Hospital, Department of Gastroenterology and Hepatology, Ogaki, Japan*, *[19]Universitá Vita-Salute, IRCCS-San Raffaele Hospital, Department of Oncology, Milan, Italy*
Email: margherita.rimini@gmail.com

Background and Aims: Despite the therapeutic advances reached in the last years, Hepatocarcinoma (HCC) remains an important health problem world-wide. Sorafenib was the first systemic therapy to be approved for the treatment of unresectable HCC. Recently, the REFLECT trial demonstrated the non-inferiority of Lenvatinib compared to Sorafenib, thus leading to the approval of new first-line standard of care, along with Sorafenib.

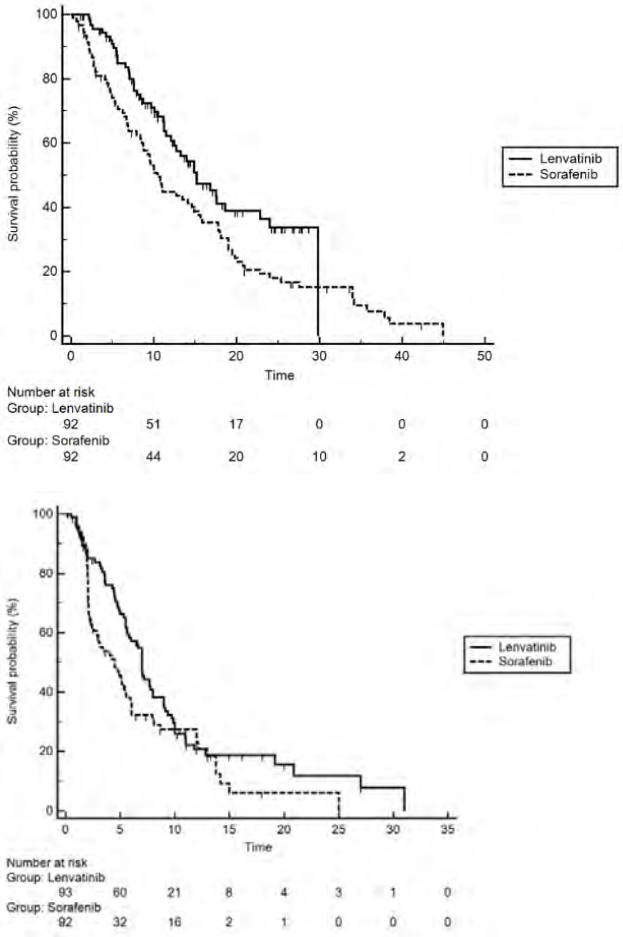
Method: With the aim to evaluate the optimal choice between Sorafenib and Lenvatinib as primary treatment in clinical practice, we performed a multicentric analysis with the propensity score matching on 184 HCC patients.

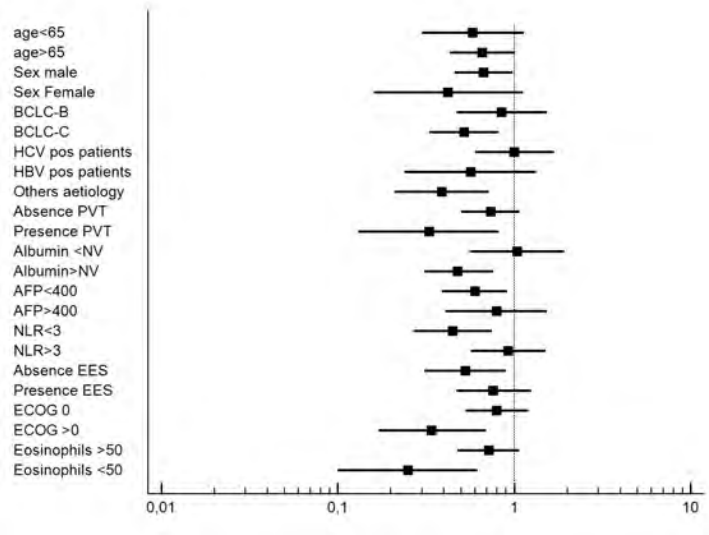
Results: 92 patients were treated with Lenvatinib and 92 patients were treated with Sorafenib. Our analysis showed median OS of 15.2 months for patients receiving Lenvatinib, and 10.5 months for patients treated with Sorafenib; a 36% reduction of death risk for patients receiving Lenvatinib versus Sorafenib was highlighted (95%CI: 0.45-0.91; p = 0.0156). Median PFS was 7.0 months for patients receiving Lenvatinib, and 4.5 months for patients treated with Sorafenib; a 29% reduction of progression risk for patients receiving Lenvatinib versus Sorafenib was highlighted (95%CI: 0.50-0.98; p = 0.0446). Patients treated with Lenvatinib showed a higher percentage of response rate (p<0.00001) and disease control rate (p=0.002) compared to patients treated with Sorafenib. Concerning the safety profile, 96.4% and 94.6% experienced at least one AEs in Lenvatinib and Sorafenib arm, respectively. Sorafenib treatment showed to be correlated with more HFS. Conversely, Lenvatinib showed to be correlated with more hypertension and fatigue. Moreover, we highlighted the prognostic role of BCLC stage, ECOG-PS, bilirubin, alkaline phosphatase and eosinophils count for patients receiving Sorafenib. Conversely, albumin, AST, alkaline phosphatase and NLR turned out to be prognostic in patients receiving Lenvatinib. Finally, by performing an

Interaction test analysis, we highlighted the positive predictive role of albumin >NV, ECOG>0, NLR<3, absence of HCV positivity, and presence of portal vein thrombosis in favour of Lenvatinib arm. On the other hand, eosinophil <50 and ECOG >0 have been showed to negatively predict the response to Sorafenib.

Conclusion: In conclusion, our analysis demonstrated the superiority of Lenvatinib over Sorafenib in a real-word setting. More researches are needed to validate the predictor factor of response to Lenvatinib rather than Sorafenib.

Figure:





Better Lenvatinib Better Sorafenib

P013YI chronic hepatitis b under tenofovir or entecavir: performance of the page-b score to predict hepatocellular **carcinoma**

Daniela Falcão[1], teresa moreira[1], joana silva[1], marta rocha[1], Luís Maia[1], Jose Manuel Ferreira[1], Isabel Pedroto[1]

[1]*centro hospitalar universitario do porto, Portugal*
Email: danielateixeirafalcao@gmail.com

Background and Aims: Long-term viral suppression and biochemical response in chronic hepatitis B (CHB) patients under nucleos(t)ide analogues (NUCs) have been linked with reduced risk of hepatocellular carcinoma (HCC). However, the likelihood of developing HCC remains uncertain and Page-B represents a promising score for the prediction of HCC. We aim to evaluate the performance of Page-B applied in a CHB population treated with entecavir (ETV) or tenofovir (TDF).

Method: Retrospective analysis of 215 patients with CHB who initiated ETV or TDF between January 2008 and June 2019.

Results: A total of 215 patients were included, most were male (67.4%), mean age of 48 years, 18% HBeAg-positive CHB, 18.5% with cirrhosis and 68% were treatment-naïve. TDF was selected in 72.5% versus ETV in 27.5%. The virologic response rate (HBV DNA level <10 IU/mL) was 86% and HBsAg-loss occurred in 6 patients (3.4%) with antiHBs-seroconversion in three patients (1,7%), after a median follow-up of 79 months, without significant differences between the two groups.
Three patients had HCC diagnosis during follow-up, cumulative-incidence of 1,7%, all male, with cirrhosis, platelets <200000/ul and two patients were HBeAg-positive. The Page-B score ≥10, presented an accuracy of 32% (95% CI, 25-40%), with a sensitivity and negative predict-value of 100%, specificity 31% and positive predict-value 2.5%. In the ROC curve analysis, the AUC of Page-B score in predicting HCC was 0.70.

Conclusion: This study confirms a persistent risk of HCC in CHB patients under NUCs regardless viral suppression and biochemical response. Our analysis showed the excellent negative predictive value of Page-B score ≥10, as previously reported, with potential implications in HCC surveillance-strategy.

P014YI Diagnosing hepatocellular carcinoma – are we ‘getting it right first time’?

Emma Saunsbury[1], elinor littlewood[1], coral hollywood[1]

[1]Gloucestershire Royal Hospital, United Kingdom
Email: emma.saunsbury@nhs.net

Background and Aims: Getting It Right First Time (GIRFT) is a national initiative designed to improve the quality of National Health Service care. A GIRFT review of hepatology services at Gloucestershire Hospitals NHS Foundation Trust highlighted a low hepatocellular carcinoma (HCC) pick up rate for our proposed population and disease burden, with higher than average metastasis outcome. This study investigated why this might be the case in order to improve our patient journey.

Method: This retrospective cohort study identified all patients diagnosed with ‘malignant neoplasm liver unspecified’, ‘liver cell carcinoma’ and ‘intrahepatic bile duct carcinoma’ between January 2017-October 2019. Electronic records were then interrogated to identify cases of confirmed HCC and to ascertain the date, method and setting of diagnosis. Cases were reviewed for evidence of pre-existing cirrhosis and HCC surveillance and their patient journey to date.

Results: Of the initial 71 patients, 42 were excluded due to non-HCC diagnoses. Of the 29 patients with confirmed HCC, only 13.8% were known to have cirrhosis prior to diagnosis (n = 4), though a further 17.2% (n = 5) were alcohol dependent. 75% (n = 3) of the cirrhotic patients were not under HCC surveillance. 52% (n = 15) of HCC patients were diagnosed from primary care investigations, 20.6% (n = 6) via outpatient clinics and 27.6% (n = 8) whilst inpatients. At the time of data collection, 65% (n=19) of patients had died, a mean of 35.4 weeks from diagnosis.

Conclusion: GIRFT has been a useful tool in highlighting gaps in our hepatology service, though not the cause. 86.2% (n = 25) of patients diagnosed with HCC in this cohort were not known to have established cirrhosis at the time of diagnosis. This may reflect issues in the data collection process, such as inadequate coding, but potentially a lack of adequate screening for cirrhosis as a precursor for HCC. Of concern, in those found to have cirrhosis only 25% were enrolled in a surveillance programme at the time of HCC diagnosis. The average time from diagnosis to death of 9 months highlights that diagnosis is often at a late stage. More work needs to be done to realise the gaps in our patient pathway. Surveillance of high-risk patients is firstly a priority, perhaps with a focus on overcoming the practical challenges of awareness, engagement and attendance in this cohort.

Figure:

P015 Modeling hepatocellular carcinoma cells dynamics by serological and imaging biomarkers in patients with response to sorafenib and/or regorafenib

Piero Colombatto[1], Coskun Ozer Demirtas[2], Gabriele Ricco[3], Luigi Civitano[1], Piero Boraschi[4], Paola Scalise[4], Daniela Cavallone[3], Filippo Oliveri[1], Veronica Romagnoli[1], Patrizia Bleve[1], Barbara Coco[1], Antonio Salvati[1], Lucio Urbani[5], Ferruccio Bonino[6], Maurizia Brunetto[7]

[1]University Hospital of Pisa, Hepatology Unit, Pisa, Italy, *[2]Marmara University, Gastroenterology Department, Istanbul, Turkey*, *[3]University Hospital of Pisa, Hepatology Unit and Department of Clinical and Experimental Medicine, Pisa, Italy*, *[4]University Hospital of Pisa, Radiodiagnostic Unit, Pisa, Italy*, *[5]University Hospital of Pisa, General Surgery Unit, Pisa, Italy*, *[6]National Research Council of Italy, Biostructure and Bio-imaging Institute , Italy*, *[7]University Hospital of Pisa, Hepatology Unit and Department of Clinical and Experimental Medicine, Biostructure and Bio-imaging Institute of National Research Council of Italy, Pisa, Italy*
Email: p.colombatto@ao-pisa.toscana.it

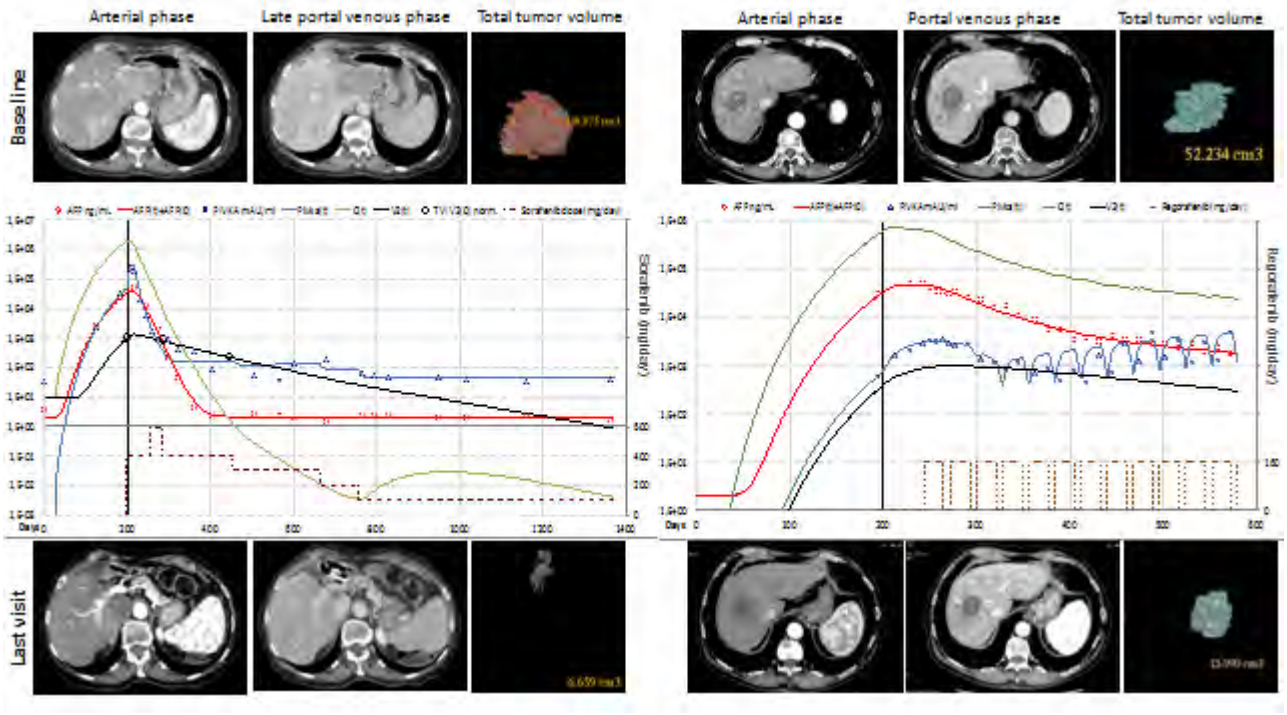
Background and Aims: In advanced HCC the overall benefits of multi-kinase inhibitors are limited, but some patients show partial response (PR) and a few achieve complete response (CR). Bio-mathematical modeling of in-vivo cancer cell dynamics by radiomics combining digital imaging and serological biomarkers would provide a better understanding of PR/CR. Aim of the study was to investigate HCC dynamics by combining data of circulating HCC biomarkers (α-fetoprotein, AFP and protein induced by vitamin K absence-II, PIVKA-II) and digital imaging in patients with CR/PR to sorafenib or regorafenib.

Method: A physic-mathematical model was set up using ordinary differential equations to describe cancer cells and vasculature dynamics in 3 patients. Case-1: 78-year-old woman with long lasting (5-years) CR to sorafenib. Case-2: 72-year-old man with PR to sorafenib. Case-3: 70-year-old with PR to regorafenib. Tumor mass and vascularization at baseline and their changes during therapy were assessed by digital imaging. AFP and PIVKA-II kinetics were fitted into the model to compute cancer cells and vascularization dynamics and drug effectiveness (Figure).

Results: The mean cancer cell life-time was about 9 days in all patients and the daily production rate was 0.36 in CR and 0.30-0.32 in PRs respectively. In the patient with CR, sorafenib lowered neo-angiogenesis to 8% and cancer cells proliferation to 37.5% of baseline; CR was explained by an additional effect of the drug on tumor vasculature decay, which became relevant when the doses of sorafenib were lowered because of side effects. In PRs anti-angiogenesis and anti-replicative activities were 4.4-3.7 and 2.0-14.3 fold lower than in CR. AFP kinetics followed that of cancer cells, whereas PIVKA-II kinetics showed time and dose dependent fluctuations caused by ischemia on both cancer cells and normal hepatocytes.

Conclusion: Pending confirmation in larger HCC series modeling the kinetics of serum AFP and PIVKA-II with standardized digital imaging features may provide a more accurate study of the response to systemic therapies and help personalization of clinical decision-making.

Figure: Model computed Cancer cells [C(t)] and Vascularitazion [V2(t)] dynamics with best fitting of AFP and PIVKA-II levels.



P016YI Profiling of cell-free DNA using Whole-exome sequencing in patients with Hepatocellular carcinoma in Thailand

Pattapon Kunadirek[1], Natthaya Chuaypen[1], Thidathip Wongsurawat[2], Piroon Jenjaroenpun[2], Intawat Nookeaw[2], Pisit Tangkijvanich[1]

[1]Center of Excellence in Hepatitis and Liver Cancer, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand, *[2]Department of Biomedical Informatics, College of Medicine, University of Arkansas for Medical Sciences, Little rock, Arkansas, United States*
Email: pkunadirek@gmail.com

Background and Aims: Hepatocellular carcinoma (HCC) was highly observed in Thailand with a high mortality rate globally. Cell-free DNA (cfDNA) is a liquid biopsy with minimally invasive and providing of tumor genetic profile. We evaluate the potential of cfDNA as a biomarker in Thai HCC patients.

Method: Paired of cfDNA and germline DNA were isolated from patients with HCC (n = 60) and chronic hepatitis (CH) (n = 17). Levels of cfDNA were quantified using Qubit fluorometry. Whole-exome sequencing (WES) was performed to investigate mutation profile of cfDNA. The mutation profile of cfDNA was compared with Thai HCC tissues and TCGA data.

Results: The level of cfDNA was significantly higher in HCC group compared with CH group. The combined of serum alpha-fetoprotein and cfDNA present high sensitivity (88.46%) and specificity (100%). All patients had identified 2,732 altered genes with a median of 49.5 variants per sample (3 - 818). The altered genes in cfDNA present a concordance with Thai HCC tissues (31%). The high mutated genes in cfDNA (ZNF814, ZNF492, HRNR, TP53, OBSCN, TTN, ADAMTS12, FLG) were covered 62% in TCGA. Additionally, co-occurrence of HRNR and TTN in cfDNA was found that HCC patients with these mutations in TCGA data were associated with shorter survival time compared with non-mutation.

Conclusion: Our study represents the first analysis of cfDNA from HCC in Thailand. The mutations of cfDNA were detected in all patients using WES. In addition, a high mutation gene could be diagnosis and prognosis in cfDNA of patients with HCC in Thailand.

P017YI Transcriptomic analyses reveal cancer-induce genes in PBMCs of Hepatocellular carcinoma for early diagnosis and prognosis

Pattapon Kunadirek[1], Natthaya Chuaypen[1], Nakarin Kitkumthorn[2], Intawat Nookeaw[3], Pisit Tangkijvanich[1]

[1]Center of Excellence in Hepatitis and Liver Cancer, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand, *[2]Department of Oral Biology, Faculty of Dentistry, Mahidol University, Bangkok, Thailand*, *[3]Department of Biomedical Informatics, College of Medicine, University of Arkansas for Medical Sciences, Little rock ,Arkansas, United States*
Email: pkunadirek@gmail.com

Background and Aims: Hepatocellular carcinoma (HCC) is the most common cause of cancer related-death. Thus, novel and sensitive biomarker is still required for predicting early HCC.

Method: We performed RNA sequencing to investigate the transcriptome profile of peripheral blood mononuclear cells (PBMCs) from patients with HCC and healthy controls (HC) and also in PBMCs co-cultured with HCC cell lines. The raw data of GSE 58208 and 49519 were downloaded from the Gene Expression Omnibus (GEO) database and the differentially expressed genes (DEGs) were identified by CU-DREAM.

Results: A total of 24 DEGs were identified in PBMCs from patients with HCC compared with healthy controls and co-cultured model, including 18 upregulated and 6 downregulated DEGs. The KEGG pathway results showed that these enriched genes were mainly associated with immune response pathways. Five candidate genes (BHLHE40, AREG, SOCS1, CCL5, and DDIT4) were selected and validated in PBMCs from 100 patients with HCC, 100 patients with chronic hepatitis B and 100 HC. These genes were significantly higher in HCC group compared with other groups. BHLHE40 and DDIT4 expression had a higher sensitivity than alpha-fetoprotein (AFP) for detecting early HCC (71% and 77% vs. 31%, respectively). In addition, BHLHE40 was identified as an independent prognostic factor of overall survival in patients with HCC.

Conclusion: Our study suggested that these two DEGs might be promising diagnostic and prognostic biomarkers for early stage of HCC.

P018YI Association between postoperative early recurrence of hepatocellular carcinoma and the expression pattern of circulating tumor cells in peripheral blood sample: a preliminary study.

Simone Famularo[1 2], Matteo Nazzareno Barbaglia[3], Livia Salmi[3], Cristina Ciulli[2 4], Francesca Carissimi[2 4], Linda Roccamatisi[2 4], Giuseppe Cordaro[2 4], Venkata Ramana Mallela[3], Luciano De Carlis[2 5], Andrea Lauterio[5], Rosalba Minisini[3], Biagio Eugenio Leone[2 6], Fabrizio Romano[2 4], Mario Pirisi[3 7 8]

[1]Humanitas University, Humanitas Clinical and Research Center, Department of Hepatobiliary and General Surgery, Italy, *[2]University of Milan-Bicocca, School of Medicine and Surgery, Italy*, *[3]Università del Piemonte Orientale, Department of Translational Medicine, Italy*, *[4]San Gerardo Hospital, Hepatobiliary Surgery Unit, Monza, Italy*, *[5]ASST Grande Ospedale Metropolitano Niguarda, , Department of General Surgery and Transplantation, Milan, Italy*, *[6]San Gerardo Hospital, Unit of Pathology, Monza, Italy*, *[7]IRCAD, Interdisciplinary Research Center of Autoimmune Diseases, novara, Italy*, *[8]"AOU Maggiore della Carità", Division of Internal Medicine, novara, Italy*
Email: simone.famularo@gmail.com

Association between postoperative early recurrence of hepatocellular carcinoma and the expression pattern of circulating tumor cells in peripheral blood sample: a preliminary study.

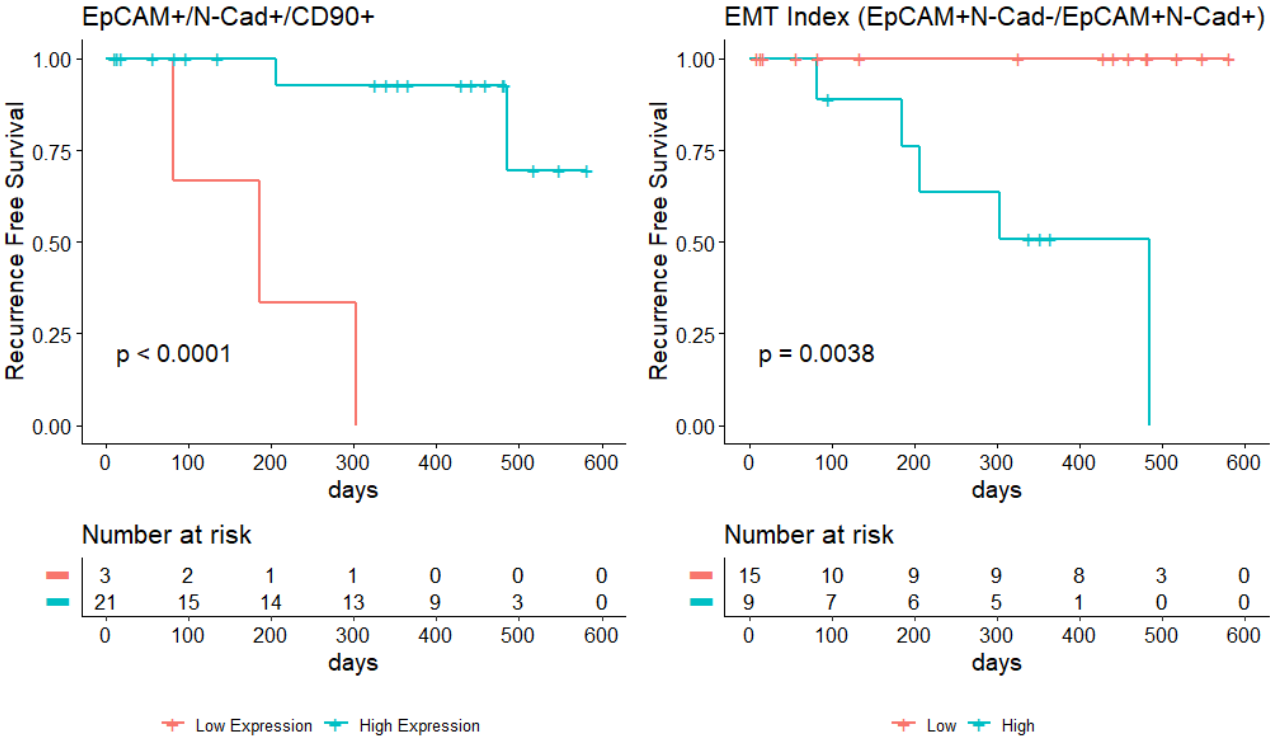
Background and Aims: Hepatocellular carcinoma (HCC) recurs in up to 60% of patients who undergo resection. Circulating tumor cells (CTC) have been advocated as promotors of the recurrence. However, their role as prognostic markers in the surgical setting is unclear. The aim of the present study has been to assess the association between CTC from peripheral blood samples and the risk of recurrence after surgery.

Method: Patients with a first diagnosis of HCC, no previous treatment for this condition, no other oncological history, and BCLC stage 0-A-B were prospectively enrolled in 2 centers. Patients were submitted to serial liquid biopsies (i.e., a 15ml peripheral blood sample on each time point) at day 0-30-90-180-365. After isolation of peripheral blood mononucleate cells, CTC were detected by FACSymphony™ and subsequent identification of the following markers: EpCAM, N-cadherin (N-cad) and CD90. Epithelial-mesenchymal transition (EMT) was analyzed by an index estimated as the ratio between the number of $EpCAM^+/N\text{-}cad^-$ and $EpCAM^+/N\text{-}cad^+$ cells (EMT Index). Patients were divided according to the recurrence status.

Results: Between 2019 and 2020, 24 patients were enrolled. The median follow-up was 365 days [95% CI: 135-481], during which HCC recurrence occurred in 5 patients (18.5%). The expressions of $EpCAM^+/N\text{-}cad^+$ and $EpCAM^+/N\text{-}cad^+/CD90^+$ cells were numerically higher in the no-relapse group at each timepoint. EMT index was significantly lower (0.10, IQR 0.06-1.33) in the no-relapse group than in the recurrence one (0.55, IQR 0.44-0.71, p=0.017) before surgery, with similar results over time. By calculating receiver-operating characteristics (ROC) areas under the curve (AUC), optimal cut-offs were established for $EpCAM^+/N\text{-}cad^+/CD90^+$ cells (AUC 0.66, accuracy 91.6%, negative-predictive value (NPV) 90.4%, positive-predictive value (PVP) 100%) and for EMT Index (AUC 0.85, accuracy 83.3%, NPV 100%, PPV 55.5%). Patients with higher expression of triple positive cells did not reach the median recurrence-free-survival (RFS), while lower expression cases had median RFS of 185 days (95% CI: 82-NA, p<0.0001). Patients with higher EMT Index before surgery had 1-year RFS of 50.8% versus 100% for lower index (p=0.0038).

Conclusion: Patients with lower expression of triple positive cells or with an increased EMT index before surgery have a higher rate of recurrence. Thus, these tests are promising candidates as useful biomarkers in HCC monitoring after surgery.

Figure:



P019YI Circulating prostaglandin E2: a novel potential prognostic biomarker in patients with hepatocellular carcinoma

Filippo Pelizzaro[1], Maria Piera Kitenge[1], Romilda Cardin[1], Barbara Penzo[1], Alberto Ponzoni[2], Umberto Cillo[3], Alessandro Vitale[3], Gianluca Businello[4], Giada Munari[4], Matteo Fassan[4], Fabio Farinati[1]

[1]University of Padua, Department of Surgery, Oncology and Gastroenterology, Gastroenterology Unit, Padova, Italy, *[2]Azienda-Ospedale Università di Padova, Radiology Unit, Padova, Italy*, *[3]University of Padua, Department of Surgery, Oncology and Gastroenterology, Hepatobiliary Surgery and Liver Transplantation Unit, Padova, Italy*, *[4]University of Padua, Department of Medicine (DIMED), Surgical Pathology and Cytopathology Unit, Padova, Italy*
Email: filippo.pelizzaro@gmail.com

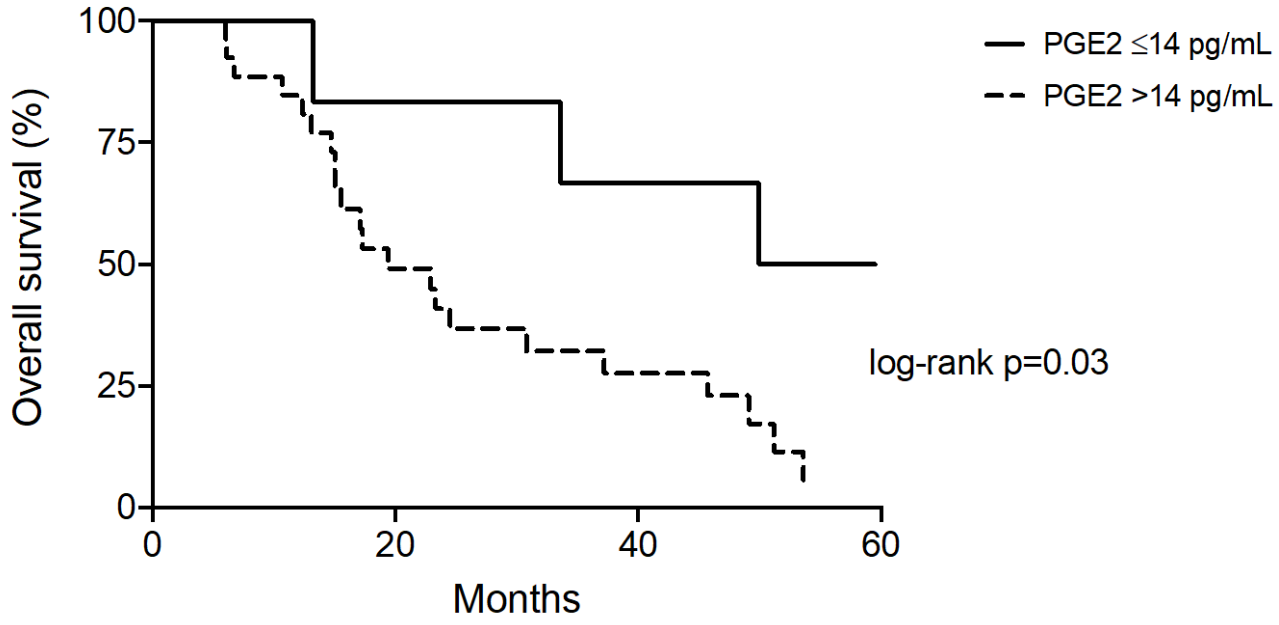
Background and Aims: Hepatocellular carcinoma (HCC) develops almost invariably on a chronic liver disease characterized by persistent inflammation and several evidences suggest that prostaglandins are involved in tumor growth. Monoacylglycerol lipase (MAGL), Cyclooxygenase-2 (COX-2) and Prostaglandin E_2 (PGE2) proved to promote tumor growth, inhibit apoptosis and enhance cell proliferation and invasion. We aimed to explore the activation of MAGL/COX-2/PGE2 axis in HCC, evaluating circulating PGE2 as prognostic biomarker.

Method: PGE2 levels were measured in serum samples from 24 cirrhotics and 34 HCC patients consecutively collected between January 2016 and December 2017. In a subgroup of patients (10 HCC patients treated with liver resection and 10 cirrhotics who subsequently underwent liver transplantation), tissue expression of MAGL mRNA and immunohistochemistry for MAGL and COX-2 were obtained. Prognostic cut-off of circulating PGE2 was established with the ROC curve method. Kaplan-Meier method and log-rank test were used to estimate and compare overall survival curves.

Results: PGE2 circulating levels were similar in HCC and cirrhotics (30.3 [22.3-39.6] and 20.4 [12.0-47.8] pg/mL; p=0.73). In HCC patients, higher levels of the marker were found in males (p<0.0001), in patients younger than 65 years (p=0.046), in multifocal disease (p=0.002) and in BCLC stage 0/A (p=0.006). Tumor tissues showed overexpression of MAGL mRNA and higher levels of both MAGL and COX-2 at immunohistochemistry. HCC patients with circulating PGE2 levels >14 pg/mL had a significantly shorter overall survival (19.4 vs. 49.9 months; p=0.03; Figure), the finding being confirmed by the multivariate analysis (HR 3.37 [95% CI 1.00 – 11.60]; p=0.05).

Conclusion: The results of this study confirm the activation of MAGL/COX-2/PGE2 axis in HCC and show that circulating PGE2 is a promising potential prognostic biomarker. Additional data in larger cohort are needed to confirm these findings.

Figure: Kaplan-Meier curves according to the PGE2 prognostic cut-off: patients with PGE2 ≤14 pg/mL had an overall survival significantly longer than patients with marker levels above the cut-off (p=0.03).


100
75
50
25
0
Overall survival (%)
0
20
40
60
Months
PGE2 ≤14 pg/mL
PGE2 >14 pg/mL
log-rank p=0.03

P020 Establishment of 3D-organoids from cryopreserved cancer tissues

Colin Rae[1], Francesco Amato[1], Maria Giuseppina Prete[1], Vicenzo Cardinale[2], Curtis Rink[3], Peter Repiscak[3], Colin Nixon[3], Nikki Paul[1], Lynn Mcgarry[1], Sabrina D'Agosta[4], Georgios Vlachogiannis[5], Guido Carpino[6], Boulter Luke[7], Fraser Duthie[1], Nigel Jamieson[1 8], David Chang[1 8], Euan Dickson[8], Colin McKay[8], Nicola Valeri[5 9], Vicenzo Corbo[4], Owen Sansom[1 3], Jen Morton[1 3], Chiara Braconi[1 10]

[1]University of Glasgow, Institute of Cancer Sciences, *[2]Sapienza University of Rome*, *[3]Beatson Institute for Cancer Research*, *[4]University of Verona*, *[5]Institute of Cancer Research, London*, *[6]University Foro Italico*, *[7]University of Edinburgh*, *[8]West of Scotland Pancreatic Unit*, *[9]Imperial College London*, *[10]Beatson West of Scotland Cancer Centre*
Email: colin.rae@glasgow.ac.uk

Background and aims: Cancer organoids are 3D-replicas of human tumours which might potentially be used to deliver precision oncology. However, the need for processing fresh tissues in a timely manner and in a highly specialized centre makes the testing of this technology within clinical trials challenging. Cryopreservation of tissue at the time of collection would facilitate the implementation of a multi-institutional trial to test the validity of organoids in the clinical management of cholangiocarcinoma (CCA). We aimed to design a protocol which allowed establishment of organoid cultures after rapid freezing of tissue in a cryopreservation solution.

Methods: Tissue from mouse cholangiocarcinoma tumours (*Ck19-CreER*; *Kras*$^{G12D/+}$; *Pten*$^{fl/fl}$) were divided into samples to be processed immediately (fresh, F) or frozen in a defined cryopreservation solution before being thawed and processed (frozen-thawed, FT). Matched F and FT organoids were cultured for up to 100 days and the morphological and functional features of organoids compared. Validation of the cryopreservation protocol was performed in human tissues.

Results: *Ck19-CreER*; *Kras*$^{G12D/+}$; *Pten*$^{fl/fl}$ gave rise to CCA that resembled the features of human CCA and were used to mimic the challenges of CCA organoid development. Organoids from F tissues were established after 7 days of culturing and were split every 7 (mean) days. FT organoids were established at day 8; the growth rate was comparable between F and FT organoids during long-term culture (up to 100 days, passage 15). Morphology of FT organoids recapitulated that of F organoids and source tissue. Immunofluorescent staining of actin with phalloidin indicated that the structure of the organoids was the same in F and FT organoids. Expression of biliary markers (CK7, CK19, CK20, PAS, SOX9) was comparable between F and FT organoids. When FT organoids were dissociated and cryopreserved, they were able to rapidly reform into organoids. F and FT tissue from a presumed distal CCA were processed according to our protocol. Final pathology concluded for pancreatic cancer extending into the common bile duct. Establishment and growth rate of F and FT organoids were comparable.

Conclusions: Our results indicate that using this method, tissue can be cryopreserved before deriving organoid culture without any alteration in the formation, growth and morphology of organoids.

P021YI Prognostic role of platelets-to lymphocytes ratio (PLR) and neutrophils-to-lymphocytes ratio (NLR) in hepatocellular carcinoma patients

Filippo Pelizzaro[1], Anna Sartori[1], Angela Imondi[1], Barbara Penzo[1], Alessandro Vitale[2], Franco Trevisani[3], Fabio Farinati[1]

[1]University of Padua, Department of Surgery, Oncology and Gastroenterology, Gastroenterology Unit, Padova, Italy, *[2]University of Padua, Department of Surgery, Oncology and Gastroenterology, Hepatobiliary Surgery and Liver Transplantation Unit, Padova, Italy*, *[3]Alma Mater Studiorum–University of Bologna, Department of Medical and Surgical Sciences, Semeiotics Unit, Bologna, Italy*
Email: filippo.pelizzaro@gmail.com

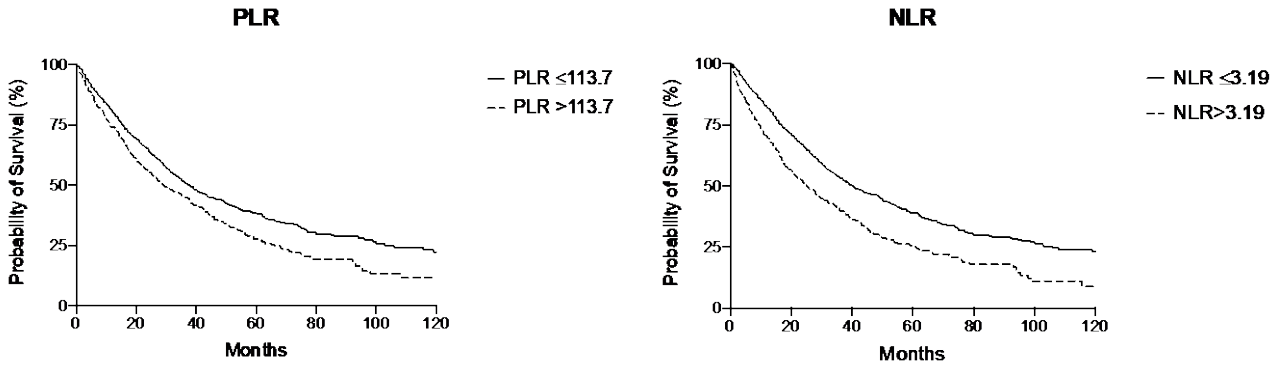
Background and Aims: Platelet-to-lymphocyte ratio (PLR) and lymphocyte-to-neutrophil ratio (NLR) have been recently proposed as biomarkers in hepatocellular carcinoma (HCC). We evaluated their prognostic role in a large cohort of patients, also according to treatment subgroups.

Method: From the Italian Liver Cancer (ITA.LI.CA) database, data of 2,513 patients with available PLR and NLR values were retrieved. Their prognostic cut-offs were established with the ROC curve method. Overall survival curves were estimated with the Kaplan-Meier method and compared with the log-rank test. Factors associated with survival and variables predictive of PLR and NLR values were identified with Cox proportional hazard and multivariable logistic regression models.

Results: A significantly longer survival was demonstrated in patients with PLR below the cut-off of 113.7 (38.0 vs. 28.0 months; p<0.0001) and NLR below the cut-off of 3.19 (40.1 vs. 25.3 months; p<0.0001) (Figure). Compared to patients with both biomarkers below their respective cut-offs (median survival 40.5 months), a shorter survival was demonstrated for those with one (31.0 months, HR=1.33, 95% CI 1.16-1.53; p<0.0001) or both markers above it (24.6 months, HR=1.69, 95% CI 1.45-1.96; p<0.0001). After adjustment for confounders, only NLR was independently associated with survival (adjusted HR=1.28, 95% CI 1.12-1.47; p=0.0005). Low levels of PLR were associated with better prognosis in patients treated with liver transplantation, ablation, intra-arterial and systemic therapies, while low levels of NLR predicted longer survival after ablation, intra-arterial therapies, sorafenib and best-supportive care. Independent predictors of high PLR values were advanced age, non-cirrhotic liver, presence of portal hypertension, increasing tumor size and presence of metastases, while high NLR values were predicted by number of liver lesions, presence of ascites and increasing MELD score.

Conclusion: PLR and NLR confirmed to be promising prognostic biomarkers, even though their predictive capacity in curative treatment settings appears to be lower in this study compared to previous literature.

Figure: Kaplan-Meier curves for overall survival according to PLR and NLR values (p<0.0001 in both cases).



P022YI Impact of hospital volume on hepatocellular carcinoma survival in Italy

Filippo Pelizzaro[1], Angela Imondi[1], Anna Sartori[1], Barbara Penzo[1], Alessandro Vitale[2], Franco Trevisani[3], Fabio Farinati[1]

[1]University of Padua, Department of Surgery, Oncology and Gastroenterology, Gastroenterology Unit, Padova, Italy, [2]University of Padua, Department of Surgery, Oncology and Gastroenterology, Hepatobiliary Surgery and Liver Transplantation Unit, Padova, Italy, [3]Alma Mater Studiorum–University of Bologna, Department of Medical and Surgical Sciences, Semeiotics Unit, Bologna, Italy
Email: filippo.pelizzaro@gmail.com

Background and Aims: Scanty data are available about the impact of hospital volume on the survival of hepatocellular carcinoma (HCC) patients. We evaluated the effect of hospital case volume on HCC patient's prognosis among the Italian Liver Cancer (ITA.LI.CA) Institutions.

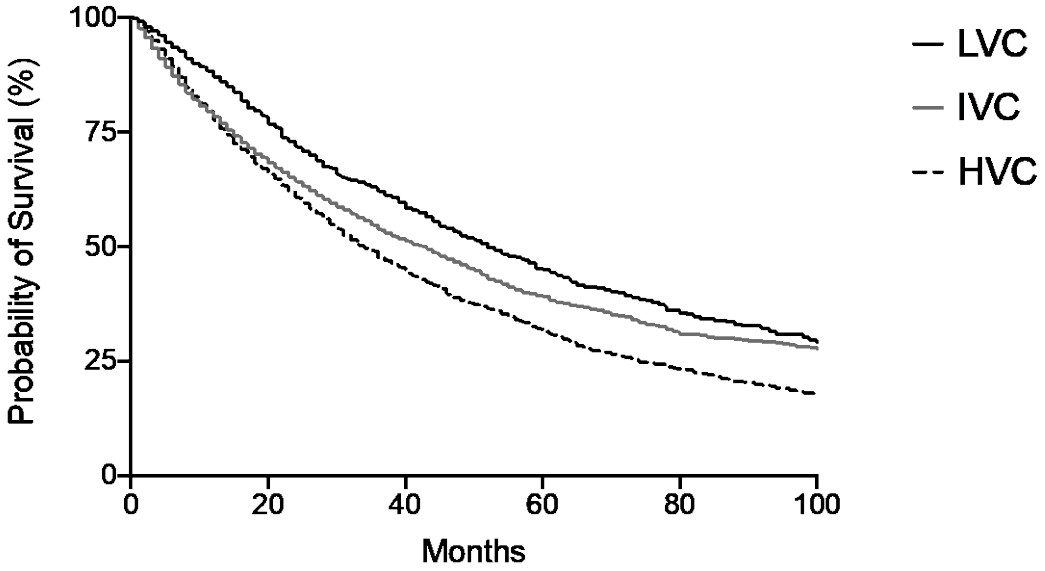
Method: From the ITA.LI.CA database, 6,704 patients diagnosed with HCC between January 2000 and December 2018 were included. After blinding, the 23 ITA.LI.CA Institutions, each of which register a median of 16 (range 6-44) newly diagnosed patients/year, were divided in: low-volume centers (LVC, ≤13 patients/year), intermediate-volume centers (IVC, 14-26 patients/year) and high-volume centers (HVC, >26 patients/year). Overall survival (OS) curves were estimated and compared with Kaplan-Meier method and log-rank test. Cox proportional hazard model was used to identify factors associated with OS.

Results: LVC, compared to IVC and HVC, showed a higher proportion of early stages tumors (p<0.0001) and a wider application of curative treatments (p<0.0001). Moreover, LVC patients were mostly Child-Pugh A (p<0.0001) and showed lower MELD score (p=0.01). The median OS of LVC patients (52.0 months, 95% CI 46.6-57.4) was higher compared to IVC and HVC patients (43.0 months, 95% CI 40.0-56.0, and 34.0 months, 95% CI 32.0-36.0, respectively; p<0.0001) (Figure). In three different time cohorts (2000-2006, 2007-2012, 2013-2018), these results were confirmed. After adjustment for several confounders among which period of diagnosis, liver function, cancer stage and treatment, hospital volume confirmed to be associated with survival (compared to LVC, the adjusted HR for mortality was 1.14, 95% CI 1.03-1.26, in IVC and 1.31, 95% CI 1.19-1.45, in HVC).

Conclusion: The unexpected inverse association between hospital volume and HCC patient survival probably depends on better liver function, earlier cancer stage and wider use of curative treatments in LVC. Overall, these findings suggest the referral of more complex and advanced stage patients to higher volume hospitals, with a "hub-to-spoke" migration.

Figure: Kaplan-Meier curves for overall survival in LVC, IVC and HVC (median survival 52.0 [95% CI 46.6-57.4], 43.0 [95% CI 40.0-56.0] and 34.0 [95% CI 32.0-36.0] months, respectively; p<0.0001).

Volume


Probability of Survival (%)
100
75
50
25
0
0
20
40
60
80
100
Months
LVC
IVC
HVC

P023YI Sarcomatoid hepatocellular carcinoma has distinct immunologic hallmarks from ordinary hepatocellular carcinoma

Ryo Morisue[1 2], Motohiro Kojima[2], Toshihiro Suzuki[3 4], Tetsuya Nakatsura[3], Motokazu Sugimoto[1], Shin Kobayashi[1], Shinichiro Takahashi[1], Naoto Gotohda[1], Atsushi Ochiai[5]

[1]National Cancer Center Hospital East, Hepatobiliary and Pancreatic Surgery, Kashiwa, Japan, *[2]Exploratory Oncology Research & Clinical Trial Center, National Cancer Center, Pathology, Kashiwa, Japan*, *[3]Exploratory Oncology Research & Clinical Trial Center, National Cancer Center, Cancer Immunotherapy, Kashiwa, Japan*, *[4]General Medical Education and Research Center, Teikyo University, Tokyo, Japan*, *[5]Exploratory Oncology Research & Clinical Trial Center, National Cancer Center, Kashiwa, Japan*
Email: rmorisue@east.ncc.go.jp

Background and Aims:
Recently, carcinomas with sarcomatoid or undifferentiated histology reportedly have had characteristic immunologic hallmarks such as high tumor-infiltrating lymphocytes and high programmed death-ligand 1(PD-L1) on tumor cells. We previously experienced and reported a case of sarcomatoid hepatocellular carcinoma (SHCC) with prominent PD-L1 expression; therefore, we sought to determine whether SHCCs also have distinct features of tumor immune microenvironment (TME) compared to ordinary HCCs (OHCCs).

Method:
SHCC defined as HCC with at least 10% of sarcomatous component and OHCC randomly matched (1:1) according to the disease stage were extracted from 1106 HCCs in the Pathology Database (1997–2019) of our hospital. The clinicopathological features and transcriptomic profiles of SHCC were compared with those of OHCC. TME was analyzed by the fluorescent multiplex immunohistochemistry to compare the density of intratumoral T cells, the level of programmed death-1 (PD-1) expression on T cells, and the frequency of PD-L1 positive (≥1%) tumor.

Results:
Fifteen SHCCs (1.4%) and fifteen OHCCs were identified from the Pathology Database. SHCC patients tended to show lower 5-year overall survival rate (16.1% versus 53.9%, p=0.056), and had significantly higher level of neutrophil-to-lymphocyte ratio (median [range], 3.67[1.61−12.6] versus 1.76[1.09−4.11],p=0.002) in the blood test. The transcriptome analysis revealed significant upregulation of gene expressions associated with epithelial-to-mesenchymal transition and inflammatory response in SHCCs (False discovery rate<0.05, respectively). The immunohistochemistry revealed a significantly higher frequency of PD-L1 positive tumor in SHCCs than in OHCCs (100% versus 47%, p=0.002), and moreover, significantly higher density of $CD4^+$ and $CD8^+$ T cells in SHCCs than in OHCCs (p=0.010 and p<0.001, respectively). The density of $CD4^+$ and $CD8^+$ T cells in the non-sarcomatous component of SHCCs was also significantly higher than that in OHCCs(p=0.019, respectively). The level of PD-1 expression on $CD4^+$ and $CD8^+$ T cells was not significantly different between SHCCs and OHCCs.

Conclusion:
The current study revealed SHCC had distinct immunologic hallmarks associated with high intratumoral T cells and high PD-L1 expression on tumor cells relative to OHCC.

P024 Characteristics of patients who received regorafenib for unresectable hepatocellular carcinoma in routine clinical practice: interim analysis of the prospective, observational REFINE study

Gianluca Masi[1], Masafumi Ikeda[2], Richard S. Finn[3], Philippe Merle[4], Ho Yeong Lim[5], Masatoshi Kudo[6], Heinz-Josef Klümpen[7], Catherine Frenette[8], Yoon Jun Kim[9], René Gerolami[10], Masayuki Kurosaki[11], Kazushi Numata[12], Hong Zebger-Gong[13], Sabine Fiala-Buskies[14], Kirhan Ozgurdal[15], Shukui Qin[16]

[1]Azienda Ospedaliero-Universitaria Pisana, Pisa, Italy, *[2]National Cancer Center Hospital East, Kashiwa, Japan*, *[3]David Geffen School of Medicine at UCLA, Los Angeles, United States*, *[4]Groupement Hospitalier Lyon Nord, Hepatology Unit, Lyon, France*, *[5]Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea, Rep. of South*, *[6]Kindai University Faculty of Medicine, Osaka, Japan*, *[7]Amsterdam University Medical Centers, Cancer Center Amsterdam, Amsterdam, Netherlands*, *[8]Scripps Green Hospital, La Jolla, United States*, *[9]Seoul National University Hospital, Seoul, Korea, Rep. of South*, *[10]CHU La Timone, Marseille, France*, *[11]Musashino Red Cross Hospital, Tokyo, Japan*, *[12]Yokohama City University Medical Center, Yokohama, Japan*, *[13]Bayer AG, Berlin, Germany*, *[14]Bayer AG, Wuppertal, Germany*, *[15]Bayer Consumer Care AG, Basel, Switzerland*, *[16]Chinese People's Liberation Army Cancer Center of Nanjing Bayi Hospital,, Nanjing, China*
Email: gl.masi@tin.it

Background and Aims: Regorafenib is approved for the treatment of patients with unresectable hepatocellular carcinoma (HCC) previously treated with sorafenib, based on results of the randomized, controlled phase 3 RESORCE trial (NCT01774344). REFINE is a large, prospective observational study designed to evaluate regorafenib in patients with unresectable HCC in real-world practice. We compared the baseline characteristics of real-world patients enrolled in REFINE with common eligibility criteria of large interventional trials of second- and later-line systemic treatments for HCC.

Method: REFINE (NCT03289273) includes patients with HCC for whom a decision to treat with regorafenib was made by the treating physician prior to enrollment according to the local health authority approved label. The primary aim is to assess safety; secondary endpoints include overall survival. We included patients from the first planned interim analysis, which was performed when the first 500 patients had been observed for ≥ 4 months.

Results: In most interventional studies of second- and later-line treatments of HCC, patients were excluded if they had certain baseline clinical parameters such as Child–Pugh class > A liver function or a certain medical history such as liver transplantation. In the observational REFINE study, of 498 patients evaluable for the interim analysis, 12% (n = 61) had Child–Pugh class B/C liver function, 5% (n = 26) had Eastern Cooperative Oncology Group performance status (ECOG PS) > 1, and 2% (n = 8) had moderate or severe ascites. A history of transplantation was reported in 2% (n = 11) of patients, 13% (n = 65) had a history of esophageal varices (full assessment of active disease is not known due to the observational nature of the study), 4% (n = 20) had a history of hepatic encephalopathy/encephalopathy, 5% (n = 25) had a history of autoimmune disorders, and 10% (n = 51) had received prior immunotherapy. Two patients (< 1%) were currently receiving or had recently received antiplatelet agents. Some patients were classified in > 1 of these categories.

Conclusion: Interim results of the observational REFINE study suggest that in real-world practice, the population of patients with unresectable HCC receiving second- or later-line systemic treatment is more varied than the populations of phase 1–3 interventional trials, and includes patients with ECOG PS > 1 and Child–Pugh class > A liver function.

P025 Radiofrequency ablation versus surgical resection for the treatment of solitary hepatocellular carcinoma 2cm or smaller: a cohort study in Taiwan

Ching-Sheng Hsu[1], Chao-Chuan Wu[1], Chih-Wei Tseng[2], Kuo-Chih Tseng[2]

[1]Taipei Tzu Chi Hospital, Buddhist Tzu Chi Medical Foundation, Liver Diseases Research Center, New Taipei, Taiwan, *[2]Dalin Tzu Chi Hospital, Buddhist Tzu Chi Medical Foundation*
Email: hsu.chingsheng@gmail.com

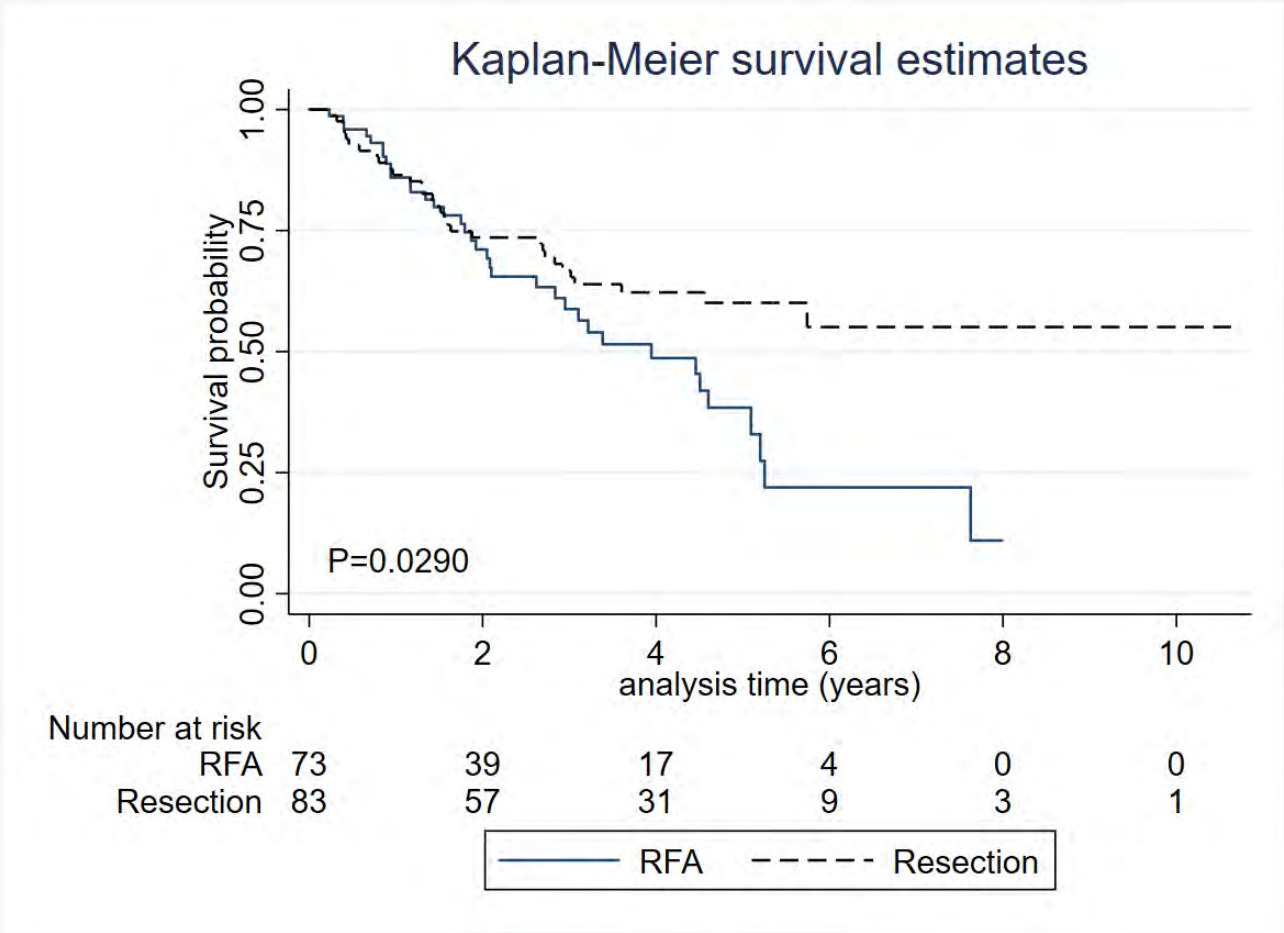
Background and Aims: Radiofrequency ablation (RFA) is increasingly being used instead of surgical resection for the treatment of hepatocellular carcinoma (HCC) tumor measuring ≦2 cm. However, the long-term outcomes of RFA, especially in comparison to surgical resection, are still debated. We compared the outcomes of surgical resection and RFA in patients with a solitary HCC tumor measuring ≦2 cm from a 10-year cohort study.

Method: From Jan 2006 to Dec 2016, 156 patients with a resectable HCC measuring ≦2 cm who underwent surgical resection (n = 83) or RFA (n = 73) at the Buddhist Tzu Chi Medical Foundation were enrolled. Patient characteristics and overall survival (OS), and recurrence-free survival (RFS) were retrospectively examined, and comparisons were made between the two groups and through subgroup analyses.

Results: The 1-year, 3-year, 5-year, and 7-year OS outcomes were comparable between the surgical resection group and the RFA group (P = 0.193), but the surgical resection group had significantly higher 1-year, 3-year, 5-year, 7-year, and 10-year RFS than the RFA group (P = 0.018). Multivariate analysis revealed that patients with lower age, Child–Turcotte-Pugh score, or albumin–bilirubin score before treatment had better OS, and patients with an HCV infection or receiving RFA treatment had higher HCC recurrence rates.

Conclusion: Antiviral therapy and the liver reserve determined the long-term OS of patients with an HCC tumor ≦ 2 cm, and surgical resection offered better RFS than RFA.

Figure: Cumulative RFS rates in patients initially treated with surgical resection or RFA



P026 Outcomes based on albumin-bilirubin grade in the phase 3 RESORCE trial of regorafenib versus placebo in patients with advanced hepatocellular carcinoma

Arndt Vogel[1], Philippe Merle[2], Alessandro Granito[3], Masafumi Ikeda[4], Marie-Aude LeBerre[5], Kirhan Ozgurdal[6], Jordi Bruix[7]

[1]Zentrum Innere Medizin, Medizinische Hochschule Hannover, Hannover, Germany, *[2]Groupement Hospitalier Lyon Nord, Hepatology Unit, Lyon, France*, *[3]University of Bologna, S. Orsola-Malpighi Hospital, Bologna, Italy*, *[4]National Cancer Center Hospital East, Kashiwa, Japan*, *[5]Bayer HealthCare SAS, Loos, France*, *[6]Bayer Consumer Care AG, Basel, Switzerland*, *[7]BCLC Group, Liver Unit, Hospital Clinic, University of Barcelona, IDIBAPS, CIBEREHD, Barcelona, Spain*

Email: vogel.arndt@mh-hannover.de

Background and Aims: In the phase 3 RESORCE trial (NCT01774344), regorafenib (REG), an oral tyrosine kinase inhibitor, significantly improved survival outcomes vs placebo (PBO) in patients with hepatocellular carcinoma (HCC) who previously failed sorafenib. Here, we evaluate outcomes based on albumin-bilirubin (ALBI) grade in the RESORCE trial.

Method: Overall, 573 patients were randomized 2:1 to REG 160 mg or PBO. Eligible adults had HCC, ≥ 1 measurable lesion, Barcelona Clinic Liver Cancer stage B/C disease, tolerated previous sorafenib and received their last dose within 10 weeks of randomization, and Child–Pugh A liver function. ALBI grade was determined by baseline ALBI score (serum albumin and total bilirubin). Subgroup analyses of Eastern Cooperative Oncology Group performance status (ECOG PS), treatment duration, overall survival (OS), and safety were conducted, stratified by ALBI grade.

Results: At baseline, 164 patients (43%) were ALBI grade 1 and 213 patients (56%) were ALBI grade 2 in the REG arm; 81 patients (42%) were ALBI grade 1 and 112 patients (58%) were ALBI grade 2 in the PBO arm. One patient in each arm was ALBI grade 3. In the REG arm, patients who were ALBI grade 1 vs 2 had better ECOG PS, longer treatment duration and OS, and fewer discontinuations due to a treatment-related adverse event (AE; Table). Median OS was longer in the REG arm vs PBO, regardless of ALBI grade. In the REG arm, incidences of grade 3/4 treatment-emergent AEs (TEAEs) by ALBI grade were similar (ALBI grade 1/2 = 66%/67%); patients who were ALBI grade 1 vs 2 had more grade 3/4 hypertension (20% vs 11%) and grade 3 hand–foot skin reaction (17% vs 9%). A lower proportion of ALBI grade 1 vs 2 had a serious TEAE (34% vs 53%).

Conclusion: REG treatment was associated with longer OS vs PBO, regardless of ALBI grade. Although grade 3/4 TEAEs were similar between ALBI grades, patients with ALBI grade 1 vs 2 had a lower rate of serious TEAEs.

Table:

	REG (n = 379)		**PBO (n = 194)**	
	ALBI grade 1	**ALBI grade 2**	**ALBI grade 1**	**ALBI grade 2**
ECOG PS 0/1, %	n = 164 70/30	n = 213 62/38	n = 81 69/31	n = 112 66/34
Median treatment duration (interquartile range)*, months	n = 163 4.6 (2.3, 10.2)	n = 209 2.9 (1.5, 6.7)	n = 81 1.7 (1.5, 3.2)	n = 111 2.3 (1.4, 4.2)

Median OS (95% confidence interval), months	n = 164 13.8 (10.3, 16.5)	n = 208 9.1 (7.4, 11.3)	n = 80 9.1 (6.6, 10.1)	n = 108 6.6 (5.1, 8.3)
Discontinuation due to a treatment-related AE, %	n = 163 7	n = 209 13	n = 81 1	n = 111 5
*Including time off drug/interruptions.				

P027YI Phenotypic characteristics of the tumour microenvironment in primary and secondary hepatocellular carcinoma.

Petros Fessas[1], Paolo Spina[2], Renzo Boldorini[3], Mario Pirisi[4], Rosalba Minisini[4], Francesco Mauri[1], Fraser Simpson[5], Paola Olivieri[5], Alessandra Gennari[4], Ching Ngar Wong[1], Abdul Siddique[1], Robert D. Goldin[6], Ayse Akarca[7], Teresa Marafioti[7], David J. Pinato[1]

[1]Imperial College London, Department of Surgery & Cancer, London, United Kingdom, *[2]Cantonal Institute of Pathology, Locarno, Switzerland*, *[3]Universitá degli Studi del Piemonte Orientale "A. Avogadro", Department of Health Sciences, Novara, Italy*, *[4]Università degli Studi del Piemonte Orientale, Department of Translational Medicine, Novara, Italy*, *[5]University College London, Department of Genetics, Evolution and Environment & Cell and Developmental Biology, London, United Kingdom*, *[6]Imperial College London, Centre for Pathology, London, United Kingdom*, *[7]University College London, Department of Histopathology, London, United Kingdom*
Email: petros.fessas93@gmail.com

Background and Aims: Intratumoral heterogeneity (ITH) is highly prevalent in HCC as shown by multi-region sequencing studies in early disease. Heterogeneity of HCC and its tumour microenvironment across primary (P) and secondary (S) disease is poorly characterized.

Method: We profiled intra-tumoural (IT) and peri-tumoural (PT) samples from 11 patients to evaluate regulatory $CD4^+/FOXP3^+$ (T-reg) and immune-exhausted $CD8^+/PD1^+$ T-cells across matched P and S deposits. Samples were stained for PD-ligands and processed for targeted next generation sequencing to derive tumour mutational burden (TMB), high-resolution T-cell receptor (TCR) sequencing to derive T-cell clonality and targeted transcriptomics with the Nanostring PanCancer Immune panel to evaluate the immune infiltrate.

Results: We analysed 24 samples from P (n=11) and S (n=13). S deposits were synchronous in 5 (45.5%) and metachronous in 6 (54.5%) patients, with 11 S lesions being extrahepatic (84.6%). Median time to relapse was 2 years. We found a negative IT/PT cell density gradient in both P (CD4+ IT/PT 13.0 vs 51.9 cells/mm^2, p=0.01; CD8+ 17.7 vs 58.2 cells/mm^2, p=0.005) and S (CD8+ IT/PT 32.5 vs 49.3 cells/mm^2, p=0.01), with evidence of higher CD4+FOXP3+ infiltrate in IT vs PT regions in P (p=0.004) but not S (p=0.18). The proportion of CD8+PD-1+ cells in IT and PT was similar in P (p=0.08) and S (p=0.92). The median histoscores for PD-L1 and PD-L2 expression were not significantly different between P and S (p=0.28, p=0.46). TMB was similar across P/S, with a median number of nonsynonymous mutations/MBp of 1.7 in P and 1.4 in S (p=0.95). TCR sequencing demonstrated higher median frequency of the most represented T-cell in S versus P (0.015 vs 0.028, p=0.02), without any significant difference in overall productive clonality (p=0.35) nor in the sums of the top 10 clones (p=0.11). T-cell productive clonality was not influenced by TMB (r=-0.34, p=0.51). Gene set analysis demonstrated uniformity of transcriptional signatures of individual immune cell types (p>0.05 throughout). Analysis of differentially regulated transcripts revealed overexpression of *COLEC12* (p=0.004), *CCL26* (p=0.02), *CD1E* (p=0.02) and *CD36* (p=0.03) and downregulation of *CXCL1* (p=0.03) in S.

Conclusion: An immune excluded phenotype is a common characteristic of the TME across P and S lesions in HCC. Transcriptional heterogeneity of the TME is substantial despite evidence of homogeneity in terms of TMB and overall T-cell clonality.

Figure: No figure associated.

P028YI Are undetected stones in the common biliary duct one of the causes of abscesses and bilomas development after percutaneous thermal ablation procedures?

Letizia Veronese[1], Francesca Torello Viera[1], Valentina Ravetta[1], Sandro Rossi[1]

[1]*III medical Division- Interventional Ultrasound, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy*
Email: letizia.veronese1@gmail.com

Background and Aims: Abscesses and bilomas are rare complications of percutaneous thermal ablation (TA). We evaluated the possible relationship between liver abscesses and bilomas complicating percutaneous TA of hepatocellular carcinoma (HCC) and pathologies of the common biliary duct which partially hindered bile outflow (i.e previously undetected stones, ab-extrinsic compressions, benign and malignant incomplete stenosis, etc.)

Method: Between January 2000 and January 2020 at our internal medicine department, 1670 patients affected by single, naïve, HCC nodule (less than or equal to 35 mm in diameter) arising in chronic liver disease [994 M; age 69 ± 7 years (range, 56 - 81 years)], underwent 1689 TA sessions. The same patients also underwent additional 1734 TA sessions for new HCC nodules detected in the follow-up. We retrospectively evaluated the incidence of abscesses/bilomas after TA sessions, their management and outcome.

Results: After 3423 TA sessions nine (0.2 %) bilomas and 12 (0.3 %) abscesses were observed in 21 patients. Of these patients, 16 (76.2 %) reported in the past medical history a cholecystectomy due to symptomatic stones and five (26.8 %) had asymptomatic gallbladder stones at the TA procedure time. Diagnosis of abscesses/bilomas was achieved by clinical and laboratory findings and by ultrasonography (US), contrast-enhanced US, spiral-computed tomography and US-guided aspiration needle. All patients underwent percutaneous US-guided drainage by placing an indwelling catheter (6 to 12 French in caliber) within the collections and antibiotic therapy for 8 - 10 days. The diagnosis of common bile duct lithiasis was done in six patients by magnetic resonance imaging (MRI), in two patients by MRI plus endoscopic US (EUS) (one of which had Mirizzi syndrome) and in the remaining 13 patients by EUS. All patients, before removing the percutaneous liver catheter, underwent endoscopic retrograde cholangiopancreatography with sphincterotomy. These combined techniques allowed the complete resolution of the complications in all patients. The five patients with gallbladder stones, within a month, also underwent laparoscopic cholecystectomy.

Conclusion: The common biliary tract of HCC patients candidates to percutaneous TA should be accurately investigated before treatment with laboratory tests and radiological imaging (US, EUS or MRI) to exclude stones, especially in those ones with previous cholecystectomy or with gallbladder stones.

Figure:

P029YI Different modalities in management of bleeding related to post-banding ulcer following endoscopic variceal ligation in cirrhotic patients with or without hepatocellular Carcinoma

Mohamed Abdel-Samiee[1], Mohammed AtefAbdelfatah Taha[1], Maha Mohamed Elsabawy[1], Nabil Abdelhamid Omar[1]

[1]National Liver Institute, Menoufia University, Hepatology and Gastroenterology, Shebin El-Kom, Egypt
Email: drmohammed100@yahoo.com

Background and Aims:
Cirrhosis leads to portal hypertension. Increase in portal pressure is initially due to a consequence of an increased resistance to flow mostly due to an architectural distortion of the liver secondary to fibrous tissue and regenerative nodules.
Despite urgent endoscopic and/or pharmacological therapy, variceal bleeding cannot be controlled in about 10%-20% of patients. An elevated HVPG >20 mmHg (measured within 24 hrs of presentation) has been shown to be predictive of treatment failure. Following band ligation, a local ulcer is commonly seen with an ensuing well-described sequence of the pathological changes. This study aims to evaluate different modalities in management of upper gastrointestinal bleeding from post band ulcers.

Method:
Patients were defined as cirrhotic patients who developed a digestive bleeding related to ulcers after spontaneous slippage of rubber band following EVL performed in emergency or prophylactic situation. This bleeding was an active one (spurting or oozing) because of the ulcers induced by the previous EVL, confirmed by a gastroscopy performed within 12 hours after the hemorrhage.

Results:
The prevalence of bleeding ulcers post EVL was 3.33 % (149 cases out of 4350 patients to have decompensated liver cirrhosis with band ligation to esophageal varices. 145 patients had bleeding post esophageal variceal band ulcers. The remaining 49 patients underwent EVL without complication (control). There was a statically significant difference between both groups concerning male gender; smoking and DM beng more comon in bleeding cases. There was no statistically significant difference between both groups regarding age and hypertension. There was a statically significant difference between both groups concerning hospital stay post intervention endoscopy being higher in bleeding cases, Pulse at bleeding and Units of blood transfusion being higher in bleeding group.

Conclusion:
Different risk factors for post band ligation bleeding as low platelet and higher pulse, urea, HCC, PVT and child score.

Figure:

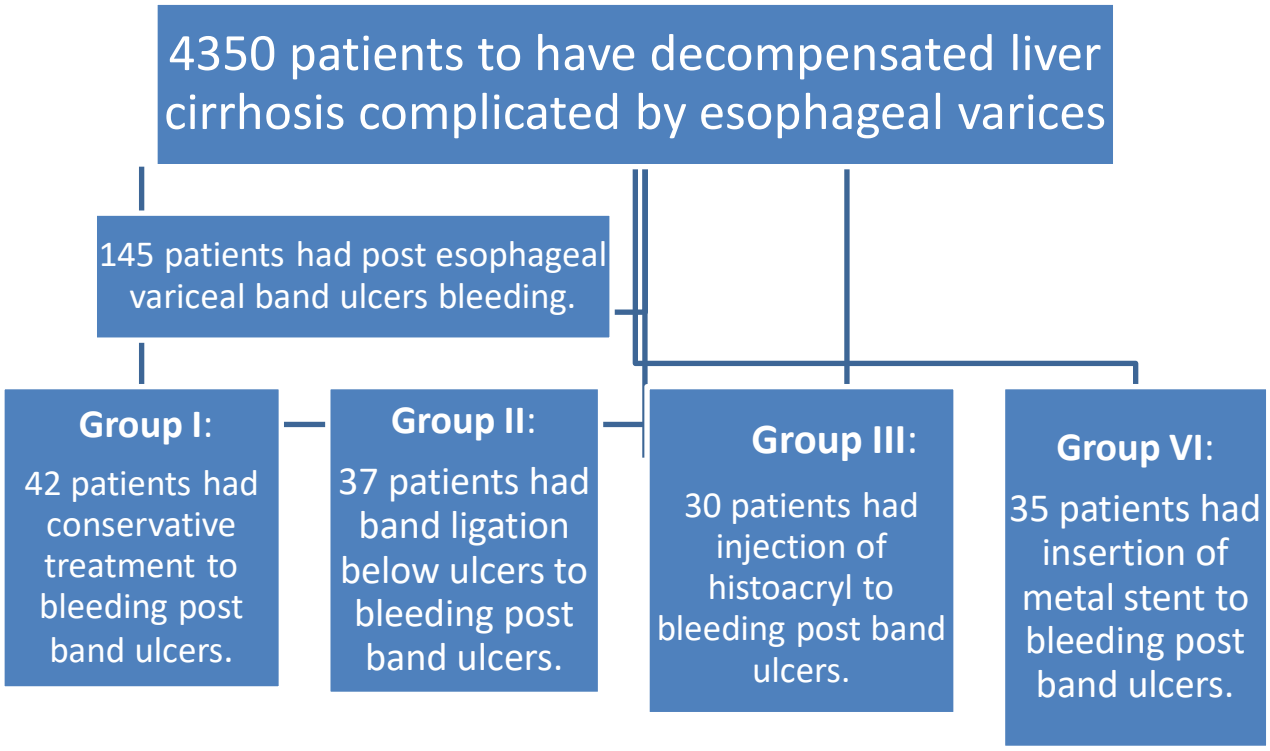


Table 1: comparision of child score; HE; HCC ;PVT; splenomegally and ascitis of the studied groups

	Group1 (case) no=145	Group2 (control) no=49	X^2	P value
	N (%)	N (%)		
Child score (mean±SD)	10.2±1.5	7.2±2.3	8.215*	0.000
Child class:			24.23	0.000
A	2.3(2.1)	13(26.5)		
B	39(26.8)	26(53.4)		
C	103(71.1)	10(20.1)		
Cirrhosis etiology:			35.02	0.000

Bilharzia HBV HCV	14(9.7) 11(8.2) 120(82.1)	3(6.1) 5(10.2) 41(83.7)		
HCC: Yes No	 87(60.0) 58(40.0)	 10(20.6) 39(79.4)	22.31	0.000
PVT: Yes No	 68(46.2) 79(53.8)	 6(12.5) 43(87.5)	18.24	0.000
Size of the spleen (mean±SD)	17.2±5.3	16.2±3.4	0.652*	0.510

*T test

P030 Copper chelators old drugs for new uses in the treatment of hepatocellular carcinoma

Silvano Junior Santini[1 2], Maria Rita Braghini[3], Anna Alisi[3], Clara Balsano[1 2]

[1]University of L'Aquila, Dept. of Life, Health and Environmental Sciences MESVA, L'aquila, Italy, *[2]Francesco Balsano Foundation, Rome, Italy*, *[3]"Bambino Gesù" Children's Hospital IRCCS, Research Unit of Molecular Genetics of Complex Phenotypes, Rome, Italy*
Email: clara.balsano@univaq.it

Background and Aims: Hepatocellular carcinoma (HCC) represents the third most frequent cause of cancer death. HCC carries an extremely poor prognosis since it is often diagnosed at advanced stages, restricting efficient therapeutic options to either surgical resection or transplantation. Hence, it is urgent to develop new and more effective therapeutic strategies to defeat HCC. Biometals, in particular copper, are emerging as important regulators of several physiological and pathological processes, including tumors. Accordingly, copper concentration correlates with incidence, malignant progression, and recurrence in HCC.

We aimed to better define the antineoplastic effects induced by the copper chelator Tetrathiomolybdate (TTM) to find new therapeutic targets in HCC.

Method: HCC cell lines, HepG2, HuH7.5 and Hep3B, have been treated with 100μM TTM. We evaluated the effects of TTM treatment on viability, cell cycle and apoptosis of HCC cell lines. We analyzed the TTM treatment effect on cell migration and invasiveness by wound healing and Boyden chamber assays. Furthermore, we performed the analysis of TTM treatment effects on the expression of a panel of 631 genes involved in stemness of HepG2 cells.

Results: HCC cell viability was strongly reduced by treatment with TTM. TTM caused an increase of HCC cells in the G2/M phase of cell cycle compared to NT cells, associated with an increment of the percentage of apoptosis. TTM treatment was also effective on cell migration. The gene expression analysis highlighted the upregulation of 56 genes, whereas 37 were downregulated. Among the studied genes, the downregulation of PTK2 gene encoding for the focal adhesion kinase (FAK) by TTM was confirmed in all HCC cell lines. Moreover, we found a deregulation of FAK also at transcriptional and functional levels under TTM treatment.

Conclusion: Our data underly the sensitivity of HCC cell lines to copper chelation. Further studies should be focalized on the evaluation of effects of TTM combined with standard-of-care locoregional therapy.

P031YI Diagnostic accuracy of a-fetoprotein (AFP), protein induced by vitamin K absence (PIVKA-II) and fibrogen like protein-2 (FGL-2) for HCC diagnosis among patients with liver cirrhosis

antonia syriha[1], petros galanis[1], Kalliopi Tabouratzi[1], Georgia Barla[1], Ioanna Stathopoulou[1], theodora biniari[1], panagiota evangelakou[2], Ioannis Elefsiniotis[1]

[1]National and Kapodistrian University of Athens, Greece, Academic Department of Internal Medicine-Hepatogastroenterology Unit, General and Oncology Hospital of Kifisia "Agioi Anargyroi", Athens, Greece, *[2]abbott laboratories greece*
Email: ielefs@nurs.uoa.gr

Background and Aims: To evaluate the diagnostic accuracy of biomarkers (AFP, PIVKA-II, FGL-2) for HCC diagnosis among patients with liver cirrhosis

Method: 103 consecutive patients with liver cirrhosis (81 males, 49 CPT score A, 54 with esophageal varices, 25 with portal vein thrombosis, 33 with diabetes) with (n=50, HCC group) or without (n=53, control group) concomitant histologically confirmed HCC were evaluated for serum levels of the three biomarkers, during a programmed outpatient visit.

Results: Patients with HCC were older (mean age=68.1 vs 58.4y, p<0.001) and presented frequently with diabetes (72.7% vs 27.3%, p=0.007) and portal vein thrombosis (92% vs 8%, p<0.001) whereas CPT scores (A: 44.9% vs 54.1%, B: 71% vs 29%, C: 46.2% vs 53.8%, p=0.33) and mean MELD scores (13.1 vs 11.7, p=0.08) were comparable between the two groups. Mean/median levels of AFP (6811.6/29 vs 5.88/3.9, p<0.001), PIVKA-II (2221.7/144.9 vs 54.8/36.5, p<0.001) and FGL-2 (5.23/3.26 vs 1.84/1.51, p=0.008) were significantly higher in HCC group compared to control group. The AUROC curve, the sensitivity and specificity for HCC diagnosis were 0.95/0.83/0.98, 0.88/0.75/0.93 and 0.79/0.71/0.77 for AFP, PIVKA-II and FGL-2, respectively. Excluding HCC patients with AFP levels above 400 ng/ml the diagnostic accuracy of PIVKA-II continued to be significant (best proposed cut-off value 144.8 mAU/ml) but the sensitivity (0.62), specificity (0.93) and the AUROC curve (0.81) for HCC diagnosis were further reduced.

Conclusion: Using the best proposed cut-off values of 9.2 ng/ml for AFP and 255.9 mAU/ml for PIVKA-II, both biomarkers showed moderate accuracy for detecting HCC in patients with liver cirrhosis. The utility of PIVKA-II for HCC diagnosis in patients with low AFP levels should be further evaluated.

Figure:

P032YI NADPH oxidase 4 (Nox4) deletion accelerates liver regeneration in mice

MACARENA HERRANZ ITÚRBIDE[1], Judit López Luque[1], Ester Gonzalez Sanchez[1], Daniel Caballero Díaz[1], Eva Crosas Molist[1], Beatriz Martín Mur[2], Marta Gut[2], Anna Esteve Codina[2], Vincent Jaquet[3], Joy Xiaosong Jiang[4], Natalie J Török[5], Isabel Fabregat[1]

[1]IDIBELL Institut d'Investigació Biomèdica de Bellvitge, L'Hospitalet de Llobregat, Spain, *[2]CNAG-CRG, Centre for Genomic Regulation, Barcelona, Spain*, *[3]University of Geneva, Geneva, Switzerland*, *[4]UC Davis Health - Gastroenterology, Sacramento, United States*, *[5]Stanford University , Gastroenterology & Hepatology, Stanford, United States*
Email: mherranz@idibell.cat

Background and Aims: Liver is a unique organ in displaying a reparative and regenerative response after acute/chronic damage or partial hepatectomy, when all the cell types must proliferate to re-establish the liver mass.

The NADPH oxidase NOX4 mediates Transforming Growth Factor-beta (TGF-β)-induced apoptosis in hepatocytes and activation of liver stellate cells to myofibroblasts, contributing to the development of liver fibrosis. Indeed, NOX4 inhibitors have been suggested as a potential anti-fibrotic therapy in the liver. Additionally, NOX4 inhibits hepatocyte growth and tumorigenesis and regulates liver tumor cell migration, being considered as a potential tumor suppressor.

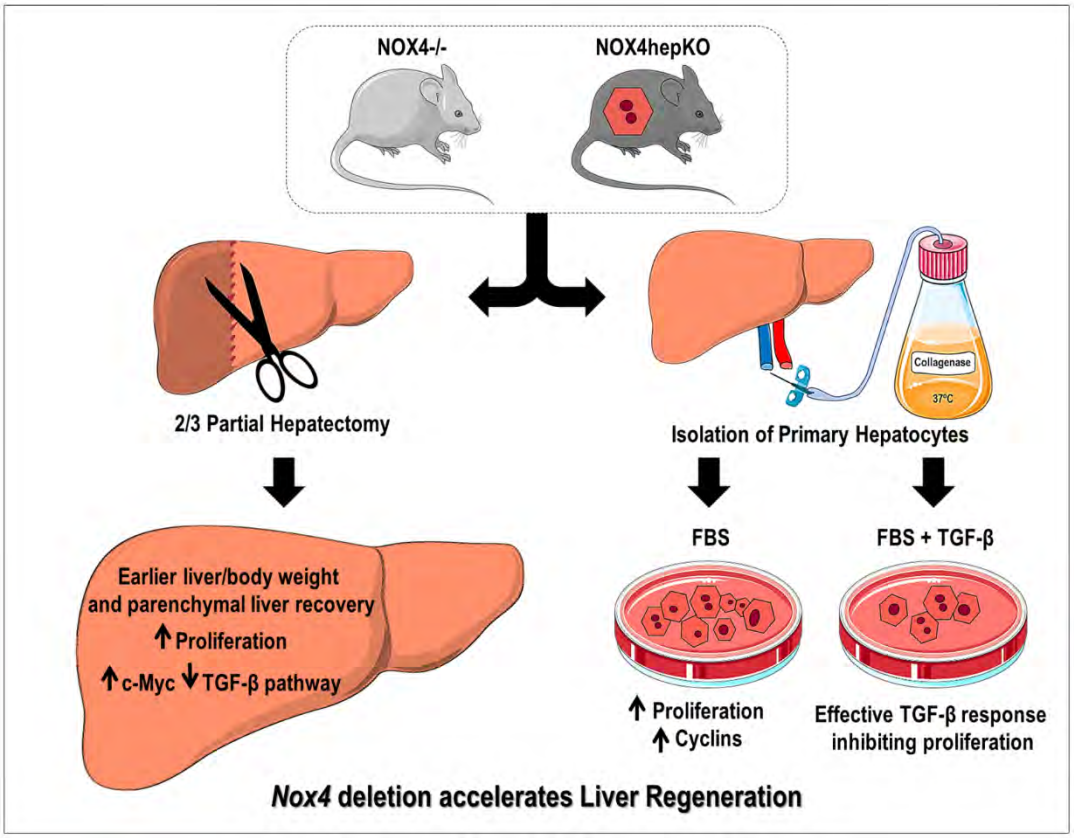
Very little is known about the role of NOXes during liver regeneration. However, considering the function of NOX4 in inhibiting hepatocyte proliferation, we previously reported that Nox4 expression is down-regulated after partial hepatectomy in mice. The aim of this work was to analyze the impact of NOX4 in liver regeneration by using two mouse models where Nox4 was deleted: 1) general deletion of Nox4 (NOX4-/-) and 2) hepatocyte-specific deletion of Nox4 (NOX4hepKO).

Method: Liver regeneration was analyzed after 2/3 partial hepatectomy (PH). Mice were euthanized at different times after surgery, and SHAM operated mice were used as control. RNA sequencing and analysis was performed in total RNA from liver samples from WT and NOX4-/- mice.
Primary hepatocytes were isolated by collagenase *in situ* perfusion, cultured in Williams' Medium E, and treated with FBS and TGF-β1.

Results: Results indicated an earlier recovery of the liver-to-body weight ratio in both NOX4-/- and NOX4hepKO mice and an increased survival, when compared to corresponding WT mice. The regenerative hepatocellular fat accumulation and the parenchyma organization recovered faster in NOX4 deleted livers. Hepatocyte proliferation, analyzed by Ki67 and phospho-Histone3 immunohistochemistry, was accelerated and increased in NOX4 deleted mice, coincident with an earlier and increased Myc expression. Primary hepatocytes isolated from NOX4 deleted mice showed higher proliferative capacity and increased expression of Myc and different cyclins in response to serum. Transcriptomic analysis through RNA-seq revealed significant changes after PH in NOX4-/- mice, and support a relevant role for Myc in a node of regulation of proliferation-related genes. Interestingly, RNA-seq also revealed changes in the expression of genes related to activation of the TGF-β pathway. In fact, levels of active TGF-β1, phosphorylation of Smads and levels of its target p21 were lower at 24 h in NOX4 deleted mice.

Conclusion: Deleting NOX4 expression, either in the total organism or specifically in hepatocytes, accelerates liver regeneration, without affecting the termination of liver regeneration.

Figure:


NOX4-/-
NOX4hepKO
2/3 Partial Hepatectomy
Collagenase
37°C
Isolation of Primary Hepatocytes
FBS
FBS + TGF-β
Earlier liver/body weight
and parenchymal liver recovery
↑Proliferation
↑c-Myc ↓TGF-β pathway
↑ Proliferation
↑ Cyclins
Effective TGF-β response
inhibiting proliferation
Nox4 deletion accelerates Liver Regeneration

P033YI Development of ex vivo and in vivo tissue models of hepatocellular carcinoma and utility for immunotherapy investigations

Amy Collins[1], Daniel Geh[1], Saimir Luli[1], Rainie Cameron[1], Helen L. Reeves[1], Fiona Oakley[1], Derek Mann[1], Jack Leslie[1]

[1]*Newcastle University, United Kingdom*
Email: a.l.collins1@newcastle.ac.uk

Development of ex vivo and in vivo tissue models of hepatocellular carcinoma and utility for immunotherapy investigations

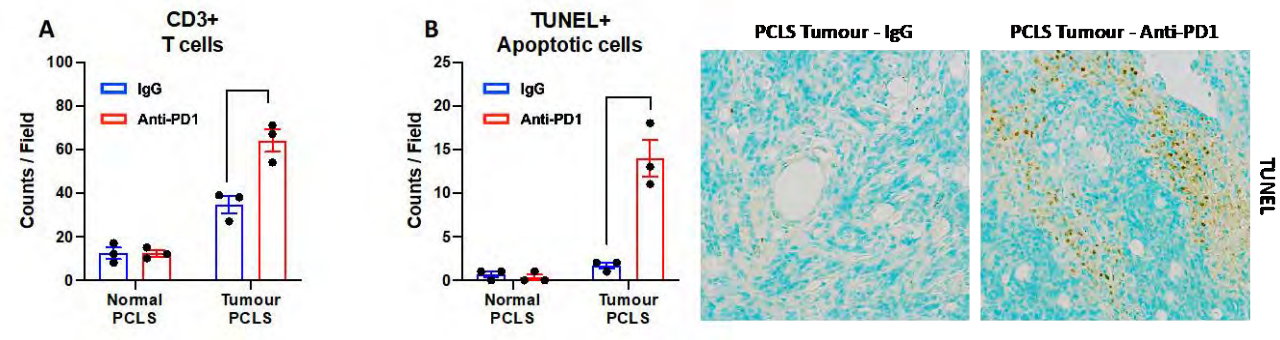
Background and Aims: Preclinical models are used extensively to study hepatocellular carcinoma (HCC) development and response therapy. Despite its promise, immune checkpoint inhibitors have only been successful in a subset of HCC patients, likely due to the complexity of the tumour-immune microenvironment. Precision cut liver slices (PCLS) retain cell composition and tissue architecture allowing investigation of complex disease processes and response to therapies in vitro. Our aim was to develop murine PCLS platforms that recapitulate the *in vivo* model to allow high throughput identification and testing of new therapies.

Method: Murine hepatoma Hep53.4 cells were implanted into the left lobe of wild type mice. Mice were treated with therapeutic administration of anti-PD1 or IgG control (Biolegend #114108 i.p. 200ug twice weekly). We are developing three PCLS based murine models of HCC. PCLS were generated from control liver tissue or Hep53.4 tumours produced *in vivo.* PCLS generated from control liver tissue were implanted with either Hep53.4 spheroids or bio-printed Hep53.4 cell suspensions. Slices were cultured for 4 days in patented BioR plates and bioreactor platform and treated with either IgG control or anti-PD1 (20 ug/mL).

Results: Hep53.4 liver tumours are immune rich with a large inflammatory infiltrate and an active stroma. *In vivo* Hep53.4 tumours, anti-PD1 therapy evoked a strong T cell response resulting in a significant reduction in tumour burden. PCLS generated from Hep53.4 liver tumours are viable, proliferative and maintain their tumour immune microenvironment when cultured *in vitro*. PCLS tumours, in line with the *in vivo* model, responded to anti-PD1 therapy. Anti-PD1 therapy induced a local activation and proliferation of T cells in the tumour slice inducing apoptosis of tumour cells (Figure A-B). This effect was limited to tumour PCLS, with PCLS generated from healthy liver tissue displaying no adverse effects. Implanted Hep53.4 derived spheroids and bio-printed Hep53.4 cell suspensions engraft onto PCLS generated from control liver. These models may be effective as an initial *in vitro* drug screening platform, prior to subsequent PCLS tumour slice and *in vivo* validation.

Conclusion: Precision cut slices are histologically and immunologically representative of the in situ tumour. We propose that these models provide the initial methodological advancement for translation into a human HCC model system with therapeutic utility.

Figure:



P034YI Methylated Septin 9 (mSEPT9) as a biomarker for hepatocellular carcinoma. Results from University Hospital Coventry and Warwickshire NHS Trust, UK

Ayman Bannaga[1 2], Rocio Alvarez[3], Michael Petchey[4], Angela Noufaily[2], Megan Hitchins[3 5], Ramesh Arasaradnam[1 2]

[1]University Hospital Coventry and Warwickshire NHS Trust , Gastroenterology and Hepatology , Coventry, United Kingdom, *[2]University of Warwick, Warwick Medical School, Coventry, United Kingdom*, *[3]Cedars-Sinai Medical Centre, Biomedical Sciences , Los Angeles, United States*, *[4]University Hospital Coventry and Warwickshire NHS Trust , Biomedical Sciences , Coventry, United Kingdom*, *[5]Cedars-Sinai Medical Center, Biomedical Sciences, Los Angeles, United States*
Email: aymanbannaga@yahoo.com

Background and Aims:
Methylated Septin 9 (mSEPT9) is associated with hepatocarcinogenesis. Following death of liver cancer cells, mSEPT9 is released into the blood stream. We investigated mSEPT9 performance as a biomarker for hepatocellular carcinoma (HCC)

Method:
A single centre case-control study that recruited 141 patients between 2013 to 2019. Ethical approval was granted from Coventry and Warwickshire and North-East Yorkshire Research Committees (Ref 18717 and Ref 260179). Multiple covariates of interest to HCC were collected. Measurement of plasma mSEPT9 was performed using the Epi proColon V2.0 kits and real time polymerase chain reaction (rtPCR). Statistical analyses were performed using SPSS software

Results:
There were 60 (42.6%) Females and 81 (57.4%) Males. There were 38 (27%) HCC cases and 103 (73%) patients without HCC (98 with liver disease and 5 without). There were 60 (42.6%) cirrhotics and 81 (57.4%) non cirrhotics. Mean age, BMI, Albumin, alpha fetoprotein (AFP) and bilirubin were 60.69 years, 28.1 Kg/m2, 42 g/l, 964 KU/L and 10 umol/l respectively. AFP sensitivity for detecting cirrhosis was 27% with specificity of 93%. AFP sensitivity for detecting HCC was 50% with specificity of 97%. mSEPT9 sensitivity for detecting cirrhosis was 63% with specificity of 80%. mSEPT9 sensitivity for detecting HCC was 89% with specificity of 81%. In addition, statistical logistic regression analyses were performed and adjusted for age, mSEPT9 showed significant ($p<0.005$) association with Albumin-Bilirubin (ALBI), Neutrophil to lymphocyte ratio (NLR) and Fibrosis 4 (FIB4) index, suggesting this marker is related directly to liver function, inflammatory status and liver fibrosis stage. Importantly as biomarker, mSEPT9 showed no association with diabetes status, BMI and gender of the patients in this study even when adjusted for age

Conclusion:
mSEPT9 has potential as HCC detection marker. The use in surveillance of liver cirrhosis could also be of potential. Results here are not applicable to the general population and would require large validatory longitudinal studies

Figure:

Logistic regression for mSEPT9 and covariates of interest in the study

Covariate	**mSEPT9**	
	p-value	Age Adjusted p-value
Gender (F/M)	0.033	**0.006**
BMI	0.010	**0.012**
HCC	0.000	0.000
Cirrhosis	0.000	0.000
AFP (>10, ≤10)	0.000	0.000
Diabetes	0.040	**0.170**
Platelets	0.001	**0.002**
Neutrophil to lymphocyte ratio (NLR)	0.000	**0.000**
Albumin	0.000	**0.000**
Bilirubin	0.003	0.005
ALBI grade	0.000	**0.000**
Fib-4 index	0.000	**0.000**

P035 Impact of NAFLD on clinical outcomes in hepatocellular carcinoma treated with sorafenib: an international cohort study

rohini sharma[1], Amit Samani[1], binish mannan[1], Saur Hajiev[1], Rebecca abdelmalak[1], Leila motedayen aval[1], Vincent Tam[2], Bettinger Dominik[3], Robert Thimme[3], Tamar Taddei[4], David E Kaplan[4], Max Seidensticker[5], Jessica Howell[6]

[1]imperial college london, *[2]University of Calgary*, *[3]University of Freiburg*, *[4]University of Pennsylvania Perelman School of Medicine*, *[5]Klinikum der Universität München*, *[6]University of Melbourne*
Email: r.sharma@imperial.ac.uk

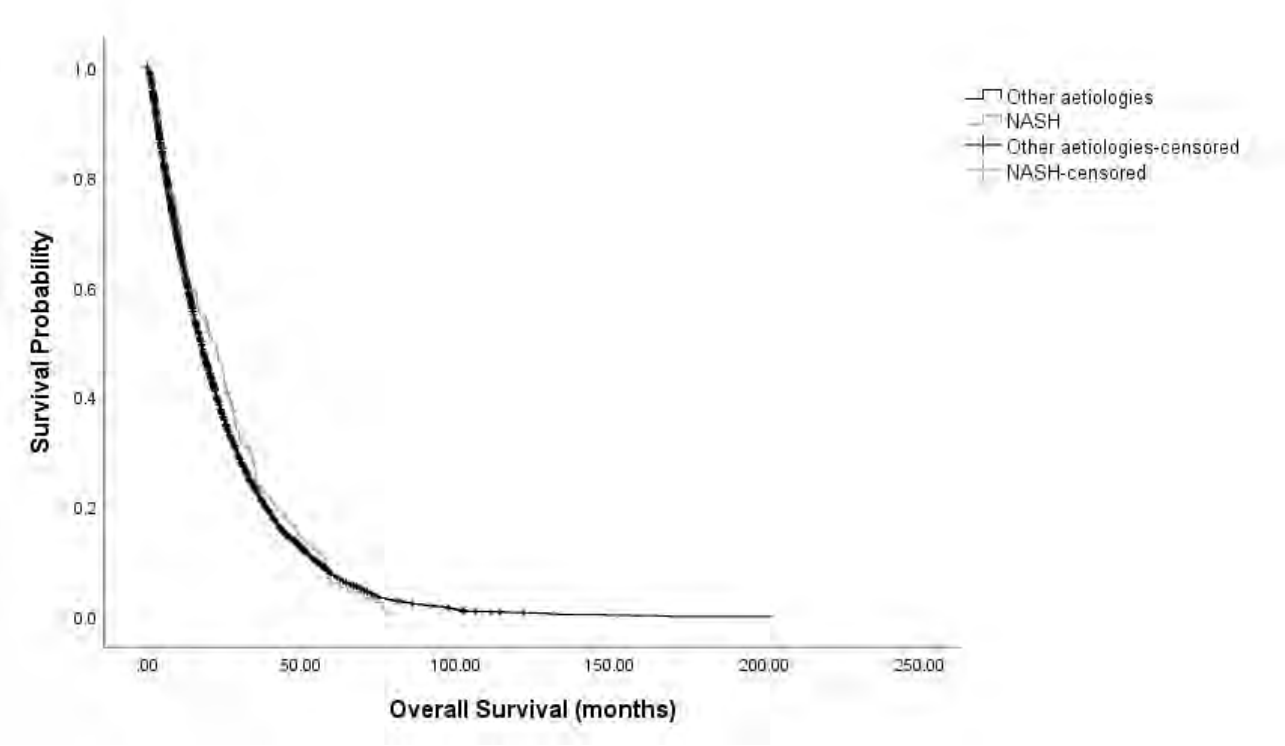
Background and Aims: The incidence of non-alcoholic fatty liver disease (NAFLD)-associated hepatocellular carcinoma (HCC) is increasing. The impact of NAFLD on overall survival (OS), treatment response and toxicity in patients with HCC treated with sorafenib is unknown. We examined the impact of NAFLD on survival and toxicity in an international cohort of patients receiving sorafenib.

Method: Clinical and demographic data were collected from patients consecutively treated at specialist centres in Europe and North America. The impact of NAFLD on OS, sorafenib-specific survival and sorafenib-related toxicity compared to other aetiologies of liver disease using multivariable Cox-proportional hazards and logistic regression modelling was assessed

Results: 5201 patients were treated with sorafenib; 183(3.6%) had NAFLD-associated HCC. NAFLD-associated HCC patients were more likely to be older women(median age 65.8 vs 63.0 years, p<0.01 and 10.4% vs 2.3%, <0.01), with a median BMI of 29.4. After controlling for known prognostic factors, no difference in OS in patients with or without NAFLD was observed(HR 0.99 (95%CI 0.84 -1.18), p=0.98). NAFLD-associated patients had more advanced stage HCC when they commenced sorafenib(BCLC C/D 70.9% vs 58.9%, p<0.01) and were more likely to be commenced on a lower starting dose of sorafenib(51.4 vs 36.4%, p<0.01). However, there was no difference in sorafenib-specific survival between NAFLD and other aetiologies(HR 0.96, 95% CI (0.79 – 1.17, p=0.96). Adverse events were similar between NAFLD and non-NAFLD HCC groups, including rates of ≥grade 2 hypertension(6.3 vs 5.8%, p=1.00). A lower rate of severe hand foot syndrome was observed in the NAFLD population(3.8 vs 12.4%, p=0.03).

Conclusion: Survival in HCC does not appear to be influenced by the presence of NAFLD. NAFLD-associated HCC derive similar clinical benefit from sorafenib compared to other aetiologies

Figure: Kaplan-Meier curves illustrating the prognostic relationship of presence of NAFLD and other aetiologies with overall survival in patients with HCC.



P036 Trans-arterial chemoembolization as a loco-regional inducer of immunogenic cell death in hepatocellular carcinoma: implications for immunotherapy.

David J. Pinato[1], Sam Murray[2], Alejandro Forner[3 4], Takahiro Kaneko[5], Petros Fessas[1], Pierluigi Toniutto[6], Beatriz Minguez[7 8], Valentina Cacciato[9], Claudio Avellini[10], Alba Díaz[11], Rosemary Boyton[2], Daniel M Altmann[12], Robert D. Goldin[13], Ayse Akarca[14], Teresa Marafioti[14], Francesco Mauri[1], Edoardo Giovanni Giannini[9], Federica Grillo[15], Edoardo Casagrande[9], Sherrie Bhoori[16 17], Vincenzo Mazzaferro[16 17]

[1]Imperial College London, London, United Kingdom, *[2]Imperial College London, Department of Infectious Disease, London, United Kingdom*, *[3]Hospital Clinic Barcelona, Barcelona Clinic Liver Cancer Group, Barcelona, Spain*, *[4]University of Barcelona, Hospital Clinic Barcelona, IDIBAPS, CIBERehd, Barcelona, Spain*, *[5]Tokyo Medical and Dental University, Faculty of Medicine, Tokyo, Japan*, *[6]Hepatology and Liver Transplantation Unit, Azienda Sanitaria Universitaria Integrata, Specialized Medicine, Udine, Italy*, *[7] Universitat Autonoma de Barcelona, Liver Unit, Hospital Universitari Vall d'Hebron., Barcelona, Spain*, *[8]Vall d´Hebron Institute of Research (VHIR), Vall d'Hebron Barcelona Hospital Campus, CIBERehd , Barcelona, Spain*, *[9]University of Genoa, Department of Internal Medicine, Genoa, Italy*, *[10]Azienda Ospedaliero-Universitaria "Santa Maria della Misericordia", Institute of Histopathology, Udine, Italy*, *[11]University of Barcelona, Pathology Department, Hospital Clínic, Barcelona, Spain*, *[12]Imperial College London, Department of Immunology and Inflammation, London, United Kingdom*, *[13]Imperial College London, Centre for Pathology, London, United Kingdom*, *[14]University College London Hospital, Department of Histopathology, London, United Kingdom*, *[15]University of Genoa, Department of Surgical Sciences and Integrated Diagnostics, Genoa, Italy*, *[16]University of Milan, Department of Oncology, Milan, Italy*, *[17]Istituto Tumori Milano, Milan,*
Email: david.pinato@imperial.ac.uk

Background and Aims: Modulation of adaptive immunity is postulated to underscore the efficacy of TACE. We evaluated the influence of TACE on T-cell function by assessing the phenotypic characteristics of lymphocyte populations from archival samples of patients who underwent surgery with (T^+) or without (T^-) prior-TACE treatment.

Method: We profiled intra-tumoural (IT), peri-tumoural (PT) and non-tumoural background tissue (NT) to evaluate regulatory $CD4^+/FOXP3^+$ (T-reg) and immune-exhausted $CD8^+/PD1^+$ T-cells across T+ (n=58) and T- (n=61). We performed targeted transcriptomics and T-cell receptor sequencing in a restricted subset of samples (n=24) evaluated in relationship with the expression of actionable drivers of anti-cancer immunity including PD-L1, IDO-1, CTLA-4, Lag-3, Tim-3 and CD163.

Results: We analyzed samples from 119 patients resected (n=25, 21%) or transplanted (n=94, 79%) for Child Pugh A (n=65, 55%) and TNM stage II (n=73, 61%) HCC. T^+ samples displayed lower IT $CD4^+/FOXP3^+$ (p=0.006), $CD8^+$ (p=0.002) and $CD8^+/PD1^+$ (p<0.001) and lower NT $CD8^+/PD1^+$ compared to T^-. Lower IT (p=0.005) and NT $CD4^+/FOXP3^+$ (p=0.03) correlated with improved recurrence free survival (RFS), with IT $CD4^+/FOXP3^+$ density predicting for RFS benefit in multivariable analyses. In a subset of samples (12 T^+, 12 T^-), transcriptomic analysis revealed differential up-regulation of genes reflective of a pro-inflammatory response in T^+. Compared to T^-, T^+ samples were significantly enriched for IRF2 expression (p=0.01), an interferon-regulated transcription factor linked to immune-responsiveness in other malignancies. Expression PD-L1, IDO-1, CTLA-4, Lag-3, Tim-3 and CD163 was not different T^+ versus T^-. T-cell clonality by ImmunoSeq assay was not different in association with TACE pre-treatment.

Conclusion: Pre-treatment with TACE is associated by lower intra-tumoral density of immune-exhausted effector and regulatory T-cells, with significant up-regulation of pro-inflammatory pathways. This highlights the pleiotropic effects of TACE in modulating the tumour microenvironment and strengthens the rationale for developing immunotherapy alongside TACE to improve outcomes of HCC patients.

Figure:

P037 PNPLA3 and Notch3 gene polymorphisms as risk factors for alcoholic cirrhosis development and its progression to hepatocellular carcinoma

Tomislav Kelava[1], Dino Šisl[1], Ana Bainrauch[2], Ivan Budimir Bekan[3], Valerija Bralic Lang[4], Andjela Krstulovic Opara[5], Ana Ostojic[2], Antonio Markotic[6], Anna Mrzljak[7]

[1] School of Medicine, University of Zagreb, Department of Physiology, Zagreb, Croatia, *[2]Merkur University Hospital, Department of Internal Medicine, Zagreb, Croatia*, *[3]Merkur University Hospital, Department of Surgery, Zagreb, Croatia*, *[4] School of Medicine, University of Zagreb, Department of Family Medicine*, *[5] School of Medicine, University of Zagreb, Zagreb, Croatia*, *[6]School of Medicine, University of Mostar, Bosnia and Herzegovina*, *[7]Merkur University Hospital, School of Medicine, University of Zagreb, Department of Internal Medicine, Zagreb, Croatia*
Email: anna.mrzljak@gmail.com

Background and Aims: Between 5 and 15% of patients with alcoholic liver disease (ALD) develop hepatocellular carcinoma (HCC). In the present study, we investigated the association of the single nucleotide polymorphisms (SNPs) for PNPLA3 (rs738409) and Notch3 (rs1043996) with the occurrence of end-stage alcoholic liver disease and with its progression towards HCC.

Method: DNA was isolated from the blood of 245 patients transplanted due to alcohol-related cirrhosis (124 with HCC and 121 without HCC) in the Merkur Transplant Centre, Zagreb, Croatia, and 70 control patients without liver disease. SNPs for PNPLA3 (rs738409) and Notch3 (rs1043996) were determined by PCR using commercially available TaqMan assays. Associations between SNPs and ALD or HCC were examined in dominant, recessive, codominant, over-dominant, and log-additive models.

Results: Genotypes were in Hardy-Weinberg equilibrium (p=0.26 for Notch3 and p=0.2 for PNPLA3). Minor allele frequency for PNPLA 3 was 41%, and for Notch3 was 27%. PNPLA3 rs738409 was associated with higher risk for occurrence of ALD in codominant (OR 95%CI = 6.15 (2.28-16.60) for GG vs CC genotype and 2.78 (1.55-4.97) for GC vs CC genotype), dominant (OR 95%CI = 3.36 (1.94-5.82) for GG/GC vs CC genotype), recessive (OR 95%CI = 3.71 (1.42-9.69) for GG vs GC/CC genotype) and log additive model (OR 95%CI = 2.60 (1.69-3.99) for G allele). Furthermore, this SNP was also associated with a higher risk for HCC occurrence among the ALD patients in codominant (OR 95%CI = 2.88 (1.37-6.03) for GC vs CC genotype), recessive (OR 95%CI = 2.72 (1.43-5.17) for GG vs GC/CC genotype) and log additive model (OR 95%CI = 1.63 (1.14-2.34) for G allele).
Notch3 rs1043996 genotype was associated with lower risk for occurrence of ALD in codominant (OR 95%CI = 0.30 (0.13-0.70) for GG vs AA genotype), recessive (OR 95%CI = 0.32 (0.14-0.72) for GG vs GA/AA genotype) and log additive model (OR 95%CI = 0.63 (0.43-0.94) for G allele). On the other hand, there was no significant association between the Notch3 genotype and risk for HCC in any of the investigated models (p>0.05).

Conclusion: Our study shows that PNPLA3 rs738409 is a considerable risk factor for the occurrence of ALD and its progression towards HCC. On the other hand, the Notch3 rs1043996 genotype is associated with a lower risk for the development of ALD but is not significantly associated with a risk of HCC occurrence.

P038YI Urinary volatile organic compounds characterization in hepatocellular carcinoma – pilot study

Ayman Bannaga[1 2], Heena Tyagi[3], Emma Daulton[3], James Covington[3], Ramesh Arasaradnam[1 2]

[1]University Hospital Coventry & Warwickshire NHS Trust, Gastroenterology and Hepatology, Coventry, United Kingdom, *[2]University of Warwick, Warwick Medical School, Coventry, United Kingdom*, *[3]University of Warwick, School of Engineering*
Email: aymanbannaga@yahoo.com

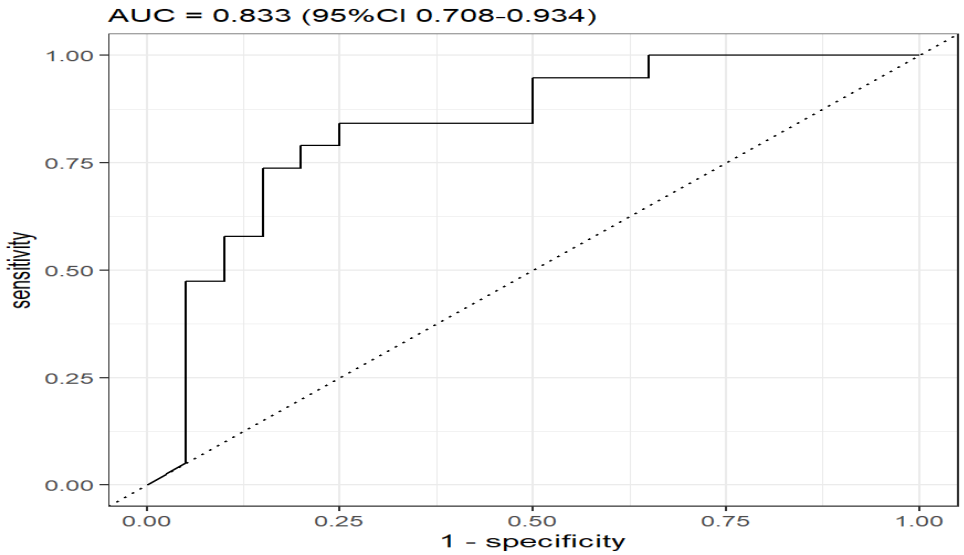
Background and Aims:
Hepatocellular carcinoma (HCC) diagnosis relies on advanced scans that are not easily accessible. Therefore, there is need for rapid noninvasive diagnostic methods for HCC. This pilot study aimed to a) assess utility of urinary volatile organic compounds (VOCs) in patients with HCC and b) characterize potential urinary chemicals present in HCC to enhance our understanding of the tumour biology of HCC

Method:
This study recruited 58 participants. There were 20 HCC cases and 38 controls. Controls included those with liver disease (fibrotic/non-fibrotic) but where HCC were excluded. 5 mL of urine was collected from participants and immediately stored at -80°C. Urine was then analysed by applying gas chromatography ion mobility spectrometry (GC-IMS, G.A.S. FlavourSpec, Germany) and time of flight mass spectrometry (TOF-MS). Data were then statistically analysed using 'R' statistical software by a linear model (Glmnet), through a 10 fold-cross validation method, with the resultant probabilities used to calculate statistical parameters. The study was approved by North-East Yorkshire NHS Ethics Committee (Ref 260179 - 14/06/2019)

Results:
Mean age was 62.8 years. Male to Female ratio was 4 to 1. In HCC, the urinary VOCs showed an AUC (area under the curve) of 0.83 (0.71-0.93, 95% CI), sensitivity of 0.84 (0.69-0.96, 95% CI), specificity of 0.65 (0.47-0.82, 95% CI), p-value 0.0001. ROC (receiver operator curve) is shown in figure 1. From this statistical analysis, we were able to tentatively identify a number of VOCs associated with separating HCC from the control group including ethanol, acetone, toluene and hydrocarbons. TOF-MS results showed further specific urinary VOCs in HCC, these were, 2-Butanone, 4-Methyl-2,4-bis(p-hydroxyphenyl)pent-1-ene (MBP), 2-Hexanone and Oxime. The literature identified multiple VOCs including the ones identified in this study as byproducts of the cytochrome polysubstrate 450 (CYP450) with some VOCs causing negative feedback leading to inhibition of the CYP450 function and accumulation of toxic/carcinogenic VOCs. Of note in this study, Limonene was not significantly abundant in the urine of HCC cases as previously reported in breath of patients with various degrees with liver disease

Conclusion:
Urinary VOCs demonstrate potential role in HCC diagnosis. The chemical identification of the urinary VOCs further enhanced our understanding on the biology of the disease

Figure: ROC curve for urinary VOC and HCC



P039YI Comparison of incidence and mortality risks due to acute kidney injury in hepatocellular carcinoma patients after liver resection, ablation, and transarterial chemoembolization: Meta-analysis of cohort studies

Boby Pratama Putra[1], Felix Nugraha Putra[2]

[1]*Medical doctor, Blitar, Indonesia*, [2]*Universitas Airlangga, Faculty of Medicine, Surabaya, Indonesia*
Email: boby_pratama_putra@yahoo.co.id

Background and Aims: Hepatocellular carcinoma (HCC) is a deadly disease whose increasing prevalence that accounts for the fifth cancer-related mortality. Liver resection, ablation, and transarterial chemoembolization (TACE) are some of the HCC treatment modalities. Previous studies showed that these procedures are associated with incidence and mortality risks due to acute kidney injury (AKI), although the results are still inconclusive. This study aims to measure the incidence and mortality risks due to AKI in HCC patients after these procedures.

Method: We did comprehensive searching in online databases of Pubmed, ScienceDirect, EMBASE, and The Cochrane Library to screen all relevant literature from 2000 - 2020. We included all cohort studies about HCC patients who underwent liver resection, ablation, or TACE, which access the incidence and mortality risks due to AKI. We excluded studies about those procedures in patients besides HCC, patients with chronic kidney disease, animal studies, and non-English publications. Bias risk was accessed by using Risk Of Bias In Non-Randomized Studies - of Interventions (ROBINS-I) tools. The analysis was performed to provide odds ratio (OR) with 95% confidence interval (CI) using random-effect heterogeneity test.

Results: Nineteen cohort studies met inclusion criteria consist of 7 studies about liver resection, 3 studies about ablation, and 11 studies about TACE. Liver resection increases AKI incidence risk although not statistically significant (pooled OR = 1.77, 95% CI = 0.92 – 3.42, p = 0.09, I^2 = 76%), but significantly increases mortality risk due to AKI (pooled OR = 2.07, 95% CI = 1.28 – 3.33, p = 0.003, I^2 = 19%). The TACE procedure significantly increases both risks of AKI incidence (pooled OR = 2.15, 95% CI = 1.36 – 3.39, p = 0.001, I^2 = 77%) and mortality (pooled OR = 3.60, 95% CI = 1.88 – 6.86, p = 0.0001, I^2 = 83%). Surprisingly, ablation procedure decreases AKI incidence risks significantly (pooled OR = 0.50, 95% CI = 0.28 – 0.90, p = 0.02, I^2 = 0%).

Conclusion: Liver resection and TACE are associated with the increased incidence and mortality risks due to AKI in HCC patients after these procedures, but ablation procedure significantly decreases AKI incidence risks. However, further studies are needed to establish the association.

P040YI Predictive value of biomarkers a-fetoprotein (AFP) and fibrogen like protein-2 (FGL-2) for HCC among patients with liver cirrhosis

antonia syriha[1], Ioanna Stathopoulou[1], petros galanis[2], Georgia Barla[1], Stavroula Michalea[1], Kalliopi Tabouratzi[1], tina voukelatou[1], Ioannis Elefsiniotis[1]

[1]National and Kapodistrian University of Athens, Greece, Academic Department of Internal Medicine-Hepatogastroenterology Unit, General and Oncology Hospital of Kifisia "Agioi Anargyroi", Athens, Greece, *[2]National and Kapodistrian University of Athens, Greece*
Email: ielefs@nurs.uoa.gr

Background and Aims: To evaluate the prognostic value of serum biomarkers (AFP and FGL-2) for HCC development among patients with liver cirrhosis.

Method: Between Jan 2015 and Oct 2020, 84 patients with liver cirrhosis (57 males, 59 CPT score A, 41 with esophageal varices, 3 with portal vein thrombosis, 21 with diabetes) without HCC at baseline were prospectively evaluated for the emerge of HCC using the standard surveillance protocol (US and serum AFP levels every 6 months). Serum AFP as well as FGL-2 levels was evaluated at baseline in all cirrhotic patients. Fourteen of them developed HCC during the follow-up period (HCC group) and 70 did not (control group). HCC diagnosis was supported by compatible dynamic CT and MRI findings and was also histologically confirmed in all cases.

Results: Patient age (mean age=65.1 vs 63.5y, p=0.6), gender (p=0.2), etiology of liver cirrhosis (p=0.9), presence of diabetes (p=0.1), presence of varices (p=0.07) or portal vein thrombosis (p=0.4) as well as MELD score (mean 10.9 vs 9.7, p=0.4) were comparable at baseline among HCC and control group. Patients from HCC group had significantly lower platelet count (mean 98.700 vs 147.400, p=0.008) and were frequently categorized as CPT B/C stage (64.3% vs 22.9%, p=0.006) at baseline compared to control group. Serum FGL-2 levels (median 3.6 vs 2.1, p=0.01) at baseline were significantly elevated in patients from HCC group compared to ones from the control group whereas baseline serum AFP levels (median 6.0 vs 4.0 ng/ml, p=0.15) were comparable among the two groups. In the multivariate (logistic regression) analysis, taking into account age, platelet count, CPT score, MELD score as well as baseline serum AFP (< or ≥ 6) and FGL-2 levels (< or ≥ 3.6) only platelets (OR=0.980, 95%CI:0.964-0.997, p=0.025) and baseline CPT score (CPT B/C vs A, OR=27.184, 95%CI:2.815-262.5, p=0.004) were significantly correlated with HCC development during follow-up period.

Conclusion: Liver disease severity according to Child-Pugh score as well as low platelet count (as probably an indirect index of clinical significant portal hypertension) seem to positively influence the appearance of HCC in cirrhotic patients, irrespective of all the other variables evaluated. The utility of serum biomarkers such as AFP and FGL-2 for the prediction of HCC should be further evaluated as it seems that their levels are influenced by cirrhotic background.

Figure:

P041YI Transarterial chemoembolization: Predictors of liver function deterioration prior to hepatocellular carcinoma refractoriness

Joel Silva[1], Helder Cardoso[1], Pedro Costa-Moreira[1], Guilherme Macedo[1]

[1]*São João Universitary Hospital Center, Porto, Portugal*
Email: jom_73@hotmail.com

Background and Aims: Transarterial chemoembolization (TACE) is the first-line treatment for patients with intermediate stage hepatocellular carcinoma (HCC). For patients without an adequate response, a therapeutic stage migration can be considered, with molecular target agents (MTA) approved for advanced stage, however, this requires a preserved liver function. The aim of this study is to evaluate possible predictors of early deterioration of hepatic reserve, prior to TACE refractoriness, in a cohort of patients treated with TACE.

Method: We performed a retrospective cohort study of 99 patients with HCC submitted to TACE during a 10-year period (2010-2020) for intermediate stage HCC or early HCC unfit for other locoregional therapies. Exclusion criteria were absence of cirrhosis, presence of Child-Pugh B/C and presence of vascular invasion or extrahepatic spread. All patients were submitted to a biochemical and medical evaluation prior to initial TACE and a every month after. Response to initial TACE was evaluated at 1 month. Statistical analysis was performed with SPSS® version 24.0.

Results: Mean age was 73 ±13.7 years, 79% male. The etiology of hepatic disease was alcohol in 42%, chronic hepatitis C in 34%, chronic hepatitis B (HBV) in 7%, non-alcoholic steatohepatitis in 5% and other in 11%. Median TACE procedures per patient was 2 (IQR 1-3). Complete response (CR) was observed in 34 patients (34%) after first TACE. Liver function deteriorated to Child-Pugh B/C in 51 (51%) patients and median time to liver function deterioration (TTLFD) was 14 (IQR 8-20) months after first TACE.

In univariable analysis, TTLFD was significantly longer in patients within vs. out up-to-7 criteria (mean, 40.9 vs. 20.3, p=0.041), albumin >38mg/dL (mean 43.7 vs. 24.5 months, p<0.001), billirrubin <2mg/dL (mean, 43.9 vs. 18.9 months, p=0.029) and patients with CR at 1 month after initial TACE vs. non-CR (mean, 56.7 vs. 27.6 months, p=0.002). Patients with HBV presented shorter TTLFD vs. all other etiologies (mean, 9.9 vs. 40.9 months, p=0.003).

In cox-regression, HBV (HR 3.79, p=0.011), outside up-to-7 criteria (HR 2.21, p=0.039), albumin >38mg/dL (HR 0.37, p=0.021) and CR to initial TACE (HR 0.45, p=0.039) associated with TTLFD.

Conclusion: Based on our findings we suggest that up-to-7 criteria and albumin are important determinants of liver function deterioration after initial TACE. HBV etiology might also be a determinant of liver function deterioration. Absence of response to initial TACE appears to present an association with early liver disfunction after first TACE. Patients with predictors of liver function deterioration prior to TACE refractoriness have a smaller window of opportunity to initiate systemic therapy. Such patients might benefit from inclusion in TACE plus MTA combination therapy or MTA monotherapy clinical trials.

P042YI Two birds one stone: a case of synchronous hepatocellular carcinoma and intrahepatic cholangiocarcinoma

Catarina Félix[1], Rui Mendo[1], Catarina O'Neill[1], João Cassis[2], Pedro Magalhães-Costa[1], Cristina Chagas[1]

[1]Centro Hospitalar Lisboa Ocidental, Gastroenterology, Portugal, *[2]Hospital da Luz*
Email: sfelixcatarina@gmail.com

Background and Aims: Hepatocellular carcinoma (CHC) and intrahepatic cholangiocarcinoma (ICC) are the two main primary liver cancers. However, synchronous double primary hepatic cancer is rarely encountered in clinical practice.

Method: A 66-year-old woman presented with two liver nodules in segment V (with 19 and 30mm), without clinical or imagiological findings of liver cirrhosis, suggestive of liver metastasis (albeit imagiological findings of the largest lesion were atypical). Her medical history was remarkable for an endometrial adenocarcinoma, treated 15 years before, without evidence of relapse. After a negative study for occult malignancy, a biopsy of both the smallest lesion and the non-tumoral liver was performed showing: a well differentiated biliary proliferation, with low proliferative index, suggestive of a biliary hamartoma, although not being possible to exclude a well-differentiated cholangiocarcinoma; the histopathological analysis of the non-tumoral liver was consistent with liver cirrhosis. A subsequent study for liver cirrhosis etiology revealed a chronic hepatitis B (previously unknown), for which antiviral treatment was started.

Results: After multidisciplinary evaluation, a right hepatectomy with lymphadenectomy was performed. Liver cirrhosis was confirmed and pathological examination of the largest nodule revealed a 45mm moderately differentiated HCC. Analysis of the smallest nodule revealed a 30mm lesion with dilated cystic bile ducts, containing bile-stained granular material, suggestive of a biliary hamartoma, with atypical cells and irregular ducts, compatible with areas of cholangiocarcinoma.

Conclusion: This case presents a synchronous double primary hepatic cancer (HCC and ICC) in the same liver lobe, an unusual situation, mostly described on patients with hepatitis C-related liver cirrhosis. Although rare, this situation should be considered as a differential diagnosis of liver tumors during the pre-operative evaluation. Also, this case presents a second peculiarity: the possible arising of a cholangiocarcinoma from a biliary hamartoma, pointing to an association between the two entities. At last, the case is paradigmatic for the need of a biopsy of the non-tumoral liver in patients without clinical or imagiological aspects of liver cirrhosis.

Figure: Iconography will be presented.

P043YI Virtual biopsy for diagnosis of steatohepatitis and chemotherapy-associated liver injuries. A combined radiomic and clinical model.

Guido Costa[1 2], Lara Cavinato[3], Chiara Masci[3], Francesco Fiz[4], Martina Sollini[1 4], Letterio Salvatore Politi[1 5], Arturo Chiti[1 4], Luca Balzarini[5], Luca Di Tommaso[1 6], Francesca Ieva[3 7], Alessio Aghemo[1 8], Guido Torzilli[1 2], Luca Vigano[1 2]

[1]Humanitas University, Department of Biomedical Sciences, *[2]Humanitas Clinical and Research Center – IRCCS, Department of Surgery, Division of Hepatobiliary and General Surgery*, *[3]MOX laboratory - Politecnico di Milano, Department of Mathematics*, *[4]Humanitas Clinical and Research Center – IRCCS, Department of Nuclear Medicine*, *[5]Humanitas Clinical and Research Center – IRCCS, Department of Radiology*, *[6]Humanitas Clinical and Research Center – IRCCS, Pathology Unit*, *[7]CADS - Center for Analysis, Decisions and Society, Human Technopole*, *[8]Humanitas Clinical and Research Center – IRCCS, Department of Gastroenterology, Division of Hepatology*
Email: luca.vigano@humanitas.it

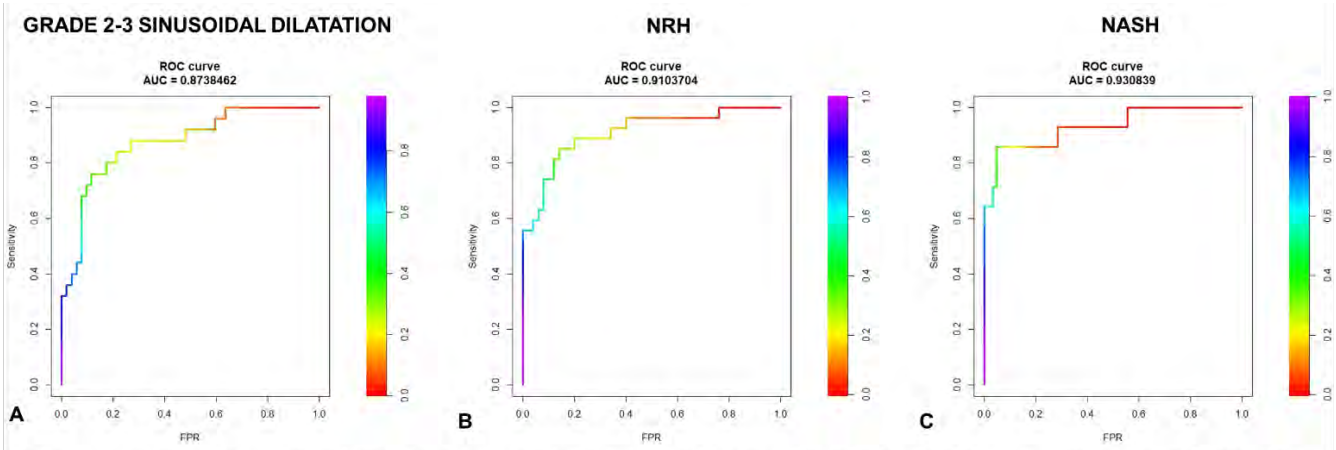
Background and Aims: Chemotherapy-associated liver injuries (CALI) have a major clinical impact, but their non-invasive diagnosis is still an unmet need. To elucidate the contribution of radiomic analyses to diagnosis of sinusoidal dilatation, nodular regenerative hyperplasia (NRH) and non-alcoholic steatohepatitis (NASH).

Methods: Patients undergoing liver resection for colorectal metastases after oxaliplatin- or irinotecan-based chemotherapy between January 2018 and February 2020 were retrospectively analyzed. Radiomic features were extracted from a standardized volume of non-tumoral liver parenchyma outlined in the portal phase of preoperative post-chemotherapy computed tomography (CT). A multivariate logistic regression model was performed to identify predictors of CALI. The model was internally validated.

Results: Overall, 78 patients were analyzed. Of these, 25 (32%) had grade 2-3 sinusoidal dilatation, 27 (35%) NRH, and 14 (18%) NASH. Three fingerprints derived from radiomic features were independent predictors of grade 2-3 sinusoidal dilatation: GLRLM_f3 (OR=12.25), NGLDM_f1 (OR=7.77), and GLZLM_f2 (OR=0.53). The combined clinical/radiomic predictive model had 82% accuracy, 64% sensitivity, and 91% specificity (AUC=0.87 vs AUC=0.77 of the model without radiomics). Three radiomic parameters were independent predictors of NRH: conventional_HUQ2 (OR=0.76), GLZLM_f2 (OR=0.05), and GLZLM_f3 (OR=7.97). The combined clinical/radiomic model had 85% accuracy, 81% sensitivity, and 86% specificity (AUC=0.91 vs AUC=0.85 without radiomic features). One radiomic feature was associated with NASH: conventional_HUQ2 (OR=0.79). Steatohepatitis was predicted with 91% accuracy, 86% sensitivity, and 92% specificity (AUC=0.93 vs AUC=0.83 without radiomic features). In the validation set, accuracy was 72%, 71%, and 91% for sinusoidal dilatation, NRH, and NASH, respectively.

Conclusions: Radiomic analysis of liver parenchyma may provide a signature that, in combination with clinical and laboratory data, improves diagnosis of CALI.

Figure:



ROC curve analysis of the combined clinical, laboratory and radiomic model for the prediction of grade 2-3 sinusoidal dilatation (A), NRH (B) and NASH (C).

P044 Pattern of progression of intrahepatic cholangiocarcinoma influences survival: implications for second-line trials

Francesco Tovoli[1 2], Ingrid Garajova[3], Massimo Iavarone[4], Piera Federico[5], Fabio Gelsomino[6], Alessandro Granito[1 2], Daniele Bruno[5], Antonella Forgione[1 2], Fabio Piscaglia[1 2]

[1]Division of Internal Medicine, IRCCS Azienda Ospedaliero Universitaria di Bologna, Italy, Italy, *[2]University of Bologna, Department of Medical and Surgical Sciences, Italy*, *[3]University Hospital of Parma, Medical Oncology Unit, Parma, Italy*, *[4]Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Division of Gastroenterology and Hepatology, Milan, Italy, Milan, Italy*, *[5]Ospedale del Mare, U.O.C. Oncologia, Napoli, Italy*, *[6]Azienda Ospedaliero-Universitaria di Modena, Dipartimento di Oncologia ed Ematologia, Modena, Italy*
Email: francesco.tovoli2@unibo.it

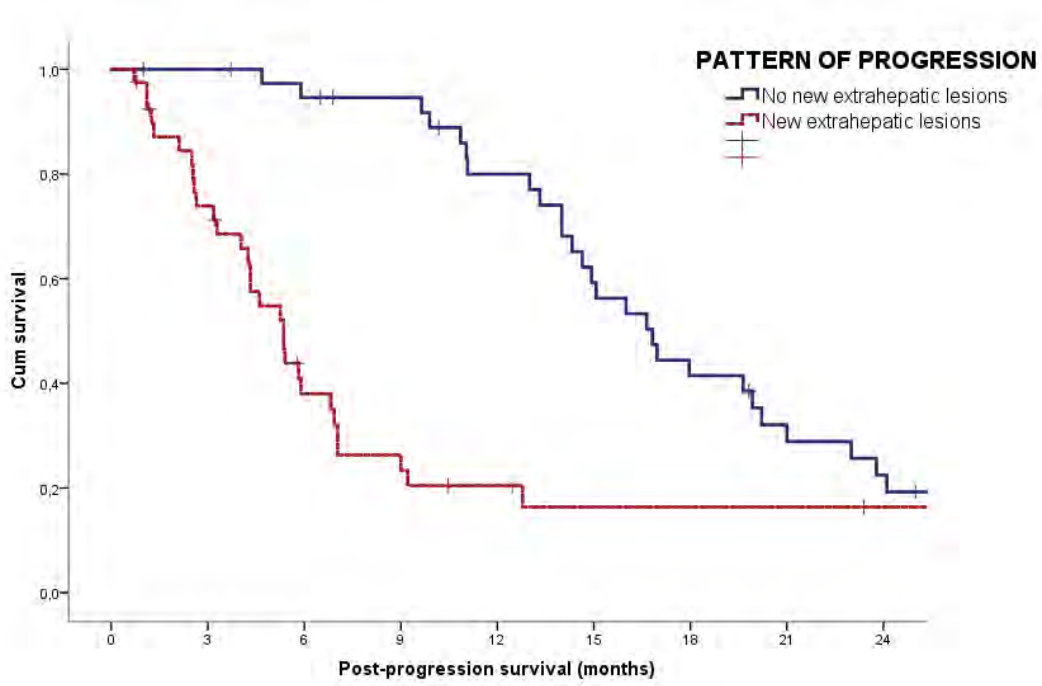
Background and Aims: Intrahepatic cholangiocarcinoma (iCCA) is the second most frequent liver cancer, with an increasing incidence worldwide. Chemotherapy regimens based on gemcitabine and cisplatin are the frontline treatment for unresectable cases, but the overall survival remains poor. For this reason, many second-line trial are ongoing. Survival predictors upon progression are not established, differently from hepatocellular carcinoma, for which postprogression survival (PPS) is driven by the progression pattern.

Method: We reported a multicenter retrospective evaluation of consecutive iCCA patients who progressed after a frontline systemic treatment.Radiological assessment of progression was evaluated according to RECIST 1.1. The progression pattern was divided into: intrahepatic/extrahepatic increase in tumor size, new intrahepatic lesion, and new extrahepatic lesion (NEH).

Results: We included 80 patients (TNM 8th edition stage: IV 32.5%, IIIb 30%, IIIa 5%, II 32.5%), of whom 12 (15%) with liver cirrhosis. The median OS from the start of systemic treatment was 20.5 months and its independent predictors (hazard ratio [HR], 95% confidence interval [CI]) were: baseline main tumour dimension 1.010 [1.003-1.017], PS 2.900 [1.537-5.470], and TTP 0.852 [0.798-0.911]. The presence of NEH was also an independent predictor of OS 2.540 [1.399-4.613] and PPS 2.715 [1.496-4.929]. Amongst the 51 patients eligible for a second-line treatment at the progression, the PPS was 16.8 and 5.9 months in cases without and with NEH, respectively (p=0.001).

Conclusion: PPS is influenced by progression pattern and this is key in prognostic prediction and second-line trial design and analysis.

Figure:


PATTERN OF PROGRESSION
No new extrahepatic lesions
New extrahepatic lesions
Cum survival
1,0
0,8
0,6
0,4
0,2
0,0
0
3
6
9
12
15
18
21
24
Post-progression survival (months)

P045YI Hepatic and extrahepatic colorectal metastases have discordant responses to systemic therapy. Pathology data from patients undergoing simultaneous resection of multiple tumor sites

Pio Corleone[1 2], Luca Viganò[1 3], Luca Di Tommaso[3 4], Shadya Sara Darwish[1], Lorenza Rimassa[3 5], Lorenzo Viggiani[3], Guido Torzilli[1 3]

[1] Humanitas Clinical and Research Center - IRCCS, Rozzano, Milan, Italy, Division of Hepatobiliary and General Surgery – Department of Surgery, Rozzano, Milan, Italy, *[2]University of Trieste, Trieste, Italy, Department of General Surgery, Trieste, Italy*, *[3] Humanitas Clinical and Research Center - IRCCS, Rozzano, Milan, Italy, Department of Biomedical Science, Pieve Emanuele, Milan, Italy*, *[4] Humanitas Clinical and Research Center - IRCCS, Rozzano, Milan, Italy, Department of Pathology, Rozzano, Milan, Italy*, *[5] Humanitas Clinical and Research Center - IRCCS, Rozzano, Milan, Italy, Medical Oncology and Hematology Unit, Rozzano, Milan, Italy*
Email: corleone.pio@gmail.com

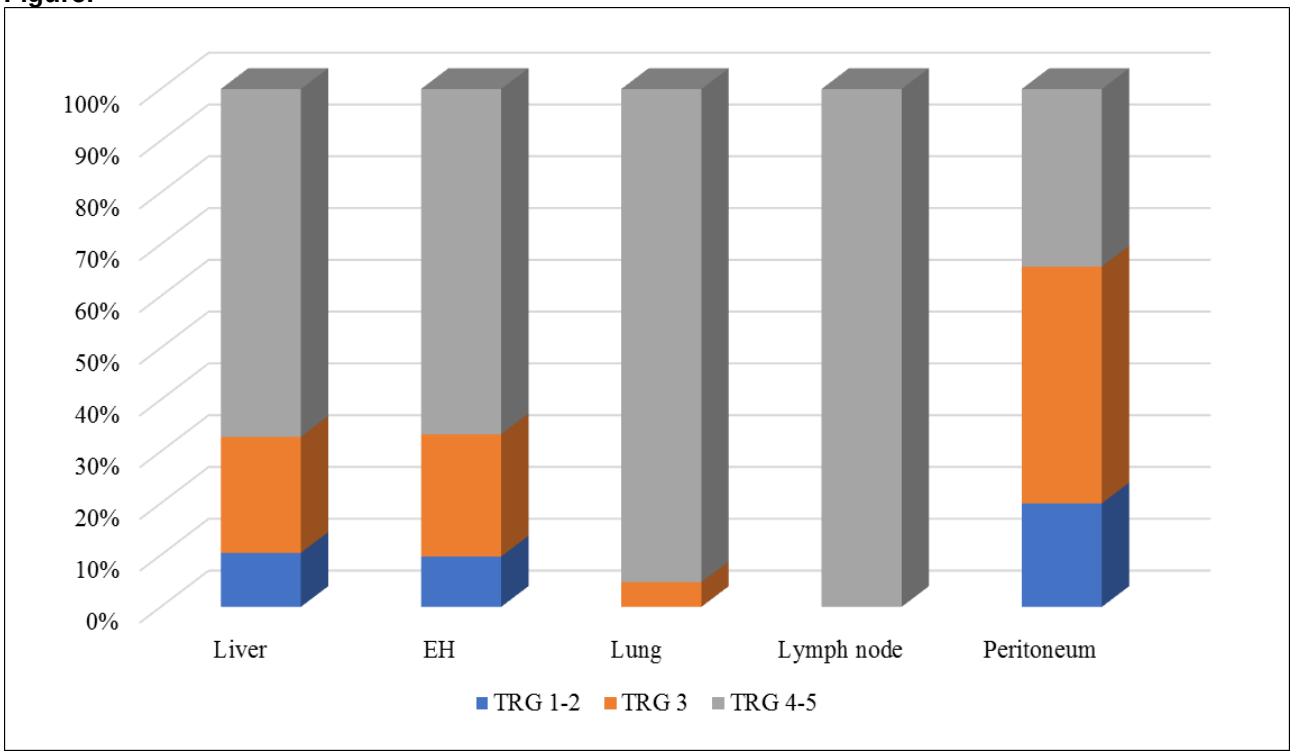
Background and Aims: Systemic therapy is the standard treatment for patients with hepatic and extrahepatic colorectal metastases. It is assumed to have the same effectiveness on all disease foci, independent of the involved organ. To compare the response rates of hepatic and extrahepatic metastases to systemic therapy.

Method: All consecutive patients undergoing resection for colorectal metastases were considered. Patients undergoing simultaneous resection of hepatic and extrahepatic metastases after preoperative chemotherapy were analyzed. Specimens of hepatic and extrahepatic metastases were reviewed. Pathological response to chemotherapy was classified according to tumor regression grade (TRG).

Results: We analyzed 45 patients undergoing resection of 134 hepatic and 72 extrahepatic metastases. Extrahepatic disease was peritoneal in 21 patients, pulmonary in 15, lymph nodal in 14, and adrenal in one (multiple organs in five patients). All patients had oxaliplatin- and/or irinotecan-based preoperative chemotherapy; 60% had associated targeted therapies. Lung and lymph node metastases had lower response rates to chemotherapy than liver metastases (TRG 4–5 in 95% and 100% vs. 67%, *P*=0.008 and *P*=0.006, respectively). Peritoneal metastases had a higher pathological response rate than liver metastases (TRG 1–3 66% vs. 33%, *P*<0.001) and non-hepatic non-peritoneal metastases (vs. 3%, *P*<0.001). Multivariate analysis identified metastases site, metastasis size and targeted therapies as predictors of pathological response to systemic therapy.

Conclusion: Response to chemotherapy of distant metastases from colorectal cancer varies in different organs. Systemic treatment is highly effective for peritoneal metastases, more so than liver metastases, while it has a very poor impact on lung and lymph node metastases.

Figure:


100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Liver
EH
Lung
Lymph node
Peritoneum
TRG 1-2
TRG 3
TRG 4-5

P046 A phase Ib study of pembrolizumab following trans-arterial chemoembolization (TACE) in hepatocellular carcinoma (HCC): PETAL.

David J. Pinato[1], Thomas Talbot[2], Robert Thomas[3], Anwar Sayed[2 4], Elias Allara[2 5], Paul Tait[3], Paul J. Ross[6], Anna Mary Young[7], Tom Cole[8], Robert Goldin[9], Caroline Ward[2], Ayse Akarca[10], Jesus Miguens Blanco[11], Teresa Marafioti[10], Saskia Killmer[12], Bertram Bengsch[12], Julian Marchesi[11 13], rohini sharma[1]

[1]Imperial College London, Department of Surgery and Cancer, London, United Kingdom, *[2]Imperial College London, Department of Surgery and Cancer, London,* , *[3]Imperial College Healthcare NHS Trust, Interventional Radiology, London, United Kingdom*, *[4]Taibah University, Department of Medical Microbiology and Immunology, Medina, Saudi Arabia*, *[5]University of Cambridge, Cardiovascular Epidemiology Unit, Cambridge, United Kingdom*, *[6]Guy's & St Thomas' NHS Foundation Trust, Department of Medical Oncology, London,* , *[7]St George's University Hospitals NHS Foundation Trust, Department of Medical Oncology, London, United Kingdom*, *[8]Imperial College London, NIHR Imperial Clinical Research Facility, London, United Kingdom*, *[9]Imperial College London, Centre for Pathology, London, United Kingdom*, *[10]University College London Hospital, Department of Histopathology, London, United Kingdom*, *[11]Imperial College London, Division of Digestive Diseases, London, United Kingdom*, *[12]University Hospital Freiburg, Department of Internal Medicine, Freiburg, Germany*, *[13]Cardiff University, School of Biosciences, Cardiff, United Kingdom*
Email: david.pinato@imperial.ac.uk

Background and Aims: The efficacy of TACE is secondary to its dual ischaemic and cytotoxic effect, which promotes immunogenic tumour cell death. We hypothesized that TACE will prime adaptive immunity and enhance pembrolizumab efficacy (pembro; anti-PD-1). The aim of this phase Ib study was to evaluate safety, preliminary activity of combination therapy and explore mechanisms of efficacy.

Method: Up to 32 patients (pts) with intermediate-stage HCC were planned to receive up to 2 rounds of TACE followed by pembro 200 mg q3w 30-days post-TACE until disease progression or unacceptable toxicity for up to 1-year. Primary endpoint was safety with dose-limiting toxicities (DLT) emerging from the combination being evaluated over a 21-day window from commencement of pembro. Secondary endpoints included progression-free survival (PFS) and evaluation of tumour and host determinants of response in tissue, blood and stool samples.

Results: Of 11 eligible pts, 82% were males, 18% HCV-positive, 55% ECOG PS 0 with a median age of 68 years. Child-Pugh (CP) class was A in 10 pts and B7 in 1 pt. Median tumour size was 4 cm, and median number of tumour nodules was 2. Six pts received pembro after 1 TACE, 5 pts after 2. Pembro yielded no synergistic toxicity with TACE and no DLTs were reported. All-grade adverse events potentially related to treatment (tx) occurred in 90% of pts most commonly skin rash (45%) and fatigue (45%). Median PFS was 9.7 months (95%CI 4.9-14.4) from TACE and 6.1 months (95%CI 3.8-8.3) from pembro initiation. Cause of withdrawal included disease progression (n=7), adverse events (n=1) worsening liver failure in the CP B7 pt, non tx-related (n=1) and withdrawal due to Covid-19 pandemic (n=2). We document dynamic changes in peripheral T-cell subsets and in stool bacterial metagenomics highlighting potential mechanisms of synergy.

Conclusion: The TACE plus pembro combination had a tolerable safety profile with no evidence of synergistic toxicity. Alongside emerging efficacy data, this encourages the clinical development of the combination in CP A pts.

Figure:

P047YI Hsa-miR-21-5p disturbs the liver metabolic network in patients with NAFLD and is increased in NASH-associated HCC, contributing to hepatocarcinogenesis in mice

Pedro Miguel Rodrigues[1 2], Marta B. Afonso[1], André L. Simão[1], Tawhidul Islam[1], Maria Manuela Gaspar[1], Colm O Rourke[3], Monika Lewinska[3], Jesper Andersen[3], Cristina Alonso[4], Enara Arretxe[4], Álvaro Santos-Laso[2], Raul Jimenez-Aguero[2], Emma Eizaguirre[2], María Jesús Pareja[5], Luis Bujanda[2], Carina PRIP-BUUS[6], Jesus Maria Banales[2 7 8], Cecília M. P. Rodrigues[1], Rui E. Castro[1]

[1]Research Institute for Medicines (iMed. ULisboa), Faculty of Pharmacy, Universidade de Lisboa, Lisbon, Portugal, *[2]Biodonostia Health Research Institute, Liver Diseases, San Sebastian, Spain*, *[3]Biotech Research and Innovation Centre, Department of Health and Medical Sciences, University of Copenhagen, Copenhagen, Denmark*, *[4]OWL Metabolomics, Bilbao, Spain*, *[5]Hospital Juan Ramón Jiménez, Huelva, Spain*, *[6]Université Paris Descartes UMR-S1016, Institut Cochin,, Paris, France*, *[7]Centre for the Study of Liver and Gastrointestinal Diseases (CIBERehd), Carlos III National Institute of Health, Madrid, Spain*, *[8]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain*
Email: pedro.rodrigues@biodonostia.org

Background and Aims: The molecular mechanisms governing progression of non-alcoholic steatohepatitis (NASH) towards hepatocellular carcinoma (HCC) remains elusive. We have recently shown that concomitant hsa-miR-21-5p ablation and obeticholic acid prevents NASH development in mice. Here, we aimed to evaluate the role of hsa-miRNA-21-5p in NASH-associated carcinogenesis.

Method: hsa-miR-21-5p levels were evaluated in 2 large international cohorts of patients with biopsy-proven NAFLD (SS cohort; n=199), HCC (TCGA cohort; n=356) and NAFLD-associated HCC (TCGA cohort; n=19), and correlated with clinicopathological findings. Liver metabolomic profiles were evaluated in NAFLD patients. Wild-type (WT) and *MiR21* KO mice were fed either a choline-sufficient, amino acid-defined (CSAA) diet or a choline-deficient, amino acid-defined (CDAA) diet for 32 and 66 weeks and processed for histological and molecular analysis. A profiler PCR array was used to evaluate expression of liver cancer-related genes.

Results: hsa-miR-21-5p expression was significantly increased with disease severity in patients with NAFLD (steatosis, lobular inflammation, ballooning, fibrosis and NAS score). Liver metabolomic analysis revealed distinct metabolomic profiles in NAFLD patients with high hsa-miR-21-5p levels, concordant with more severe disease stages, when compared with patients with lower hsa-miR-21-5p expression. WT CDAA-fed mice displayed increased hsa-miR-21-5p expression and progressively developed NASH, fibrosis and preneoplastic nodules, presenting hyperplastic foci, anisokaryosis, highly proliferative hepatocytes and deregulated cancer-related pathways. Contrariwise, *MiR21* KO mice presented with increased and activated levels of hsa-miR-21-5p target PPARα, augmented mitochondrial activity and decreased serum fatty acid levels. Strikingly, hsa-miR-21-5p deficiency ameliorated disease pathogenesis, decreasing liver injury and NAS (<5), with the pro-inflammatory/fibrogenic milieu reversed to baseline levels, paralleling a reduction in hepatocyte proliferation and oncogenic pathways, thus hampering NASH-associated carcinogenesis. Finally, the hsa-miRNA-21-5p/PPARα pathway was significantly deregulated in patients with HCC and NASH-associated HCC, correlating with tumor markers and worse prognosis.

Conclusion: Activation of the miR-21 pathway contributes to NASH-associated carcinogenesis. Targeting miR-21 may constitute an appealing therapeutic approach to ameilorate NASH and its progression towards HCC. (PTDC/MED-PAT/31882/2017, FCT, Portugal and EU H2020 Marie Sklodowska-Curie 722619 grant).

Figure:

P048YI Balloon occluded TACE (B-TACE) vs DEM-TACE for HCC. A single center retrospective case control study.

Pierleone Lucatelli[1], Bianca Rocco[1], Leonardo Teodoli[1], Claudio Trobiani[1], Simone Zilahi De Gyurgyokai[1], Stefano Cipollari[1], Alessandro Cannavale[1], Pier Giorgio Nardis[1], Mario Corona[1], Mario Bezzi[1]

[1]*University La Sapienza, Rome, Policlinico Umberto I, Radiology and Interventional Radiology , Rome, Italy*
Email: pierleone.lucatelli@gmail.com

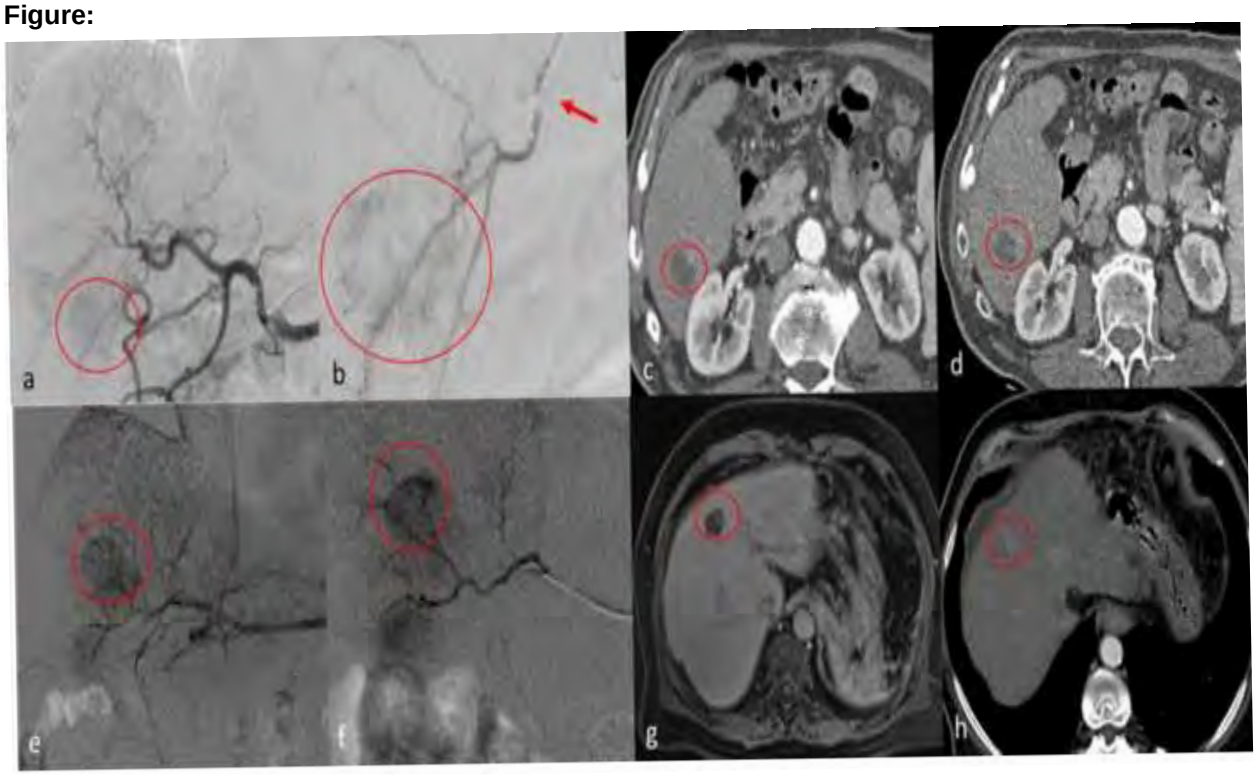
Background and Aims: To compare oncological results and safety profile of balloon micro-catheter transarterial chemoembolization (b-TACE) and drug-eluting-microsphere (DEM-TACE) in patients with hepatocellular-carcinoma (HCC)

Method: This is a case-control, retrospective, single-center study. Between January-2015/March-2019, 149 patients (131 males [87.9%]) with 226 HCC were treated, 22 patients (35 HCC; 19 [86.4%] males) with b-TACE and 127 with DEM-TACE (191 HCC, 112 [88.2%] males). Embolization protocol was standardized (sequential 100±25 and 200±25 μm microspheres). Results were evaluated by modified-response-evaluation-criteria-in-solid-tumor [mRECIST] at 1, 3-6 and 9-12 months and time to recurrence after complete response [TTR] at 1 year. Cox's regression weighted with tumor dimensions was performed. Adverse events (AEs) were recorded.

Results: mRECIST oncological response at all time points (1, 3-6 and 9-12 months) for both treatments were similar, with the exception of Objective response rate at 9-12months. Objective response at 1 and 3-6 months between b-TACE vs DEM-TACE [23/35 (65.7%) vs 119/191 (62.3%), 21/29 (72.4%) vs 78/136 (57.4%) (p>0.05), respectively]. On the contrary, at 9-12 months, it was significantly higher in b-TACE subgroup than DEM-TACE (15/19 [78.9%] vs 48/89 [53.9%], p=0.05). TTR for complete response at 1 year had a better trend for b-TACE vs DEM-TACE (278.0 days [196.0-342.0] vs 219.0 days [161.0-238.0], OR 0.68 [0.4-1.0], p=0.10). The use of balloon micro-catheter reduced the relative risk of the event of recurrence by 0.63 [CI95% 0.38-1.04]; p=0.07). No significant differences were found in AEs rate.

Conclusion: b-TACE showed a trend of better oncological response over DEM-TACE with and longer TTR with a similar adverse events rate, in patients presenting with larger tumors.

Figure:



P049YI Perfusion analysis with dynamic contrast enhanced ultrasound for the diagnosis of hepatocellular carcinoma: a pilot study of a novel parameter aiming to improve the detection of wash-out

Lorenzo Mulazzani[1], Antonella Forgione[2], Chiara Di Bonaventura[2], Eleonora Terzi[1], Laura Gramantieri[1], Andrej Lyshchik[3], Fabio Piscaglia[1 2]

[1]IRCCS Azienda Ospedaliero Universitaria di Bologna, Division of Internal Medicine, Bologna, Italy, *[2]University of Bologna, Department of Medical and Surgical Sciences, Bologna, Italy*, *[3]Thomas Jefferson University Hospital, Department of Radiology, Philadelphia, United States*
Email: lorenzo.mulazzani2@studio.unibo.it

Background and Aims:
Wash-out is a hallmark for the diagnosis of hepatocellular carcinoma (HCC) but may be missed by contrast-enhanced ultrasound (CEUS) operators since typically of mild degree.

Dynamic contrast enhanced ultrasound (DCE-US) quantifies ehnacement signals and could improve the detection of wash-out.

The aim is to verify whether DCE-US increases the sensitivity in the detection of wash-out in small HCC.

Method:

We took advantage of a series of 119 patients at risk of HCC with 138 nodules (diameter 5-50mm) who had been prospectically submitted to CEUS within a validation study of the LI RADS system. For our DCE-US study we selected those with available CEUS video clip in the late phase of contrast enhancement. Diagnostic reference was histology or CT/MRI within 4 weeks plus follow up.

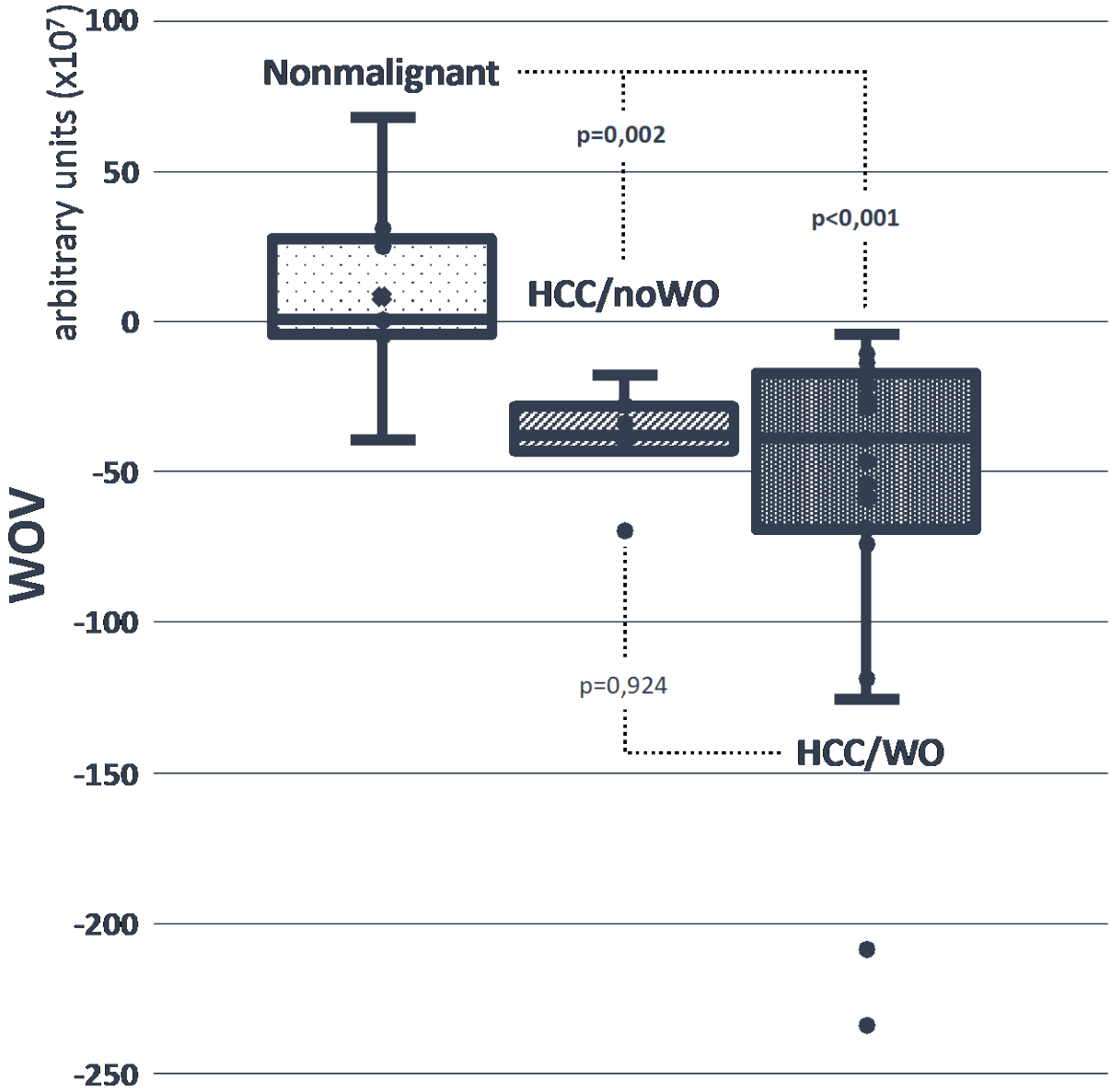
The final study population included 39 nodules: 30 HCC and 9 lesions classified as follows: 4 TC/MRI LI-RADS 4, 2 LI-RADS 3 and 3 not visible with CT/MRI.

DCE-US was carried out with VueBox® to produce time/intensity curves. The median value of all frame-by-frame differences in nodule – parenchyma signal intensity was calculated and named wash-out value (WOV): negative values indicate occurrence of wash-out.

Results:
CEUS showed wash-out in 23/30 HCC according to the operator who performed the examination. DCE-US detected wash-out (WOV<0) in 30/30 HCC and also in 5/9 of the remaining nodules. Median WOV values were lower in HCC (figure).

Conclusion:
This preliminary study demonstrates that WOV obtained with DCE-US might be a promising tool for the detection of wash-out of HCC, potentially improving CEUS sensitivity.

Figure: Box and whisker plots of WOV according to the nature of the lesion and the presence of wash-out at CEUS (WO/noWO) as judged by the operator at the time of examination.



	N	Median	min	max
Nonmalignant	9	0,5 x10^7	-39,7 x10^7	67,8 x10^7
HCC/noWO	7	-38,2 x10^7	-69,2 x10^7	-18 x10^7
HCC/WO	23	-38,9 x10^7	-234 x10^7	-4,7 x10^7

HCC/noWO: HCC without wash-out at CEUS. **HCC/WO:** HCC with wash-out at CEUS.

P050 Chronic disruption of the late cholesterol synthesis leads to female-prevalent liver cancer

Kaja Blagotinšek Cokan[1], Ziga Urlep[2], Gregor Lorbek[2], Madlen Matz-Soja[3], Cene Skubic[1], Martina Perše[4], Jera Jeruc[5], Peter Juvan[1], Tadeja Rezen[1], Damjana Rozman[6]

[1]University of Ljubljana, Faculty of Medicine, Centre for Functional Genomics and Bio-Chips, Institute of Biochemistry and Molecular Genetics , Ljubljana, Slovenia, *[2]University of Ljubljana, Faculty of Medicine, CFGBC, Institute of Biochemistry , Ljubljana, Slovenia*, *[3]University of Leipzig, Faculty of Medicine, Rudol-Schönheimer-Institute of Biochemistry, Divison of General Biochemistry, Leipzig , Germany*, *[4]University of Ljubljana, Faculty of Medicine, Medical Experimental Centre, Institute of Pathology, Ljubljana, Slovenia*, *[5]University of Ljubljana, Faculty of Medicine, Institute of Pathology, Ljubljana , Slovenia*, *[6]University of Ljubljana, Faculty of Medicine, Centre for Functional Genomics and Bio-Chips, Institute of Biochemistry and Molecular Genetics , Ljubljana , Slovenia*
Email: damjana.rozman@mf.uni-lj.si

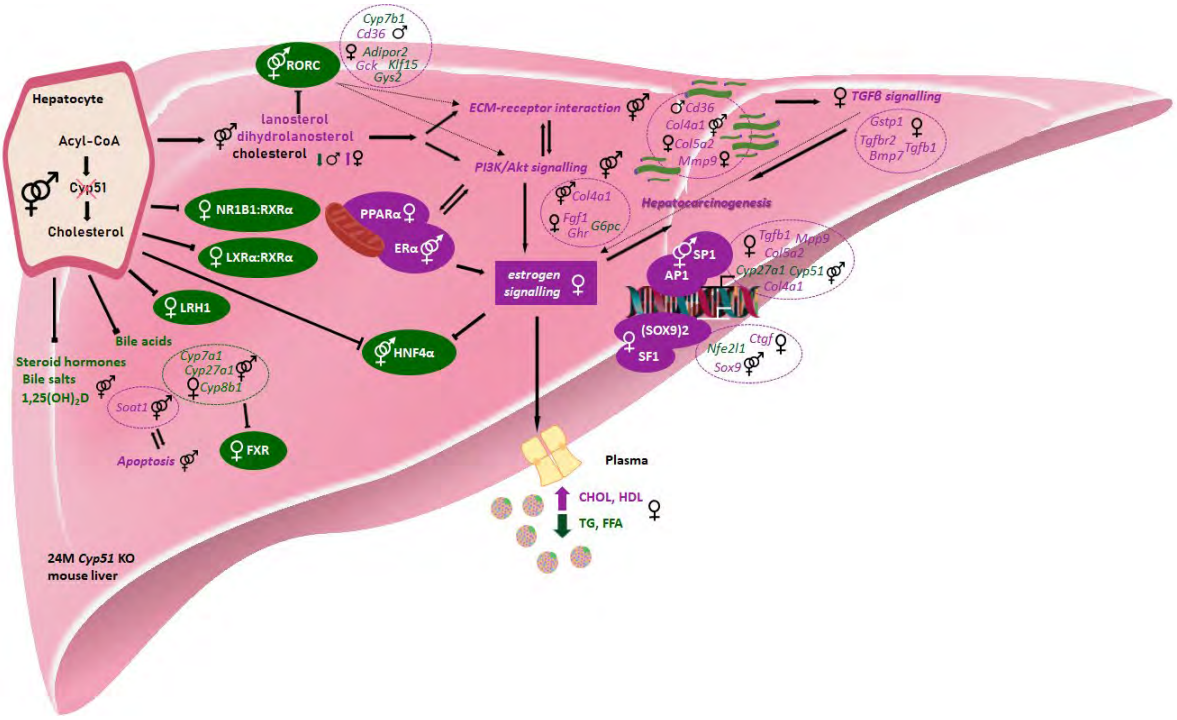
Background and Aims:
The sex disparity in liver pathologies is far from conclusive. There is increasing evidence that after the menopause MAFLD, the metabolic associated liver disease that can progress to hepatocellular carcinoma (HCC), occurs at a higher rate in women. However, HCC generally prevails in the males. The aim of this study was to unravel the development of cholesterol associated and sex-dependent mechanism of hepatocellular carcinoma in a mouse model with defected cholesterol synthesis during the aging, in light of increased incidence of liver cancers in the post-menopausal women.

Methods:
The creation of liver conditional knockout mice (*Cyp51* LKO) was described earlier (*Lorbek et al., Sci. Rep., 2015*). Livers and blood plasma of 1–2 year old mice were examined histologically and by multiple biochemical and statistical approaches. Metabolic and transcription factor networks were deduced from the liver transcriptome data (R/limma controlling false discovery rate at α=0.05), combined by sterol metabolite and blood parameter analyses. KEGG and TRANSFAC databases were used for functional enrichment studies and interpreted with relevance to humans.

Results:
Tumors develop in knock-out mice after year one, with 2:1 prevalence in the females. Female knock-outs show increased plasma cholesterol and HDL, dampened lipid-related transcription factors FXR, LXRα:RXRα, and importantly, crosstalk between reduced LXRα and activated TGF-β signalling, indicating a higher susceptibility to HCC in the aging females. PI3K/Akt signalling and ECM-receptor interaction are common pathways that are disturbed by sex-specific altered genes. Additionally, transcription factors (SOX9)2 and PPARαwere recognized as important for female hepatocarcinogenesis, while overexpressed *Cd36*, a target of nuclear receptor RORC, is a new male-related regulator of ECM-receptor signalling in hepatocarcinogenesis.

Conclusion:
We uncover the sex-dependent metabolic reprogramming of cholesterol-related pathways that predispose for hepatocarcinogenesis in aging females. This is important in light of increased incidence of liver cancers in post-menopausal women.

Figure: Sex-dependent metabolic and transcriptional changes after disrupted *Cyp51* from cholesterol synthesis align with hepatocarcinogenesis in humans. Arrows- connections between enriched genes, pathways, TFs. Blocked arrows- repression. Hatched arrows – regulation. Violet- positively; Green - negatively enriched.



P051 Real-life clinical data of cabozantinib for unresectable hepatocellular carcinoma: efficacy, safety, and prognostic factors.

Francesco Tovoli[1], Vincenzo Dadduzio[2], Stefania De Lorenzo[3], Lorenza Rimassa[4 5], Gianluca Masi[6], Massimo Iavarone[7], Fabio Marra[8], Ingrid Garajova[9], Maria Pia Brizzi[10], Daniele Bruno[11], Franco Trevisani[12], Carlo Messina[13], Francesco Di Clemente[14], Sara Pini[15], Giuseppe Cabibbo[16], Alessandro Granito[1], Mario Domenico Rizzato[2 17], Vittorina Zagonel[2], Giovanni Brandi[1], Tiziana Pressiani[4 5], Piera Federico[11], Caterina Vivaldi[6], Irene Bergna[7], Claudia Campani[8], Fabio Piscaglia[1]

[1]Division of Internal Medicine, IRCCS Azienda Ospedaliero Universitaria di Bologna, Italy, Italy, *[2]Istituto Oncologico Veneto, IRCCS, Padova, Italy., Medical Oncology Unit 1, Padova, Italy*, *[3]Oncologia Medica, IRCCS Azienda Ospedaliero Universitaria di Bologna, Italy, Italy*, *[4]Humanitas Cancer Center, Humanitas Clinical and Research Center-IRCCS, Rozzano, Milan, Italy., Medical Oncology and Hematology Unit,, Milano, Italy*, *[5]Humanitas University, Pieve Emanuele, Milan, Italy., Department of Biomedical Sciences, Milano, , [6]Pisa University Hospital, Department of Medical Oncology, , Pisa, Italy*, *[7]Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, Division of Gastroenterology and Hepatology, Milano, Italy*, *[8]University of Florence, Department of Experimental and Clinical Medicine,, Firenze, Italy*, *[9]University Hospital of Parma, Medical Oncology Unit,, Parma, Italy*, *[10]A.O.U. S.Luigi Gonzaga, 10043 Orbassano (Torino), Italy., Torino, Italy*, *[11]Ospedale del Mare, U.O.C. Oncologia, Napoli, Italy*, *[12]Semeiotica Medica, IRCCS Azienda Ospedaliero Universitaria di Bologna, Italy, Italy*, *[13]Santa Chiara Hospital, Oncology Unit, Trento, Italy*, *[14]Unità Operativa Semplice Dipartimentale (UOSD) "Oncologia medica" del Valdarno, Italy, Italy*, *[15]AUSL Romagna, Division of Oncology, Rimini, Italy*, *[16]University of Palermo, Department of Health Promotion Sciences Maternal and Infant Care, Section of Gastroenterology & Hepatology, Internal Medicine and Medical Specialties, PROMISE, Palermo, Italy*, *[17]Unversity of Padua, Department of Surgery, Oncology and Gastroenterology, Padua, Italy*
Email: francesco.tovoli2@unibo.it

Background and Aims: Cabozantinib has been approved by the European Medicine Agency (EMA) as a second or third-line therapy for hepatocellular carcinoma (HCC) previously treated with sorafenib. Cabozantinib is also being tested in combination with immune checkpoint inhibitors in the frontline setting. Real-life clinical data of cabozantinib for HCC are still lacking. Moreover, the prognostic factors for HCC treated with cabozantinib have not been investigated..

Method: We evaluated clinical data and outcome of HCC patients who received cabozantinib in the legal context of named patient use in Italy.

Results: Ninety-six patients from 15 centres received cabozantinib. All patients had preserved liver function (Child-Pugh A), mostly with an advanced HCC (77.1%) in a third-line setting (75.0%). Prevalence of performance status (PS)>0, macrovascular invasion (MVI), extrahepatic spread, and alpha-fetoprotein (AFP)>400 ng/ml was 50.0, 30.2, 67.7, and 44.8%, respectively. Median overall and progression-free survivals were 12.1 (95% CI 9.4-14.8) and 5.1 months (3.3-6.9), respectively. Most common treatment-related adverse events (AEs) were fatigue (67.7%), diarrhoea (54.2%), anorexia (45.8%), HFSR (43.8%), weight loss (24.0%), and hypertension (24.0%). Most common treatment-related Grade 3-4 AEs were: fatigue (6.3%), HFSR (6.3%), and increased aminotransferases (6.3%). MVI, ECOG-PS>0, and AFP>400 ng/ml predicted a worse OS. Discontinuation for intolerance and no new extrahepatic lesions at the progression were associated with better outcomes.

Conclusion: In a real-life Western scenario (mostly in a third-line setting), cabozantinib efficacy and safety data were comparable with those reported in its registration trial. Data regarding the prognostic factors might help in patient selection and design of clinical trials.

Figure:

P052YI Human antigen R (HuR) SUMOylation is a fine-tuner of hepatocellular carcinoma (HCC) progression via the modulation of key mitochondrial mRNAs

Sofia Lachiondo-Ortega[1], Jorge Simón[1], Fernando Lopitz Otsoa[2], Teresa Cardoso Delgado[1], Naroa Goikoetxea[1], Marina Serrano-Macia[1], Maria Mercado-Gómez[1], Irene González-Recio[1], Rubén Rodríguez Agudo[1], David Fernández Ramos[2], Ruba Al-Abdulla[3], Alejandro Velázquez-Cruz[4], Mikel Azkargorta[5], Joris Guyon[6], thomas daubon[6], Cesar Augusto Martín[7], Juan Diego Riveros Zalamea[8], Nicola G A Abrescia[8], Felix Elortza[5], Ana María Aransay[9], Myriam Gorospe[10], Amaia Lujambio[11], Luis Alfonso Martinez-Cruz[1], Antonio Diaz-Quintana[4], Irene Diaz-Moreno[4], Manuel S Rodríguez[12], María Luz Martínez-Chantar[1]

[1]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Liver Disease Lab, Derio, Spain, *[2]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Precision Medicine and Metabolism Lab, Derio, Spain*, *[3]Biomedical Research Institute of Salamanca (IBSAL), Laboratory of Experimental Hepatology and Drug Targeting (HEVEFARM), Salamanca, Spain*, *[4]Universidad de Sevilla, Instituto de Investigaciones Químicas (IIQ) - Centro de Investigaciones Científicas Isla de la Cartuja (cicCartuja), Sevilla, Spain*, *[5]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Proteomics Platform, Derio, Spain*, *[6] Institut national de la santé et de la recherche médicale (Inserm), Angiogenesis and Cancer Lab, Pessac, France*, *[7]Centro Superior de Investigaciones Científicas (CSIC) and University of the Basque Country, Biofisika Institute, Leioa, Spain*, *[8]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Molecular Recognition and Host-Pathogen Interactions Programme, Derio, Spain*, *[9]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Genome Analysis Platform, Derio, Spain*, *[10]National Institute on Aging Intramural Research Program, National Institutes of Health (NIH), Laboratory of Genetics and Genomics, Baltimore, United States*, *[11]Icahn School of Medicine at Mount Sinai, Department of Oncological Sciences, New York, United States*, *[12]Advanced Technology Institute in Life Sciences (ITAV)-CNRS-IPBS, UbiCARE, Toulouse, France*
Email: mlmartinez@cicbiogune.es

Background and Aims: As we await the results from ongoing phase III clinical trials, we sought to identify additional approaches for the management of hepatocellular carcinoma (HCC). Considering the numerous and complex molecular mechanisms underlying the malignant transformation of a healthy liver into HCC, drugs against more than one signaling route would need to be developed in order to stop the progression of the disease. In this context, the posttranslational modification (PTM) of proteins, which controls the specificity, timing, duration and amplitude of virtually all cellular processes, has emerged as a robust and multidimensional therapeutic strategy in cancer. Interestingly, PTMs are key mechanisms that regulate the function of the RNA-binding protein Human antigen R (HuR), which is known to be involved in HCC transformation, in addition to playing a role in liver physiology. Moreover, a Gene Set Enrichment Analysis (GSEA) based on The Cancer Genome Atlas (TCGA) mRNA expression repository associated HuR expression with the ubiquitin-like PTM SUMOylation in HCC. Given that SUMOylation has never been described for HuR, the main aim of the present work is to elucidate the pathophysiological role of this PTM in liver cancer.

Method: A novel protein pulldown technology based on GST-tagged SUMO Binding Entities (SUBEs) was used to capture the native SUMO-interacting proteome from human and mouse liver tissue and cell line extracts, in combination with Western Blotting and Liquid Chromatography-tandem Mass Spectrometry (LC-MS/MS) for protein identification purposes. Additionally, human HCC HuH-7 cell lines that stably express the wild-type and the SUMOylation mutant versions of HuR were developed in-house and their phenotypes studied.

Results: HuR is SUMOylated in the tumor sections of HCC patients in contrast to the surrounding tissue, as well as in the *MYC-luc;sg-p53* genetically engineered mosaic mouse model of liver cancer, and in human hepatoma cell lines. SUMOylation of HuR promotes major cancer hallmarks, namely proliferation and invasion, in the HuH-7 cell line. Conversely, the absence of HuR SUMOylation results in a senescence-like phenotype with damaged mitochondrial structure and function. Regarding the mechanism of action, we propose that HuR SUMOylation might drive HCC progression by modulating mitochondrial structural integrity and functionality through the regulation of the stability and translation of mRNAs encoding key mitochondrial proteins.

Conclusion: SUMOylation constitutes a novel mechanism of HuR regulation that could be potentially exploited as an aptamer-based therapeutic strategy for liver cancer, thus highlighting the importance of PTMs as disease targets. Furthermore, understanding the effects of HuR SUMOylation in hepatocarcinogenesis will provide new functional insights into the relatively unknown role of SUMOylation in cancer.

P053 GALAD Score shows improved detection of early stage hepatocellular carcinoma in a Caucasian cohort of chronic Hepatitis B and C

Clemens Schotten[1], Bastian Ostertag[1], Paul Peter Manka[1], Gudrun Hilgard[1], Claudio Marquardt[1], Lars Bechmann[2], Ali Canbay[2], Christian Lange[1], Philip Johnson[3], Heiner Wedemeyer[1 4], jan best[1 5]

[1]Essen University Hospital, Department of Gastroenterology and Hepatology, Essen, Germany, *[2]University Hospital Knappschaftskrankenhaus Bochum GmbH, Department of Internal Medicine, Bochum, Germany*, *[3]University of Liverpool, Department of Molecular and Clinical Cancer Medicine, Liverpool, United Kingdom*, *[4]Medizinische Hochschule Hannover, Department of Gastroenterology, Hepatology and Endocrinology, Hannover, Germany*, *[5]Otto von Guericke University Magdeburg, Department of Gastroenterology, Hepatology and Infectious Diseases, Magdeburg, Germany*
Email: clemens.schotten@uk-essen.de

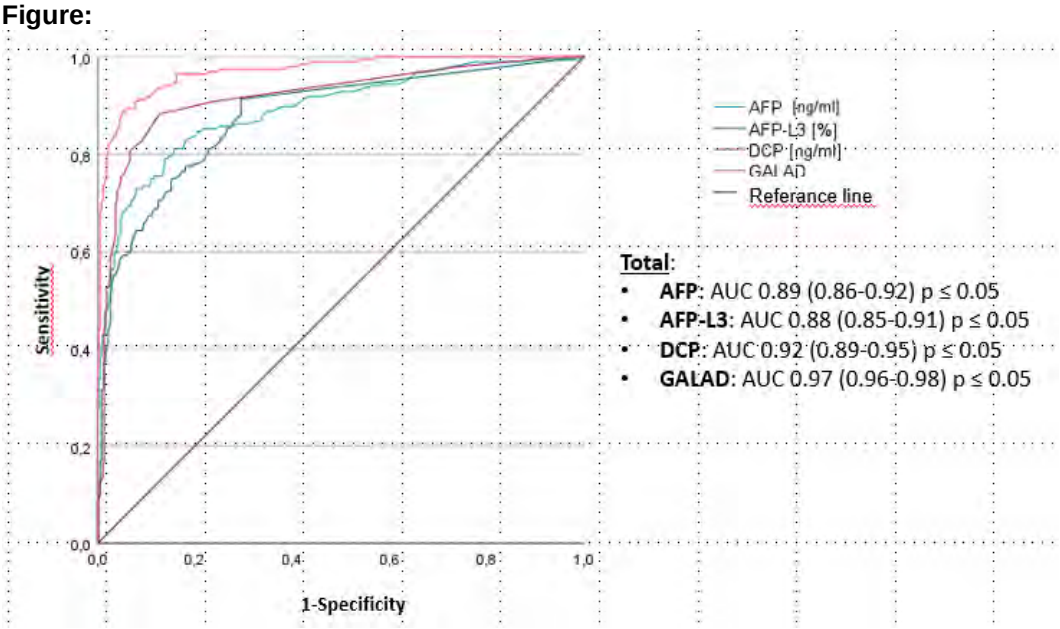
Background and Aims: Despite Hepatitis B (HBV) vaccination programs and effective direct antiviral treatment agents for chronic hepatitis C (HCV), the incidence of virus-related hepatocellular carcinoma (HCC) remains high, while detection rate for early stage HCC is continuously poor. To address this insufficiency, we set out to characterize, whether GALAD score, which incorporates gender, age, and serum levels of AFP, AFP isoform L3 (AFP-L3), and des-gamma-carboxy prothrombin (DCP) can improve early stage HCC detection in a Caucasian HBV/HCV cohort.

Method: In a retrospective German single-center study between 2008 and 2020 182 patients with chronic HBV, 223 with chronic HCV and 186 with other etiology (OE) of chronic liver disease (CLD) were enrolled. In 52 HBV, 84 HCV and 60 OE CLD patients HCC was confirmed by biopsy or imaging. The diagnostic performance of the single biomarkers was compared to the GALAD model.

Results: In all 3 subgroups, at initial diagnosis majority of patients was at early BCLC 0 (n=14/7%) or A (n=56/29%) or intermediate stage BCLC B (n=93/47%) HCC. In the total cohort, GALAD score exhibited an AUC of 0.97 to discriminate HCC from non-HCC *vs*. AFP (AUC, 0.89), AFP-L3 (AUC, 0.88) or DCP (AUC, 0.92). In the HBV population GALAD achieved an AUC of 0.96, in HCV 0.98 and in OE 0.99, again clearly superior to the biomarkers alone. Of note, in HCV patients GALAD Score showed a significantly higher specifity (89%) *vs.* AFP (64%) alone.

Conclusion: The GALAD showed superior performance in detection of early stage HCC compared to AFP alone, while exhibiting higher specifity in HCV patients. We conclude that GALAD score shows promising potential for future HCC surveillance in Caucasian HBV/HCV patients.

Figure:


1,0
0,8
0,6
0,4
0,2
0,0
Sensitivity
0,0
0,2
0,4
0,6
0,8
1,0
1-Specificity
AFP [ng/ml]
AFP-L3 [%]
DCP [ng/ml]
GALAD
Referance line
Total:
AFP: AUC 0.89 (0.86-0.92) p ≤ 0.05
AFP-L3: AUC 0.88 (0.85-0.91) p ≤ 0.05
DCP: AUC 0.92 (0.89-0.95) p ≤ 0.05
GALAD: AUC 0.97 (0.96-0.98) p ≤ 0.05

P054YI Evaluation of cardiovascular events in patients with hepatocellular carcinoma treated with sorafenib in the clinical practice: CARDIO-SOR study.

Lorena Carballo Folgoso[1], Miriam Celada Sendino[1], Álvaro Gejo Benéitez[1], Andrés Castano-Garcia[1], Javier Cuevas Pérez[2], Rut Álvarez-Velasco[2], Rebeca Lorca[2 3], Carmen Álvarez-Navascués[1], Maria Luisa Gonzalez Dieguez[1], Valle Cadahía-Rodrigo[1], Manuel Rodríguez[1 3 4], Maria Varela[1 3 4 5]

[1]Hospital Universitario Central de Asturias, Liver Unit, Oviedo, Spain, *[2]Hospital Universitario Central de Asturias, Department of Cardiology, Oviedo, Spain*, *[3]ISPA, Oviedo, Spain*, *[4]Universidad de Oviedo, Medicine, Oviedo,* , *[5]IUOPA, Oviedo,*
Email: lorenacarballo93@gmail.com

Background and Aims: The effectiveness of systemic treatment in hepatocellular carcinoma (HCC) depends on the selection of suitable patients, carefully management of cirrhosis complications and expertise to treat adverse events. The aim of this study is to assess the cardiovascular (CV) events in a cohort of HCC patients treated with sorafenib (SOR).

Method: Observational retrospective study including all HCC patients treated with SOR from 2007 to 2019 in a western tertiary centre, except those included in clinical trials or treated after liver transplantation. Before starting SOR, baseline features including CV risk factors and prior CV history were recorded. Patients were visited at week 2, 4, 8, 12 and every 2 months thereafter, with dialy control of blood pressure. EKG was done at baseline and every 2-3 months. Adverse events, dosing and outcome data were collected during the follow-up.

Results: 299 HCC patients, median age 66 years, Child A 85%, BCLC-C 73%, ECOG-PS 0 67%, cirrhosis 90% (mostly alcohol, 43%), diabetes 32%, arterial hypertension 42%, dyslipidaemia 25%, obese 28%, smokers 53%. Median overall survival was 11.1 months (IQR 5.6-20.5); ECOG-PS 0 vs PS 1-2 (16.0 vs 5.3, p<0.001). Median treatment duration 7.4 months (IQR 3.3-14.7). Over the 13.6 months' median follow-up (IQR 5.9-24.2), 33 patients (11%) suffered a major CV event [heart failure (n=11), acute coronary syndrome (n=11), acute cerebrovascular accident (n=12) and peripheral vascular ischemia (n=8)], with median time from SOR-start 12.7 months (IQR 4.5-28.0). These major CV events forced to temporal and permanent discontinuation of SOR in 18% and 52% of patients, respectively. Ninety-nine of all patients (33%) had a minor CV event: increase arterial hypertension (n=81), long QT (n=20) and new atrial fibrillation (n=11). Multivariate cox regression analysis found age as the only independent factor associated to CV event during follow-up (HR 1.07; 95% CI 1.03-1.12; p=0.002). The main reason for SOR cessation was CV event in 19 patients (6.3%). Only 2 died due to CV event (1 acute coronary syndrome; 1 cerebrovascular accident).

Conclusion: The incidence of CV events in HCC patients treated with SOR in clinical practice is high, appears late, and independently associated with age. The currently approved TKI options share molecular targets involved in activity and toxicity, so the awareness of CV events may allow a successful sequential therapy for tumor progression.

P055YI Treatment of the chemotherapy induced hepatotoxicity

Igor Ivanikov[1], Yulia Grigorieva[1], Mariia Zharova[1 2], Nataliya Vinogradova[1], Ol Krasenkow[1]

[1]Tsentral'naya Klinicheskaya Bol'nitsa S Poliklinikoy, Moskva, Russian Federation, *[2]Moscow, Gastroenterology, Moscow, Russian Federation*
Email: zharowa.mariya@yandex.ru

Background and Aims:
Treatment of chemotherapy-induced hepatotoxicity (CIH) is a pressing challenge for oncology and gastroenterology. To study the efficacy of several hepatoprotectors in CIH.

Method: 144 patients with a tumor of one of the three localizations (breast cancer, colorectal cancer, and prostate cancer) received 912 courses of chemotherapy. Chemotherapy was suspended in patients if the AST and ALT were 5 times the ULN, and the bilirubin level was three times the ULN. For CIS treatment, hepatoprotectors S-adenosylmethionine or essential phospholipids with glycyrrhizic acid, Sterofundin solution, and ursodeoxycholic acid were used for at least four weeks.

Results: 50 (35%) patients had an elevation in the liver function tests ranged from 2 to 20 times ULN. Thirty-six patients had moderate (grade 2) and four patients severe (grade 3) hepatotoxicity.
We used S-adenomethionine in 39 patients and essential phospholipids in combination with glycyrrhizic acid in 11 patients. Ursodeoxycholic acid was added in the presence of cholestatic syndrome in 22 patients.
Sterofundin infusion, in combination with hepatoprotectors for ten days and subsequent oral administration of the latter, led to a significant decrease in liver function tests in all patients in 7-30 days.

Conclusion:
The treatment regimens we used have shown their effectiveness in treating CIH in cancer patients with moderate and severe hepatotoxicity.

Figure:

P056YI Serum miRNA profiling for the differential diagnosis of distal cholangiocarcinoma from pancreatic diseases

André F. Tomás[1], André L. Simão[1], Luis Muñoz-Bellvis[2], Ainhoa Lapitz[3], Luis Bujanda[4], Pedro Miguel Rodrigues[3], Jesus Maria Banales[3], Jose Marin[5], Cecília M. P. Rodrigues[1], Rocio IR Macias[5], Rui E. Castro[1]

[1]Research Institute for Medicines (iMed.ULisboa), Faculty of Pharmacy, Universidade de Lisboa, Lisbon, Portugal, *[2]Service of General and Gastrointestinal Surgery, University Hospital of Salamanca, IBSAL, CIBERONC, Salamanca, Spain*, *[3]Department of Liver and Gastrointestinal Diseases, Biodonostia Health Research Institute, Donostia University Hospital, University of the Basque Country (UPV/EHU), CIBERehd, Ikerbasque, San Sebastian, Spain*, *[4]National Institute for the Study of Liver and Gastrointestinal Diseases (CIBERehd, Carlos III Health Institute), Madrid, Spain*, *[5]Experimental Hepatology and Drug Targeting (HEVEPHARM), University of Salamanca, Biomedical Research Institute of Salamanca (IBSAL), CIBERehd, Salamanca, Spain*
Email: adlsimao@ff.ulisboa.pt

Background and Aims: When distal cholangiocarcinomas (dCCAs) cross the pancreatic head, it is clinically challenging to distinguish them from pancreatic ductal adenocarcinomas (PDACs) or even benign pancreatic diseases (BPDs) by using currently available imaging techniques or established biomarkers. Moreover, all these disease conditions share similar clinical symptoms. In that regard, serum microRNAs (miRNA/miRs) have been emerging as novel potential biomarkers for diseases. Thus, we aimed to profile global serum miRNA expression changes in human patients with dCCA, PDAC and BPDs in order to identify potential candidates for the differential diagnosis of these diseases, which would have a direct impact on their specific treatment.

Method: Serum samples were obtained from patients with histopathologic confirmation of dCCA (n=35) or PDAC (n=38), as well as from BPD patients (with cysts (n=22) or with chronic pancreatitis (n=20), or from healthy subjects (n=45), with complete anthropometric, biochemical and clinical characterization. Serum miRNAs were isolated using the miRNeasy Serum Advanced Kit and sample pools from each group were analyzed using TaqMan Advanced miRNA Human Serum array cards. cel-miR-39-3p was used as an exogenous spike-in control. Selected miRNAs were validated by TaqMan Advanced Real-Time RT-PCR.

Results: Real time RT-PCR array analysis revealed distinct serum miRNA expression profiles between all groups of patients. Upon individual validation, miRNAs found to be mostly differentially expressed in dCCA included miR-95-3p and miR-154-5p, up- and down-regulated, respectively, comparing with PDAC, but also with BPD and healthy controls. Of note, miR-204-5p and miR-200c-3p were particularly elevated in BPD patients with cysts or with chronic pancreatitis, respectively, comparing with dCCA. In addition, the levels of these miRNAs were found to be independent of the patient gender or the presence of selected liver disease risk factors. Moreover, serum miR-154-5p levels negatively correlated with GGT, while both miR-204-5p and miR-200c-3p positively correlated with patients' age. Of note, in dCCA patients, miR-95-3p levels positively correlated with carbohydrate antigen19-9 (CA19-9), a non-specific tumor marker commonly used to help in the diagnosis of CCA and PDAC.

Conclusion: In conclusion, analysis of serum levels of specific miRNAs, including miR-95-3p and miR-154-5p, may allow for the differential diagnosis of dCCAs from both benign and malignant pancreatic diseases, as well as correlations with clinical features of patients. Further validation in independent cohorts could translate into the use of miRNAs as accurate biomarkers for dCCA early diagnosis. (OLD-HEPAMARKER, 0348_CIE_6_E; V Beca de Investigación Carmen Delgado/Miguel Pérez-Mateo, Spain; PTDC/MED-PAT/31882/2017, Portugal; COST Action CA18122, EURO-CHOLANGIO-NET)

Figure:

P057 Hepatocellular Carcinoma Surveillance in Cirrhotic patients.

Shreyashee Sengupta[1], Catherine Rowan[1], Binish Baburaj[1], Zahid Idrees[1], Javeria Tariq[1], Erica Stapleton[1], Murat Kirca[1]

[1]Midlands Regional Hospital Mullingar, Department of Gastroenterology , Mullingar, Ireland
Email: torasengupta92@gmail.com

Background and Aims:
Cirrhosis is a significant risk factor for hepatocellular carcinoma (HCC) (1). Regular surveillance of patients with cirrhosis at 6 monthly intervals helps to detect early-stage HCC, which is potentially curable.(2) Our aim was to assess our compliance with international guidelines on HCC surveillance in our cohort of patients with cirrhosis.(3, 4)

Method:
A retrospective database of patients with a diagnosis of cirrhosis, admitted to MRHM from 2010-2020, was created. This was collected using the Hospital In-Patient Enquiry (HIPE) database.
Data collected included:

1. First radiological documentation of cirrhosis
2. Number of ultrasound (US) scans booked/attended since initial diagnosis
3. Number of alpha-fetoprotein (AFP) measurements since initial diagnosis
4. Primary team of the patient
5. If a GI referral was made or not
6. diagnosis of HCC
7. Mortality, Date of death
8. Survival diagnosis until death

Patients with a HIPE diagnosis of cirrhosis without radiological features of cirrhosis/ portal hypertension were subsequently excluded.

Results:
125 patients were included based on HIPE data. 37 patients were excluded due to the absence of radiological evidence of cirrhosis. 88 patients were included in the final cohort.

12.5% (n=11) had a diagnosis of HCC with concomitant cirrhosis.

The median age of our cohort was 69 years, with 54.5% male (n=48).

The mortality rate was 59.1% (n=52), of which 22 patients died within 6 months of diagnosis of cirrhosis.

40% (n=35) of the cohort had completed <50% of recommended US and 68% (n=60) has <50% of recommended AFP measurements.
There was significantly higher mortality rate in the cohort with HCC when compared to the group with no HCC, (p=0.047)

The overall mortality and 6-month mortality were significantly higher in cirrhotic patients under the care of non-gastroenterology specialists (p=0.002 and p=0.006 respectively).

Conclusion:
From our audit it is evident that

1. Patients with cirrhosis have a high mortality risk, which increases with the diagnosis of HCC.
2. Compliance with established HCC surveillance recommendations for patients with cirrhosis does not meet international standards. Furthermore, it is evident that there is a significant benefit to providing specialised gastroenterologist/hepatologist- led care to patients with cirrhosis.

References

1. Flemming JA, Yang JD, Vittinghoff E, Kim WR, Terrault NA. Risk prediction of hepatocellular carcinoma in patients with cirrhosis: the ADRESS-HCC risk model. Cancer. 2014;120(22):3485-93.
2. Bruix J, Sherman M. Management of hepatocellular carcinoma: an update. Hepatology. 2011;53(3):1020-2.
3. Harrison P, Hogan BJ, Floros L, Davies E. Assessment and management of cirrhosis in people older than 16 years: summary of NICE guidance. Bmj. 2016;354:i2850.
4. EASL Clinical Practice Guidelines: Management of hepatocellular carcinoma. J Hepatol. 2018;69(1):182-236.

P058YI Dynamic risk profile of hepatocellular carcinoma recurrence after curative intent liver resection

Tommy Ivanics[1], Carla Fiorella Murillo Perez[1 2], Marco Claasen[1], Madhukar Patel[1], Chaya Shwaartz[1 3], Luckshi Rajendran[3], Grainne O'Kane[4], Bettina Hansen[2 5], Sean Cleary[6], Gonzalo Sapisochin[1 3]

[1]University Health Network, Multi-Organ Transplant Program, Toronto, Canada, *[2]University Health Network, Toronto Centre for Liver Disease, Toronto, Canada*, *[3]University Health Network, Division of General Surgery, Toronto, Canada*, *[4]University Health Network, Medical Oncology and Hematology, Toronto, Canada*, *[5]University of Toronto - St. George Campus, Institute of Health Policy, Management and Evaluation, Toronto, Canada*, *[6]Mayo Clinic, Division of Hepatobiliary and Pancreatic Surgery, Rochester, United States*
Email: tommy.ivanics@uhn.ca

Dynamic risk profile of hepatocellular carcinoma recurrence after curative intent liver resection

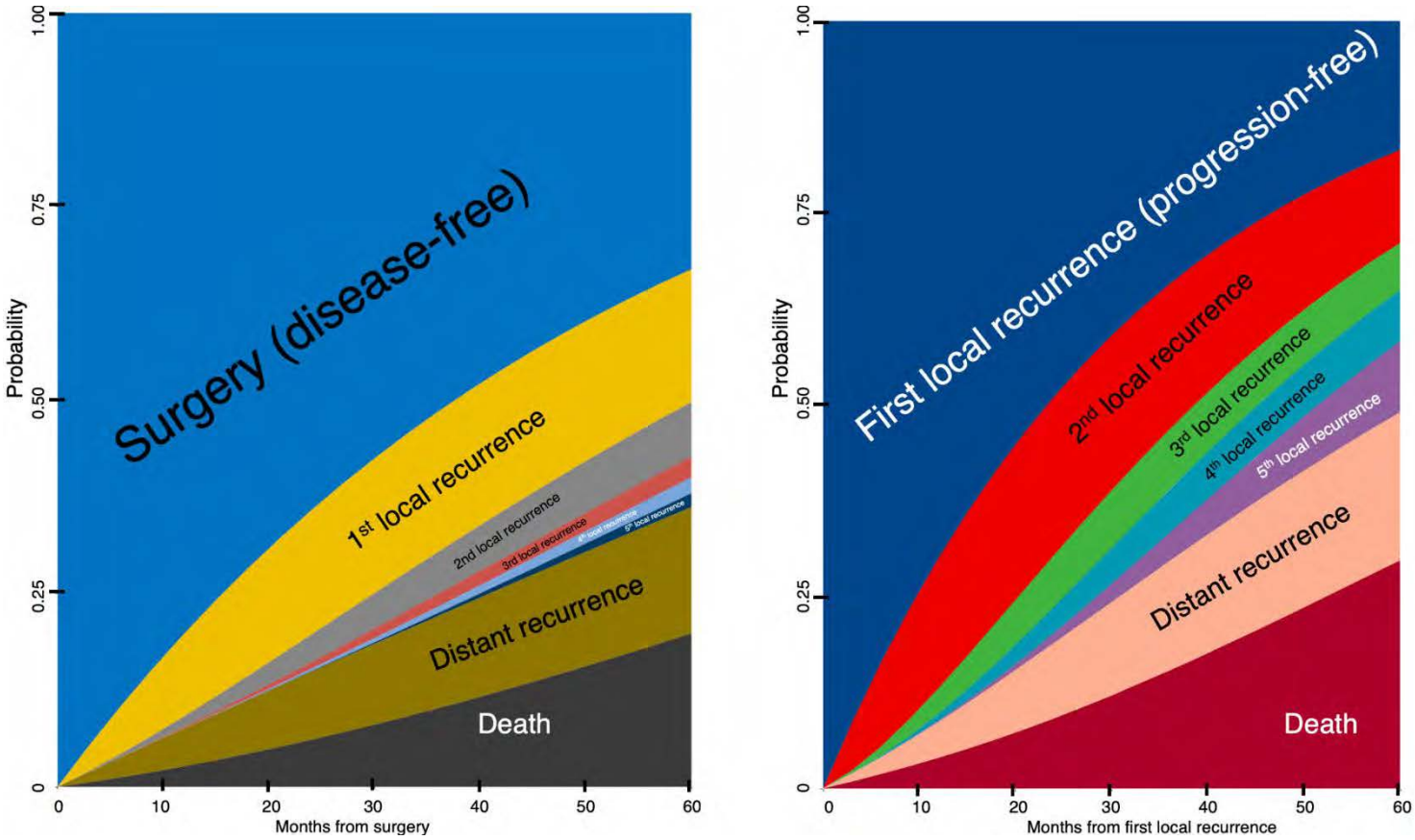
Background and Aims:
Following curative-intent liver resection (LR) for hepatocellular carcinoma (HCC), the likelihood of survival is dynamic, in that multiple recurrences and/or metastases are possible, each with distinct location and varying impact on outcomes. We sought to evaluate the natural progression, pattern, and timing of the various disease states after LR for HCC using multi-state modeling.

Method:
Adult patients undergoing LR for HCC between Jan-2000 and Dec-2018 were retrospectively identified at a single center. Multistate data analysis was employed to model post-LR tumor progression by describing transitions between distinct disease states. In the selected model, the states included surgery, local recurrence (1^{st}, 2^{nd}, 3^{rd}, 4^{th}, 5^{th}), distant metastasis with or without local recurrence, and death.

Results:
A total of 486 patients were included with a median follow-up of 41.8 (IQR 19.1-84.2) months. Of these, 169 (34.8%) patients remained recurrence free throughout follow-up, 205 (42.2%) developed local recurrence, 80 (16.5%) developed distant metastasis, and 32 (7%) patients died. The 1-year local-recurrence free probability was 76.2% after a 1^{st} local recurrence, 58.2% after a 2^{nd} local recurrence, 48.7%, after a 3^{rd}, 39.9% after a 4^{th} and 54.6% after a 5^{th}. For a typical patient having undergone curative intent liver resection, 33.1% remained disease-free, 31.0% had at least one local recurrence, 16.3% had distant metastasis, and 19.8% had mortality in the first 60 months after surgery. The probability of transitioning from surgery to a first local recurrence, without a subsequent transition to a next state, increased from 3% at 3 months to 17.4% at 30 months, and to 17.2% at 60 months (**Figure 1**). Factors identified that could modify these probabilities included tumor number, size, satellite lesions, and microvascular invasion.

Conclusion:
Multistate modeling provides meaningful prognostic information to patients undergoing surveillance as outcomes following curative-intent LR for HCC are highly variable depending on the timing and pattern of disease recurrence and/or metastasis. Accordingly, taking into account the various disease states that could occur after cancer treatment may provide a more realistic representation of outcomes than standard time-to-one event estimates.

Figure 1. Probability of making a transition and reaching each state over the first 5 years from the **a)** surgery state (post-curative intent surgery) **b)** first local recurrence state



P059YI Targeting tumor-initiating cells and compensatory YAP pathway activation to overcome sorafenib-induced resistance in hepatocellular carcinoma

Darko Castven[1 2], Carolin Czauderna[1 2], Diana Becker[2], Sharon Pereira[2], Kai Breuhahn[3], Jennifer Schmitt[3], Jovana Hajduk[1 2], Friederike Keggenhoff[2], Marcus-Alexander Wörns[2], Snorri Thorgeirsson[4], Peter Galle[2], Jens Marquardt[1 2]

[1]Universitätsklinikum Schleswig-Holstein, Campus Lübeck, Department of Medicine I, Lübeck, Germany, *[2]Universitätsmedizin Mainz, Department of Medicine I, Mainz, Germany*, *[3]University Hospital Heidelberg, Institute of Pathology, Heidelberg, Germany*, *[4]National Cancer Institute, NIH, Bethesda, United States*
Email: castvendarko@gmail.com

Background and Aims: Induction of chemoresistance upon systemic therapy is frequently observed in the majority of HCC patients. Accumulating evidence suggests that tumor-initiating cells (TICs) may contribute to the acquisition of resistance in many solid tumors, but their exact role for HCC remains to be defined. Here, we evaluate the importance of TICs in the development of resistance and relapse formation after exposure to sorafenib in HCC and define concomitant adaptive molecular targets.

Method: Four HCC cell lines and two primary HCC isolates were exposed to sorafenib for a total of 14 days. The treatment effects on TICs were estimated by sphere forming capacity *in vitro* and tumor-initiating potential *in vivo*, as well as the side-population (SP) approach. Expression of key CSC marker EpCAM was assessed by flow cytometry. Furthermore, whole transcriptome analyses were performed across the cell lines and identified potential targets, which were further validated by immunohistochemistry, western blot and administration of specific inhibitors.

Results: Treatment with sorafenib effectively reduced oncogenic properties in all investigated HCC cells. However, sustained anti-proliferative effect after treatment was observed in half of the cell lines, while initial treatment effect in other lines was followed by rapid re-growth thereby resembling the responses observed in patients. Anti-oncogenic effects in sensitive cell lines were associated with significant reduction in sphere-forming and tumor-initiating capacity, CSC marker EpCAM as well as SP cells, while resistant cell lines showed transient induction in TIC properties. Acquired resistance uniformly developed in cell lines suggesting that common molecular mechanisms might be operative. These adaptive molecular changes involved signaling pathways known to be associated to cell survival, proliferation and cell cycle regulation (RAS, IL6, MYC, E2F3). Furthermore, the resistant cell lines showed compensatory upregulation of key oncogenic molecules such as EGFR, as well as YAP. Validation on authentic HCC samples confirmed enrichment of YAP signaling in the patients with worst response to sorafenib treatment. Conclusively, combined treatment including sorafenib and specific YAP inhibitor showed beneficial effects in resistant cell lines which resulted in complete response to the therapy.

Conclusion: Our model recapitulates features of drug resistance observed in human HCC patients. Resistance to anti-angiogenic therapy might be fueled by transient expansion of TICs. Therefore, specific targeting of TICs as well as pro-oncogenic compensatory signaling pathways might be an effective therapeutic strategy to overcome resistance in HCC.

Figure:

P060 Immunosuppressive Drug Resistant Armored TCR T cells for immune-therapy of HCC in liver transplant patients

Morteza Hafezi[1], Meiyin Lin[1], Adeline Chia[1], Alicia Chua[2], Zi Zong Ho[2], Royce Fam[2], Damien Tan[3], Joey Aw[3], Andrea Pavesi[3], Thinesh Krishnamoorthy[4], Wan Cheng Chow[4], Wenjie Chen[5], Qi Zhang[5], Lu-En Wai[2 6], Sarene Koh[2 6], Anthony Tan[1], Antonio Bertoletti[1 6]

[1]Duke-NUS Medical School, Singapore, Singapore, *[2]Lion Tcr Pte. Ltd., Singapore, Singapore*, *[3]Institute of Molecular and Cell Biology (IMCB), Singapore, Singapore*, *[4]Singapore General Hospital, Singapore, Singapore*, *[5]The Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, China*, *[6]Singapore Immunology Network, Singapore*
Email: anthony.tan@duke-nus.edu.sg

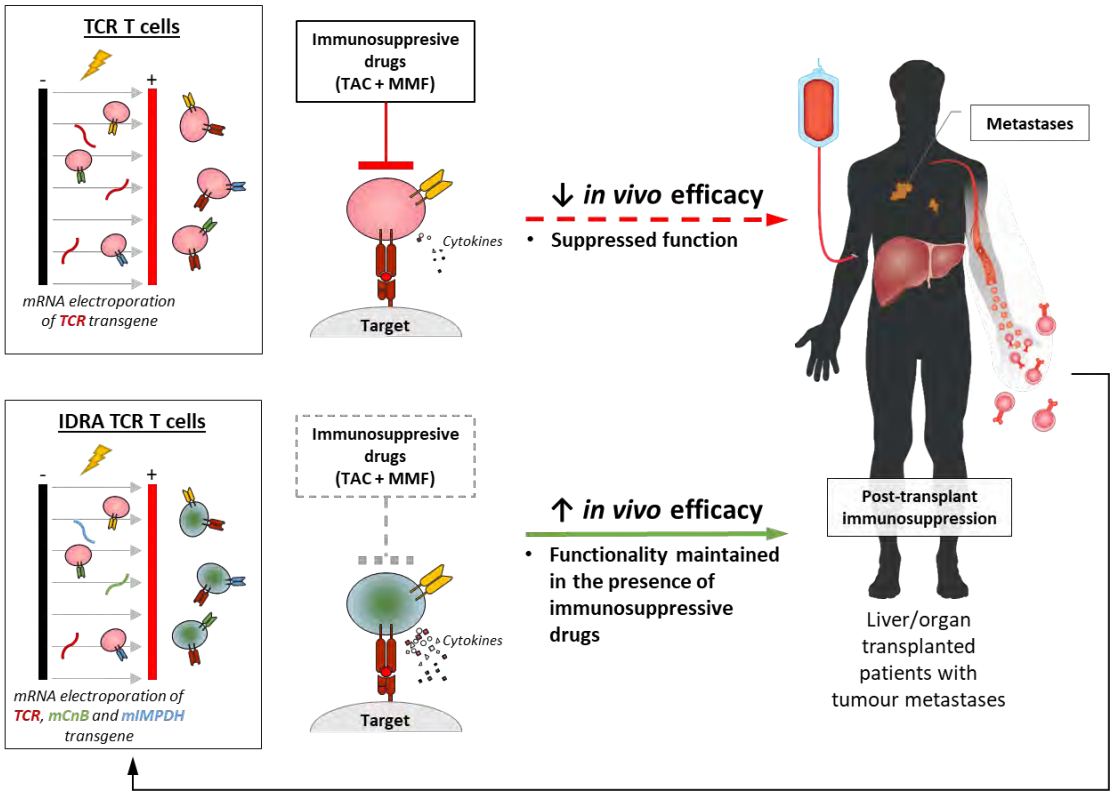
Background and Aims:
HBV-specific T cell receptor (HBV-TCR) engineered T cells have the potential for treating hepatocellular carcinoma (HCC) relapses after liver transplantation, but their efficacy can be hampered by the concomitant immunosuppressive treatment required to prevent graft rejection. Our aim is to molecularly engineer TCR-T cells that could retain their polyfunctionality in such patients while minimising the associated risk of organ rejection.

Method:
We first analysed how immunosuppressive drugs can interfere with the in vivo function of TCR-T cells in liver transplanted patients with HBV-HCC recurrence receiving HBV-TCR T cells, and in vitro in the presence of clinically relevant concentrations of immunosuppressive Tacrolimus (TAC) and Mycophenolate Mofetil (MMF). Immunosuppressive Drug Resistant Armored (IDRA) TCR-T cells of desired specific (HBV or EBV) were then engineered by concomitantly electroporating mRNA encoding specific-TCRs and mutated variants of calcineurin B (CnB) and inosine-5′-monophosphate dehydrogenase (IMPDH), and their function was assessed through intracellular cytokine staining and cytotoxicity assays in the presence of TAC and MMF.

Results:
Liver transplanted HBV-HCC patients receiving different immunosuppressant drugs exhibited varying levels of activated (CD39+ Ki67+) PBMCs post HBV-TCR T cell infusions that positively correlates with clinical efficacy. In vitro experiments with TAC and MMF showed a potent inhibition of TCR-T cell polyfunctionality. This inhibition can be effectively negated by the transient overexpression of mutated variants of CnB and IMPDH. Importantly, the resistance only lasted for 3-5 days after which sensitivity was restored.

Conclusion:
We engineered TCR-T cells of desired specificities that transiently escape the immunosuppressive effects of TAC and MMF. This finding has important clinical applications for the treatment of HBV-HCC relapses and other pathologies occurring in organ transplanted patients.

Figure:


TCR T cells
mRNA electroporation of TCR transgene
Immunosuppresive drugs (TAC + MMF)
Cytokines
Target
↓ in vivo efficacy
• Suppressed function
Metastases
Post-transplant immunosuppression
IDRA TCR T cells
mRNA electroporation of TCR, mCnB and mIMPDH transgene
Immunosuppresive drugs (TAC + MMF)
Cytokines
Target
↑ in vivo efficacy
• Functionality maintained in the presence of immunosuppressive drugs
Liver/organ transplanted patients with tumour metastases

P061YI Circular RNA expression is modulated by RNA-binding proteins in hepatocellular carcinoma

Rok Razpotnik[1], Valeriya Musina[1], Petra Ivanuša[1], Anže Šumah[1], Tanja Kunej[2], Damjana Rozman[1], Tadeja Rezen[1]

[1]University of Ljubljana, Faculty of Medicine, Institute of Biochemistry and Molecular Genetics, Centre for Functional Genomics and Bio-Chips, Ljubljana, Slovenia, *[2]University of Ljubljana, Biotechnical Faculty, Department of Animal Science, Domžale, Slovenia*
Email: tadeja.rezen@mf.uni-lj.si

Background and Aims: Circular RNA (circRNA) can act as oncogenes or tumor suppressors also in hepatocellular carcinoma (HCC). Many circRNA are dysregulated in cancer and the drivers of differential expression of circRNA remain unknown. Transcriptome analyses have shown that circRNA/mRNA transcripts from the same host gene locus can have different patterns of expression. Current hypothesis is that the expression of circRNA in different processes is actively regulated by trans-acting factors, such as RNA-binding proteins (RBPs), and independently from linear transcripts. Aim of this study was to identify the regulators of circRNA expression in HCC.

Methods: We analysed transcriptome data from GEO (circRNA) and TCGA-LIHC (mRNA) and compared the concordance of expression across circRNA-mRNA pairs from the same host gene locus. Using MEME Suite software, we identified the enrichment of RNA splicing factor motifs around circRNA splice sites. Next, we analysed publicly available eCLIP and ChIP-Seq datasets in ENCODE to identify confirmed binding sites of the splicing regulators in HepG2 HCC model cell line. Finally, we performed *in vitro* analyses in Huh-7 HCC model cell line, where we overexpressed or silenced the expression of selected splicing factors and measured the expression of candidate circRNA using RT-qPCR.

Results: We identified statistically significant differentially expressed circRNA and mRNA in HCC and divided the mRNA-circRNA pairs in groups according to concordance of their expression. Next, we identified the enriched RNA splicing factor motifs around circRNA splice and aligned them with published binding sites in HCC model cell lines. From this list of potential RNA splicing/binding factors we selected those which were differentially expressed in TCGA-LIHC cohort and are significantly correlated with the patient's survival. Finally, we performed *in vitro* analyses in Huh-7 HCC model cell line. We overexpressed (*HNRNPK, NONO, PCBP2*) or silenced *(ESRP2, PCBP2*) the expression of selected splicing factors and measured the expression of candidate circRNAs confirming that RBPs can modulate the expression of circRNA.

Conclusion: Identified regulators of circRNA expression will result in better understanding of HCC pathogenesis and could also open new possibilities for therapeutic approaches.

P062 The impact of ultrasonographic blind spots for detecting early-stage hepatocellular carcinoma during surveillance

HA IL KIM[1], Junghwan Lee[2], Soomin Noh[3], Li-Chang Hsing[3]

[1]Kyung Hee University Hospital at Gangdong, Internal medicine, Seoul, Korea, Rep. of South, *[2]Asan Medical Center, Internal medicine, Seoul, Korea, Rep. of South*, *[3]Asan Medical Center, Gastroenterology, Seoul, Korea, Rep. of South*
Email: mondosewan@gmail.com

Background and Aims:
Abdominal ultrasonography (US) is a backbone of surveillance for hepatocellular carcinoma (HCC). The methodology of HCC surveillance has been controversial for decades, because of the suboptimal detecting power of US. Although numerous studies have been evaluated the clinical factors related to surveillance failure, there have been no studies about US blind spots *per se*.

Method:
We included, from a hospital-based registry, 706 adults who were surveilled 6 months interval with US and the serum alpha-fetoprotein (AFP) test and eventually diagnosed with early-stage HCC. Based on surveillance results, these patients were divided into two groups: US-detected (positive for US with or without positive AFP test; n=484) and US-missed (positive for AFP only; n=222). The definition of "blind spots" consists of 4 locations (hepatic dome, caudate lobe or around inferior vena cava, beneath ribs <1cm, and the surface of left lateral segment). We evaluated how US blind spots affect tumor stage, treatment method, and survival when the surveillance test detected HCC.

Results:
The size of HCC in US-missed group was significantly higher than US-detected group (1.5±0.5 vs. 2.2±1.2 cm, *P*<0.001). The proportion of ≥ 2cm HCC detected on blind spot was significantly higher than HCC on non-blind area (10.6% vs. 26.9%, *P*<0.001). US detected group underwent more radiofrequency ablation, otherwise, hepatectomy performed more in US-missed group (All *P* values <0.05) regardless of HCC location. The proportion of curative-intent treatment applied both groups without significant differences. Kaplan-Meier curve showed that US-detected group had significantly better overall survival than the US-missed group in patients with HCC on blind spot (log-rank test, *P*=0.036). When the tumor size restricted to < 2cm, there was no survival difference between the two groups regardless of HCC location.

Conclusion:
In the surveillance test of HCC, US blind spots could affect the initial tumor stage, treatment modality, and overall survival. Unfortunately, AFP test could not fully compensate for detecting HCC <2cm in current surveillance. Additional research is needed to overcome the inherent limitation of US for detecting early-stage HCC.

P063 Prognostic factors and post-treatment survival in patients enrolled onto second-line trials for unresectable hepatocellular carcinoma after sorafenib: a multicentre Italian cohort study

Nicola Personeni[1 2], Tiziana Pressiani[2], Antonio D'Alessio[2], Maria Giuseppina Prete[2], Andrea Casadei Gardini[3], Antonella Cammarota[2], Martina Valgiusti[4], Vincenzo Dadduzio[5], Vittorina Zagonel[6], Laura Giordano[7], armando santoro[1 2], Lorenza Rimassa[1 2]

[1]Humanitas University, Department of Biomedical Sciences, Pieve Emanuele, Italy, *[2]Humanitas Clinical and Research Center-IRCCS, Medical Oncology and Hematology Unit, Rozzano, Italy*, *[3]San Raffaele Hospital IRCCS, Department of Medical Oncology, Milano, Italy*, *[4]Istituto Scientifico Romagnolo per lo Studio e la Cura dei Tumori (IRST) IRCCS, Meldola, Italy*, *[5]Veneto Institute of Oncology (IOV) - IRCCS, Unit of Medical Oncology 1, Padova, Italy*, *[6]Veneto Institute of Oncology (IOV) - IRCCS, Unit of Medical Oncology 1, Padova , Italy*, *[7]Humanitas Clinical and Research Center-IRCCS, Biostatistic Unit, Rozzano, Italy*
Email: nicola.personeni@hunimed.eu

Background and Aims: Second-line treatments following sorafenib are standard of care for advanced hepatocellular carcinoma (HCC) patients with preserved liver function. Determinants of treatment benefit and prognostic factors influencing post-treatment survival (PTS) remain unknown.
We evaluate predictors for overall survival (OS), time to treatment failure (TTF) and PTS calculated from the last day of second-line treatment to death or last follow-up.

Method: We included 174 patients intolerant or progressing on first-line sorafenib. 80 patients received either targeted agents or immune checkpoint inhibitors granted subsequent regulatory approval (AT), while 94 received agents not approved for HCC (OT). Univariate and multivariate analyses using Cox proportional hazards method established relationships among treatments, clinical variables, and OS.

Results: Median OS from the beginning of second-line treatment was 9.7 months (8.47-11.07). OS was independently predicted by treatment received, portal vein thrombosis and disease extent. Greater survival benefit from AT was predicted by extrahepatic spread (EHS) and low neutrophil-to-lymphocyte ratio (NLR; *P* of interaction = 0.005 and 0.032, respectively). Median TTF for patients receiving AT was 4.30 months, and 3.63 months with OT (Hazard ratio 0.69; 0.51-0.94; *P* = 0.020). Median PTS was 4.0 months (2.79-5.32), resulting from 5.3 (3.97-6.54) months for progressors, 5.0 (0.03-8.18) months for those with adverse events, and 0.9 (0.53-1.38) months in decompensated patients (*P* <0.001). Alpha-fetoprotein levels, albumin-bilirubin grade, and enrolment onto subsequent trials independently predicted PTS.

Conclusion: Besides established prognostic factors, this retrospective analysis indicates EHS and NLR as predictive factors useful to gauge the benefit of currently approved second-line treatments.

P064YI FGFR2 fusion protein-driven mouse models of intrahepatic cholangiocarcinoma unveil a necessary role for Erk signaling

Giulia Cristinziano[1], Manuela Porru[1], Dante Lamberti[1], Simonetta Buglioni[2], Francesca Rollo[2], Carla Azzurra Amoreo[2], Isabella Manni[3], Diana Giannarelli[4], Cristina Cristofoletti[5], Giandomenico Russo[5], Mitesh J Borad[6], Gian Luca Grazi[7], Maria Grazia Diodoro[2], Silvia Giordano[8], Mattia Forcato[9], Sergio Anastasi[1], Carlo Leonetti[3], Oreste Segatto[1]

[1]IRCCS Regina Elena National Cancer Institute, Unit of Oncogenomics and Epigenetics, Rome, Italy, *[2]IRCCS Regina Elena National Cancer Institute, Department of Pathology, Rome, Italy*, *[3]IRCCS Regina Elena National Cancer Institute, SAFU, Rome, Italy*, *[4]IRCCS Regina Elena National Cancer Institute, Unit of Biostatistics, Rome, Italy*, *[5]IRCCS Istituto Dermopatico dell'Immacolata, Rome, Italy*, *[6]Mayo Clinic, Division of Hematology and Oncology, Scottsdale, United States*, *[7]IRCCS Regina Elena National Cancer Institute, Division of Hepatobiliary Pancreatic Surgery, Rome, Italy*, *[8]University of Torino, Department of Oncology, Turin, Italy*, *[9]University of Modena and Reggio Emilia, Department of Life Sciences, Italy*
Email: giulia.cristinziano@ifo.gov.it

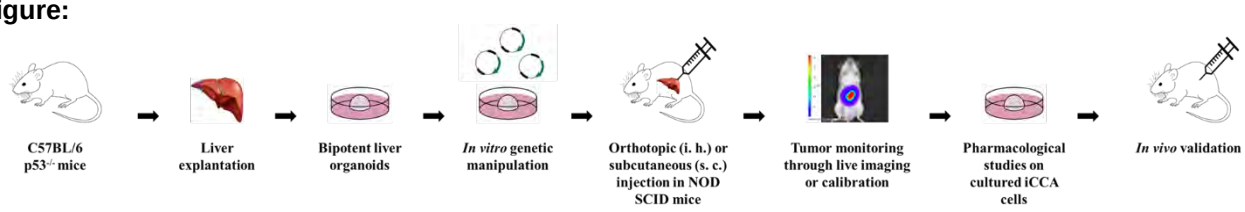
Background and Aims: About 15% of intrahepatic cholangiocarcinoma (iCCA) express fibroblast growth factor receptor 2 (FGFR2) fusion proteins (FFs), most often in concert with mutationally inactivated TP53, CDKN2A or BAP1. In FFs, FGFR2 residues 1-768 are fused to sequences encoded by any of a long list of partner genes (>60), a configuration that ignites oncogenic FF activation. While FGFR-specific tyrosine kinase inhibitors (F-TKI) provide clinical benefit in FF+ iCCA, responses are partial and/or limited by resistance mechanisms, including FF tyrosine kinase domain mutations, prominent among which is the V565F substitution in the FGFR2 gatekeeper residue. Improving on FF targeting in iCCA is therefore a pressing need. Herein, we present the generation of FF-driven murine iCCA models and their exploitation for discovering actionable FF-associated dependencies.

Method: Four iCCA FFs carrying different fusion sequences were expressed in *Tp53*$^{-/-}$ mouse liver organoids. Tumorigenic properties of genetically modified liver organoids were assessed by their intrahepatic or subcutaneous transplantation in immuno-deficient mice. Cellular models derived from neoplastic lesions were exploited for pre-clinical studies.

Results: Tumors diagnosed as CCA were obtained upon transplantation of FF-expressing liver organoids. The penetrance of this tumorigenic phenotype was influenced by FF identity. Tumor organoids and 2D cell lines derived from CCA lesions were addicted to FF signaling via Ras-Erk, regardless of FF identity or V565F mutation. Double blockade of FF-Ras-Erk pathway by concomitant pharmacological inhibition of FFs and Mek1/2 provided greater therapeutic efficacy than single agent F-TKI *in vitro* and *in vivo*.

Conclusion: FF-driven iCCA pathogenesis was successfully modeled in murine *Tp53*$^{-/-}$background, revealing biological heterogeneity among structurally different FFs. Double blockade of FF-Erk signaling deserves consideration for improving on precision-based approaches against human FF+ iCCA.

Figure:



P065 Phase 1 study of autologous hepatitis B virus (HBV)-specific T cell receptor (TCR) T-cells, LioCyx-M in unresectable HBV-related hepatocellular carcinoma (HCC)

Fu-Sheng Wang[1], Fanping Meng[1], Jiehua Jin[1], Yuanyuan Li[1], Regina Wanju Wong[2], Anthony Tanoto Tan[3], Tingting Wang[2], Antonio Bertoletti[2 3]

[1]5th Medical Center, PLA General Hospital, Department of Infectious Diseases, *[2]Lion TCR Pte Ltd*, *[3]Duke-NUS Medical School, Emerging Infectious Diseases*
Email: fswang302@163.com

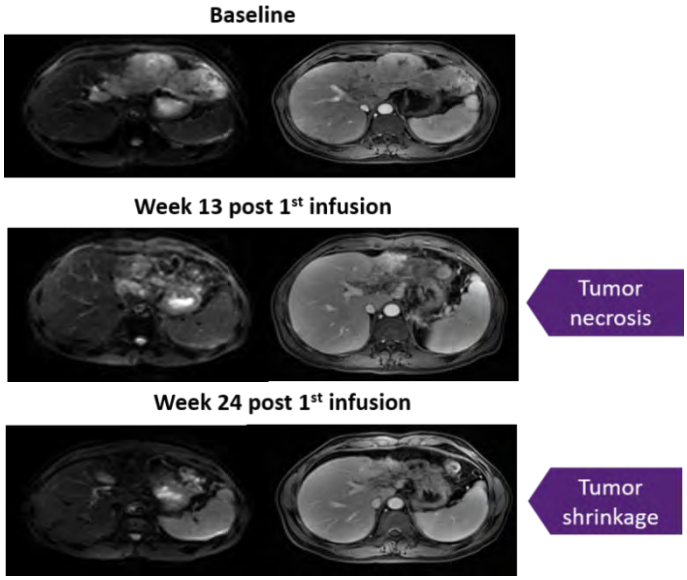
Background and Aims: LioCyx-M is an autologous T cell product transiently modified with *in-vitro* transcribed mRNA encoding Hepatitis-B-virus (HBV)-specific T cell receptor (TCR). The transient TCR expression allows for a conservative dose escalation to closely monitor for potential toxicities. Here, we assessed the safety and tolerability of LioCyx-M in patients with unresectable HBV-related hepatocellular carcinoma (HCC) without curative treatment options.

Method: Eligible patients had matched HLA class I and BCLC B or C HCC (Child-Pugh < 7 points). The treatment regimen consists of 2 treatment cycles. Treatment Cycle 1 consists of 4 weekly escalating doses of 1x10E04 cells/kg, 1x10E05 cells/kg, 1x10E06 cells/kg, 5x10E06 cells/kg bodyweight (BW). This is followed by a 1-month safety assessment according to NCI CTCAE V4.0.3. If there were no dose associated toxicities, patients were eligible to receive Treatment Cycle 2 consisting four weekly infusions at dose of 5 x10E06 cells/kg BW. Tumor response per RECIST 1.1 criteria and survival time were assessed.

Results: As of data cutoff on 30 April 2020, 8 patients of a median age of 53 (range: 49 - 67) were enrolled. A median number of 6 (range: 4 - 12) infusions were administrated to each patient. Of the 7 patients evaluable for tumour response, 3 patients showed stable disease with ≥ 3 months duration (range: 3.0 - 9.5 months) as per RECIST criteria 1.1. A remarkable reduction in the size of liver tumor of > 30% which maintained for 30 months was observed in one patient (Please see Figure). Notably, this tumour shrinkage was accompanied by transient Grade 3 elevations in ALT, GGT, AST and bilirubin after receiving second dose of LioCyx-M at dose level of 1x10E05 cells/kg BW, indicating the on-target effects of LioCyx-M. Grade 1 transient fever was reported in another patient after receiving 4th, 5th and 6th infusions at dose level 5x10E06 cells/kg BW. There were no treatment-related adverse events such as cytokine release syndrome or neurotoxicity. Median time to progression was 2.97 months (90% CI: 0.23 - 9.53 months). Median overall survival was 33.57 months (90% CI: 2.53 months - NA). Four patients were alive at data cutoff.

Conclusion: Our data have demonstrated the on-target ability of LioCyx-M with promising clinical outcome and good safety profile. Further efficacy exploration of LioCyx-M for advanced HBV-HCC is required in a Phase 2 study. Funding: Lion TCR. Clinical trial information: NCT03899415

Figure:


Baseline
Week 13 post 1st infusion
Tumor necrosis
Week 24 post 1st infusion
Tumor shrinkage

P066YI The combination of alcohol and metabolic syndrome is a fast track to hepatic tumorigenesis

Raquel Benedé[1], Chaobo CHEN[2], Olga Estévez[1], Feifei Guo[2], Arantza Lamas Paz[2], Laura Moran Blanco[2], Johanna Reißing[3], Tony Bruns[3], kang Zheng[2 4], Maria Isabel Peligros[5], Javier Vaquero[6 7 8], Christian Trautwein[9], Christian Liedtke[9], Rafael Bañares[6 7 8], Francisco Javier Cubero[2 4], Yulia Nevzorova[2 4 9]

[1]Complutense University of Madrid, Genetics, Physiology and Microbiology , Madrid, Spain, *[2]Complutense University of Madrid, Immunology, Ophthalmology and ENT, Madrid, Spain*, *[3]University Hospital RTWH Aachen, Department of Internal Medicine III, Germany*, *[4]12 de Octubre Health Research Institute (imas12), Madrid, Spain*, *[5]Hospital General Universitario Gregorio Marañón, Servicio de Anatomía Patológica, Madrid, Spain*, *[6]Hospital General Universitario Gregorio Marañón, Servicio de Aparato Digestivo, MADRID, Spain*, *[7]Instituto de Investigación Sanitaria Gregorio Marañón (IiSGM), Madrid, Spain*, *[8]Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas (CIBEREHD) , Madrid, Spain*, *[9]University Hospital RTWH Aachen, Department of Internal Medicine III, Aachen, Germany*
Email: rabenede@ucm.es

Background and Aims: Patients with clinical features of **b**oth alcoholic and non-**a**lcoholic **s**teato**h**epatitis are classified as **BASH.** Recent clinical studies report that intermediate alcohol use in combination with metabolic syndrome (MS) associated with an increased risk of steatohepatitis, cirrhosis and cancer. However, BASH still remains an unexplored area of great interest, given the increasing number of patients affected. In the present study, we aimed to develop a novel preclinical model reflecting BASH in mice.

Method: We applied two different models of BASH-associated tumorigenesis: 1. prolonged BASH model: C57BL6/J mice received BASH diet (10%v/v alcohol in sweetened drinking water in combination with Western diet (WD)) for 52 weeks, 2. BASH+DEN model: animals received a single dosage of diethylnitrosamine (DEN) at the age of 14 days and, 8 weeks later, were placed on a BASH or control chow diet for following16 weeks.

Results: After 52 weeks of BASH diet the murine livers became extraordinarily enlarged, whitish-yellow in colour, with pronounced scar tissue on the surface and multiple nodules**.** Additionally, animals developed splenic enlargement, and significantly increased serum levels of AST, ALT, LDH and cholesterol. H&E and SR stainings demonstrated intense immune infiltration, significant fatty changes, extensive collagen deposition and well-differentiated micronodules, surrounded by massive fibrotic connective tissue extending between portal regions. Notably, GS staining, a broadly used marker of HCC differentiation, was absent inside the nodules, characterizing these lesions as preneoplastic. Macroscopic evaluation of the second model revealed that all DEN+BASH mice exhibited single or multiple tumor nodules larger than 0,5 cm. In contrast, only 1 out of 9 mice treated with DEN+chow had visible nodules. Moreover, the BASH diet significantly increased the hepatosomatic ratio, ALT, AST and LDH in DEN-treated mice. Pathological examination revealed well-circumscribed lesions with compressed adjacent parenchyma, loss of lobular architecture and moderate fatty changes and negative GS staining.

Conclusion: In summary, our new hybrid BASH model functions as an excellent tumor promoter for liver tumorigenesis in mice. This observation supports the importance of identifying patients with excess alcohol consumption and MS as they are at a higher risk of liver-related cancer.

P067YI Thyroid disease and hepatocellular carcinoma survival. A Danish nationwide cohort study

Linda Skibsted Kornerup[1], Frederik Kraglund[1], Ulla Feldt-Rasmussen[2], Peter Jepsen[1], Hendrik Vilstrup[1]

[1]Aarhus University Hospital, Dept. of Hepatology and Gastroenterology , Aarhus N, Denmark, *[2]Rigshospitalet, Dept. of Endocrinology, Copenhagen, Denmark*
Email: lindajen@rm.dk

Background and Aims: Hepatocellular carcinoma (HCC) is the fourth leading cause of cancer mortality worldwide. Recent animal studies suggest that thyroid hormone treatment improves HCC prognosis.

The aim of this study was to describe the association between thyroid disease and HCC prognosis in humans.

Method: We performed a nationwide cohort study including all persons with an HCC diagnosis from 2000-2018. Patients' age, sex, HCC treatment, and diagnoses of thyrotoxicosis, nontoxic goitre, and myxoedema, were obtained from Danish national healthcare registries. We used regression models to examine the association between thyroid disease and mortality hazard and restricted mean survival time after HCC diagnosis, adjusting for confounding by sex and age.

Results: We included 4,812 patients with HCC and 107 patients with thyroid disease. Median follow-up time was 5 months (total 5,985 person-years). The adjusted mortality hazard ratio was 0.68 (95% CI 0.47-0.96) for thyrotoxicosis and 0.60 (95% CI 0.41-0.88) for nontoxic goitre. The restricted mean survival time during the five years following HCC diagnosis was 6.8 months (95% CI 1.1–12.6) longer for HCC patients with thyrotoxicosis than for patients without thyroid disease, and it was 6.9 months (95% CI 0.9–12.9) longer for HCC patients with nontoxic goitre than for patients without thyroid disease.

Conclusion: In this large nationwide cohort study, thyrotoxicosis and nontoxic goitre were associated with prolonged HCC survival.

Figure:

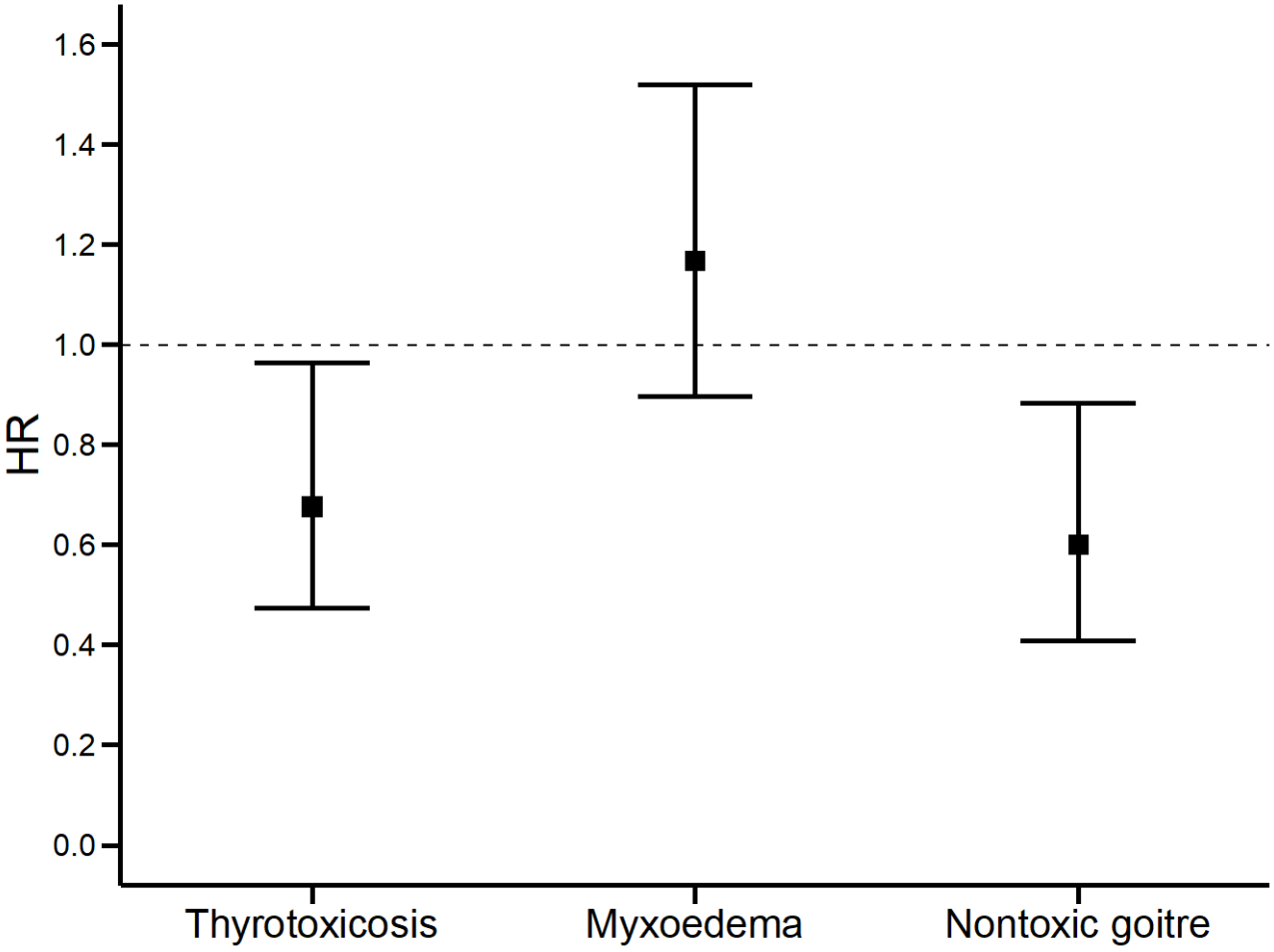


Figure 1. The effects of thyroid diseases on the hazard of all-cause mortality adjusted for sex and age.

P068YI Usefulness of circulating tumor cells in the management of patients with hepatocellular carcinoma

Elena Vargas Accarino[1], Monica Higuera[1], Maria Torrens[1], Gloria Torres[1], Joan Martínez-Campreciós[1], Monica Pons[1], Maria Teresa Salcedo[2], Serres Xavier[3], Itxarone Bilbao[4], Beatriz Minguez[1 5]

[1]Vall d'Hebron Institute of Research, Liver Diseases Research Group, Barcelona, Spain, *[2]Vall d'Hebron Universitary Hospital, Pathology Department, Barcelona, Spain*, *[3]Vall d'Hebron Universitary Hospital, Radiology Department, Barcelona, Spain*, *[4]Vall d'Hebron Universitary Hospital, General and Digestive Surgery Department, Barcelona, Spain*, *[5]Vall d'Hebron Universitary Hospital, Liver Unit, Barcelona, Spain*
Email: bminguez@vhebron.net

Background and Aims:
Circulating tumor cells (CTCs) circulate through the bloodstream after being shed from the primary tumor. Previous studies have demonstrated a correlation between the number of detected CTCs in blood and the prognosis of cancer patients. However, there are few studies that, in addition to counting CTCs, evaluate molecular information derived from them. The objective of this study is to develop a method counting and isolating CTCs, in order to capture molecular information.

Method:
Thirty-six patients with hepatocellular carcinoma (HCC) undergoing curative treatments (surgical resection and radiofrequency ablation) in our center have been prospectively included to date. Blood samples were obtained at the moment of the curative treatment and since then, every 3 months during the regular follow up. 10ml of blood were collected and the mononucleated cell fraction was extracted by centrifugation. CD45 positive cells were eliminated from this fraction and the rest of the cells were labeled with 3 antibodies against proteins described as HCC markers. Once marked, the cells were counted, separated and collected by fluorescence-activated cell sorting (FACS).

Results:
CTCs are given as a ratio referred to CD45 negative cells, expressed in percentage. Mean CTCs ratio in the 36 basal samples (before treatment) was 3.05%; significantly higher than what was obtained in the healthy controls (0.11%). The mean CTCs ratio in the samples after the curative treatment was significantly lower than in the basal samples; being 3.05% before treatment and 0.67%, 0.66% and 0.1% 3, 6 and 9 months after treatment.
Cells positive for these antibodies and negative for CD45 were visualized by confocal microscopy. They had a round morphology and a high nuclear-cytoplasmic ratio, typical from CTCs.

Conclusion:
Counting and isolating CTCs by cell sorting is possible in HCC patients. CTCs ratio in HCC patients drops after curative treatments supporting that counting CTCs could be useful in HCC patients´ follow up. The detection and monitoring of CTCs is a minimal invasive marker for HCC patients that could be used as a liquid biopsy. Furthermore, the CTCs isolation by cell sorting is allowing us to characterize the isolated cells by confocal microscopy and to perform molecular studies. This is a recently started prospective study that will provide valuable information about the diagnostic and prognostic utility of CTCs in HCC.

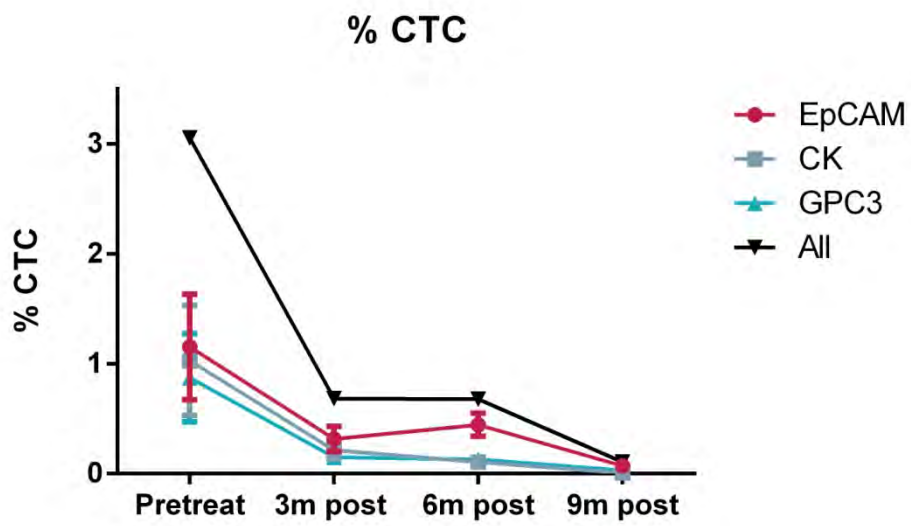

% CTC
% CTC
3
2
1
0
Pretreat
3m post
6m post
9m post
EpCAM
CK
GPC3
All

Figure:

P069YI Prognostic value of metabolic imaging data of ^{11}C-choline PET/CT in patients undergoing hepatectomy for hepatocellular carcinoma

Jacopo Galvanin[1], Matteo Donadon[1 2], Egesta Lopci[3], Simone Giudici[1], Daniele Del Fabbro[1], Ezio Lanza[4], Vittorio Pedicini[4], Arturo Chiti[2 3], Guido Torzilli[1 2]

[1]Humanitas Research Hospital IRCCS, Division of Hepatobiliary and General Surgery, Rozzano, Milano, Italy, *[2]Humanitas University, Department of Biomedical Science, Pieve Emanuele, Milan, Italy*, *[3]Humanitas Research Hospital IRCCS, Department of Nuclear Medicine, Rozzano, Milano, Italy*, *[4]Humanitas Research Hospital IRCCS, Department of Radiology, Rozzano, Milano, Italy*
Email:

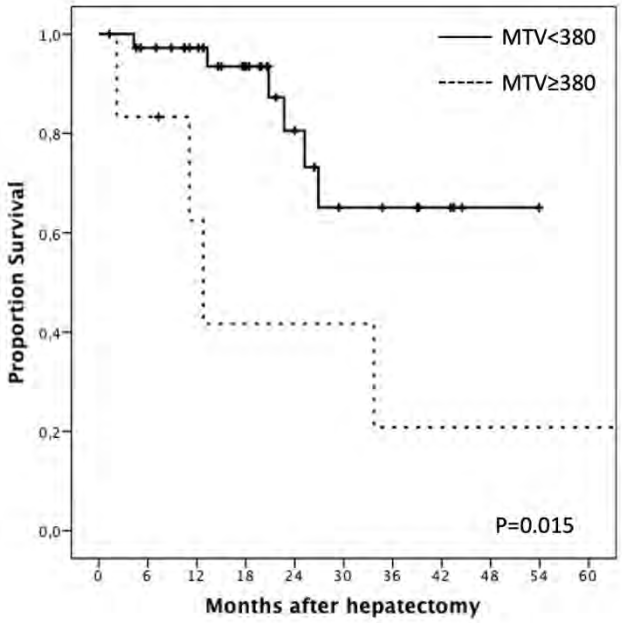
Background and Aims:
^{11}C-choline positron emission tomography/computed tomography (PET/CT) has been used for imaging patients with some types of solid cancers, but to date few data are available in patients with hepatocellular carcinoma (HCC). The aim of this study was to analyze the clinical significance of metabolic imaging data of ^{11}C-choline PET/CT in patients with HCC investigated before hepatectomy.

Method:
Our prospective institutional database was queried for patients with HCC preoperatively staged with ^{11}C-choline PET/CT. This imaging modality was performed in addition to standard abdominal CT or magnetic resonance imaging. Seven parameters were recorded for PET/CT: maximum standardized uptake value (SUVmax), mean standardized uptake value (SUVmean), liver standardized uptake value (SUVliver), metabolic tumor volume (MTV), photopenic area, metabolic burden (MTVxSUVmean), and SUVratio (SUVmax/SUVliver). Multivariate analysis was performed to identify parameters that could be predictors of overall survival (OS).

Results:
The study included 60 patients. Fourteen (23%) patients were in stage 0-A, 37 (62%) in stage B, and 9 (15%) in stage C of the Barcelona classification. The Cox regression for OS showed that Barcelona stages (HR=2.94; 95%CI=1.41-4.51; p=0.003) and MTV (HR=2.11; 95%CI=1.51-3.45; p=0.026) were the only factors independently associated with survival. Receiver operating characteristics curve analysis revealed valuable MTV ability in predicting survival (AUC=0.77; 95%CI=0.57-097; p<0.001), with a cutoff value of 380 (Se=82%; Sp=44%). Patients with MTV ≥380 had significant worse survival (p=0.015) (Figure 1).

Conclusion:
The use of ^{11}C-choline PET/CT allows for better prognostic refinement in patients undergoing hepatectomy for HCC. Incorporation of such metabolic modality into HCC staging system should be considered.

Figure:


MTV<380
MTV≥380
Proportion Survival
1,0
0,8
0,6
0,4
0,2
0,0
P=0.015
0
6
12
18
24
30
36
42
48
54
60
Months after hepatectomy

P070 First real-life experience with atezolizumab plus bevacizumab in the treatment of advanced hepatocellular carcinoma

Martin Schoenlein[1], Thorben Fruendt[2], Johann von Felden[2], Kornelius Schulze[2], Berit Behrends[3], Thomas Renne[3], Gerhard Adam[4], Ansgar W. Lohse[2], Henning Wege[2 5]

[1]University Medical Center Hamburg-Eppendorf, Oncology and Hematology, Hamburg, Germany, *[2]University Medical Center Hamburg-Eppendorf, Gastroenterology and Hepatology, Hamburg, Germany*, *[3]University Medical Center Hamburg-Eppendorf, Institute of Clinical Chemistry and Laboratory Medicine, Hamburg, Germany*, *[4]University Medical Center Hamburg-Eppendorf, Diagnostic and Interventional Radiology, Hamburg, Germany*, *[5]Cancer Center Esslingen, Esslingen, Germany*
Email: m.schoenlein@uke.de

Background and Aims:
In the recently published phase 3 IMbrave150 study, the PD-L1-inhibitor atezolizumab plus bevacizumab (Atezo+Bev) significantly extended the overall and progression-free survival (PFS) in comparison to sorafenib (6.8 versus 4.3 months) in the treatment of unresectable hepatocellular carcinoma (HCC). In addition, an objective radiological response rate of 33% was reported (13% in sorafenib) employing HCC-specific mRECIST. The aim of this study is to report first real-life experience with Atezo+Bev in the treatment of HCC.

Method:
Between January and September 2020, we treated 14 patients with advanced HCC (BCLC stage C or B with new lesions following TACE) with Atezo+Bev in a compassionate use program. Atezo+Bev was administered every three weeks. Patients were monitored by 3-weekly outpatient visits and laboratory work up, including AFP, AFP-L3, and DCP. After every 4 cycles (±1 cycle), imaging studies were performed and evaluated per mRECIST. Adverse events were graded using CTCAE 5.0.

Results:
9 patients (7 male, 2 female) with a median age of 66 (range 47 to 76) years who received Atezo+Bev as first line systemic treatment were included in this analysis (the other 5 patients received Atezo+Bev as a latter treatment line). Cirrhosis in these patients was due to hepatitis C (1), alcoholic (2) or non-alcoholic (4) steatohepatitis, or without known cause (2). At start of Atezo+Bev, 7 patients fulfilled strict eligibility criteria (SEC) and 2 patients were Child-Pugh B7. In total, we administered 64 cycles (total observed treatment time 48 months) with 3 patients having received 10 or more cycles until data analysis. Up until data cut in September 2020, 2 patients have presented with progressive disease (PD) after 6.3 and 2.2 months, whereas 1 patient showed a partial response (PR) after 4.2 months, and the remaining patients presented with stable disease (SD). Interestingly, DCP values in- or decreased already after 1-2 cycles before radiologically observed PD or PR, respectively. 5 out of 9 patients experienced grade 1 or 2 adverse events. The only grade 3 event reported was hypothyroidism.

Conclusion:
In this first real-life experience, including patients with Child-Pugh score B7, Atezo+Bev was well tolerated with only one grade 3 adverse event and no treatment interruption due to adverse events. Objective response rates were less frequent than expected. Finally, DCP emerged as promising marker for early response prediction.

P071 Extracellular signal-regulated kinase 5 regulates the malignant phenotype of cholangiocarcinoma cells

Alessandra Gentilini[1], Giulia Lori[1], Alessandra Caligiuri[1], Chiara Raggi[1], Giovanni Di Maira[1], Benedetta Piombanti[1], Tiziano Lottini[1], Jesus Maria Banales[2], Sabina Di Matteo[3], Domenico Alvaro[4], Jesper Andersen[5], Lea Duwe[5], Alessandro Tubita[6], Elisabetta Rovida[6], Fabio Marra[1]

[1]University of Florence, Department of Experimental and Clinical Medicine, Florence, Italy, *[2]Health Research Institute, CIBERehd, Ikerbasque, Department of Liver and Gastrointestinal Diseases, San Sebastian, Spain*, *[3]Children's Hospital, IRCCS, Department of Immunology, Bambino Gesù , Rome, Italy*, *[4]Sapienza University of Rome, Department of Internal Medicine and Medical Specialties, Rome, Italy*, *[5]University of Copenhagen, Biotech Research and Innovation Centre (BRIC), Dept. of Health and Medical Sciences, Copenagen, Denmark*, *[6]University of Florence, Department of Experimental and Clinical Biomedical Sciences "Mario Serio", Florence, Italy*
Email: alessandra.gentilini@unifi.it

Background and Aims:
Cholangiocarcinoma (CCA) is characterized by a high resistance to chemotherapy and a poor prognosis. Several oncogenic pathways converge on activation of extracellular signal-regulated kinase 5 (ERK5), whose role in CCA has not been explored. The aim of this study was to investigate the role of ERK5 in the biology of CCA.

Method:
Two lines of human intrahepatic cholangiocarcinoma (HuCCT-1 and CCLP-1) and two primary human iCCA cells (iCCA58 and iCCA60) were used. Western blotting technique, RTPCR, cell cycle analysis, MTT assay, cell count, migration and invasion assays, Huvec assay and xenograft model were employed in this study.

Results:
ERK5 expression was detected in two lines of human intrahepatic cholangiocarcinoma (HuCCT-1 and CCLP-1) and two primary human iCCA cells (iCCA58 and iCCA60). ERK5 phosphorylation was increased in CCA cells exposed to EGF. Growth, migration and invasion of CCA cells was decreased when ERK5 was silenced using specific shRNA. The inhibitory effects on migration and invasion results were recapitulated by cell treatment with small molecule inhibitors targeting ERK5. In addition, expression of the angiogenic factors VEGF and Angiopoietin 1 was reduced after ERK5 silencing, and conditioned medium (CM) from ERK5-silenced cells had a lower ability to induce tube formation by HUVEC and to induce migration of myofibroblasts and monocytes/macrophages. In mice, subcutaneous injection of CCLP-1 cells silenced for ERK5 resulted in less frequent tumor development and smaller size of detectable xenografts compared to cells transfected with non-targeting shRNA.

Conclusion:
ERK5 is a key mediator of growth and motility of CCA cells, and mediates a pro-tumoral cross-talk with the microenvironment of CCA.

P072YI Alteration of miRNA content in extracellular vesicles in Sorafenib-treated liver cancer cells. miRNA prognostic value in plasma from patients with advanced HCC treated with Sorafenib

Patricia de la Cruz-Ojeda[1 2], Elena Navarro-Villarán[1 3], María Isabel Gómez-Espejo[4], Inés María Fernández-Luque[4], Manuela Fátima Domínguez[5], Víctor Sapena[6], Juan Manuel Praena-Fernández[7], Félix Royo[8], María Azparren-Angulo[8], Francisco Javier Castell-Monsalve[9], Concepción Meana[5], Loreto Boix[3 6], Álvaro Giráldez[5], María Reig[3 6], Juan Manuel Falcón-Pérez[8 10], Jordi Bruix[3 6], María Teresa Ferrer[5], Jordi Muntané[1 2 3 4]

[1]Institute of Biomedicine of Seville (IBiS), Hospital University "Virgen del Rocío"/CSIC/University of Seville, Spain, *[2]Department of Surgery, University of Seville, Seville, Spain*, *[3]Biomedical Research Center for Hepatic and Digestive Diseases (CIBERehd), Madrid, Spain*, *[4]Department of General Surgery, Hospital University "Virgen del Rocío"/CSIC/University of Seville/IBIS, Seville, Spain*, *[5]Department of Gastroenterology, Hospital University "Virgen del Rocío"/CSIC/University of Seville/IBIS, Seville, Spain*, *[6]BCLC Group, Liver Unit, Hospital Clinic, University of Barcelona, IDIBAPS, CIBEREHD, Barcelona, Spain*, *[7]Department of Nursery, CSIC-JA-University of Seville-Institute of Biomedicine of Seville, Seville, Spain*, *[8]Exosomes Laboratory and Metabolomics Platform, CIC bioGUNE-BRTA, CIBERehd, Bizkaia Technology Park, Derio, Spain*, *[9]Department of Radiodiagnosis, Hospital University "Virgen del Rocío"/CSIC/University of Seville/IBIS, Spain*, *[10]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain*
Email: patricia.cruz.ojeda@gmail.com

Background and Aims:
Sorafenib is the second-line systemic therapy in patients with advanced hepatocellular carcinoma (HCC). Treatment resistance is related to the heterogeneity of tumoral genetic alterations and upregulation of survival pathways. Extracellular vesicles (EVs) participate in the intracellular signaling, cellular transformation and tumor microenvironment. The study assessed the alteration of EVs release and their miRNA signature in Sorafenib-treated HepG2 cells. The functional relevance of the identified miRNA was also assessed, and their content determined in plasma from two independent cohorts of patients with advanced HCC treated with Sorafenib.

Method:
Differentially expressed miRNAs in Sorafenib treated HepG2 cells were analyzed by TaqMan® OpenArray® technology. The impact of these miRNAs on proliferation, apoptosis, autophagy, migration and invasiveness was assessed through functional assays using mimics or inhibitors. EVs were isolated through differential ultracentrifugation and they were characterized by Western-blotting, Nanoparticle Tracking Analysis and cryo-electron microscopy analysis. miRNA profile was determined in cell lysate, culture medium, and large, small and very small EVs in Sorafenib-treated HepG2 (6 and 24 hours), as well as in plasma obtained from two independent cohorts of patients with advanced HCC treated with Sorafenib (pre- and 1 month after treatment) (study cohort n=23, validation cohort n=80)

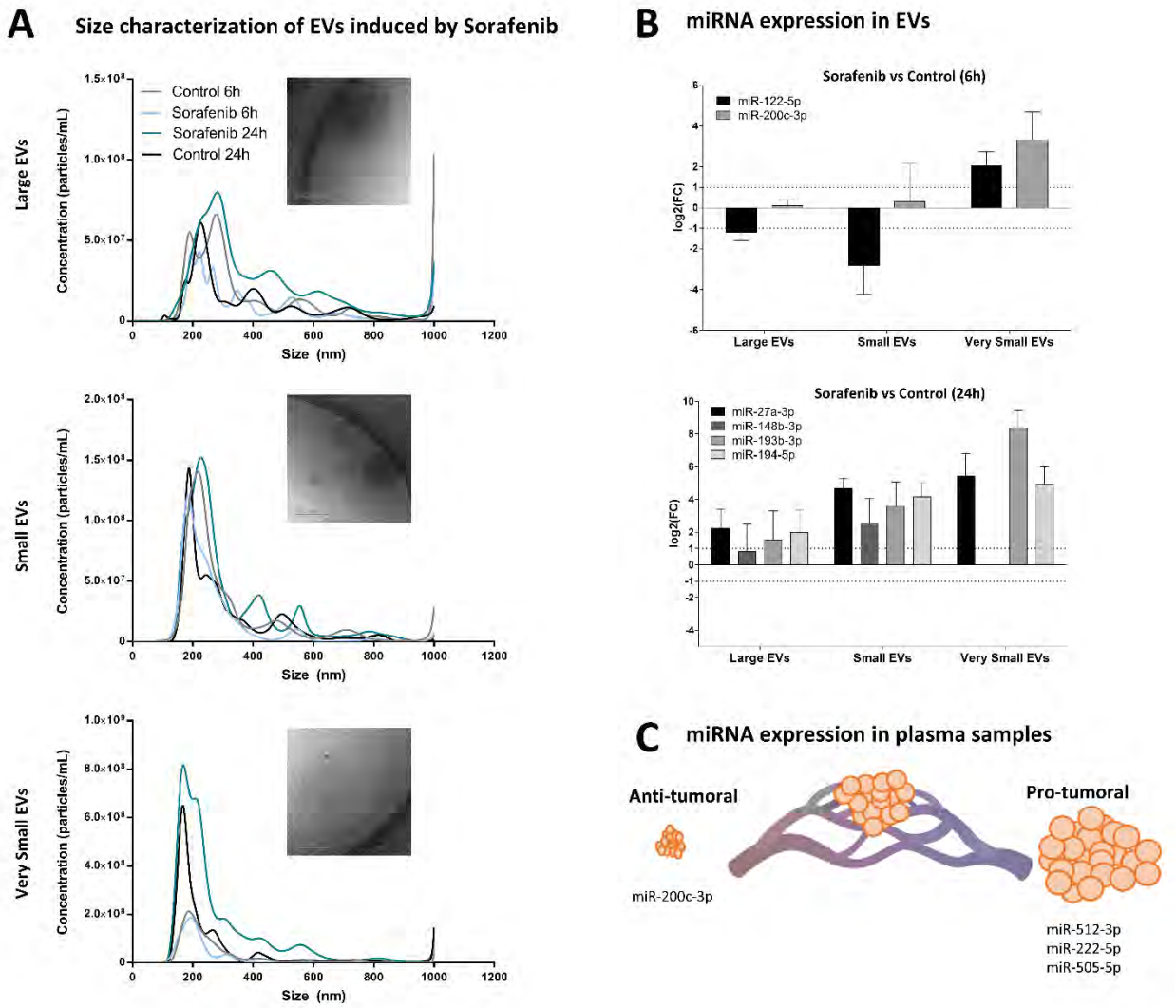
Results:

The functional analysis showed that miR-27a-3p, miR-148-3p, miR-194-5p, miR-200c-3p and miR-512-3p reduced proliferation, migration and invasiveness in Sorafenib-treated HepG2 cells. By contrast, miR-222-5p, miR-505-5p and miR-122-5p exerted protumoral activity in HepG2 cells. Electron microscopy and Nanoparticle Tracking analysis revealed that Sorafenib shifted the release of EVs from very small to larger sized fractions and increased the secretion of extracellular proteins (Figure 1A). miRNA analysis of EVs showed that Sorafenib particularly enriched miR-122-5p and miR-200c-3p (6 hours) and miR-27a-3p, miR-193b-3p and miR-194-5p (24 hours) in the very small EV fraction from cultured HepG2 cells (Figure 1B). The study in patients showed that patients with increased levels of miR-200c-3p have a lower risk of death/rapid progression, and those with miR-222-5p, miR-505-5p and miR512-3p have a higher risk of disease progression

Conclusion:

The antitumoral properties of Sorafenib were associated with the alteration of EVs release, as well as their miRNA content in HepG2 cells. Sorafenib induced the expression of miRNA that exerted antitumoral properties, while other miRNAs might be related to treatment resistance. The clinical study showed potentially relevant role of miRNA as a prognostic value of Sorafenib responsiveness in patients in advanced stage of HCC

Figure:



P073 Virological and oncological factors of HCC recurrence after liver trasplantation in a single transplant center

Martina Gambato[1], Alberto Ferrarese[1], Alberto Zanetto[1], Monica Pellone[1], Sarah Shalaby[1], Sara Battistella[1], Giacomo Germani[1], Marco Senzolo[1], Francesco Paolo Russo[1], Patrizia Burra[1]

[1]*Multivisceral Transplant Unit and Gastroenterology, Department of Surgery, Oncology and Gastroenterology, Padua University Hospital, Padua, Italy., Padua, Italy*
Email: martina.gambato@gmail.com

Background and Aims:
After the advent of prophylaxis with nucleot(s)ide analogues (NUCs) and anti-HBs globulins (HBIG), the risk of HBV recurrence after liver transplantation (LT) dropped down. The role of HBV serological factors in HCC recurrence after LT is still unclear. The aims were to evaluate the association between virological and oncological factors and HCC recurrence after LT. The role of HCC in the recurrence of HBV infection after LT was also assessed.

Method:
The study included all HBsAg positive patients transplanted in the Liver Transplant Centre of Padua University-Hospital from January 2007 to December 2018. HCC recurrence and HBV reinfection after LT were registered. Pre-LT virological variables (HBV DNA, HBeAg, HCV/HDV co-infection) and oncological variables (AFP, HCC features at explant pathology) were considered. The antiviral therapy (NUCs and HBIG) before and after LT was evaluated.

Results:
133 patients transplanted for HBV-related disease were included. 79 patients (59%) had HCC. Patients were followed for a median time of 46 months (range 1-157 months). 10% of patients had HCC recurrence after a median time of 14.5 months (2-63) post-LT. At the univariate analysis, no virological factors were associated with an increased risk of HCC recurrence. The larger diameter of HCC nodules and the presence of vascular invasion were associated with an increased risk of tumor recurrence (p=0.0003 and p=0.0085, respectively). HBV reinfection occurred in 6% (n=8) after a median time of 15 months (3-22) after LT. HCC recurrence was associated with an increased risk of HBV recurrence after LT (p=0.0005).

Conclusion:
No virological factors related to HBV infection had an impact on HCC recurrence after LT. The most aggressive histopathological features of HCC were the main determinants of HCC recurrence. On the other hand, HCC recurrence was a risk factor for HBV reinfection of the graft. Therefore, post-LT HBV prophylaxis should be continued (NUCs and HBIG) in LT recipients transplanted for HCC.

Figure: none

P074YI Dynamics of endothelial progenitor cells in patients with advanced hepatocellular carcinoma

Claudia Campani[1], Manuela Capone[2], Francesco Liotta[2], Umberto Arena[2], Chiara Di Bonaventura[2], Stefano Colagrande[3], Francesco Annunziato[2], Fabio Marra[2]

[1]Università degli Studi di Firenze, Dipartimento di Medicina Sperimentale e Clinica, Firenze, Italy, *[2]Università degli studi di Firenze, Dipartimento di Medicina Sperimentale e Clinica*, *[3]Università degli studi di Firenze, Dipartimento di Scienze Biomediche, Cliniche e Sperimentali*
Email: claudiacampani.cc@gmail.com

Background and Aims:
Angiogenesis inhibitors are used for treatment of advanced hepatocellular carcinoma. Previous studies have highlighted the importance of circulating endothelial progenitor cells (EPC) as predictors of tumor vascularization and disease progression. However, only limited information is available on the levels of EPC and their dynamics upon treatment in HCC

Method:
We prospectively analyzed the levels of different populations of circulating EPC (totalCD34+, CD34+/CD133+, CD34+/KDR+, CD34+/133+/KDR+, CD34+/133-/KDR-) in patients with advanced HCC candidate to therapy with sorafenib. Patients were studied before the start of therapy (T0) and after two (T2) and eight weeks (T8) using high-performance flow-cytometry. Tumor response was evaluated at T8 according to mRECIST criteria. Patients were divided in: progressive disease (PD) and clinical benefit (all other responses).

Results:
Sixteen patients (15 men, mean age 71 years) were enrolled. The median alpha-fetoprotein at T0 was 129 ng/ml. Eight patients were intolerant to sorafenib and stopped the treatment before T8. At T8 five patients had CB and three had PD. The median OS was 132 days. At baseline, frequencies of CD34+KDR+ and CD34+CD133+KDR+ were strongly correlated with platelet count ($r=0.788$, $p<0.001$ and r 0.734, respectively, $p=0.001$). A borderline correlation between CD34+CD133+KDR+ cells and BMI was also found ($r=0.45$, $p= 0.08$). No correlation with other clinical features at baseline were found. Frequencies of all EPCs subpopulation declined at T8 compared with T0. Levels of CD34+/CD133+ were higher at T0 in patients with CB compared to patients with PD ($p=0.05$). Moreover, mean frequency of CD34+ cells significantly declined in patients with CB comparing T0/T2 and T0/T8.

Conclusion:
In patients with advanced HCC treated with sorafenib EPC levels are directly correlated with platelet count, suggesting a common activation of selected bone marrow pathways. Levels of a subset of EPC are higher at baseline in patients responding to an angiogenesis inhibitor.

P075YI Tumor Burden Score: a novel tool to predict immune-related liver injury during immunotherapy for hepatocellular carcinoma

Antonio D'Alessio[1 2], Nicola Personeni[1 2], Antonella Cammarota[1 2], Maria Giuseppina Prete[1 2], Giuseppe Ferrillo[2 3], Vittorio Pedicini[3], Tiziana Pressiani[1], valeria smiroldo[1], silvia bozzarelli[1], Laura Giordano[4], Armando Santoro[1 2], Lorenza Rimassa[1 2]

[1]Humanitas Clinical and Research Center - IRCCS, Humanitas Cancer Center, Department of Medical Oncology and Hematology, Rozzano, Milan, Italy, *[2]Humanitas University, Department of Biomedical Sciences, Pieve Emanuele, Milan, Italy*, *[3]Humanitas Clinical and Research Center - IRCCS, Department of Radiology, Rozzano, Milan, Italy*, *[4]Humanitas Clinical and Research Center - IRCCS, Humanitas Cancer Center, Department of Biostatistics, Rozzano, Milan, Italy*
Email: antonio.dalessio@cancercenter.humanitas.it

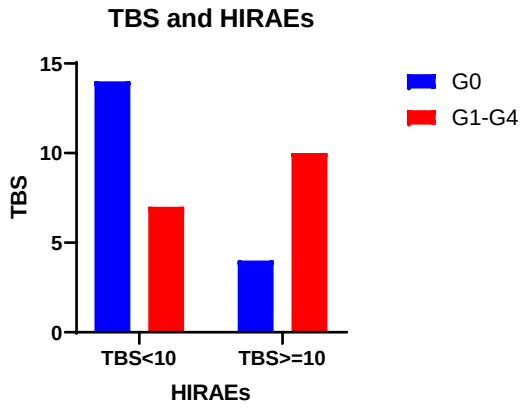
Background and Aims:
Treatment with immune checkpoint inhibitors (ICI) associated with the development of hepatic immune-related adverse events (HIRAEs) in approximately 9-20% of cases. While the risk factors for these adverse events are largely unknown, we aimed to evaluate the role of tumor burden as a determinant for development of HIRAEs.

Method:
Our ongoing analysis has included so far 36 patients with hepatocellular carcinoma (HCC) treated with a monoclonal antibody (mAb) targeting the programmed cell death receptor-1 or its ligand (PD-1/PD-L1) as single agent (16 patients, 44%) or in combination with a mAb against the cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) (20 patients, 56%). The pretreatment Tumor Burden Score (TBS) was calculated considering both the total number of liver nodules (a) and the maximum diameter (b) according to the following formula: $TBS^2 = a^2 + b^2$. Also, we used a ROC curve to set a TBS threshold which could be used to predict the onset of HIRAEs. HIRAEs were categorized according to the Common Terminology Criteria for Adverse Events (v. 5.0).

Results:
18 patients (50%) developed any grade HIRAEs, of whom 5 (18%) developed G3-G4 HIRAEs. No G5 AEs were registered. No patients permanently discontinued treatment because of HIRAEs. Patients who developed any grade HIRAEs tended to have significantly higher mean values of TBS compared to patients with no HIRAEs (13.4 [95% CI 8.3 - 18.5] vs 7.3 [95% CI 3.7 - 11.1], p = 0.048). We did not find a significant correlation between the mean baseline TBS value and the development of G3-G4 HIRAEs (12.0 vs 10.1, p = 0.70, in patients who developed G3-G4 HIRAEs vs those with a lower grade liver toxicity, respectively). From the analysis of the ROC curve, we chose a TBS threshold of 10, with an area under the curve of 0.70 (95% CI 0.52-0.87, p = 0.046).
Patients with a TBS of 10 or more had a significantly higher risk of developing any grade HIRAEs compared to patients with TBS < 10 (Odds ratio = 5 [95% CI 1.1-17.8], p = 0.041). Median overall survival did not significantly differ between patients with TBS<10 and patients with TBS≥10 (7.9 months vs 6.3 months, p = 0.60). Updated results will be presented.

Conclusion:
In HCC patients treated with ICI, hepatic tumor burden could be a risk factor for the development of any grade HIRAEs. TBS is a useful tool to measure hepatic tumor burden and could be helpful to predict the risk of HIRAEs. The role of TBS in predicting HIRAEs needs to be prospectively confirmed and validated in larger cohorts of patients.

Figure:



P076 Phase 3 KEYNOTE-937 Trial: Adjuvant Pembrolizumab for Hepatocellular Carcinoma and Complete Radiologic Response After Surgical Resection or Local Ablation

Arndt Vogel[1], Andrew Zhu[2], Ann-Lii Cheng[3], Thomas Yau[4], Jian Zhou[5], Raul N. Uppot[2], Eunhee Kim[6], Usha Malhotra[6], Abby B. Siegel[6], Masatoshi Kudo[7]

[1]Hannover Medical School, Hannover, Germany, *[2]Massachusetts General Hospital Cancer Center, Harvard Medical School, Boston, MA, United States*, *[3]National Taiwan University Cancer Center and National Taiwan University Hospital, Taipei, Taiwan*, *[4]University of Hong Kong, Queen Mary Hospital, Hong Kong, China*, *[5]Zhongshan Hospital Fudan University, Shanghai, China*, *[6]Merck & Co., Inc., Kenilworth, NJ, United States*, *[7]School of Medicine, Kindai University, Osaka, Japan*
Email: vogel.arndt@mh-hannover.de

Background and Aims:
Surgical resection and local ablation are potentially curative options for patients with hepatocellular carcinoma (HCC); however, tumor recurrence is not uncommon. There is an unmet medical need for standard-of-care adjuvant therapy to prevent disease recurrence and improve overall survival. Pembrolizumab, a programmed death receptor-1 blocking antibody, has shown evidence of a favorable benefit-to-risk profile in the adjuvant setting in many tumor types but has not been investigated in HCC. KEYNOTE-937 (NCT03867084) will evaluate the safety and efficacy of pembrolizumab versus placebo as adjuvant therapy in patients with HCC who had a complete radiologic response after surgical resection or local ablation.

Method:
KEYNOTE-937 is a randomized, double-blind, phase 3 study. Adults with confirmed HCC, complete radiologic response after surgical resection or local ablation, Eastern Cooperative Oncology Group performance status of 0 or 1, and Child-Pugh liver class A are eligible. Patients with past or ongoing hepatitis C or controlled hepatitis B virus infection may be enrolled if they meet prespecified criteria. Patients will be randomly assigned 1:1 to receive pembrolizumab 200 mg intravenously or placebo intravenously every 3 weeks for up to 17 cycles or until disease recurrence, unacceptable toxicity, or withdrawal, stratified by geographic region, prior local therapy (resection vs ablation), recurrence risk, and alpha-fetoprotein level at diagnosis. Coprimary end points are recurrence-free survival and overall survival; secondary end points are safety and tolerability (graded per National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.0) and health-related quality of life; and exploratory end points include distant metastases-free survival; time to recurrence; and genomic, metabolic, and/or proteomic biomarkers. Tumor imaging will be assessed until recurrence, and adverse events will be recorded up to 30 days after the last dose (90 days for serious adverse events).

Results:
Recruitment began in May 2019, and the planned sample size is 950 patients.

Conclusion:
KEYNOTE-937 will elucidate the efficacy and safety of pembrolizumab as adjuvant treatment in patients with HCC who had a complete radiologic response after surgical resection or local ablation.

P077YI PRPF8 regulates FAK/AKT pathway via fibronectin 1 splicing modulation to promote hepatocellular carcinoma aggressiveness

Juan Luis López-Cánovas[1 2 3], Natalia Hermán-Sánchez[1 2 3], Mercedes del Rio-Moreno[1 2 3], Antonio C. Fuentes-Fayos[1 2 3], Marina E. Sánchez-Frias[2 4], Victor Amado[4 5 6], Rubén Ciria[4 7], Javier Briceño[4 7], Manuel de la Mata[4 5 6], Manuel Rodriguez-Perálvarez[4 5 6], Raúl M. Luque[1 2 3], Manuel D. Gahete[1 2 3]

[1]Maimónides Institute of Biomedical Research of Córdoba, Department of Cell Biology, Physiology and Immunology, University of Córdoba, Córdoba, Spain, *[2]Reina Sofía University Hospital, Córdoba, Spain*, *[3]CIBER Pathophysiology of Obesity and Nutrition (CIBERobn), Córdoba, Spain*, *[4]Maimónides Institute of Biomedical Research of Córdoba, Córdoba, Spain*, *[5]Reina Sofía University Hospital, Department of Hepatology and Liver Transplantation, Córdoba, Spain*, *[6]CIBER Hepatic and Digestive Diseases (CIBERehd), Córdoba, Spain*, *[7]Reina Sofía University Hospital, Unit of Hepatobiliary Surgery and Liver Transplantation, Córdoba, Spain*
Email: b32locaj@uco.es

PRPF8 regulates FAK/AKT pathway via fibronectin 1 splicing modulation to promote hepatocellular carcinoma aggressiveness

Background and Aims:
A vast number of tumor pathologies, including hepatocellular carcinoma (HCC), are associated with the expression of aberrant splice variants involved in tumor development and/or progression. A dysregulation of the machinery responsible for the splicing process (spliceosome and splicing factors) could be responsible for these alterations. In fact, the expression of PRPF8, an essential component of the spliceosome, is dysregulated in some tumors; however, the role of PRPF8 has not been described in HCC. Thus, we aimed to analyze the expression of PRPF8 in different HCC cohorts, and to characterize its putative role in tumor development/progression.

Method:
PRPF8 expression (mRNA and protein) was analyzed in a retrospective cohort of patients with HCC (n=172: HCC and non-tumor tissues) and validated in two different in silico cohorts (TCGA and CPTAC). Functional and molecular consequences of PRPF8 silencing (using specific siRNAs) were evaluated in liver cancer (HepG2, Hep3B and SNU-387) cell lines and in Hep3b-induced xenograft tumors. Moreover, RNAseq and eCLIP data generated in HepG2 cells were analyzed.

Results:
This study shows that PRPF8 is elevated (mRNA/protein) in different HCC cohorts and associated with: i) increased tumor aggressiveness (tumor size, patient survival, etc.), ii) the expression of HCC-related splicing variants and, iii) the modulation of critical genes related to different cancer pathways. PRPF8 silencing ameliorated *in vitro* aggressiveness (reducing proliferation, migration, tumorospheres and colonies formation, while increasing apoptosis) and reduced tumor size *in vivo*. CLIPseq data in HepG2 demonstrated that PRPF8 binds preferentially to exons of protein-coding genes, and RNAseq analysis showed that PRPF8-silencing alters numerous splicing events, mainly exon skipping, of multiple genes. Integrated analysis of CLIPseq and RNAseq and *in vitro* experiments revealed that PRPF8-silencing modulates fibronectin (FN1) splicing, thus promoting the exclusion of exon 40.2, which is paramount for binding to integrins. Consistently, PRPF8 silencing reduced the FAK/AKT phosphorylation and blunted stress-fibres formation. Indeed, HepG2 cells exhibited lower invasive capacity in membranes treated with media from PRPF8-silenced cells compared to that observed with media from scramble-treated cells.

Conclusion:
PRPF8 is overexpressed and associated with aggressiveness in HCC, and exerts important roles in hepatocarcinogenesis, by altering FAK/AKT activation and stress-fibres formation through the modulation of FN1 splicing.

P078YI Retrospective analysis of one-center experience in diagnosis and treatment of liver cancer

Igor Chekmazov[1], Igor Ivanikov[1], Georgi Sapronov[1], Natalia Kirillova[1], Natalia Vinogradova[1], Yulia Grigorieva[1], Mariia Zharova[1 1 1]

[1]Tsentral'naya Klinicheskaya Bol'nitsa S Poliklinikoy, Moskva, Russian Federation
Email: zharowa.mariya@yandex.ru

Background and Aims:
We analyze the medical histories of patients with liver cancer of FSBI from 1981 to 2015.

Method:
In the analysis, we included patients diagnosed with the biliary tract and primary liver tumors (ICD-C22) between 1981 and 2015. We analyzed demographic data (gender, age), used diagnostic tests, mortality, and morbidity.

Results:
Over 35 years, 227 cases of primary liver tumors were diagnosed (66% men). The median age was 71.6 years. Intravital morphological verification of liver cancer ranged from 64% to 75%. Hepatocellular carcinoma was diagnosed more frequently than cholangiocarcinoma (4: 1). We found a steady tendency towards a decrease in morbidity and mortality in both sexes during 35 years. The incidence of liver cancer in the FSBI is higher than the national ones, but the mortality rate is much lower. Overall one-year mortality from liver cancer was 61.5%, and in the last five years, one-year mortality was 51.6%. We found that if cancer was found during a routine check-up, the 3-year survival rates were 20%, but if cancer was diagnosed in patients with primary hepatobiliary complaints, 3-year survival rates were 10% ($p<0,0001$).

Conclusion:
Active examination of patients from the high-risk group of the liver and hepatobiliary cancer, even without liver disease's primary symptoms, may decrease liver cancer mortality.

Figure:

P079 Analysis of risk factors associated with hepatocellular carcinoma development in patients with hepatitis B virus infection

Stavros Kanaloupitis[1], Evaggelos Zazas[1], Katerina Karaivazoglou[2], Maria Kalafateli[1], Konstanitnos Zisimopoulos[1], Ioanna Aggeletopoulou[1], Eirini-Eleni-Konstantina Kottaridou[1], Aspasia Antonopoulou[1], Dimosthenis Drakopoulos[1], Georgia Diamantopoulou[1], ANGELIKI TSINTONI[3], Konstantinos Thomopoulos[1], Christos Triantos[1]

[1]University Hospital of Patras, Division of Gastroenterology, Department of Internal Medicine, Patras, Greece, *[2]University Hospital of Patras, Department of Psychiatry, Patras, Greece*, *[3]University Hospital of Patras, Department of Internal Medicine, Patras, Greece*
Email: chtriantos@hotmail.com

Background and Aims:
Hepatitis B virus (HBV) infection constitutes a common cause of hepatocellular carcinoma (HCC) development. The identification of HCC development risk factors is essential for early diagnosis and treatment. The aim of the current retrospective study is to evaluate common clinical risk factors associated with HCC in HBV infected patients.

Method:
Nine hundred forty-three consecutive adult patients (n=943) [mean age: 50 years (range: 16-87)] with HBV, referred to our outpatients' Hepatology clinic between January 1993 and September 2020 were evaluated. Clinical data were evaluated as potential risk factors for HCC occurrence and Page-B score was calculated.

Results:
Fifty-eight patients (6.15%) presented with a baseline HCC, whereas thirty-seven patients (4.1%) developed HCC during follow-up. Thirty-two out of 37 patients were men. Mean age for HCC development was 62 years (range: 44-80) and mean BMI was 27 (range: 22.6-33.8). 60% of patients reported increased alcohol consumption at first presentation. 75.7 % had liver cirrhosis at the time of HCC diagnosis and 82% had chronic hepatitis B (CHB). Thirty-two patients received treatment during their follow up (37.5 % IFN, 62.5% NUCs). 70% of patients had a complete viral and biochemical response during the first year of their treatment. In 30 patients, due to treatment discontinuation or modification, a second treatment regimen with NUCs was used. Mean time to HCC development was 85 months (range: 1-180) and 70 months (range: 4-165) from treatment onset. Mean page-B score was 19 (range: 12-23), (60% high and 40% intermediate risk for HCC development).

Conclusion:
Most patients who developed HCC were overweight men, with liver cirrhosis and history of alcohol consumption. They developed HCC although they received a treatment regimen. Page- B scores appears to be a reliable prognostic factor for the evaluation of HCC development risk in HBV patients.

P080YI Serum levels of CXCL13 are an independent predictor of survival following resection of biliary tract cancer

Sven Loosen[1], Anjali A. Röth[2], Patrick Hamid Alizai[3], Jan Bednarsch[2], Christian Trautwein[4], Tom Florian Ulmer[2], Christoph Roderburg[1], Ulf Neumann[2], Tom Luedde[1]

[1]University Hospital Düsseldorf, Clinic for Gastroenterology, Hepatology and Infectious Diseases, Düsseldorf, Germany, *[2]University Hospital RWTH Aachen, Department of General, Visceral and Transplant Surgery, Aachen, Germany*, *[3]University Hospital RWTH Aachen, Department of General, Visceral and Transplant Surgery*, *[4]RWTH Aachen, Medicine III*
Email: sloosen@ukaachen.de

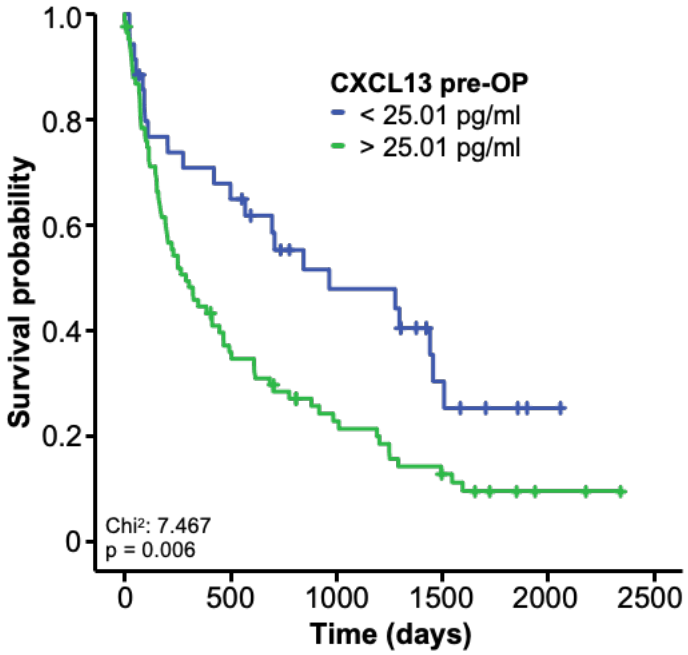
Background and Aims: The prognosis of biliary tract cancer (BTC) has remained poor. Although tumor resection represents a potentially curative therapy for selected patients, disease recurrence is common and 5-year survival rates remain below 50%. As stratification algorithms comprising parameters of the individual tumor biology are missing, the identification of the ideal patients for extensive liver surgery is often challenging. The CXC chemokine family exerts decisive functions in cell-cell interactions and has only recently been associated with cancer. However, only very little is known on their role in BTC. Here, we aim at evaluating a potential role of circulating CXCL1, CXCL10 and CXCL13 in patients with resectable BTC.

Method: Serum levels of CXCL1, CXCL10 and CXCL13 were measured by multiplex immunoassay in a cohort of 119 BTC undergoing tumor resection as well as 50 healthy control samples.

Results: Circulating levels CXCL1, CXCL10 and CXLC13 were all significantly elevated in BTC patients compared to healthy controls and increased the diagnostic power of established tumor markers when used in combination. Importantly, elevated levels of CXCL13 both before and after tumor resection identified a subgroup of patients with a significantly impaired outcome following tumor resection. As such, BTC patients with initial CXCL13 levels above the ideal prognostic cut-off value (25.01 pg/ml) had a median OS of 290 days compared to 969 days for patients with low initial CXCL13 levels. The prognostic value of circulating CXCL13 was further confirmed by uni- and multivariate Cox-regression analyses. Finally, the individual kinetic of CXCL13 before and after tumor resection was also indicative for patients' outcome.

Conclusion: Our data suggest a fundamental role of the CXC chemokine family in BTC and identified circulating levels of CXCL13 as a previously unrecognized parameter for the prediction of outcome following resection of BTC.

Figure1: At the optimal cut-off values, preoperative CXCL13 identify BTC patients with a significantly impaired postoperative overall survival.



P081YI

Validation of the aMAP risk score for the development of hepatocellular cancer in patients with cirrhosis in Glasgow

Chris Curran[1], Stephen Barclay[2]

[1]Queen Elizabeth University Hospital, Glasgow, United Kingdom, *[2]Glasgow Royal Infirmary, Department of Gastroenterology, Glasgow, United Kingdom*
Email: chriscurran152@gmail.com

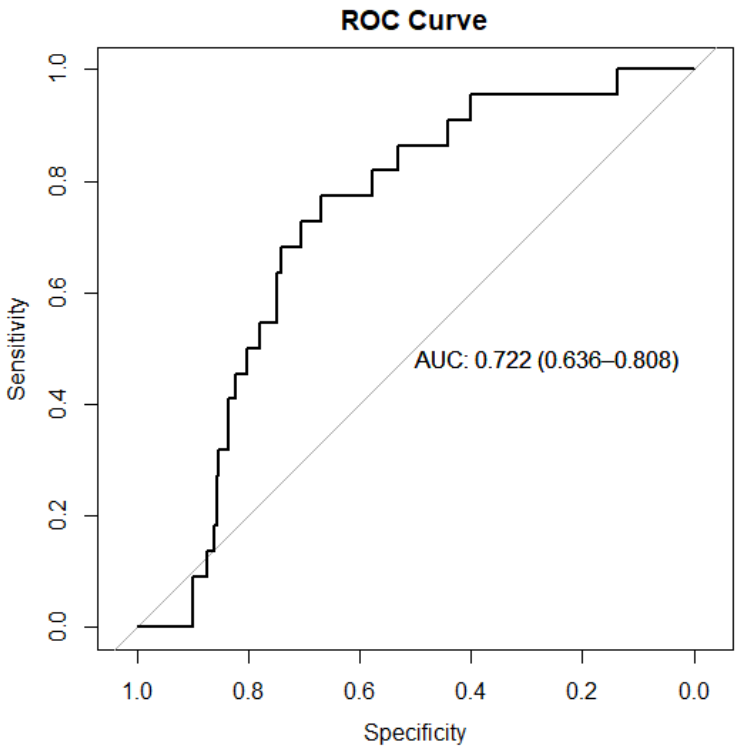
Background and Aims: Current European and American guidelines suggest surveillance for hepatocellular carcinoma (HCC) in cirrhotic patients is warranted in those with an annual risk of 1.5% or greater per year irrespective of aetiology. Multiple risk scores have been developed to stratify the risk of patients developing HCC with the potential aim of developing individualised screening strategies. The aMAP risk score, developed from a large cohort of patients with viral and non-viral hepatitis, consists of age, sex, albumin, bilirubin and platelets. It demonstrated excellent discrimination in assessing 5-year HCC risk in its original development and validation cohort. We aimed to see how the aMAP score performed in a cohort of mixed aetiology in Glasgow.

Method: Data was collected on 482 patients with cirrhosis who attended at least one clinic appointment between January 2013 and December 2014. Patients were followed up until 31/12/2019. The aMAP score was calculated based on blood results at the index clinic visit. Patient scores were then dichotomised into low, medium and high-risk groups as in the original paper. The receiver operator characteristic (ROC) curve was drafted to identify the value of the aMAP score in predicting HCC. Statistical analysis was carried out using R.

Results: The leading cause of cirrhosis in this cohort was alcohol with 262 (54.4%) patients having it as part of their aetiology. This compared with 143 (29.7%) and 93 (19.3%) patients with HCV and NAFLD respectively. At the end of follow up 22 (4.6%) patients had developed HCC. The area under the ROC curve for the aMAP score to predict HCC was 0.722 [95% CI 0.636 – 0.808]. The cumulative incidence of HCC in the low, medium and high risk groups was 0%, 0.73% and 7.34% respectively. No patient in the low risk group developed HCC over the course of follow up.

Conclusion: In this cohort of patients with cirrhosis from a broad range of aetiologies the aMAP score performed well and showed good predictive ability. The high negative predictive value of the lower risk groups suggests the potential of using the aMAP score as a tool to identify patients who may be able to undergo less intensive surveillance.

Figure:



P082YI In vivo quantification of micro-balloon interventions (MBI) advantage. Cohort, retrospective, bi-centric study of DEB-TACE vs b-TACE and SIRT vs b-SIRT

Pierleone Lucatelli[1], Gianluca De Rubeis[1], Sara Ungania[2], Bianca Rocco[1], Claudio Trobiani[1], Simone Zilahi De Gyurgyokai[1], Marica Masi[2], Irene Pecorella[3], Federico Cappelli[4], Giulio Vallati[4]

[1]University "Sapienza", Rome, Policlinico Umberto I, Vascular and Interventional Radiology, Rome, Italy, [2]Istituto Regina Elena Istituto di Ricovero e Cura a Carattere Scientifico, Physics, Rome, Italy, [3]Department of Radiological Sciences, Oncology and Anatomical Pathology, Policlinico Umberto I, University of Rome "Sapienza", Rome, Italy, Anatomical Pathology, Rome, Italy, [4]Istituto Regina Elena Istituto di Ricovero e Cura a Carattere Scientifico, Interventional Radiology Unit, Rome, Italy
Email: pierleone.lucatelli@gmail.com

Background and Aims:
This cohort, retrospective, bi-centric study purpose was to quantify in vivo the micro-balloon role by comparing TACE and SIRT procedures performed with and without balloon-microcatheter for HCC.

Method:
We treated 84 patients with hepatocellular carcinoma (HCC) using trans-arterial loco-regional therapy. 53/84 patients were treated with TACE, divided into 26/53 DEB-TACE and 27/53 b-TACE. 31/84 patients were treated with SIRT, divided into 24/31 SIRT and 7/31 b-SIRT.
Impact of balloon micro-catheter on trans-arterial loco-regional treatment was analyzed using: post-procedural cone beam CT (CBCT) after TACE/b-TACE, 2D and 3D dosimetry in SPECT after SIRT/b-SIRT and histological count of the bead following orthotopic liver transplantation (OLT) in the subgroup of TACE/b-TACE.

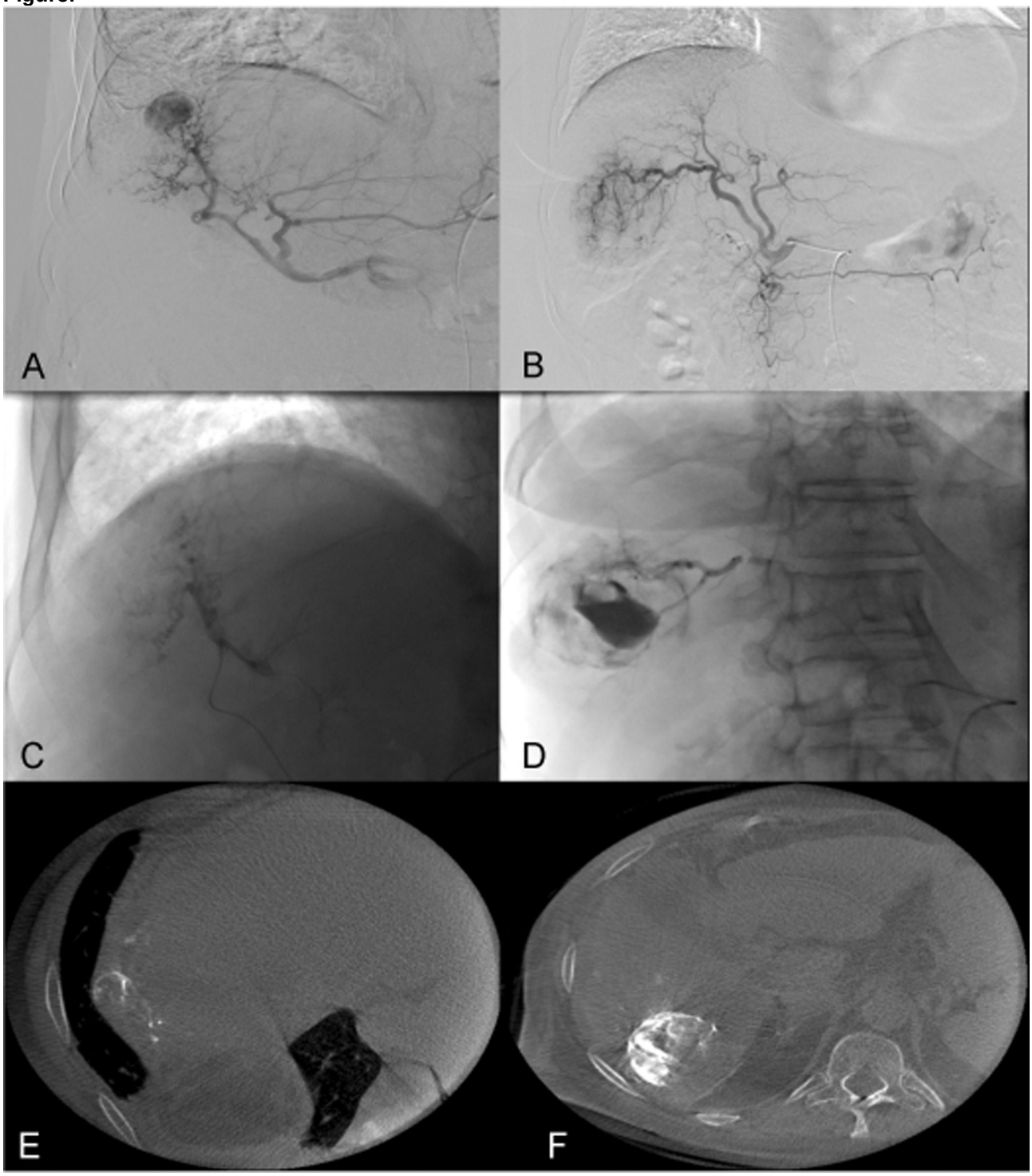
Results:
Fifty-three patients were analysed in TACE group. Contrast, signal to noise ratio, and contrast to noise ratio were significantly higher in b-TACE subgroup compared with DEB-TACE.

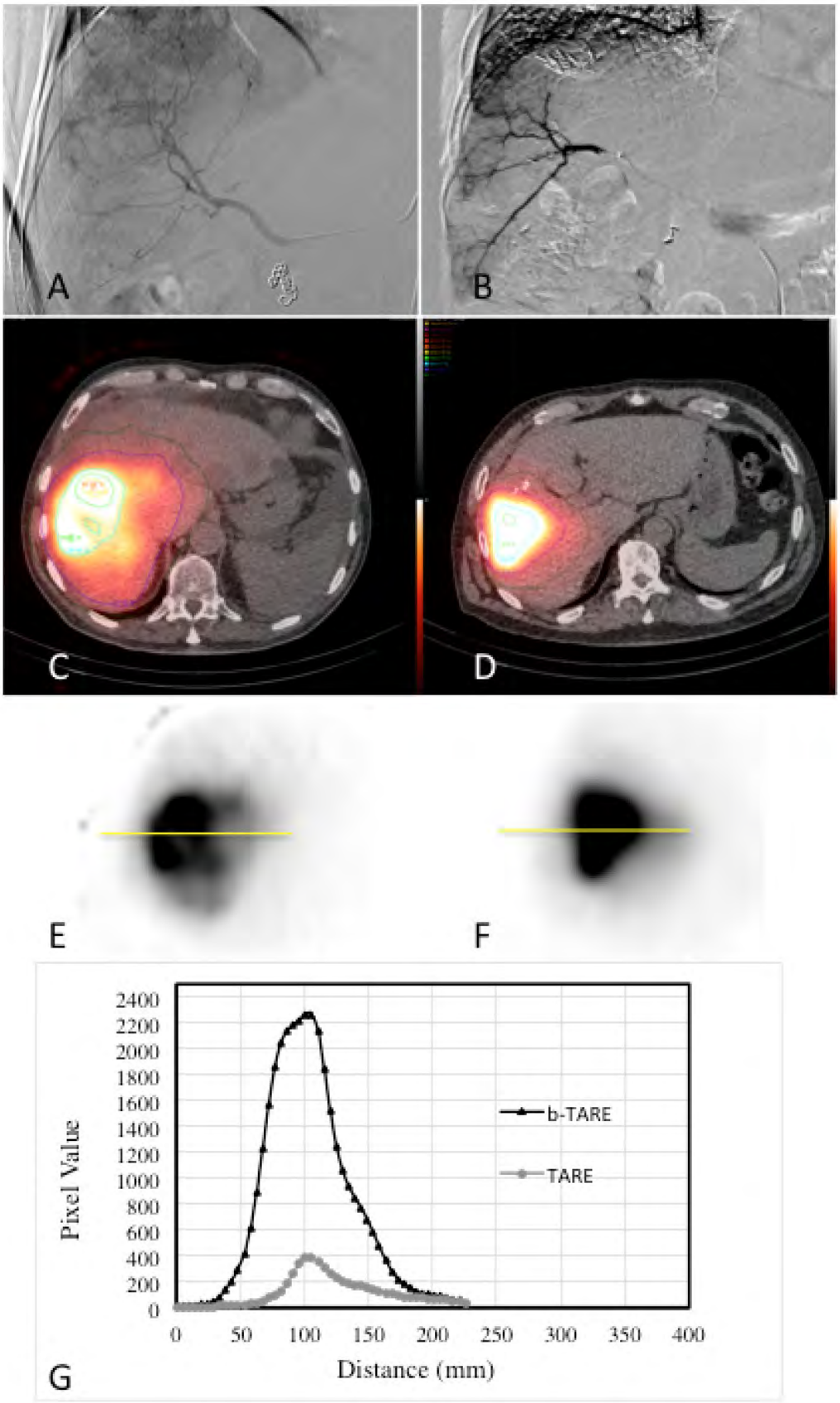
Thirty-one patients were analysed in SIRT group. b-SIRT had a better dosimetry profile both in 2D and 3D analysis. Concerning 2D evaluation, the activity intensity peak was significantly higher in the b-SIRT subgroup compared with SIRT. Regarding 3D dose analysis, the mean dose administered to the treated lesions was significantly higher in b-SIRT group than SIRT with almost no increase of the mean dose delivered to the normal liver.

Regarding the specimen analysis, there was a trend for higher intra-tumoral localization of PEG microsphere for b-TACE in comparison with DEB-TACE.

Conclusion:
The results of the present study quantify in vivo, thanks to the use of three different methods, the ameliorative embolization profile (measured as higher target lesion signal at non enhanced CBCT post b-TACE, higher signal at SPECT/CT post b-SIRT and higher lesion absorbed dose post b-SIRT) of oncological interventions performed with balloon-micro catheter.

Figure:




A
B
C
D
E
F
2400
2200
2000
1800
1600
1400
1200
1000
800
600
400
200
0
Pixel Value
b-TARE
TARE
0
50
100
150
200
250
300
350
400
Distance (mm)
G

P083 Comparison of Liver Transplant Criteria for Hepatocellular Carcinoma

Volkan INCE[1], Sertac Usta[1], Brian Carr[1], Veysel Ersan[1], Cemalettin Koc[1], fatih ozdemir[1], Harika G Bag[2], sezai yilmaz[1]

[1]Inonu University, Liver Transplantation Institute, General Surgery, Malatya, Turkey, *[2]İnönü Üniversitesi, Medical School, Biostatistics, Malatya, Turkey*
Email: volkanince@gmail.com

Comparison of Liver Transplant Criteria for Hepatocellular Carcinoma

Background and Aims:
To investigate which liver transplant (LT) criteria provide the longest overall and disease-free survival and lowest recurrence rates for patients with hepatocellular carcinoma (HCC).
We aimed to compare the results of liver transplantation, performed for HCC in our institute which is a high-volume liver transplant center, by analyzing it according to other published defined criteria.
In addition, this study is the first comprehensive comparison of liver transplant criteria for HCC.

Method:
A total of 430 patients were analyzed, who had HCC and were transplanted at the Liver Transplantation Institute of Inonu University, Malatya, Turkey, between 2006–2020.
Inclusion criteria: Patients with HCC in the explanted whole liver specimen were included. Patients with pre-transplant history of loco-regional therapy were also included
Exclusion criteria: The only exclusion criteria were patients with post-transplant follow up of less than 90 days (n=75) to focus on oncologic outcomes.
The remaining 355 LT patients who were transplanted with HCC were included in the study and analyzed**.** All of the recorded laboratory parameters were the last results before transplantation (within 2 weeks before-LT). Tumor morphology data were recorded according to the explant pathology report. We applied all LT criteria to our cohort. A total of 25 LT criteria from 10 countries were defined in the literature (Table 1).

Results:
Post-transplant 5-year overall, DFS and recurrence rates of the 355 patients were 67.8%, 68.7%, 19.1% respectively. Survivals and recurrence rates according to transplant criteria are summarized in Table 2. Some of the extended criteria such as, UpTo7, AFP-Model, AFP-TTD, Samsung, MoRAL classification, Metroticket-2.0 and 5-5-500 criteria exclude some patients within Milan criteria from LT. So, these criteria should not be extended criteria. Because they reduce the Milan Criteria**.** The recently published criteria of Extended Malatya Criteria provide 78.8% 5-year DFS and a 35.4% extension rate of Milan criteria while the reference criteria, Milan, provide 82.2% 5-year DFS.

Conclusion:
Reasonable extended criteria are the Extended Malatya criteria and Berlin criteria with 35.6%, 42.4% extension rate and 5-year DFS 78.8%, 79.1%, respectively (Figure).

Table 1: All of the transplant criteria for hepatocellular carcinoma

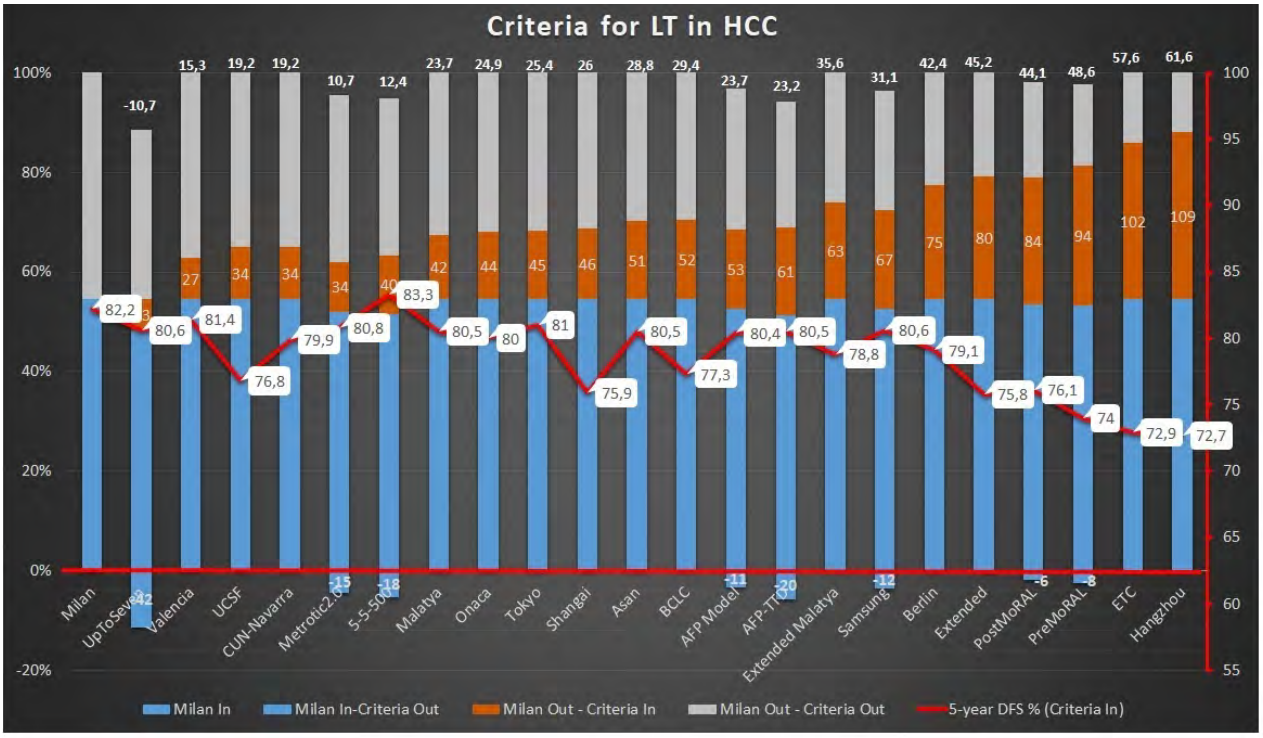
Criteria	Year	Features	Country	Author
Milan	1996	Single tm (≤5 cm) or ≤ 3 tm with the largest ≤3 cm*	Italy	Mazzaferro
UCSF	2001	Single tm (≤6.5 cm) or ≤ 3 tm with the largest ≤4.5 cm Total tm diameter ≤ 8 cm*	USA	Yao
BCLC	2002	Single tm (≤7 cm)- Three tm (≤5 cm)- Five tm (≤3cm)- Maintained response after downstaging to Milan Crit.	Spain	Bruix
Extended Criteria	2004	Single tm (≤7.5 cm) or multiple tm < 5 cm (each) **(>5cm nodule with poor diff were also excluded)**	Canada	Kneteman
Berlin	2007	No limit in tumor number MTD ≤ 6 cm and TTS ≤ 15 cm	Germany	Jonas
Kyoto	2007	Total number of tm ≤10-All tm size ≤ 5 cm- DCP ≤400	Japan	Ito
Tokyo (5-5-500 rule)	2007	Tm size ≤5 cm- Total number of tm≤ 5	Japan	Sugawara
Onaca	2007	Single tm ≤ 6 cm- 2-4 tm with largest tm ≤ 5 cm	USA	Onaca
Hangzhou	2008	Total tm diameter≤ 8 cm or Total tm diameter >8 cm with grade I-II tumor biopsy and AFP ≤ 400	China	Zheng
Asan	2008	Largest tm diameter≤ 5 cm- Total number of tm ≤ 6	S.Korea	Lee
CUN (Navarra)	2008	Single tm (≤6 cm) or ≤ 3 tm with the largest ≤5 cm	Spain	Herrero
Valencia	2008	Total tm diameter ≤ 10 cm- 1-3 tm ≤ 5 cm	Spain	Silva
Shanghai	2009	Single tm (≤9 cm)- ≤ 3 tm with the largest ≤5 cm Total tm diameter≤9 cm*	China	Fan
Up-to-Seven	2009	Total tm diameter≤ 7 cm-Total number of tm ≤ 7*	Italy	Mazzaferro
TTV/AFP Model	2009	TTV > 115 cm3 or AFP > 400	Canada	Toso
Kyushu University	2011	Any number of tm with diameter < 5 cm or DCP <300	Japan	Shirabe
Extended Toronto	2011	No limit in tm size/number- No *-No cancer related symptoms-biopsy of largest tm not poorly differentiated if beyond Milan	Canada	DuBay
AFP Model	2012	Largest tm size (point): ≤3 cm (0), 3-6 cm (1) >6 cm (4) Total number of tm (point): 1-3 tm (0) ≥4 tm (2); AFP (point): ≤100 (0), 100-1000 (2) >1000 (3)	France	Duvoux
AFP-TTD	2012	Total tm diameter ≤ 8 cm- AFP ≤ 400	Italy	Lai
Samsung	2013	Total number of tm ≤7- Largest tm ≤ 6 cm- AFP ≤ 1000	S.Korea	Kim
MORAL	2017	Pre-Moral: NLR ≥5, AFP > 200, Size >3 cm; Post-Moral: grade 4 HCC's, vascular invasion, size >3cm and number>3	USA	Halazun
Metroticket 2.0	2018	Total tm number and size: (≤ 7- AFP <200) or (≤ 5- AFP: 200-400) or (≤ 4- AFP: 400-1000)	Italy	Mazzaferro
5-5-500 Rule	2019	Tm size ≤5 cm- Total number of tm≤ 5- AFP ≤ 500	Japan	Shimamura
Malatya Criteria	2020	Largest tm diameter ≤ 6 cm- AFP ≤ 200- GGT≤104- Well/Moderate Tm differentiation	Turkey	Ince
Extended Malatya Criteria*	2020	Largest tm diameter ≤ 10 cm- AFP ≤ 200- GGT≤104	Turkey	Ince

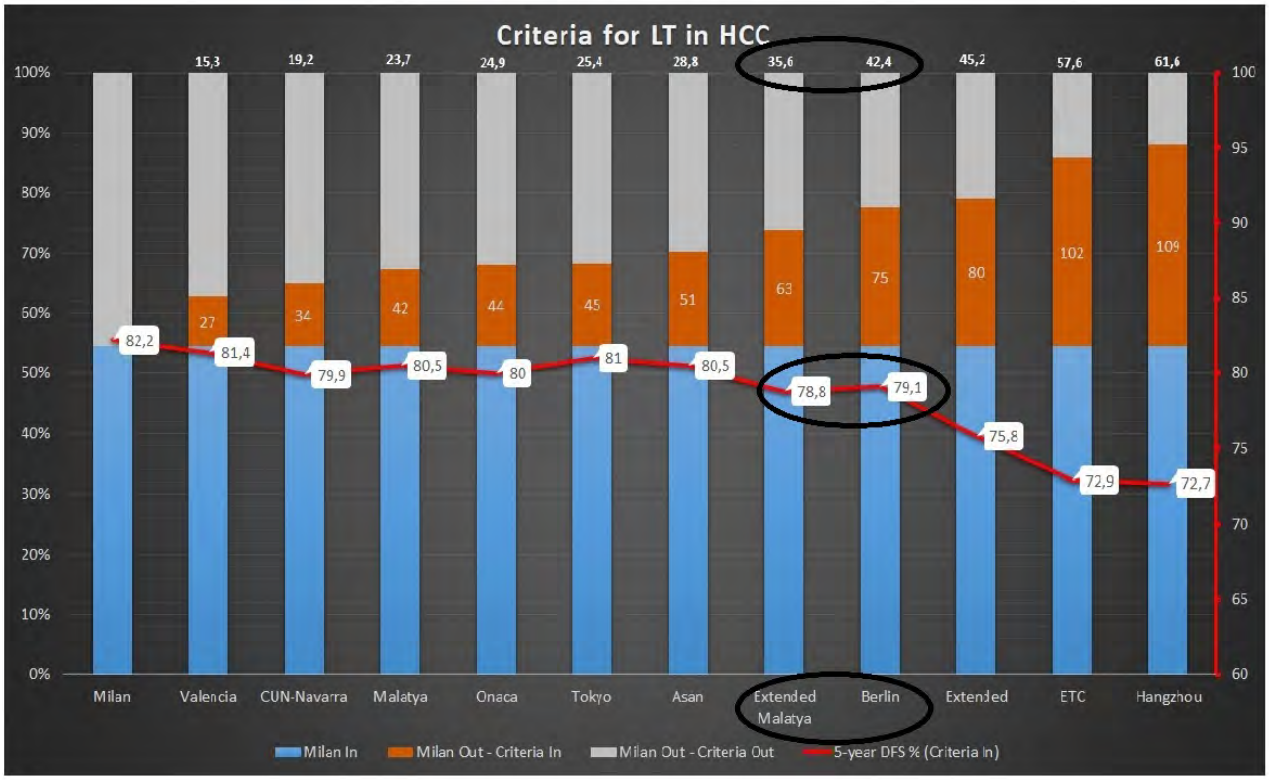
*Accepted for publication (World journal of Gastrointestinal Surgery 2020, (in press))

Table 2: Application of all transplant criteria to our total cohort (n=355).

Within Criteria	Expansion rate of Milan	5 year DFS	Recurrence Rate %	Milan in Criteria out Patient number	Milan in Criteria out Patient 5-year DFS	Milan in Criteria out Pts Rec (n)	Milan in Criteria out Pts Rec %	Milan out Criteria in Patient number	Milan out Criteria in Patient 5-year DFS	Milan out Criteria in Pts Rec %
All Cohort,		68.7	19.1							
Milan	Reference Criteria	82.2	3.4		82.2	11	5.2		53	27
UCSF	19.2	76.8	5.2					34	47.8	14.7
BCLC	29.4	77.3	5.2					52	58.6	11.5
Extended	45.2	75.8	7.4					80	61.9	16.3
Berlin	42.4	79.1	5.6					75	71.9	10.7
Tokyo	25.4	81	4.1					45	75	6.7
Onaca	24.9	80	4.5					44	69.1	9.1
Hangzhou	61.6	72.7	9.4					109	58	19.3
Asan	28.8	80.5	3.9					51	73.8	5.9
CUN-Navarra	19.2	79.9	4.3					34	65.7	8.8
Valencia	15.3	81.4	3.4					27	73.6	3.7
Shangai	26.0	75.9	6.3					46	51.2	17.4
UpToSeven	-10.7	80.6	3.8	42	86.4	2	4.8	23	78.1	8.7
ETC	57.6	72.9	9.7					102	58	20.6
AFP Model	23.7	80.4	3.2	11	77.1	2	18	53	73.2	5.7
AFP-TTD	23.2	80.5	3.2	20	74.1	5	25	61	70.9	9.8
Samsung	31.1	80.6	4.3	12	72.2	1	8.3	67	75	7.5
PostMoRAL	44.1	76.1	6.3	6	100	1	17	84	65.7	13.1
PreMoRAL	48.6	74	8.4	8	68.6	2	25	94	58.6	19.1
Metrotic2.0	10.7	80.8	2.6	15	78.3	2	13.3	34	71.4	2.9
5-5-500	12.4	83.3	1.0	18	70	4	22.2	40	81.4	0
Malatya	23.7	80.5	3.2					42	74.8	2.4
Extended Malatya	35.6	78.8	4.6					63	70.1	7.9

Figure: X axis is Tx criteria,Y axis on the left shows Tx-Patient percent,Y axis on the right shows DFS percent of patients within the criteria.The numbers on the top of the columns are extension rate of Milan Criteria.The numbers in orange column are patient numbers that beyond Milan but within the criteria.





P084 LDH as early prognostic markers for response in advanced hepatocellular carcinoma treated with Nivolumab

Birgit Schwacha-Eipper[1], Marcel Hug[1], Pompilia Radu[1], Jean-Francois Dufour[1 2]

[1]Inselspital, University Clinic for Visceral Surgery and Medicine, Bern , Switzerland, *[2]University of Bern, Hepatology, Department of Clinical Research, Bern, Switzerland*
Email: radupompilia@yahoo.com

Background and Aims:
Nivolumab, a programmed death (PD)-1 (PD-1) inhibitor, has shown encouraging results in advanced hepatocellular carcinoma (HCC) patients, mainly after sorafenib failure. In recent trials with immune checkpoint inhibitors elevated baseline lactat dehydrogenase (LDH) had been shown to correlate with poor survival and poor response rates. Identifying biochemical factors that may predict response to Nivolumab in HCC patients is still a matter of debate.

Method:
From May 2017 to April 2020, 31 HCC patients staged as BCLC B and BCLC C had received nivolumab in Inselspital, Bern. After exclusion of the patients who had less than 3 cycles of nivolumab, 25 patients were included. Based on the modified Response Evaluation Criteria in Solid Tumors (m RECIST), patients were classified into 2 groups: a responder group (patients with complete response (CR), partial response, or stable disease) and a non-responder group (progressive disease). LDH and AFP pre-treatment serum levels were assessed in our patients. Univariate and multivariate analysis of progression free survival (PFS) and overall survival (OS) were performed.

Results:
After median follow-up of 24.8 months, the overall response rate was 32% (8/25), including two CR. Median progression-free survival (PFS) and overall survival (OS) rates were 3.7 and 17.2 months, respectively. Multivariate analysis revealed that the presence of macrovascular invasion (MVI) and elevated LDH were independent factors associated with impaired OS [HR=0.139 (95% CI=0.035-0.553), p=0.005] and [HR=1.009 (95% CI=1.001-1.017), p=0.023], respectively. Elevated LDH and the presence of MVI were associated with poor PFS [HR= 1.011 (95% CI=1.002-1.021), p=0.021] and [HR=0.307 (95% CI=0.099-0.951), p=0.041], respectively.

Conclusion:
Pre-treatment serum LDH value might be a potential marker for response and OS in advanced HCC patients treated with nivolumab.

Figure:

Fig1. Kaplan Meier curves of overall survival according to low (below median) and high (above median) pretreatment LDH (p=0.005).

Fig 2. Kaplan Meier curves of PFS according to low (below median) and high (above median) pretreatment LDH values (p<0.001)

P085YI DNA Methyltransferases as Potential Biomarkers for Egyptian Patients with HCV Related Hepatocellular Carcinoma

Mona M Hassona[1], Mary Naguib[1], Enas M Radwan[2], Mohamed Abdel-Samiee[3], Suzanne Estaphan[4], Eman Abdelsameea[3]

[1]National Liver Institute, Menoufia University, Clinical and Chemical Pathology, Shebin El-Kom, Egypt, *[2]National Cancer Institute, Cairo University, Clinical pathology, Cairo, Egypt*, *[3]National Liver Institute, Menoufia University, Hepatology and Gastroenterology, Shebin El-Kom, Egypt*, *[4]Faculty of Medicine, Cairo University, Physiology, Cairo, Egypt*
Email: drmohammed100@yahoo.com

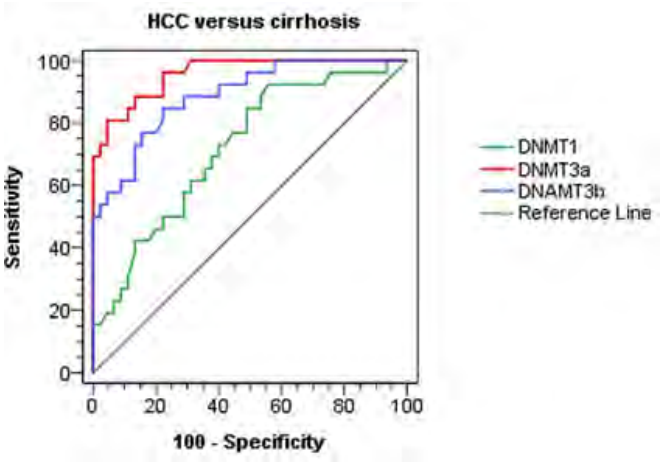
Background and Aims:
Several major risk factors for hepatocellular carcinoma (HCC) have been identified, including chronic infection of hepatitis B virus (HBV) and hepatitis C virus (HCV). Nevertheless, only a fraction of infected patients develops HCC during their lifetime suggesting that genetic factors might modulate HCC development. The alteration in DNA methylation that was observed in HCC patients. We aimed to study the change in the expression of DNA methyltransferases (DNMTs) in chronic HCV infected patients as biomarker for diagnosis of HCC.

Method:
All enrolled study participants we divided into 4 groups: 26 patients with HCC, 45 patients with liver cirrhosis, 20 chronic HCV patients and 20 apparently healthy individuals as a control group. Real-Time Quantitative Reverse Transcription PCR (qRT-PCR) was performed in the studied groups.

Results:
We observed a significant difference in the expression of DNMTs among the studied groups. According to receiver operating characteristics (ROC) curve analysis, it revealed that at a cutoff value of 3.16 for DNMT 3A expression, the sensitivity was 80.8% and the specificity was 95.6% respectively and area under curve (AUC) was 0.958, p< 0.001 for discriminating HCC among post hepatitis C cirrhotic patients. Moreover, DNMT 3B relative expression cutoff value of 3.10 revealed 84.6% sensitivity and 77.8% specificity and AUC was 0.888, p< 0.001. On the contrary, cutoff value 0.65 for DNMT1 relative expression showed 92.3% sensitivity and 44.4% specificity and AUC was 0.72, p= 0.002. DNMT1, DNMT 3A and DNMT 3B had significant positive correlation with the level of AFP (p-value = 0.003, 0.004 and 0.008 respectively). The relative DNMT3B expression was significantly correlated with focal lesion size (p-value = 0.015). High DNMTs expression was significantly associated with the presence of multiple focal lesions but not associated with the Child Pugh Score (p> 0.05).

Conclusion:
The mRNA levels of different DNMTs could be used as a potential biomarker for early HCC detection.

Figure:



P086YI Survival analysis of the most frequent Single Nucleotide Variants in Hepatocellular Carcinoma

Jimmy A. Daza Barragan[1], Timo Itzel[1], Matthias Ebert[1 2], Andreas Teufel[1 2]

[1]Universitätsmedizin Mannheim, Department of Medicine II, Section of Hepatology, Mannheim, Germany, *[2]Universitätsmedizin Mannheim, Center for Preventive Medicine and Digital Health Baden-Württemberg (CPDBW),, Mannheim, Germany*
Email: jimmy.daza@medma.uni-heidelberg.de

Background and Aims:
Hepatocellular carcinoma (HCC) is the most common type of liver cancer and its incidence is rising. The introduction of new systemic therapies, including immune-based therapies and biomarker driven therapies, has improved survival in patients at advanced stages. However, overall survival is still poor, and recent advances in understanding of the molecular alterations of HCC have not translated yet into novel biomarkers. Over the past decade, major advancements in 'omic' technologies have enabled monitoring of a variety of molecular and organismal processes. A comprehensive analysis of single gene mutations in HCC might lead to detect biomarkers that improve our prognosis and treatment.

Method:
We developed a bioinformatics pipeline capable of analyzing genomic data to identify key regulatory molecular changes in HCC development and their influence in patient's prognosis.

Results:
By looking at genetically determined subgroups of HCC in the TCGA Liver Cancer dataset, we managed to obtain 15 genes frequently affected by oncogenic mutations and analyzed their influence in patient's survival, identifying CSMD1 as a prognostic biomarker candidate. Nevertheless, the validation in the ICGC HCC database showed that it did not have any statistically significant influence in overall survival.

Conclusion:
This work reveals that the most frequent single gene mutations are not enough for significant survival changes in HCC and that we should focus our efforts in integrative analysis of clinical information and multi-omics to maximize our clinical benefits in this devastating disease.

Figure:

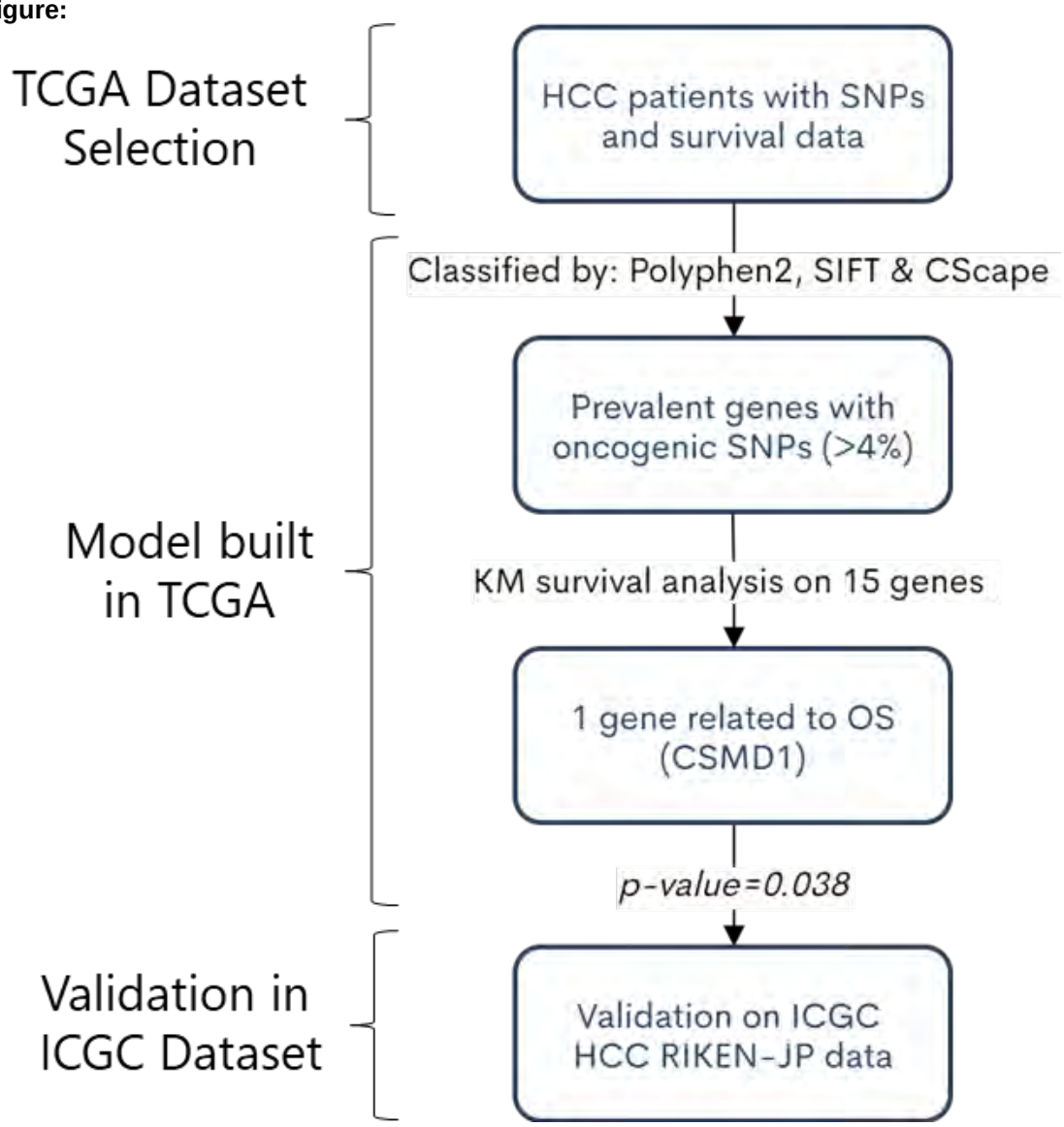


Figure 1. Workflow of the process used to find potential genomic biomarkers in HCC.

P087YI Se and Zn supplements attenuates HCC among recovered Hepatitis C Obese population in Middle Belt Nigeria.

OLAJUYIN OLALEYE[1], Ayodeji Faremi[2]

[1]Federal Medical Centre, Makurdi, Benue State Nigeria., Department of Medical Laboratory Science, Makurdi, Benue State, Nigeria, *[2]The government hospital, ART Clinic, Laboratory Section, Ede Nigeria, Department of Medical Laboratory Services, Osun State, Nigeria*
Email: olajuyine@gmail.com

Se and Zn supplements attenuate HCC among recovered Hepatitis C Obese population in Middle Belt Nigeria.

Background and Aim:
With trends emanating from most cancers registry in Nigeria, Hepatitis C virus has continued to cause serious damage on the liver hepatocytes with little information on its associated importance with liver steatosis and hepatocellular carcinoma. How ever the public health significance of vital trace elements in suppressing the replication and multiplication of positive anti-HCV antibody which could speed up the rate of recovery in Hepatitis C Obese patients in combination with other therapy have not been explored much.

Hence the aim of this study was to evaluate the clinical importance of selenium and zinc supplements among recovered Hepatitis C Obese populace in our environs in line with previous studies.

Method:
Informed consent and ethical approval was appropriately sought for. A supplement composition that contains 15mg and 70mcg was provided daily for 6months to 25 patients (experimental group) already diagonised with HCV-related CLD (genotype 1 and 3a) with evidence of visceral obesity in addition to their ongoing pharmaceutical prescription. Peripheral blood counts, liver related biochemical parameters, serological makers of liver fibrosis, HCV-RNA loads and serum level of zinc and selenium were evaluated before and after supplement administration including the 25 patients(control group) who are not willing to take the supplements.

Results:
During the six month intervention and follow up, the blood selenium concentration and zinc activity of the subjects in the experimental group were increased and had significant difference as compared with those of the control group ($P < 0.01$). Almost all the control group subjects had prolong Prothrombin time and was associated with increase risk of developing HCC in univariate (hazard ratio[HR]=2.6;95%[CI]1.3-8.4;p=.04) and multi variable analysis(HR=3.1;95%CI=1.6-9.7;P=.02) and low platelet count (HR=1.9;95%CI=1.2-2.1;P<.02) were significantly associated with increased risk of developing HCC in multivariable analysis. Alphafeto protein in control group was statistically significantly higher than the experimental group (p=.03). Same statistical trend was observed in serum ALP and ALT in the same manner (p=.04 and p=.02)

Conclusion:
The results confirms that the supplements administered to the experimental populations might be effective in the prevention of liver cancer and the findings further suggest that selenium and polaprezinc exert great anti inflammatory effect and tumorigenesis prevention on the liver of HCV-related CLD patients with favourable liver enzymes outcome. This study can be optimised for a more nutritional support that can lead to a better treatment response.

P088YI CAMD score predicts the risk of hepatocellular carcinoma in Tunisian patients with chronic hepatitis B on antiviral therapy

Myriam Ayari[1], Rym Ennaifer[1], Fatma Ben Farhat[1], Bochra Bouchabou[1], Houda Ben Nejma[1]
[1]*Mongi Slim Hospital, Hepatology, La Marsa, Tunisia*
Email: ayari.myriam@hotmail.fr

Background and Aims:
Chronic hepatitis B (CHB) is widespread disease leading to significant morbidity and mortality. Currently the nucleos(t)ide analog inhibitors represent the treatment of choice as they are highly effective in the suppression of HBV replication and thus aiming to prevent HCC development. However HCC risk remains obviously despite long-term oral antiviral therapy. Many scores have been developed to identify patients requiring close monitoring of HCC. The aim of our study is to assess the predictive value of a new born scoring system: the CAMD score in CHB infected Tunisian patients.

Method:
We performed a single-center retrospective study analyzing data of patients with CHB from 2007 to 2018. We enrolled all CHB patients either cirrhotic or no, receiving Entecavir (ETV) therapy since at least one year. Patients with co-infection C, D, VIH or associated nonalcoholic steatohepatitis were excluded. With a scoring range from 0 to 19 points, CAMD score was calculated for each patient based on cirrhosis, age, sex and diabetes mellitus. A score <8 and >13 points identified patients at distinctly low and high risks. Area under the curve (AUC) were calculated to assess the accuracy of CAMD score in predicting HCC.

Results:
A total of 81 consecutive patients were included: 59 men and 22 women (sex ratio M/F = 2.68). The mean age at diagnosis was 48 ± 10.50 years (range 22-73 years). Most of patients (80%) were infected by mutant virus B and 55 % were cirrhotic. HCC occurred in 34 patients with an average period of 48 months ± 14.70. Univariate analysis identified: male gender (p=0.028), older age (p=0.001), low platelet count (p=0.046), cirrhosis (p=0.001), high ALAT level at 1 year of treatment (p= 0.05) and positive Hbe Ag (p=0.034) as factors associated with HCC occurrence. At multivariate analysis only cirrhosis was an independent predictor of HCC. According to CAMD score, low risk was noted in 26.6% of patients and 40.5% were considered at high risk of HCC. None of patients at low risk had developed HCC while in patients considered at high risk the incidence of HCC was of 75%. When analyzing the receiver operating characteristic (ROC) curve, AUC of CAMD score in predicting HCC was 0.872 (95% CI: 0.795–0.948, p<0.0001).

Conclusion:
CAMD score is a reliable, useful and easy to calculate tool for prediction of HCC in HBV patients on ETV treatment and thus allows a better stratification of patients and limiting screening efforts to those at highest need. In our study, patients with CAMD score < 8 did not developed HCC and therefore would not require close monitoring.

P089 Centrality of Estrogen Receptor 1 in Modulating Hepatocellular Carcinoma Progression through Wnt signaling

Mamatha Bhat[1], Elisa Pasini[1], Chiara Pastrello[2], Marc Angeli[1], Max Kotlyar[2], Igor Jurisica[2]
[1]University Health Network, Multi Organ Transplant, Toronto, Canada, *[2]Krembil Research Institute, University Health Network , Toronto, Canada*
Email: mamatha.bhat@uhn.ca

Background and Aims:
Hepatocellular carcinoma (HCC) is a high-fatality and sexually dimorphic cancer. There is a limited understanding of the crosstalk that occurs between pathways to modulate hepatocarcinogenesis.

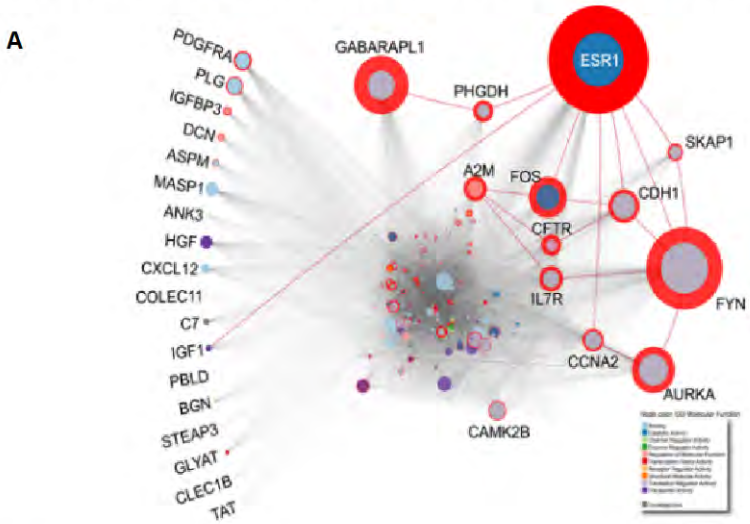
Method:
We integrated and analyzed all publicly available, high-throughput gene expression data in HCC, followed by mapping of the differentially expressed genes into a physical protein-protein interaction network. The most central gene was identified using betweeness centrality algorithm. *In vivo* validation in mice (both male and female) was performed inducing HCC through hydrodynamic tail vein injection of plasmids coding for pro-oncogenes (K -ras and -β-catenin) and the mice were exposed to the agonist or antagonist for ESR1. Cancer pathways affected by ESR1 modulation were identified by microarray followed by pathway enrichment analysis. *In vitro* validation of the pathways identified to be down modulated by ESR1 agonist (Wnt and β-catenin signaling) was performed transfecting HepG2 cells with ESR1 vector in the presence or absence of estrogen. Confocal microscopy was performed to monitor the nuclear translocation of ESR1 and its interaction with β-catenin. The transcriptional effects of ESR1-β-catenin interactions were monitored by Real Time on β-catenin target genes, such as cyclinD1 and c-myc.

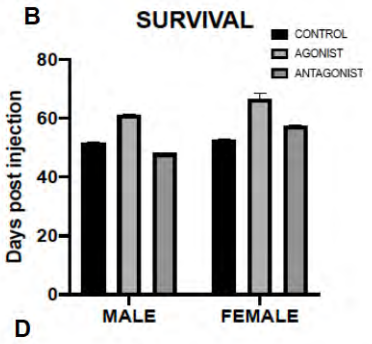
Results:
We identified Estrogen Receptor-1 (ESR1) to be the most central protein in the network using the betweenness centrality algorithm (Figure 1A). Treatment of a genetic mouse model of HCC with ESR1 agonist significantly improved survival (Figure 1B), decreased tumor burden (***p<0.01) (Figure 1C) and inhibited Wnt, Ras/Raf/MAPK and mTOR pathways based on transcriptomics (Figure 1D). Analysis of The Cancer Genome Atlas dataset revealed ESR1 to be protective for overall survival, with hazard ratio of 0.45 (95%CI 0.32-0.64, p=4.4 e-06), which was more pronounced in women. In vitro experiments revealed colocalization of ESR1 with B-catenin at the nuclear level in the presence of Estrogen with inhibitory effects on transcription of pro-oncogenic genes such as c-myc (Fold Change=0.06, p=0.0022) and cyclinD1 (Fold Change =0.3, p=0.0031).

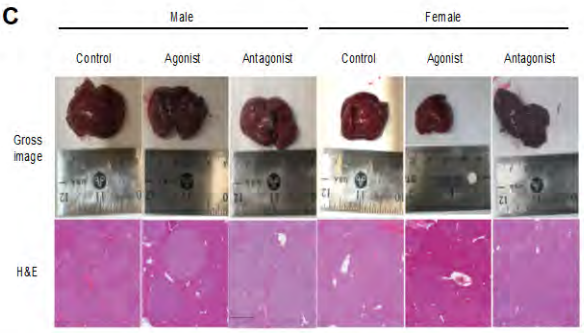
Conclusion:
The centrality of ESR1 and the significant impact of its activation on Wnt signalling provides a biological rationale for sex-specific differences in HCC incidence and biological behavior.

Figure:

Figure 1: ESR1 is Central to the Protein-Protein Interaction Network of HCC, reflecting its importance in HCC pathogenesis. (A) ESR1 is Central to the Protein-Protein Interaction Network of HCC, reflecting its importance in HCC pathogenesis. The diameter of the red circle is proportional to the centrality of the protein. The size of the circles is proportional to the degree of the protein. Molecular functions of these proteins according to Gene Ontology are highlighted in the color legend. B) ESR1 agonist treatment significantly improved survival. C) and decreased tumor burden in female mice. D) ESR1 agonist treatment decreases expression of genes involved in Wnt, Ras/Raf/MAPK and mTOR pathways







D

GENES DOWN REGULATED (log2FC<-1) n=293				
Pathway Source	Pathway Name	p-value	q-value (FDR: BH-method)	Genes identified (n)
REACTOME	Wnt /beta-catenin	3.99E-11	1.53E-07	57
REACTOME	DNA replication initiation	4.64E-06	2.09E-04	22
KEGG	Hippo signaling	1.88E-03	1.58E-02	45
ACSN2	PI3K_AKT_MTOR	5.39E-03	3.44E-02	57

P090YI Hepatoprotective effects of pirfenidone in early stages of hepatocellular carcinoma

Jorge Silva-Gomez[1], Christian Monroy-Ramirez[1], Marina Galicia[1], ana soledad sandoval-rodriguez[1], arturo santos[2], juan armendariz-borunda[1]

[1]University of Guadalajara, Molecular Biology and Genomics, Guadalajara, Mexico, *[2]Tecnológico de Monterrey, Zapopan, Mexico*
Email: armdbo@gmail.com

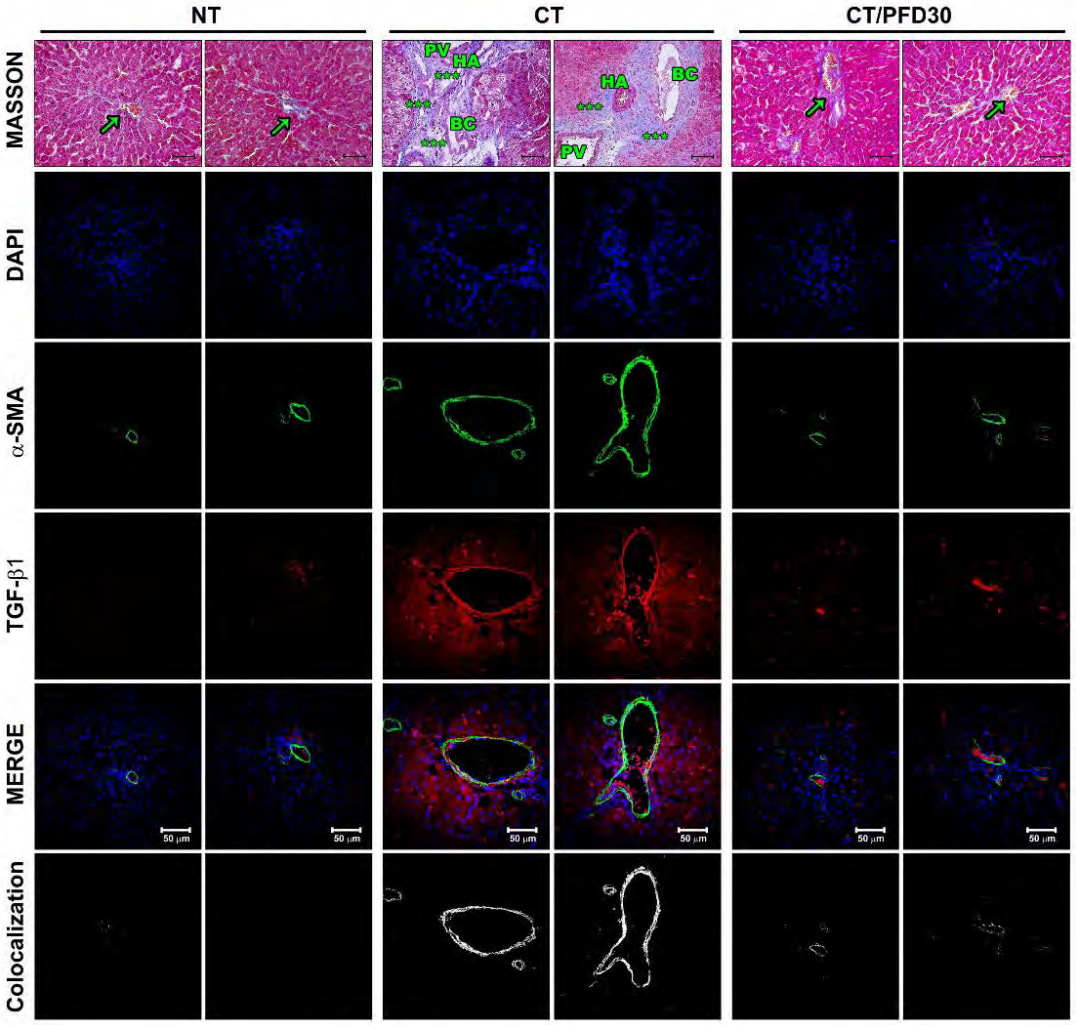
Background and Aims:
Hepatocellular carcinoma (HCC) is a primary liver neoplasm with high recurrence and mortality rate. HCC is the fourth most common cause of cancer-related death and the sixth in terms of incidence. The etiological factors are hepatitis B and/or C virus infections, non-alcoholic steatohepatitis, alcohol consumption, and aflatoxin b1 exposition. These factors promote inflammation, fibrosis, and cirrhosis, and alter the expression of genes and molecular mechanisms, initiating hepatocarcinogenesis. The modified resistant hepatocyte model (MRHM) has been established which simulates the stages of carcinogenesis. Pirfenidone (PFD) has shown antifibrotic, anti-inflammatory and antioxidant effects in liver damage models, so the aim was to evaluate the administration of PFD on histopathological alterations and the expression of key proteins in the development of hepatocarcinogenesis in MHRM.

Method:
Longitudinal experimental design. A total of 30 Fisher rats were divided into 3 groups: control group (NT), carcinogenic treatment group (CT), and carcinogenic treatment plus daily administration of PFD (CT/PFD30). Rats from CT and CT/PFD30 groups were subjected to MRHM. Physical and clinical data of animals were analyzed at 30 days Masson's and H&E staining were performed for histopathological analysis. TGF-beta1 and alpha-SMA expression were evaluated as fibrosis markers. NF-kB cascade in cytoplasmic and nuclear fraction were evaluated as inflammation process. Proteins involved in apoptosis, proliferation, tumor promotion/suppression, and cell metabolism were performed using Western-Blot and microscopy confocal. Data were analyzed and plotted in GraphPad Prism7.

Results:
Liver from CT group exhibited a dense, pale brown and inflamed appearance compared to CT and CT/PFD30 groups. PFD administration was effective to prevent histopathological damage and TGF-beta1 and alpha-SMA overexpression. Anti-inflammatory PDF effects correlate with IKK decrease and reduction in both, IkB-phosphorylation/NF-kB p65 expression and p65-translocation into the nucleus. Pro-apoptotic PFD-induced effects are related with p53 expression, Caspase-3 p17 activation and PARP-1-cleavage. PFD was effective to re-establishes proteins in cellular metabolism regulation such as PPAR-alpha and PPAR-gamma.

Conclusion:
PFD prevents chemical-induced carcinogenic damage in MMRH by preventing fibrosis, reducing inflammation, and promoting apoptosis in MHRM.

Figure:


NT
CT
CT/PFD30
MASSON
DAPI
α-SMA
TGF-β1
MERGE
Colocalization
PV
HA
BC
50 μm

P091 Real-world experience of hepatocellular carcinoma patients after sorafenib treatment: eligibility to subsequent lines and prognostic factors.

Leonardo Gomes da Fonseca[1], Paulo Amaral[1], Lucas Uratani[1], Gabriela Soares[1], Guilherme Marta[1], Jorge Sabbaga[1]

[1]*Instituto do Cancer do Estado de Sao Paulo, Clinical Oncology, Sao Paulo, Brazil*
Email: l.fonseca@fm.usp.br

Background and Aims:
In the past years, novel drugs were adopted in hepatocellular carcinoma (HCC) after sorafenib failure. Placebo-controlled trials demonstrated improved overall survival (OS) with regorafenib (RESORCE trial), cabozantinib (CELESTIAL) and ramucirumab (REACH-2). In addition, immune checkpoint inhibitors were approved based on durable response rates in phase II studies. However, strict eligibility criteria in clinical trials limit the adoption of these drugs in the practice. In this study, we aimed to characterize a real-world cohort of HCC patients after progression to sorafenib and apply trial eligibility criteria for subsequent therapies.

Method:
HCC patients treated with sorafenib between Jan/2017 and Nov/2019 were included. We retrospectively assessed clinical and laboratorial data at the time of progression to sorafenib and evaluated the eligibility to CELESTIAL, RESORCE, REACH-2 and to phase II immunotherapy trials (KEYNOTE 244 and CHECKMATE-040). Median OS after sorafenib progression was estimated by Kaplan Meier methods and prognostic factors were determined using a Cox regression model.

Results:
125 patients were included: 69.2% male, 92.8% Child-Pugh A, 62.3% HCV etiology, 81.6% BCLC stage C and 50.4% received previous locoregional therapies. Median sorafenib duration was 6.5 months (IQR: 4.7-8.4), median OS was 10.2 (CI95% 8.5-11.1) and the median post sorafenib-progression survival (mPPS) was 4.9 months (CI95% 2.9-5.8). According to trial eligibility, 28.8% would be eligible for cabozantinib, 24% for regorafenib, 11.2% for ramucirumab and 29.6% for immune checkpoint inhibitors. Of those eligible for any second-line trial (n=49, 39.2%) mPPS was 8.0 months (CI95% 3.1-18.9) vs 1.7 months (CI95% 0.4-3.6) for those ineligible (p<0.001). Prognostic factors independently associated with PPS were: ECOG PS 0-1 *vs* 2-4 (p<0.001), Child-Pugh A-B7 *vs* >B7 (p<0.001), AFP<200 ng/ml (0.04) and absence of new extrahepatic lesion *vs* progression with new metastasis (p<0.001).

Conclusion:
According to trial eligibility, subsequent therapies after sorafenib are restricted. Liver function, AFP and the pattern of progression can help to stratify patients in prognostic subgroups. Real-world data will be important to determine if subgroups not included in clinical trials may benefit from novel agents.

P092 Hepatotoxicity in patients with hepatocellular carcinoma on treatment with immune checkpoint inhibitors

Nicola Personeni[1 2], Tiziana Pressiani[3], Antonio D'Alessio[3], Luca Di Tommaso[2 4], Maria Giuseppina Prete[3], silvia bozzarelli[3], valeria smiroldo[3], Arianna Dal Buono[5], Antonio Capogreco[5], Alessio Aghemo[2 5], Ana Lleo[2 5], Romano Fabio Lutman[6], Laura Giordano[7], armando santoro[2 3], Lorenza Rimassa[2 3]

[1]Humanitas Clinical and Research Center-IRCCS, Medical Oncology and Hematology Unit, Italy, *[2]Humanitas University, Department of Biomedical Sciences, Pieve Emanuele, Italy*, *[3]Humanitas Clinical and Research Center-IRCCS, Medical Oncology and Hematology Unit, Rozzano, Italy*, *[4]Humanitas Clinical and Research Center-IRCCS, Pathology Unit, Rozzano, Italy*, *[5]Humanitas Clinical and Research Center-IRCCS, Department of Gastroenterology, Rozzano, Italy*, *[6]Humanitas Clinical and Research Center-IRCCS, Department of Radiology, Rozzano, Italy*, *[7]Humanitas Clinical and Research Center-IRCCS, Biostatistic Unit, Rozzano, Italy*
Email: nicola.personeni@hunimed.eu

Background and Aims:
Risk factors for hepatic immune-related adverse events (HIRAEs) in patients with advanced/unresectable hepatocellular carcinoma (HCC) receiving immune checkpoint inhibitors (ICI) are poorly understood. Here, we investigated: (i) clinical predictors of HIRAEs, (ii) their relationship with subsequent treatment outcomes, (iii) morpho-pathological correlates.

Method:
58 HCC patients and preserved liver were included in this retrospective analysis. HIRAEs were graded according to National Cancer Institute's Common Terminology Criteria for Adverse Events. In 27 pretreatment tumor biopsies, we evaluated intratumoral tertiary lymphoid structures, expression of Glutamine Synthase, CD3 and CD79 as surrogate biomarkers of T-cell exclusion.

Results:
20 patients (34%) received anti-programmed cell death protein 1 (PD-1)/PD ligand 1 (PD-L1) antibodies alone, and 38 (66%) in combination with anticytotoxic T-lymphocyte-associated protein 4 antibodies and/or tyrosine kinase inhibitors. After a median time of 0.9 months, 9 patients (15.5%) developed grade ≥3 hepatitis, not associated to any etiologic nor clinical parameter, but higher baseline ALT levels (p = 0.037). ICI were safely resumed in 6 out of 9 patients. Time to treatment failure (TTF) was not significantly different in patients developing grade ≥3 hepatitis vs lower grades (3.25 vs 3.91 months, respectively; p = 0.81). Though not significant, biomarkers of T-cell exclusion were more frequently observed in patients developing grade ≥3 hepatitis.

Conclusion:
Grade ≥3 hepatitis in advanced HCC patients has a benign course that does not preclude ICI reintroduction, without any detrimental effect on TTF. In pretreatment biopsies, biomarkers of T-cell exclusion could be related to high-grade hepatitis and warrant further investigation.

P093YI Identification of a prognostic 3-protein score of childhood liver cancer by proteomic profiling

Juan Carrillo[1], Marina Simon-Coma[1], Mikel Azkargorta[2], Laura Royo[1], Laura Guerra[3], del Rio Alvaro[1], Akira Akuchi[4], Marta Garrido[5], Francisco Hernandez Oliveros[6], Eduard Porta-Pardo[7], Irene Oi-Lin NG[8], Montserrat Domingo-Sàbat[1], Manuel López-Santamaría[6], Bruce Morland[9], Margaret Childs[10], Piotr Czauderna[11], Rudolf Maibach[12], Alfonso Valencia[13], Ramon Planas[14], Margarita Sala[15], Felix Elortza[2], Marie-annick Buendia[16], Maria-Rosa Sarrias[17], Carolina Armengol[18]

[1]Childhood Liver Oncology Group (c-LOG), Health Sciences Research Institute Germans Trias i Pujol (IGTP), Badalona, *[2]Proteomics Platform, CIC bioGUNE - ProteoRed-ISCIII, Derio*, *[3]Pathology Department,Universitary Hospital La Paz, Madrid*, *[4]Department of Molecular Biology and Biochemistry, Osaka University, Suita, Japan.* , *[5]Pathology Department, Universitary Hospital Vall d'Hebron, Barcelona*, *[6]Pediatric Surgery Department, Universitary Hospital La Paz, Madrid*, *[7]*, *[8]Department of Pathology, The University of Hong Kong, Hong Kong, China*, *[9]Department of Oncology, Birmingham Women's and Children's Hospital, Birmingham, UK*, *[10]Nottingham Clinical Trials Unit, Nottingham, United Kingdom*, *[11]Department of Surgery and Urology for Children and Adolescents, Medical University of Gdansk, Gdansk, Poland*, *[12]International Breast Cancer Study Group Coordinating Center, Bern, Switzerland*, *[13]Barcelona Supercomputing Centre (BSC), Barcelona, Spain*, *[14]CIBERehd, Gastroenterology Department, Hospital Universitari Germans Trias i Pujol Hospital, Badalona, Spain*, *[15]CIBERehd, Gastroenterology Department, Hospital Universitari Josep Trueta, Girona, Spain*, *[16]INSERM, UMR 1193, Paul-Brousse Hospital, Hepatobiliary Centre, Villejuif, France*, *[17]Innate Immunity Group, IGTP, Badalona, Spain*, *[18]Childhood Liver Oncology Group (c-LOG), CIBERehd, Health Sciences Research Institute Germans Trias i Pujol (IGTP), Badalona*
Email: carmengol@igtp.cat

Background and Aims:
Hepatoblastoma (HB) is the main liver tumor in childhood. However, it is a rare tumor with an annual incidence of 1 case per million children. Despite good response to chemotherapy, still 20% of HB patients do not survive. Gene expression and methylation profiling studies revealed two HB molecular prognostic subtypes but no proteomic studies have been conducted to date. Aim: Perform a proteomic study of HB and identify new prognostic biomarkers/pathways.

Method:
Twenty-two snap-frozen samples from 14 HB patients were profiled by 2D-DiGE-MALDI-TOF/TOF and Label Free. Pathway analysis was performed using random walk with restart and fast gene set enrichment analysis. Prognostic biomarkers were validated by immunohistochemistry (IHC) in an independent set of 509 FFPE samples from 170 pediatric patients with liver cancer.

Results:
We identified 136 tumor deregulated proteins involved on activation of cell-cycle checkpoints, extracellular matrix, EIF2, SLIT/ROBO and RUNX1 pathways. The unsupervised analysis revealed two tumor proteomic clusters (PC-1, PC-2) and from their 345 differently expressed proteins, we identified 3 proteins which expression by Western blot and IHC was associated with overall survival in training and validation sets (log rank=0.0083 and $1.59x10^{-14}$), respectively. The 3-protein score was identified as an independent prognostic factor (HR:12.02, IC:4.19-34.51, p<0.0001) for pediatric liver cancer patients and it was also associated with survival of HCC, cholangiocarcinoma, lung, kidney, prostate, thyroid and thymus cancer patients.

Conclusion:
We identified a prognostic 3-protein score that could be easily applied at the clinical practice to improve clinical management of patients with liver cancer.

P094YI Pattern of macrovascular invasion in patients with hepatocellular carcinoma influences treatment and outcome.

Maria Guarino[1], Alessandro Cucchetti[2], Giuseppina Pontillo[1], Fabio Farinati[3], Francesca Benevento[4], Gian Ludovico Rapaccini[5], Mariella Di Marco[6], Eugenio Caturelli[7], Marco Zoli[8], Rodolfo Sacco[9], Giuseppe Cabibbo[10], Fabio Marra[11], Andrea Mega[12], Antonio Gasbarrini[13], Gianluca Svegliati-Baroni[14], Francesco Giuseppe Foschi[15], Gabriele Missale[16], Alberto Masotto[17], Gerardo Nardone[18], Giovanni Raimondo[19], Francesco Azzaroli[20], Gianpaolo Vidili[21], Filippo Oliveri[22], Franco Trevisani[23], Edoardo Giovanni Giannini[24], Filomena Morisco[1]

[1]Department of Clinical Medicine and Surgery, Gastroenterology Unit, University of Napoli Federico II", Italy, *[2]Department of Medical and Surgical Sciences, General Surgery of the Morgagni-Pierantoni Hospital Forlì; Alma Mater Studiorum - University of Bologna, Italy*, *[3]Department of Surgery, Oncology and Gastroenterology, University of Padova,, Italy*, *[4]Division of Internal Medicine, Azienda Ospedaliero-Universitaria di Bologna, Italy*, *[5]Gastroenterology Unit, Fondazione Policlinico Universitario A. Gemelli, IRCCS, Roma, Italy*, *[6]Medicine Unit, Bolognini Hospital, Seriate, Italy*, *[7]Gastroenterology Unit, Belcolle Hospital, Viterbo, Italy*, *[8]Department of Medical and Surgical Sciences, Internal Medicine Unit, University of Bologna, Bologna, Italy*, *[9]Gastroenterology and Digestive Endoscopy Unit, Foggia University Hospital, Foggia, Italy*, *[10]Department of Health Promotion, Mother & Child Care, Internal Medicine & Medical Specialties, PROMISE, Gastroenterology & Hepatology Unit, University of Palermo, Palermo, Italy*, *[11]Department of Experimental and Clinical Medicine, Internal Medicine and Hepatology Unit, University of Firenze, Firenze, Italy*, *[12]Gastroenterology Unit, Bolzano Regional Hospital, Bolzano, Italy*, *[13]Internal Medicine and Gastroenterology Unit, Policlinico Gemelli, Università Cattolica del Sacro Cuore, Roma, Italy*, *[14]Gastroenterology Unit, Polytechnic University of Marche, Ancona, Italy*, *[15]Department of Internal Medicine, Ospedale per gli Infermi of Faenza, Faenza, Italy*, *[16]Infectious Diseases and Hepatology Unit, Azienda Ospedaliero-Universitaria di Parma, Parma, Italy*, *[17]Gastroenterology Unit, Ospedale Sacro Cuore Don Calabria, Negrar, Italy*, *[18]Department of Clinical Medicine and Surgery, HepatoGastroenterology Unit, University of Napoli "Federico II", Napoli, Italy*, *[19]Department of Clinical and Experimental Medicine, Clinical and Molecular Hepatology Unit, University of Messina, Italy*, *[20]Department of Surgical and Medical Sciences, Gastroenterology Unit, Alma Mater Studiorum–Università of Bologna, Italy*, *[21]Department of Medical, Surgical and Experimental Sciences. Clinica Medica Unit, University of Sassari, Azienda Ospedaliero-Universitaria di Sassari, Italy*, *[22]Department of Clinical and Experimental Medicine, Hepatology and Liver Physiopathology Laboratory and Internal Medicine, University of Pisa, Pisa, Italy*, *[23]Department of Medical and Surgical Sciences, Semeiotics Unit, Alma Mater Studiorum–University of Bologna, Bologna, Italy*, *[24]Department of Internal Medicine, Gastroenterology Unit, University of Genova, IRCCS Policlinico San Martino, Genova, Italy*
Email: maria.guarino86@gmail.com

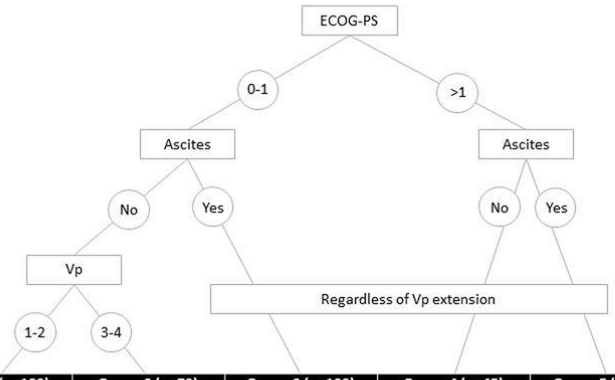
Background and Aims:
In patients with hepatocellular carcinoma (HCC), macro-vascular invasion (MaVI) limits treatment options and decreases survival. However, detailed data on the relationship between MaVI extension and patients' characteristics, and its impact on patients' outcome are limited. We evaluate the prevalence and extension of MaVI in a large cohort of consecutive HCC patients, analysing its association with liver disease and tumour characteristics, as well as with treatments performed and patients' survival.

Method:
We analysed data of 4,774 patients diagnosed with HCC recorded in the Italian Liver Cancer (ITA.LI.CA) database (2008-2018). Recursive partition analysis (RPA) was performed to evaluate interactions between MaVI, clinical variables and treatment, aiming to explore the inter-relationship determining overall survival.

Results:
The prevalence of MaVI was 11.1%, and median survival of these patients was 6.0 months (95% Confidence Interval, 5.1–7.1). MaVI presence was associated with young age at diagnosis, presence of symptoms, worse Performance Status (PS) and liver function, high alpha-fetoprotein and large HCCs. MaVI extension was associated with worse PS, ascites, and greater impairment in liver function. RPA identified patients' categories with different treatment indications and survival, ranging from 2.4 months in those with PS >1 and ascites, regardless MaVI extension (receiving best supportive care in 90.3% of cases) to 14.1 months in patients with PS 0-1, no ascites and Vp1-Vp2 MaVI (treated with surgery in 19.1% of cases) (figure 1).

Conclusion:
MaVI presence and extension, together with PS and presence of ascites, significantly affect patients' survival and treatment selection. The decision-tree based on these parameters may help assess patients' prognosis and inform therapeutic decisions.

Figure:
Multivariable recursive partitioning analysis performed in 553 patients with hepatocellular carcinoma and macro-vascular invasion identified 5 groups with significantly different probability of survivals.



	Group 1 (n=183)	Group 2 (n=72)	Group 3 (n=129)	Group 4 (n=45)	Group 5 (n=124)	*P*
Overall median survival (months)	14.1 (11.0-21.1)	8.0 (5.0-14.1)	6.1 (4.1-7.4)	4.0 (2.9-6.1)	2.4 (2.0-3.0)	*0.001*
Treatment received						*0.001*
Surgery	35 (19.1%)	1 (1.4%)	4 (3.1%)	2 (4.4%)	0 (0.0%)	
Loco-regional therapy	56 (30.6%)	8 (11.1%)	12 (9.3%)	4 (8.9%)	6 (4.8%)	
Sorafenib	56 (30.6%)	44 (61.1%)	35 (27.1%)	8 (17.8%)	6 (4.8%)	
Best supportive care/Other	36 (19.7%)	19 (26.4%)	78 (60.5%)	31 (68.9%)	112 (90.3%)	

P095 Quantification of extracellular matrix features and its implications in hepatocellular carcinoma patients after curative liver resection

Kai-Wen Huang[1]

[1]*National Taiwan University, Surgery of Department, Taiwan*
Email: skyntuh@gmail.com

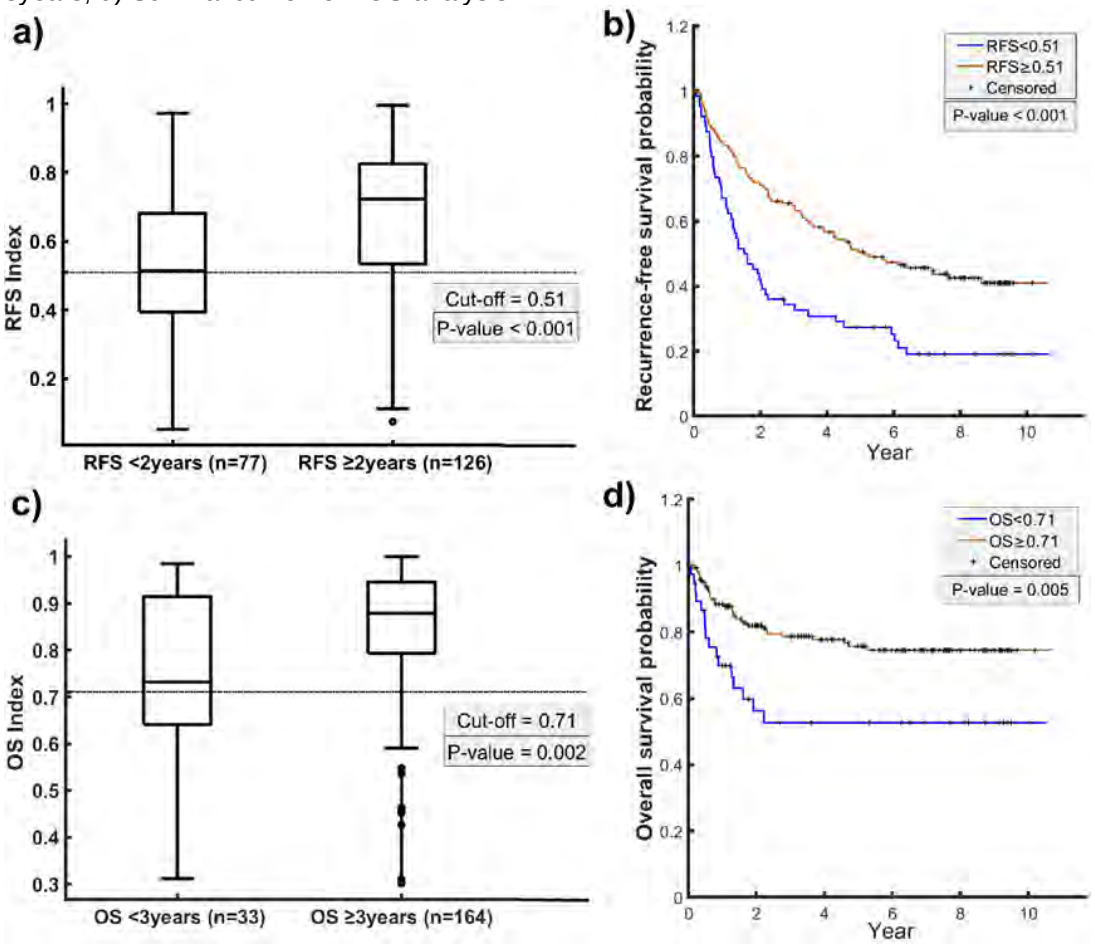
Background and Aims:
The recent increase in the availability of treatment options for hepatocellular carcinoma (HCC) has allowed for gradual improvement in patients' survival. The advent of precision medicine warrants a need for a more personalized treatment based on efficacy and costs. qFibrosis, a digital pathological system has recently been validated in drug development for Nonalcoholic steatohepatitis. The aim is to demonstrate a histopathological evidence-based approach by utilizing qFibrosis to examine dynamics of extracellular matrix (ECM) to fulfil this need.

Method:
Normal liver tissue and liver tumor from 203 patients with HCC who underwent curative tumor resection were imaged and assessed using qFibrosis system, which later generated a total of 33 and 156 collagen parameters from normal liver tissue and tumor part, respectively. We used these collagen parameters to build two models, (RFS-index and OS-index) for prediction of patient's recurrence-free survival (RFS) and overall survival (OS) years. The models were validated using leave-one-out method.

Results:
The RFS-index can differentiate the patients with RFS>2 years (n=126) and RFS≤2 years ($p<0.001$) with a cut-off value RFS-index=0.51. The OS-index can also differentiate the patients with OS>3 years (n=164) and OS≤3 years ($p=0.005$) with a cut-off value OS-index=0.71.

Conclusion:
We have established a histopathological evidence-based evaluation on HCC patient outcome. Quantification of ECM features from HCC patients appear to be a significant parameter, together with other clinical and biochemical data including tumor staging, alpha-fetoprotein, etc. We propose that these could help to build a cost-effective system for a personalized treatment platform.

Figure:
Validation models; a) Boxplot differentiating patients with RFS < 2years from RFS ≥ 2years; b) Survival curve from RFS analysis; c) Boxplot differentiating patients with OS < 3years from OS ≥ 3years; d) Survival curve from OS analysis.



P096YI Enhanced mitochondrial activity and immunometabolic reprogramming, by silencing Methylation Controlled J protein (MCJ), accelerates liver tumorigenesis in a hepatocellular carcinoma (HCC) mice model

Naroa Goikoetxea-Usandizaga[1], Leire Egia-Mendikute[2], Teresa Cardoso Delgado[1], Marina Serrano-Macia[1], Iraia Ladero[3], Elena Molina[3], Maria Mercado-Gómez[1], Sofia Lachiondo-Ortega[1], Irene González-Recio[1], Rubén Rodríguez Agudo[1], Claudia Gil-Pitarch[1], Jorge Simón[1], Silvia Marin[4], Marta Cascante[4 5], Juan Anguita[6 7], Natalia Elguezabal[3], Asís Palazón[2 7], María Luz Martínez-Chantar[1]

[1]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Liver Disease Lab, Derio, Spain, *[2]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Derio, Spain, Cancer Immunology and Immunotherapy Lab, Derio, Spain*, *[3]NEIKER-Instituto Vasco de Investigación y Desarrollo Agrario, Animal Health Department, Derio, Spain*, *[4]Institute of Biomedicine of University of Barcelona, Faculty of Biology, Universitat de Barcelona, Department of Biochemistry and Molecular Biomedicine , Barcelona, , [5]Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas (CIBEREHD) and Metabolomics node at Spanish National Bioinformatics Institute (INB-ISCIII-ES-ELIXIR), Instituto de Salud Carlos III (ISCIII), Madrid, , [6]Center for Cooperative Research in Biosciences (CIC bioGUNE), Basque Research and Technology Alliance (BRTA), Derio, Spain, Inflammation and Macrophage Plasticity Laboratory, Derio, Spain*, *[7]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain*
Email: mlmartinez@cicbiogune.es

Background and Aims: Cancer metabolism is no longer based on the exclusive Warburg effect. Indeed, recent evidence supports the need of a mitochondria-based metabolism for tumor growth. Silencing of MCJ, an endogenous negative regulator of mitochondrial complex I, has proven to accelerate mitochondrial respiration, increase ATP synthesis and enhance liver proliferation, without affecting ROS production. Mitochondrial metabolism also plays a critical role in the survival and activation of immune cells, therefore, metabolic changes in the tumor microenvironment might also alter the immune response. This work aims to study the metabolic reprogramming of cancer cells at different stages, to prove increased malignancy in mitochondria-based tumors, and to analyze the differential immune response driven by metabolic changes.

Method: Mice were injected intraperitoneally with 25 mg/kg body weight of diethylnitrosamine (DENA) at 14 days of age and they were monthly monitored, tumor growth was followed by ultrasound analysis and blood samples were extracted. DENA-treated mice, and the corresponding controls, were sacrificed at 5, 8, and 12 months post injection (a minimum of 5 mice per group). Mortality, tumor number and size, liver metabolism, mitochondrial activity and tumor infiltrating immune cell analysis were then assessed.

Results: The initial *in silico* approach using UALCAN revealed reduced *Mcj* expression in stage 4 HCC patients. *In vivo*, at 5 and 8 months after DENA injection, the amount of liver tumors was higher in MCJ KO mice (33% Wt, 70% MCJ KO and 60%Wt, 100% MCJ KO, respectively), and at 12 months they showed a 20% higher mortality rate. Fluxomics analysis using [U-$_{13}$C]glucose or [U-$_{13}$C]glutamine will provide information about the metabolic reprogramming during tumorigenesis. Furthermore, elevated hepatic levels of NADPH and NAD$^+$ were measured in DENA-treated MCJ KO mice, matching with energetic, redox and anabolic demands of cancer metabolism. We then studied tumor infiltrating immune cells in the liver of DENA-treated mice and T lymphocytes (CD4$^+$ and CD8$^+$), neutrophils (GR1$^+$CD11b$^+$) and B lymphocytes (CD19$^+$) were mainly detected. A further analysis of specific infiltrating T cells showed a reduction in effector CD4$^+$ T lymphocytes (CD44+ CD62L-) in MCJ KO mice, 5 months after DENA injection; a similar trend was also visible at 12 months, along with significantly reduced PD-1.

Besides, highly ROS producing and cytotoxic neutrophils were also found in Wt mice. Serum analysis of cytokines highlighted reduced levels of inflammatory cytokines in MCJ KO mice; altogether suggesting a deficient anti-tumoral immune response in these mice.

Conclusion: Overall, an accelerated mitochondrial respiration, due to the lack of MCJ, increases tumorigenesis, hepatic biomass and mortality rate; along with increased nicotinamide metabolism and deficient anti-tumoral immune response.

P097YI Absence of a complete response after two trans-arterial chemo-embolizations during liver transplantation waiting list: should we persist or change?

Edoardo Poli[1], Marc Antoine Allard[2], Ilias Kounis[1], Alina Pascale[1], Cyrille Feray[1], Catherine Guettier[3], Maite Lewin[4], Jean-Charles Duclos-Vallée[1], Audrey COILLY[1], Eric Vibert[2], Daniel Cherqui[2], Antonio Sa Cunha[2], Didier Samuel[1], René Adam[2], Olivier Rosmorduc[1]

[1]Paul Brousse Hospital, Hepatology, Centre Hépatobiliaire, Villejuif, France, *[2]Paul Brousse Hospital, Gastroenterological Surgery, Centre Hépatobiliaire, Villejuif, France*, *[3]Bicetre Hospital, Pathology , Le Kremlin-Bicêtre, France*, *[4]Paul Brousse Hospital, Radiology, Centre Hépatobiliaire, Villejuif, France*
Email: edoardo.poli88@gmail.com

Background and Aims:
Hepatic trans-arterial chemoembolization (TACE) is a widely-used treatment to prevent hepatocellular carcinoma (HCC) progression in patients enlisted for liver transplantation (LT). Several sessions can be performed but the clinical benefit of repeated TACE remains uncertain. Our study aimed to assess the impact of TACE number and efficacy on the risk of pre-LT tumor-related delisting or of post-LT HCC recurrence.

Method:
All patients enlisted for LT who prospectively received at least one TACE for HCC within the LT criteria (French AFP score ≤ 2) in a single institution, from 2013 to 2018, were included. Treatment failure was defined as pre-LT tumor-related delisting or of post-LT HCC recurrence. Factors associated with this treatment failure were analysed in the entire cohort. TACE response was evaluated using the mRECIST criteria to identify the absence of a complete radiological response (CRR).

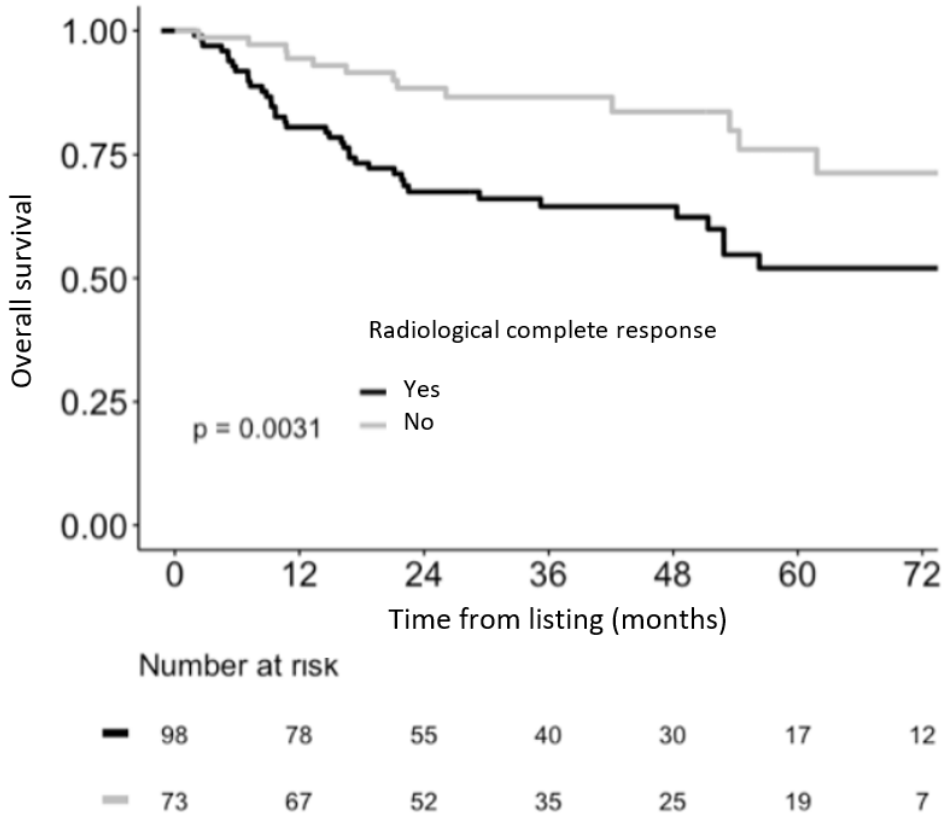
Results:
A total of 172 patients were included in the study. At last follow-up, 127 (73.8%) patients were transplanted, 37 (21.5%) were delisted because of HCC progression and 14 presented post-LT HCC recurrence (11% of transplant recipients). At univariate analysis, factors associated with treatment failure were: higher pre- and post-TACE serum AFP, absence of complete radiological response in post -TACE, number of TACE sessions before LT > 2 (Table 1). The risk of treatment failure was 38% in patients who received > 2 TACE *vs.* 20% in patients who received 1 or 2 TACE (p = 0.02). At multivariate analysis, being treated with > 2 TACE was highly predictive of treatment failure [RR 3.3 (1.52-7.31), p = 0.003] after adjusting for serum AFP, Child-Pugh score, maximum HCC size and number of HCC nodules. Moreover, the overall survival was significantly decreased in patients without a CRR compared to those with a CCR after 1 (if unique) or 2 TACE (67% vs 88% at 2 years from the date of enlisting; p = 0.003; Figure 1).

Conclusion:
Enlisted patients who receive more than 2 TACE without complete response are at higher risk of poor outcome with a high risk of pre-LT tumor-related delisting or of post-LT HCC recurrence. An alternative strategy should be considered to avoid additional deleterious TACE in this setting.

Table 1

	Univariate analysis		
	Delisting of post-LT HCC recurrence		
	NO N=127	YES N=45	p
Age, years	62 [33-74]	60 [42-70]	0.358
Male gender, n (%)	109 (85.8%)	39 (86.7%)	1.000
Cirrhosis aetiology: -Virus -Alcohol -NASH -other	 51 (40.2%) 46 (36.2%) 18 (14.2%) 12 (9.45%)	 16 (35.6%) 22 (48.9%) 4 (8.89%) 3 (6.67%)	0.520
MELD score	8 [6-20]	9 [6-18]	0.441
AFP pre-TACE (ng/ml)	**7.40 [2.00-700]**	**17.0 [3.00-780]**	**0.002**
AFP post-TACE (ng/ml)	**6.00 [1.60-519]**	**41.0 [1.40-25979]**	**<0.001**
Number of HCC	2 [1-6]	2 [1-9]	0.708
Maximum HCC size, mm	23 [7-60]	24 [12-50]	0.651
Absence of CRR	**67 (52.8%)**	**43 (95.6%)**	**<0.001**
Number of TACE sessions	2 [1-5]	2 [1-6]	0.068
>2 TACE, n (%)	**36 (28.3%)**	**22 (48.9%)**	**0.020**

Figure:



P098 Prediction of early recurrence of hepatocellular carcinoma after resection: international validation of the ERASL risk models

Berend Beumer[1], Kosei Takagi[2], Bastiaan Vervoort[1], Stefan Buettner[1], Yuzo Umeda[2], Takahito Yagi[2], Toshiyoshi Fujiwara[2], Ewout Steyerberg[3], Jan IJzermans[1]

[1]Erasmus MC, University Medical Centre Rotterdam, Rotterdam, The Netherlands Erasmus MC Rotterdam, Department of Surgery, Division of HPB & Transplant Surgery, *[2]Okayama University Graduate School of Medicine, Dentistry, and Pharmaceutical Sciences, Okayama, Japan Okayama University Hospital, Department of Gastroenterological Surgery, Okayama, Japan*, *[3]Leiden University Medical Centre, Leiden, the Netherlands, Department of Biomedical Data Science*
Email: b.beumer@erasmusmc.nl

Prediction of early recurrence of hepatocellular carcinoma after resection: international validation of the ERASL risk models

Background and Aims:
Recently, novel preoperative (ERASL-pre) and postoperative (ERASL-post) risk scores were published [1], which predict early recurrence of hepatocellular cancer after surgical treatment. To date, these models have not yet been externally validated by an independent research group. We examined the predictive performance of the ERASL models.

Method:
Our independent external validation was based on data from 279 patients from the Netherlands and 392 patients from Japan. All patients received first time resection with curative intent and were diagnosed with hepatocellular carcinoma (HCC) on pathology. Performance was assessed according to discrimination (concordance (C) statistic) and calibration (correspondence between observed and predicted risk, with recalibration in a Weibull model).

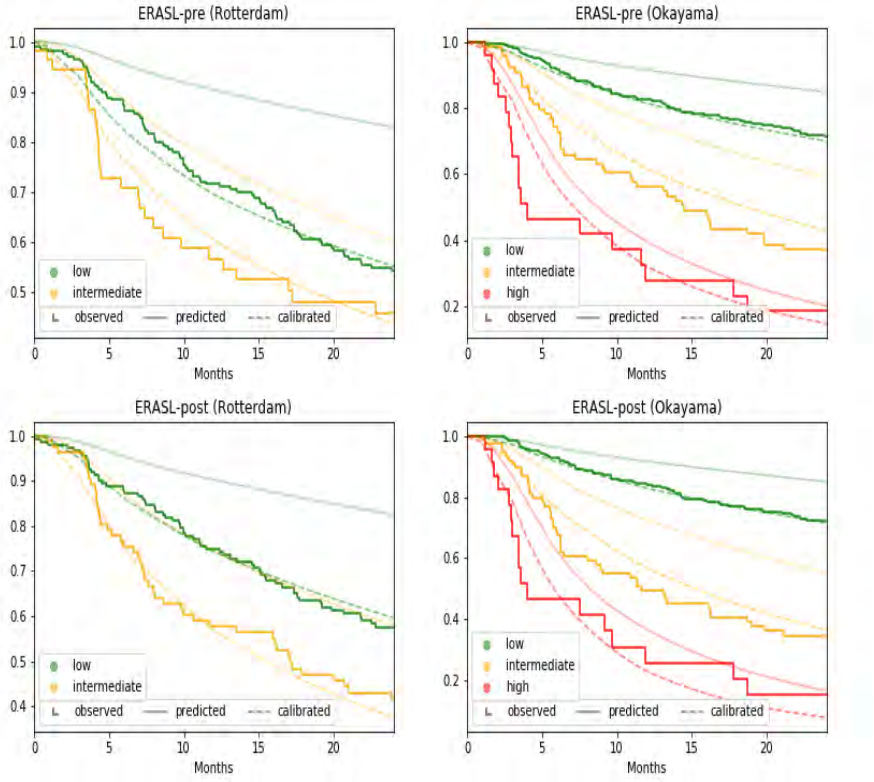
Results:
The discriminatory power of both the pre and postoperative models was lower in the Netherlands compared to Japan (C 0.57 [95%CI 0.52; 0.62] vs 0.69 [95%CI 0.65; 0.73]) for the ERASL-pre model and 0.62 [95%CI 0.57; 0.67] vs 0.70 [95% CI 0.66; 0.74] for the ERASL-post model. Further, the probabilistic estimates of the ERASL model systematically too optimistic for the low-risk and intermediate-risk groups. Updated ERASL risk scores improved local accuracy while leaving the relative risk information from the original models unchanged.

Conclusion:
The discrimination of ERASL risk scores may be limited in the Netherlands and other western patients, in contrast to japan, where good performance similar to the Hong Kong derivation cohort may be found. Absolute risk prediction requires local re-calibration. Calibration using a Weibull model was successful to largely remedy the mismatch between predicted and observed survival probabilities. Further validation of the re-calibrated ERASL risk scores is necessary before application in clinical practice can be advised.

Reference:
1. Chan AW, Zhong J, Berhane S, Toyoda H, Cucchetti A, Shi K, et al. Development of pre and post-operative models to predict early recurrence of hepatocellular carcinoma after surgical resection. Journal of hepatology. 2018;69(6):1284-93

Figure:

Prediction and recalibration



The smooth solid lines represent the average predictions per risk group from the original model. The dashed curves represent the calibrated survival probabilities.

P099 Equal overall survival in elderly patients with hepatocellular carcinoma receiving palliative treatment

Thorben Fründt[1], Christian Casar[2], Johann von Felden[1], Jenny Krause[1], Ines Gil-Ibanez[1], Ulrike Schöler[1], Maximilian Priebe[1], Jenny Kraczyk[1], Gerhard Adam[3], Ansgar W. Lohse[1], Henning Wege[1 4], Kornelius Schulze[1]

[1]University Medical Center Hamburg-Eppendorf, I.Department of Medicine, Hamburg, Germany, *[2]University Medical Center Hamburg-Eppendorf, Bioinformatics Facility , Hamburg, Germany*, *[3]University Medical Center Hamburg-Eppendorf, Department of Diagnostic and Interventional Radiology and Nuclear Medicine, Hamburg, Germany*, *[4]Klinikum Esslingen, Cancer Center Esslingen, Esslingen, Germany*
Email: tfruendt@uke.de

Background and Aims:
Hepatocellular carcinoma (HCC) is the most common primary liver cancer with an increasing incidence worldwide, especially in elderly patients. Diagnosis is often made in an advanced stage, rendering curative treatment impossible. Although therapeutic options for patients in a palliative setting have improved in recent years, treatment of elderly patients is often complicated by challenging comorbidities and frailty. Since data about the outcome and overall survival (OS) in elderly HCC patients receiving palliative therapy is limited, we conducted a retrospective study at a tertiary referral center.

Method:
Data from 987 patients with HCC, who were treated at the University Medical Center Hamburg-Eppendorf between January 2007 and October 2017, were analyzed. Tumor stage and liver function were rated according to the Barcelona Clinic Liver Cancer (BCLC) classification and Child-Pugh-Turcotte score (CPS), and therapy related adverse events (AE) were assessed via CTCAE 5.0. Patients were grouped according to their age as young (< 60 years; YP), intermediate (60-70 years; IP) or elderly (> 70 years; EP). Administration of tyrosine kinase inhibitor (TKI) and transarterial chemoembolization (TACE) was defined as palliative treatment. Patients receiving curative intended treatment (e.g. liver transplantation, resection or microwave ablation following TACE) were excluded from this analysis.

Results:
Out of 987 patients, n=330 were considered for curative treatment and excluded, n= 657 received palliative treatment: YP n=194; IP n=241; EP n=222. 82.5% (n=542) were male, median age was 67 (range 23 – 87) years. All patients had underlying liver cirrhosis with a larger proportion of impaired liver function in the YP cohort (CPS A/B/C [%]: YP 53/28/18 vs. IP 61/26/12 vs. EP 66/28/6; p=0.01). Patients performance status according to Eastern Cooperative Oncology Group (ECOG) was comparable between YP, IP, and EP (ECOG 0/1/2/3 [%]: 40/37/16/4 vs. 36/40/14/6 vs. 33/41/17/6; p=0.85). OS in patients receiving TACE, including CPS A and B patients, was 16 vs. 16 vs. 20 months for YP, IP, and EP, respectively; p=0.38. In TKI-treated patients, OS was 13 vs. 15 vs. 13 months; p=0.73. The rate of AE in TKI-patients was comparable with 47 vs. 43 vs. 49 %, and the most prevalent AE in all three groups was diarrhea.

Conclusion:
In this study, OS was equal in elderly patients with liver cirrhosis receiving palliative treatment for HCC with TACE or TKI. Furthermore, the rate of AE was comparable in younger and elderly patients. Therefore, we propose regular palliative treatment stratification in spite of high age of patients.

P100 PD-1 expression, polymorphisms in PDCD1 and hepatocellular carcinoma (HCC) in patients with non-alcoholic fatty liver disease (NAFLD).

nardeen eldafashi[1 2], Rebecca Darlay[3], Misti McCain[1 3 4], robyn watson[1 3 4], nikki mcstraw[1], João Maurício[1 5], Moustafa Fathy[2], michael Atef Fawzy[2], Heather Cordell[3], Ruchi Shukla[1 6], Jean-Francois Dufour[7], Luca Valenti[8], Quentin Anstee[5], Helen Louise Reeves[1 3 4]

[1]Newcastle University Centre for Cancer, Newcastle University, Newcastle upon Tyne, United Kingdom, *[2]Minia University, Biochemistry Department, Minia, Egypt*, *[3]Newcastle University Translational and Clinical Research, Newcastle University, Newcastle upon Tyne, United Kingdom*, *[4]The Freeman Hospital Liver Unit, Newcastle upon Tyne Hospitals NHS Foundation Trust, Newcastle upon Tyne, United Kingdom*, *[5]Newcastle University Translational and Clinical Research Institute, Newcastle University, Newcastle upon Tyne, United Kingdom*, *[6]Newcastle University Biosciences , Newcastle University, Newcastle upon Tyne, United Kingdom*, *[7]INSELSPITAL University Clinic , Bern, Switzerland*, *[8]University of Milan, Department of Pathophysiology, Milan, Italy*
Email: h.l.reeves@ncl.ac.uk

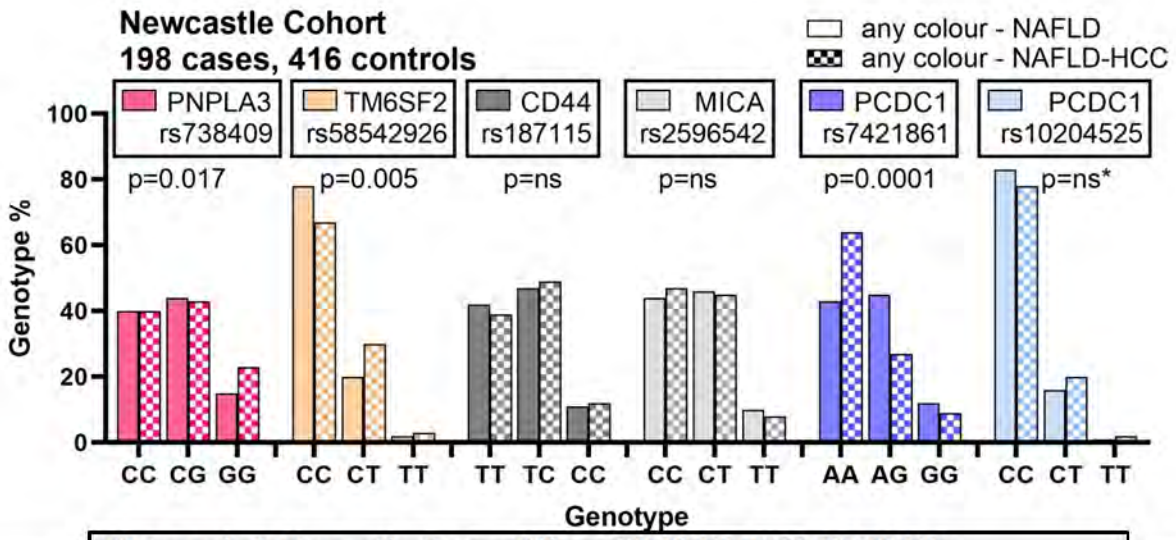
Background and Aims:
Non-alcoholic fatty liver disease (NAFLD) is now one of the major causes of hepatocellular carcinoma (HCC). Patients with NAFLD cirrhosis have 6 monthly abdominal ultrasound for HCC surveillance, although whether this is cost effective is debatable. For NAFLD patients without cirrhosis, with a lesser risk of HCC, the pathogenesis of HCC is poorly understood and no surveillance test is advised. Improved surveillance tools are needed. Evidence suggests that an immune response to fat accumulation lies at the heart of NAFLD-HCC risk, with genetic variation in fat regulatory genes (*PNPLA3*; *TM6SF2*) associated with elevated steatosis and HCC development. Here, we have explored single nucleotide polymorphisms (SNPs) in candidate immunoregulatory genes, including *PDCD1*, which encodes the T cell receptor, PD-1.

Method:
SNPs in the candidate genes (*PNPLA3* rs738409; *TM6SF2* rs2596542; *MICA* rs2596542; *CD44* rs187115; *PD-1* rs7421861 and rs10204525) determined by taqman assay or GWAS, were analysed in a Newcastle cohort (416 NAFLD, 198 NAFLD-HCC patients). Associations with age, sex, cirrhosis and diabetes status, were sequentially included as covariates using logistic regression (PLINK). Data from two additional cohorts (Berne: 76 NAFLD, 84 NAFLD-HCC; Milan: 102 NAFLD, 109 NAFLD-HCC) enabled a fixed-effect meta-analysis, performed on the log Odds Ratios (lnORs) and standard errors of all three cohorts. PD-1 expression was examined by immunohistochemistry (IHC) in NAFLD patients with (n=30) and without (n=16) HCC.

Results:
Analysis of the Newcastle cohort (Figure 1) confirmed significant HCC associations for the variant alleles of *PNPLA3* and *TM6SF2*. A novel association for PD-1 rs7421861 allele A was also identified ($p<0.001$), which was independent of age, sex, cirrhosis and diabetes (P=1.53E-03, lnOR -0.72). Surprisingly, despite a lack of univariate significance for other immunoregulatory SNPs, a significant association for the 'T' allele of PD-1 rs10204525 with NAFLD-HCC emerged when gender was entered into the regression – attributed to a risk in females with HCC. In the combination meta-analyses (Newcastle, Berne, Milan), only rs10204525 remained independently significant, more so when conditioned on *PNPLA3* and *TM6SF2*. *In silico* studies and published literature indicate that both PD-1 SNPs promote expression of PD-1. PD-1 IHC revealed scant expression in T cells in NAFLD patients without cancers, although levels were increased stepwise from steatosis, to NASH to Cirrhosis. Patients with NAFLD-HCC had significantly more PD-1 expression in non-tumour liver, in both the presence and absence of cirrhosis.

Conclusion:
Factors which promote PD-1 expression, such as regulatory SNPs in PDCD1, may be associated with an elevated risk of NAFLD-HCC. Monitoring PD1 expression in NAFLD may inform surveillance strategies.

Figure:



Meta-analysis Newcastle, Berne, Milan – 391 NAFLD-HCC, 594 NAFLD						
Gene	rs identity			Conditioned on PNPLA3+TM6SF2		
Univariate analyses		p	OR		p	OR
PNPLA3 C>G	rs738409	0.044	1.20		NA	NA
TM6SF2 C>T	rs58542926	0.019	1.37		NA	NA
PCDC1 A>G	rs7421861	0.026	0.79		0.045	0.81
PCDC1 C>T	rs10204525	0.123	1.30		0.137	1.30
Regression Age, Sex						
PNPLA3 C>G	rs738409	0.010	1.38		NA	NA
TM6SF2 C>T	rs58542926	0.602	1.10		NA	NA
PCDC1 A>G	rs7421861	0.159	0.82		0.162	0.82
PCDC1 C>T	rs10204525	0.044	1.67		0.011	1.96
Regression Age, Sex, Cirrhosis						
PNPLA3 C>G	rs738409	0.195	1.19		NA	NA
TM6SF2 C>T	rs58542926	0.626	0.91		NA	NA
PCDC1 A>G	rs7421861	0.278	0.85		0.278	0.85
PCDC1 C>T	rs10204525	0.0178	1.95		0.007	2.19
Regression Age, Sex, Cirrhosis, T2DM						
PNPLA3 C>G	rs738409	0.232	1.18		NA	NA
TM6SF2 C>T	rs58542926	0.655	0.91		NA	NA
PCDC1 A>G	rs7421861	0.182	0.82		0.172	0.81
PCDC1 C>T	rs10204525	0.024	1.90		0.009	2.13

P101YI Is first- and second-line immunotherapy sequence the optimal strategy for unresectable hepatocellular carcinoma?

Giuseppe Cabibbo[1], Ciro Celsa[1], Salvatore Battaglia[2], Marco Enea[3], Giacomo Emanuele Maria Rizzo[1], Anita Busacca[1], Paolo Giuffrida[1], Caterina Stornello[1], Antonio Craxi[1], Calogero Camma[1]

[1]University of Palermo, Section of Gastroenterology and Hepatology, PROMISE Department, Palermo, Italy, *[2]University of Palermo, Department of Economics, Business, and Statistics, University of Palermo, Italy, Palermo, Italy*, *[3]University of Palermo, PROMISE department, Palermo, Italy*
Email: calogero.camma@unipa.it

Background and Aims:
The combination of Atezolizumab plus Bevacizumab has showed to be the best performing first-line approach for unresectable hepatocellular carcinoma (u-HCC). However, to date, the best sequential strategy after every first-line failure (for progression or intolerance) remains elusive, and options for retreating patients after Atezolizumab plus Bevacizumab failure with multi-kinase inhibitors (MKI) or immune checkpoint inhibitor (ICI) are yet undefined.

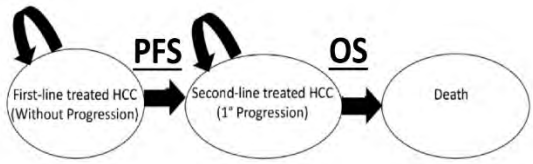
Method:
A Markov model was developed to analyze simulated-Overall Survival (s-OS) of two different systemic treatment sequences for u-HCC over a lifetime horizon (Figure Panel A). For first-line therapy, PFS of Atezolizumab plus Bevacizumab was extracted from Imbrave 150 trial and it was used as endpoint since it is not influenced by post-progression survival. For second-line retreatment, pooled OS of MKIs (Regorafenib from RESORCE trial and Cabozantinib from CELESTIAL trial), or ICIs (Nivolumab from CheckMate-040 trial and Pembrolizumab from KeyNote-240 trial) were adopted. Survival estimates for sequential settings considered the proportion of patients who did not receive second-line therapy due to death during first-line therapy. Individual patient survival data were extracted from PFS and OS Kaplan-Meier curves of the above trials. Each reconstructed survival curve was inspected for accuracy and was compared with originally published curves.

Results:
The s-OS curves of Atezolizumab plus Bevacizumab followed by ICIs or MKIs are showed in Figure Panel B. First-line Atezolizumab plus Bevacizumab followed by ICIs turned on from the model as the best sequential strategy (median s-OS 24 months; 95% Confidence Interval (CI) 23-26 months) and extends survival when compared Atezolizumab plus Bevacizumab followed by MKIs (median s-OS 20 months; 95% CI 19-21 months).

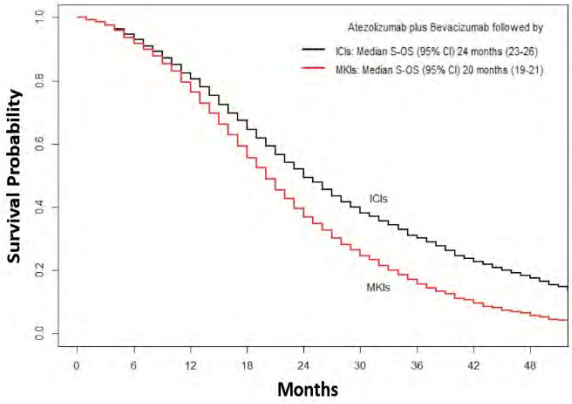
Conclusion:
To our knowledge and given the absence of adequately designed sequential Randomized Controlled Trials (RCTs), this is the first model to date which suggests, with a proper methodological approach, an accurate estimate of outcome of patients with u-HCC treated by sequential systemic therapies. In patients with u-HCC failing first-line Atezolizumab plus Bevacizumab, modelling estimates of s-OS for each retreatment strategies may assist in choosing the most promising sequences in order to plan appropriate RCTs.

Figure: Panel A. General structure of the Markov Model. Panel B. Simulated overall survival of Atezolizumab plus Bevacizumab followed by ICIs or MKIs in patients with unresectable HCC

A)



B)



P102YI Circulating miR 373 but not miR 210 as a new promising preoperative predictor of response to superselective transarterial chemoembolization bridging therapy in Egyptian patients with hepatocellular carcinoma on top of hepatitis C virus infection

Omneya Abdelkarem[1], Amel Abdel Fatthah Kamel[1], Moyassar Ahmad Zaki[1], Ahmed Salah Eldin Tork[1], Reham Abdel Haleem Abo Elwafa[2], Omar Sameh Elaassar[3]

[1]Medical Research Institute, Chemical Pathology Department, Alexandria, Egypt, *[2]Faculty of Medicine, Clinical and Chemical Pathology, Alexandria, Egypt*, *[3]Faculty of Medicine, Radiodiagnosis and Intervention Radiology, Alexandria, Egypt*
Email: omneyaahmed@alexu.edu.eg

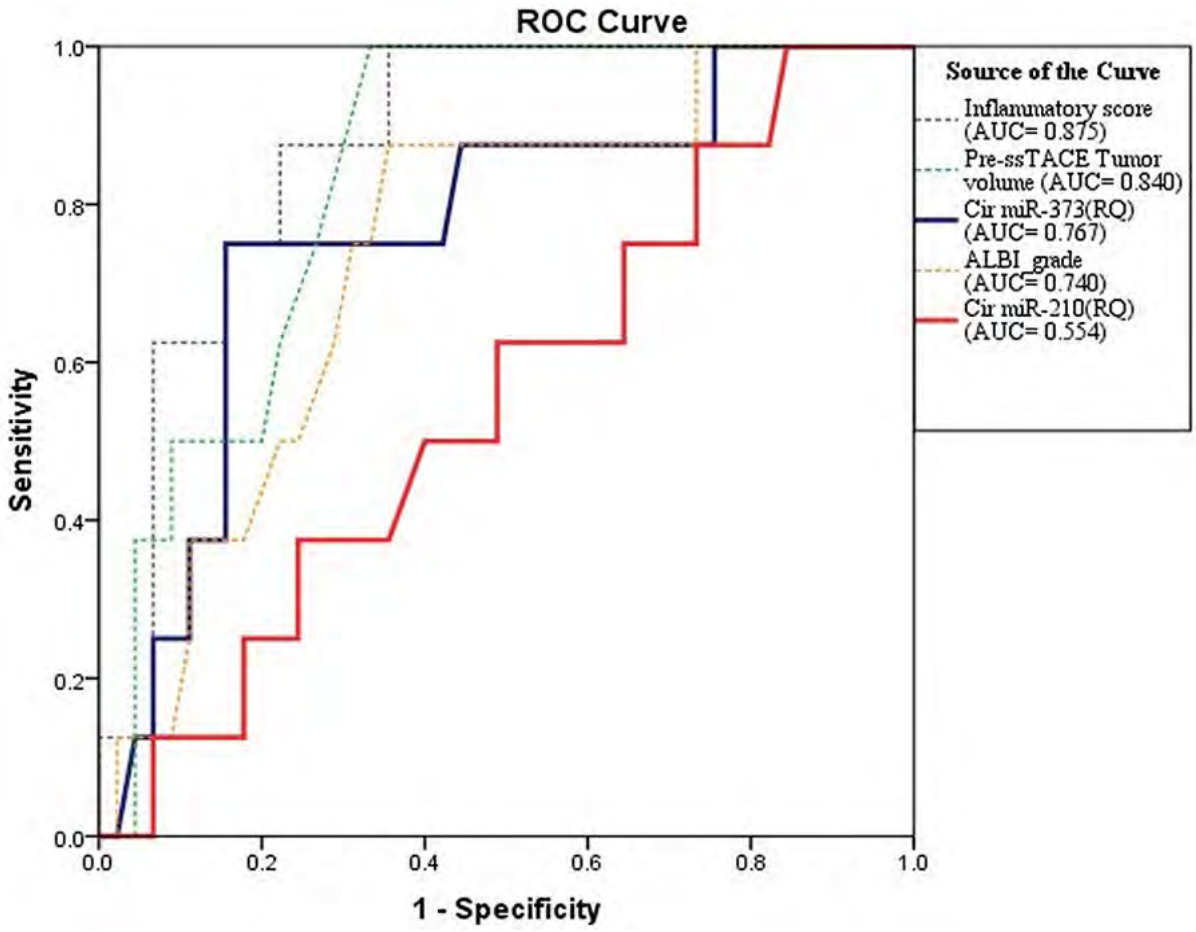
Background and Aims:
Super selective transarterial chemoembolization (ssTACE) has emerged as a bridging therapy for early stages of hepatocellular carcinoma (HCC) awaiting liver transplantation.Various non-invasive liver reserve markers were proposed to predict the prognosis of HCC patients subjected to TACE. No single marker has agreed upon as a surrogate prognostic marker. ALBI grade was introduced as a simple and objective method in assessing liver functional reserve for HCC patients. The inflammatory score was suggested to assess inflammation in all HCC patients. Additionally,it was used as a guide for HCC prognosis. Novel biomedical discoveries in epigenetics have revolutionized the understanding of carcinogenesis. Hypoxia regulated miRNAs(HRMs) were expressed in response to hypoxia. This study aims at assessing the expression profiles of circulating miR-210 and -373 as potential HRMs predictors of ssTACE bridging therapy response in Egyptian HCC patients awaiting liver transplantation.

Method:
After approval of the ethical committee, 53 HCC patients referred for ssTACE were enrolled in the study. After a 3 months follow up, they resulted in 45 responders and 8 non-responders based on mRECIST. Twenty healthy individuals,a reference group, were included for gene expression calculation. Pre and post ssTACE tumor viability were assessed before and three months after completing the scheduled ssTACE sessions respectively using a triphasic CT scan. The ssTACE response was evaluated based on mRECIST.Pre-ssTACE miR210 and 373were determined using reverse transcription quantitative polymerase chain reaction. The inflammatory score and ALBI grade were calculated for HCC cases.

Results:
Circulating pre ssTACE miR-373 but not 210 was significantly higher in non-responders than responders. ROC curves analyses were performed for the significant variables by univariate analysis miR-373, pre-ssTACE tumor volume, inflammatory score and ALBI grade revealed highest sensitivity (100%) for pre-ssTACE tumor volume and highest specificity for pre-ssTACE miR-373 (84.44%). A combined ROC curve series approach starting with pre-ssTACE tumor volume followed by miR-373, achieved an overall sensitivity and specificity of 75% and 97.8% respectively. Multivariate logistic regression revealed pre ssTACE miR-373 as a significant independent predictor of response. miR-373 has survived the multivariate analysis' adjustment for confounding effect of pre ssTACE tumor volume. The odds of being a non-responder to ssTACE would increase by 1.054 for every unit increase in miR-373 level.

Conclusion:
This study highlights the value of circulating miR-373 determination as a predictor marker of response to ssTACE bridging therapy in early HCC patients awaiting liver transplantation. Subsequently,tailoring ssTACE candidates selection based on the pre ssTACE miR-373 expression level.

Figure:


ROC Curve
Sensitivity
1 - Specificity
0.0
0.2
0.4
0.6
0.8
1.0
Source of the Curve
Inflammatory score (AUC= 0.875)
Pre-ssTACE Tumor volume (AUC= 0.840)
Cir miR-373(RQ) (AUC= 0.767)
ALBI_grade (AUC= 0.740)
Cir miR-210(RQ) (AUC= 0.554)

P103 Systematic review and meta-analysis of validated prognostic models for resected hepatocellular carcinoma patients

Berend Beumer[1], Stefan Buettner[1], Boris Galjart[1], Jeroen van Vugt[1], Robert De Man[2], Jan IJzermans[1], Bas Groot Koerkamp[1]

[1]Erasmus Medical Center, Department of Surgery, Division of HPB & Transplant Surgery, Rotterdam, Netherlands, *[2]Erasmus Medical Center, Department of Gastroenterology, Rotterdam, Netherlands*
Email: b.beumer@erasmusmc.nl

Systematic review and meta-analysis of validated prognostic models for resected hepatocellular carcinoma patients

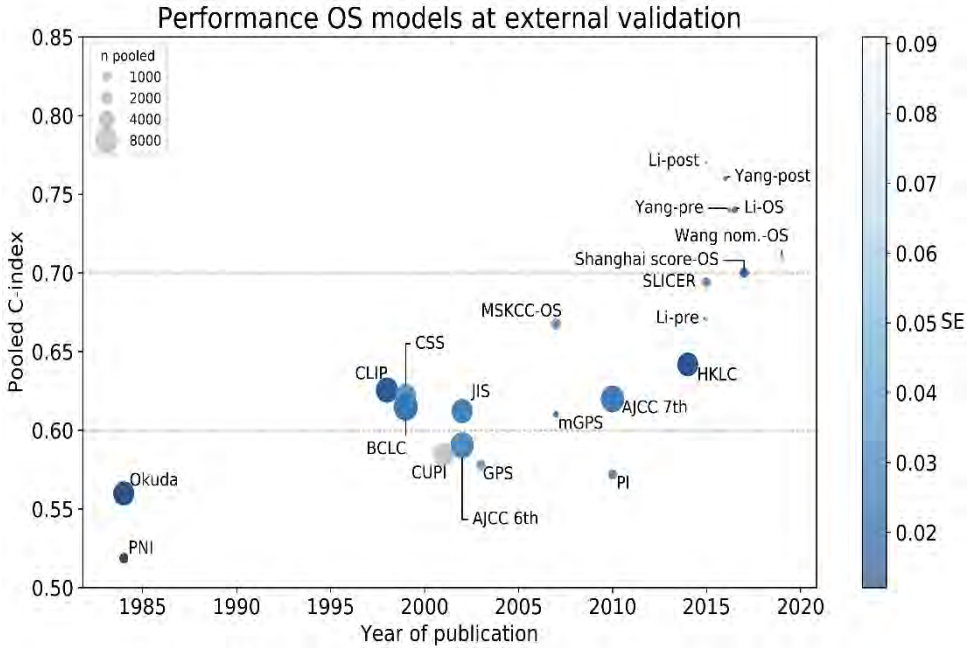
Background and Aims:
Many prognostic models for hepatocellular carcinoma (HCC) have been developed to inform patients and doctors about individual prognosis. Previous reviews of these models were qualitative and did not assess performance at external validation.

Method:
We systematically reviewed all externally validated models predicting survival for patients with resected HCC. After selection, we extracted descriptive statistics and aggregated c-indices using meta-analysis.

Results:
Thirty-eight validated prognostic models were included. Models used on average 7 (IQR:4-9) prognostic factors. Traditional tumor characteristics such as tumor size, tumor number, and vascular invasion were always included. Alpha-fetoprotein (AFP) was commonly incorporated since 2007. Recently, the more subjective items ascites and encephalopathy have been dropped. Eight established models performed poor to moderate at external validation, with a pooled C-index below 0.7; including the Barcelona Clinic Liver Cancer (BCLC) system, the American Joint Committee on Cancer (AJCC) 7th edition, the Cancer of the Liver Italian (CLIP) Program, and the Japan Integrated Staging (JIS) score. Out of 24 prognostic models predicting OS, only 6 (25%) had good performance at external validation with a pooled C-index above 0.7; the Li-post (0.77)[1], Li-OS (0.74)[2], Yang-pre (0.74)[3], Yang-post (0.72)[3], Shanghai-score (0.70)[4], and Wang-nomogram (0.71)[5]. Models improved over time, but overall model performance and study quality remained low.

Conclusion:
Six validated prognostic models demonstrated a good performance for predicting survival after resection of HCC. These models can guide patients and doctors and are a benchmark for future models incorporating novel biomarkers.

Figure:



Each point represents the pooled c-index of a model. The size corresponds to the total number of patients in which the model is validated. The color represents standard error (SE) of the estimate; the darker the color the more precise the estimate. Lastly, the horizontal dashed lines represent the performance thresholds.

Reference:

1. Li Y(2015) 221(5):962-974. e964.
2. Li J (2016) 62:86-95.
3. Yang P (2016) 263(4):778-786.
4. Sun (2017) 130(22):2650-2660.
5. Wang Y (2019).

P104 Levels of circulating PD-L1 are decreased in patients with resectable biliary tract cancer (BTC)

Sven H Loosen[1], Tom Luedde[2], Christoph Roderburg[1]

[1]Medical Faculty of Heinrich Heine University Düsseldorf, Clinic for Gastroenterology, Hepatology and Infectious Diseases, Düsseldorf, Germany, *[2]Medical Faculty of Heinrich Heine University Düsseldorf, Clinic for Gastroenterology, Hepatology and Infectious Diseases, Germany*
Email: christoph.roderburg@med.uni-duesseldorf.de

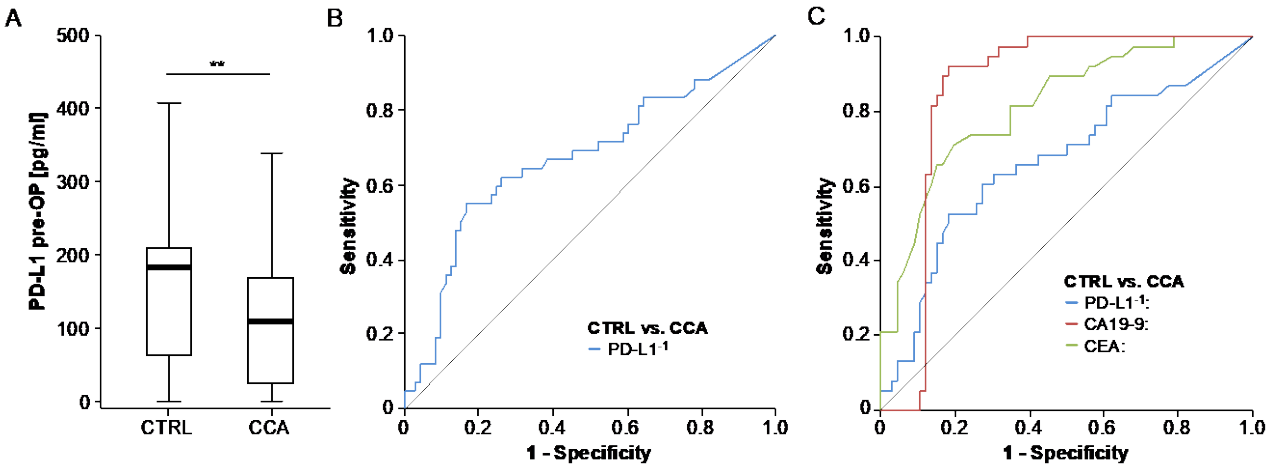
Background and Aims:
Tumor resection represents the only curative treatment option for patients with biliary tract cancer (BTC). However, many patients develop early tumor recurrence and are unlikely to benefit from surgery. Therefore, markers to identify ideal surgical candidates are urgently needed. Circulating programmed cell death 1 ligand 1 (PD-L1) has recently been associated with different malignancies, including pancreatic cancer which closely resembles BTC in terms of patients´ prognosis and tumor biology. Here, we aim at evaluating a potential role of circulating PD-L1 as a novel biomarker for resectable BTC.

Method:
Serum levels of PD-L1 were analyzed by ELISA in 73 BTC patients as well as 42 healthy controls.

Results:
Circulating levels of PD-L1 were significantly lower in patients with BTC compared to controls. In contrast, serum levels of circulating PD-L1 were independent of patient or tumor characteristics. Patients with low PD-L1 levels displayed a strong trend towards an impaired prognosis and circulating PD-L1 negatively correlated with experimental markers of pro-malignant tumor characteristics such as CCL1, CCL21, CCL25 and CCL26. For 37 out of 73 patients postoperative PD-L1 levels were available. Interestingly, after tumor resection circulating PL-L1 raised to almost normal levels. Notably, patients with further decreasing PD-L1 concentrations after surgery showed a trend towards an impaired postoperative outcome.

Conclusion:
Circulating PD-L1 represents a novel biomarker for the diagnosis of patients with resectable BTC, which might help to identify patients who will particularly benefit from surgery.

Figure:
Serum levels of PD-L1 are elevated in patients with CCA



P105YI Endothelial Angiopoietin-2 overexpression in explanted HCCs identifies subjects at high risk of recurrence after liver transplant

Simone Lasagni[1], Filippo Leonardi[2], Lorenza Di Marco[1], Alessandra Pivetti[1], Fabiola Milosa[1], Rosina Maria Critelli[1], Nicola De Maria[1], Paolo Magistri[3], Federico Casari[4], Cristian Caporali[4], Veronica Bernabucci[1], María Luz Martínez-Chantar[5], Daniela Cannamela[1], Dante Romagnoli[1], Fabrizio Di Benedetto[3], Erica Villa[1]

[1]Policlinico di Modena Azienda Ospedaliero-Universitaria di Modena, Gastroenterology, Modena, Italy, *[2]Ospedale Papa Giovanni XXIII , Gastroenterology, Bergamo, Italy*, *[3]Policlinico di Modena Azienda Ospedaliero-Universitaria di Modena, Liver Transplant Unit, Modena, Italy*, *[4]Policlinico di Modena Azienda Ospedaliero-Universitaria di Modena, Radiology, Modena, Italy*, *[5]Bizkaia Science and Technology Park, Liver Diseases Lab, Derio, Spain*
Email: erica.villa@unimore.it

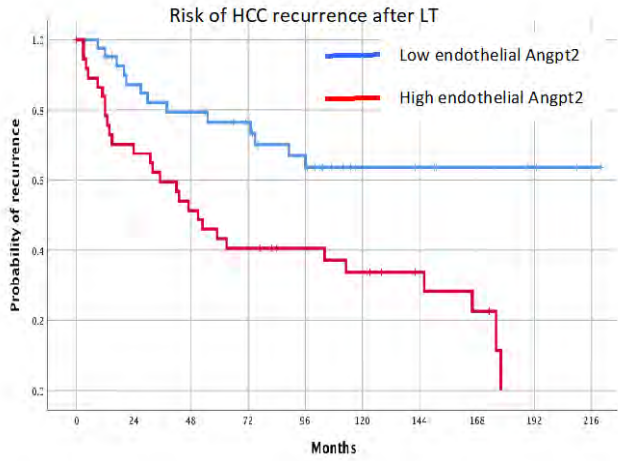
Background and Aims:
HCC recurrence (HCC-R) after liver transplant (LT) remains a relevant problem. To identify the predictive factors associated with HCC-R and survival we analyzed clinical,histopathological and biological features of LT patients with HCC-R or no-HCC-R focusing on angiopoietin-2 expression as a molecular marker of biologic aggressiveness.

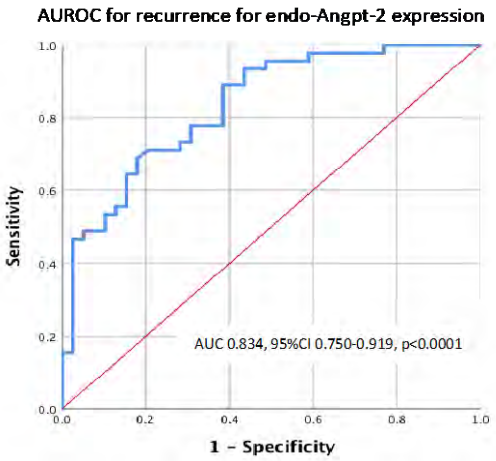
Method:
We analyzed 94 LT patients (47 with HCC-R, matched by age and LT date with 47 no-HCC-R) followed up for median 6.2 years. Probability of survival and recurrence was assessed by Kaplan-Maier curves. Predictive power for recurrence and survival of gender,BMI, alpha-fetoprotein at LT, portal vein thrombosis (PVT) at LT, down staging treatments, predictive scores (Milan,Upto7,Metroticket_Afp), and features of explanted liver (tumor size, residual tumor vitality, presence of microvascular invasion, and endothelial angiopoietin-2 expression [endo-angpt-2]), were assessed by multivariate Cox model.

Results:
HCC-R occurred significantly more often in males (p=0.027, Fisher exact test), with no differences in etiology. At the time of last visit, 48 patients (51.1%) had died. Deaths were significantly higher in HCC-R vs. no-HCC-R patients (80.9% vs 19.1% respectively, p<0.0001), being related in 86.8% of HCC-R to progression of recurrent HCC and in 66.7%) of no-HCC-R patients with sepsis. Endo-angpt-2 was significantly associated with recurrence at KM analysis (Fig. 1).
Univariate analysis for survival identified PVT, residual tumor vitality, Upto7 score, endothelial angiopoietin-2 expression as significant, all but Upto7 being independently associated with survival at multivariate analysis (PVT: HR 3.532,95%CI 1.044-11.945); residual nodule vitality: HR 4.081, 95%CI 1.353-12.354; endo-angpt-2 expression: HR 3.910; 95%CI 1.671-9.150). At univariate Cox analysis, individual clinical scores, gender, BMI, residual tumor vitality, and endothelial angiopoietin-2 expression predicted recurrence. At multivariate analysis, angiopoietin-2 expression only independently predicted recurrence (HR: 3.910, 95%CI 1.577-9.691). AUROC for endo-Angpt-2 expression for recurrence was 0.834 (95%CI 0.750-0.919)(Fig. 1).

Conclusion:
Overexpression of endo-Angpt-2 in explanted HCCs is a strong predictor for mortality, together with PVT and residual nodule vitality, but is the only independent factor for HCC recurrence after LT, further confirming its negative prognostic role in HCC (doi.org/10.1136/gutjnl-2014-308483). Its ability to predict recurrence should be tested prospectively before LT as the high mortality associated with endo-angpt-2-positive HCCs drastically reduces the transplant benefit.

Figure:


Risk of HCC recurrence after LT
Low endothelial Angpt2
High endothelial Angpt2
Probability of recurrence
Months
0
24
48
72
96
120
144
168
192
216


AUROC for recurrence for endo-Angpt-2 expression
Sensitivity
1 – Specificity
AUC 0.834, 95%CI 0.750-0.919, p<0.0001
0.0
0.2
0.4
0.6
0.8
1.0

P106YI Role of LI-RADS indeterminate observations in the prediction of HCC occurrence after direct antiviral therapy for hepatitis C virus infection.

Francesco Matteini[1], Federica Vernuccio[2], Roberto Cannella[3], Giuseppe Cabibbo[4], Silvia Greco[5], Massimo Midiri[3], Calogero Cammà[6], Vito Di Marco[7], Giusseppe Brancatelli[8]

[1]University of Palermo , BiND Department: Biomedicine, Neuroscience and Advanced Diagnostic, Palermo, Italy, *[2]University of Palermo, BiND Department: Biomedicine, Neuroscience and Advanced Diagnostic, , Palermo, Italy*, *[3]University of Palermo, BiND Department: Biomedicine, Neuroscience and Advanced Diagnostic, Palermo, Italy*, *[4]University of Palermo, Department of Health Promotion Sciences Maternal and Infant Care, Section of Gastroenterology & Hepatology, Internal Medicine and Medical Specialties, PROMISE, Palermo, Italy*, *[5]University of Palermo, BiND Department: Biomedicine, Neuroscience and Advanced Diagnostic, Palermo, Italy*, *[4]University of Palermo, Department of Health Promotion Sciences Maternal and Infant Care, Section of Gastroenterology & Hepatology, Internal Medicine and Medical Specialties, PROMISE, Palermo, Italy*, *[4]University of Palermo, Department of Health Promotion Sciences Maternal and Infant Care, Section of Gastroenterology & Hepatology, Internal Medicine and Medical Specialties, PROMISE, Palermo, Italy*, *[3]University of Palermo, BiND Department: Biomedicine, Neuroscience and Advanced Diagnostic, Palermo, Italy*
Email: dott.francescomatteini@gmail.com

Background and Aims:
To assess whether the presence of Liver Imaging Reporting and Data System (LI-RADS) indeterminate observations (LR-3 or LR-4) have an impact on HCC (LR-5) occurrence in HCV patients treated with direct acting antiviral (DAA) therapy.

Method:
This retrospective study included HCV patients at risk for HCC treated with DAA therapy who achieved sustained virologic response (SVR) between 2015 and 2019 and submitted to CT/MRI follow-ups with a minimum interval time of six months before and after DAA. Two blinded readers reviewed CT/MRI to categorize observations according to LI-RADSv2018. Differences in rate of LR-5 before and after SVR were assessed. Predictors of HCC after DAA and time to LR-5 occurrence were evaluated by using the Cox proportional hazard model, Kaplan-Meier method, and log-rank test.

Results:
Our final study population comprised 115 patients (median age 72 years) with a median CT/MRI follow-up of 47 months (IQR 26-77 months). Twenty-nine (25.2%) patients were diagnosed with LR-5 after DAA. The cumulative incidence of LR-5 after DAA was 10.4% (12/115) at 1 year and 24.3% (28/115) at 4 years. The rate of LR-5 after DAA therapy was significantly lower compared to pre-DAA (n = 29, 25.2% *vs*. n = 52, 45.2%; p = 0.001). Presence of LR-3 or LR-4 observations was a significant predictor of LR-5 after DAA at univariate (p = 0.030) and multivariate analysis, (p = 0.048), and was associated with higher incidence of LR-5 (p = 0.024)

Conclusion:
The presence of LR-3 and LR-4 observations increases HCC risk following the eradication of HCV infection.

Figure:

P107YI lncRNA-H19 as a marker of liver disease progression: Hepatocellular Carcinoma

Ángela Rojas[1 2], Antonio Gil-Gomez[1 3], Patricia de la Cruz-Ojeda[4], Rocio Munoz Hernandez[1 2], Yolanda Sánchez[5], Rocío Gallego-Durán[1 2], M. Carmen Rico[1], Maria Teresa Ferrer[5], Raquel Millan[1], Rocío Montero-Vallejo[1], Sheila Gato Zambrano[1], Francisco Javier Padillo[6], Jordi Muntané[4], Javier Ampuero[1 2 5], Manuel Romero Gomez[1 2 5]

[1]Seliver Group. Instituto De Biomedicina De Sevilla (IBiS), Hospital Universitario Virgen Del Rocío/Csic/Universidad De Sevilla. , Sevilla, Spain, *[2]Centro De Investigación Biomédica En Red De Enfermedades Hepáticas y Digestivas (CIBEREHD). , Spain*, *[3]Centro De Investigación Biomédica En Red De Enfermedades Hepáticas y Digestivas (CIBEREHD). , Sevilla, Spain*, *[4]Laboratorio de Cirugía oncológica, terapia celular y trasplante de órganos. Instituto De Biomedicina De Sevilla (IBiS), Hospital Universitario Virgen Del Rocío/Csic/Universidad De Sevilla., Spain*, *[5]Servicio de Digestivo, Hospital Universitario Virgen Del Rocio, Sevilla, España. , Sevilla, Spain*, *[6]Servicio de Cirugía General, Hospital Universitario Virgen Del Rocío, Sevilla, España. , Sevilla, Spain*
Email: mromerogomez@us.es

Background and Aims:
Epigenetics play an important role in the liver progression to hepatocellular carcinoma (HCC). Liver cancer stem cells (LCSCs) could be responsible for the carcinogenesis, recurrence, metastasis and chemoresistance of HCC. The main aims of this study were a) to analyze the epigenetic profile of LCSC c) Validation of H19 as a biomarker in HCC patients

Method:
a) In vitro EpCAM-CD133-LCSC were isolated from a Huh7.5 cell line by FACS and functional assays and epigenetic profile were analysed, b)H19 levels were determined in 12 liver tissues from cirrhotic patients, with HCC (n=6) or without (n=6). Circulating H19 was evaluated in 85 patients; 28 with liver cirrhosis without HCC at baseline (of them, 8 developed HCC during follow-up) and 57 suffering from HCC. Seventeen patients with HCC underwent therapy were follow-up during at least 3 months (6 with complete response, 7 partial response and 4 null responder). Total RNA was isolated from plasma and H19 quantified by ddPCR.

Results:
a) $EpCAM^{+}CD133^{+}$ cells presented stem cells phenotype, self-renewal capacity (an increase in size and number of spheroids was observed (p=0.004 and p<0.001); upregulation of pluripotent genes expression NANOG, POU5F1, Sox-2 (p=0.05) and H19 (p=0.003). Suppression of H19 by antisense oligo treatment significantly reduce self-renewal capacity (p<0.001)

b) H19 was found significantly increased in liver tissue from HCC patients compared to cirrhotic patients (fold- 4.45±1.003 p=0.0082). In addition, H19 was found upregulated in plasma from HCC vs. cirrhotic patients without development of HCC (Cirrhotic=1.02±0.58 vs. HCC: 2.86±2.05; p<0.0001; AUC 0.841 (CI95%: 0.751-0.930 p<0.0001). By multivariate analysis, we confirmed the association of the male sex (OR: 119.792 (1.145-12533.8) p=0.044), age (OR: 1.23 (1.038-1.460) p=0.017 and lncRNA-H19 (OR: 9.644 (1.175-79.188) p=0.035) with the development of HCC. In addition, a positive correlation was found between lncRNA-H19 levels and BCLC stages (p<0.05). Cirrhotic patients who developed HCC during the follow-up showed higher levels of H19 compared to non-HCC cirrhotic patients(p=0.0025) (AUC: 0.856, 95%CI: 0.637–1.00) p =0.004). H19 plasma levels decrease significantly in those patients who achieved complete and partial response after 3 months of follow-up (p=0.046 and 0.028 respectively).

Conclusion:
lncRNA-H19 was found to be increased in LCSC, promoters of the carcinogenesis. Also, it was found upregulated in liver tissue and plasma from HCC patients. H19 was upregulated in cirrhotic patients who developed HCC during the follow-up. In addition, H19 decreased after 3 months of follow up in those patients with partial and complete response. Therefore, H19 could constitute a biomarker of disease progression and HCC diagnosis.

P108 Nanobody-targeted liposomes for photodynamic therapy of Cholangiocarcinoma

Bárbara Mesquita[1], Michal Heger[1 2], Sabrina Oliveira[1 3]

[1]Utrecht Institute for Pharmaceutical Sciences, Utrecht University, Pharmaceutics Department, Utrecht, Netherlands, *[2]College of Medicine, Jiaxing University, Department of Pharmaceutics, China*, *[3]Division of Cell Biology, Neurobiology and Biophysics, Utrecht University, Cell Biology Department, Utrecht, Netherlands*
Email: s.oliveira@uu.nl

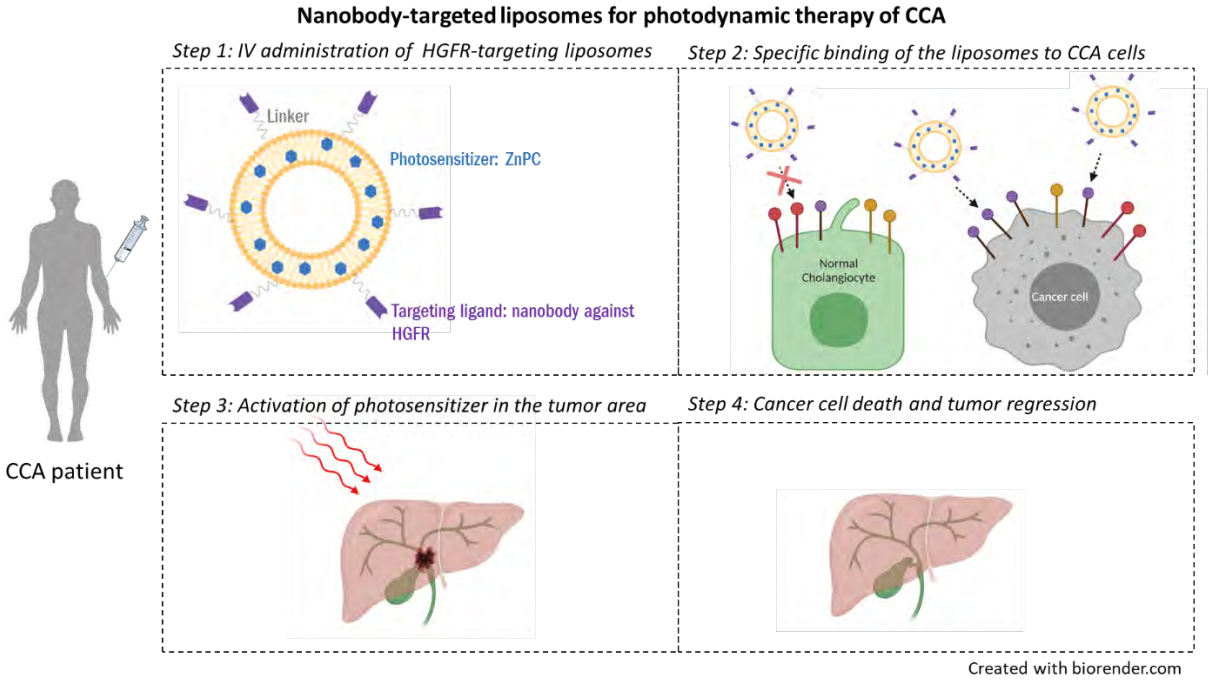
Background and Aims:
Photodynamic therapy (PDT) is a promising alternative to currently employed treatments for cholangiocarcinoma (CCA) (Quyn, A.J., *et al.* 2009). However, clinical implementation is still limited due to abundant skin phototoxicity, forcing patients with a short life expectancy to remain inside and shielded from light. To reduce skin photosensitization, we developed liposomes encapsulating zinc phthalocyanine (ZnPC) and decorated with nanobodies against hepatocyte growth factor receptor (HGFR) to increase specificity of ZnPC to CCA cells and prevent extravasation from cutaneous microcirculation.

Method:
Liposomes encapsulating ZnPC (ZnPC-L) at 0.3% mol of total lipid were prepared by lipid film hydration. Nanobodies targeting HGFR (named G2 (Heukers, Raimond, *et al.* 2014)) were conjugated to the surface of liposomes via maleimide-thiol click chemistry. Z-average hydrodynamic diameter and size distribution were measured by dynamic light scattering and the G2 conjugation efficiency was evaluated using SDS-PAGE. To investigate specific binding to CCA cells, TFK1 cell line (with elevated expression of HGFR compared to THLE2, a hepatocyte cell line) was exposed to G2-targeted ZnPC-L at 4 ˚C for 1 hour. A competition group consisting of G2-targeted ZnPC-L and an excess of free nanobody was used to verify specific interaction. Confocal microscopy was employed to observe the localization of the ZnPC and confirm specific binding of the G2-targeted ZnPC-L to HGFR. The phototoxicity of nanobody-targeted and non-targeted ZnPC-Lwas tested *in vitro* by exposing TFK1 to a range of ZnPC-loaded liposome concentrations. After incubation and washing, the cells were illuminated with a LED device. Cell viability was measured following overnight incubation with a sulforhodamine B assay.

Results:
G2-targeted and non-targeted ZnPC-L had a size of 168 ± 1 nm and 158 ± 3 nm, respectively.
G2-targeted ZnPC-L exhibited specific binding to TFK1 cells through HGFR and, upon illumination, induced higher phototoxicity compared to non-targeted liposomes.

Conclusion:
CCA-targeting liposomes containing ZnPC were developed that selectively bind HGFR overexpressed on cholangiocytes. The liposomes are not cytotoxic in the dark but induce extensive cell death upon exposure to resonant light. The formulation constitutes a first-in-line photonanomedicine for CCA that will next be validated in vivo.

Figure:


Nanobody-targeted liposomes for photodynamic therapy of CCA
Step 1: IV administration of HGFR-targeting liposomes
Step 2: Specific binding of the liposomes to CCA cells
Linker
Photosensitizer: ZnPC
Targeting ligand: nanobody against HGFR
Normal Cholangiocyte
Cancer cell
CCA patient
Step 3: Activation of photosensitizer in the tumor area
Step 4: Cancer cell death and tumor regression
Created with biorender.com

P109YI Effectiveness of hepatocellular carcinoma treatments with curative intent: a nationwide study

Frederik Kraglund[1], Gerda Elisabeth Villadsen[1], Peter Jepsen[1]

[1]*Aarhus University Hospital, Department of Hepatology and Gastroenterology, Aarhus N, Denmark*
Email: frekra@clin.au.dk

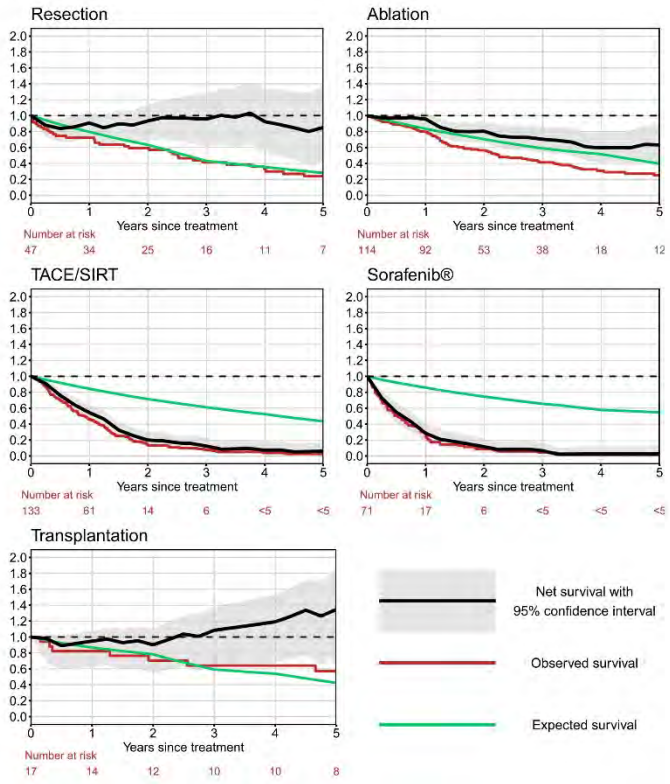
Background and Aims:
Most patients with hepatocellular carcinoma (HCC) have cirrhosis, which in itself is associated with a high mortality. Resection and ablation are curative-intent treatments for HCC, aiming to restore to the patient to the survival they had without HCC. We examined the extent to which this aim was fulfilled.

Method:
Using the Danish health registries, all patients with HCC and all HCC treatments in 2000-2018 were identified. Patients were followed from the date of their first HCC resection or ablation, and recurrence was defined as renewed HCC treament of any kind or referral to palliative care >3 months later. Survival was measured using Kaplan-Meier estimation, and recurrence risk was modelled with death without recurrence as a competing event. Net survival was estimated using the Ederer II method in a control cohort of patients with cirrhosis due to alcohol-related liver disease.

Results:
After resection, the 5-year survival increased from 38.0% (95% CI: 32.9−43.1) on the date of surgery to 59.4% (46.2−70.4) at 5 years after the surgery, conditional on remaining alive. On the date of ablation, the 5-year survival was 28.8% (95% CI: 22.9−34.9), and despite increasing age, it remained stable over time since the procedure. After resection, the 5-year recurrence risk decreased markedly from 38.5% (95% CI: 33.5−43.5) on the date of surgery to 1.9% (95% CI: 0.2−8.6) at 5 years after the resection, conditional on remaining alive and recurrence-free. After ablation, the 5-year recurrence risk decreased from 50.6% (95% CI: 44.1−56.8) to 11.4% (95% CI: 1.8−30.7). Net survival decreased gradually over 5 years to 0.85 (95% CI: 0.42−1.37) after resection and to 0.63 (95% CI: 0.40−0.89) after ablation. In contrast, net survival dropped to close to zero after a few years following life-prolonging HCC treatments, and it increased after liver transplantation (**Figure**).

Conclusion:
After curative-intent HCC treatments, overall survival is low and, during the first months, recurrence risk is high. Even so, the majority of patients are restored to the survival they had without HCC.

Figure:


Resection
Ablation
TACE/SIRT
Sorafenib®
Transplantation
Years since treatment
Number at risk
47 34 25 16 11 7
114 92 53 38 18 12
133 61 14 6 <5 <5
71 17 6 <5 <5 <5
17 14 12 10 10 8
Net survival with 95% confidence interval
Observed survival
Expected survival

P110YI Epidemiological trends of hepatocellular carcinoma in patients with Metabolic-Dysfunction-Associated Fatty Liver Disease (MAFLD) in Italy.

Alessandro Vitale[1], Gianluca Svegliati-Baroni[2], Giulio Marchesini Reggiani[3], Monica Cucco[4], Umberto Cillo[1], Franco Trevisani[3], Luca Miele[5], Fabio Farinati[1]

[1]University of Padua, Department of Surgery, Oncology and Gastroenterology, Padua, Italy, *[2]Polytechnic University of Marche, Liver Injury and Transplant Unit, Ancona, Italy*, *[3]Alma Mater Studiorum–University of Bologna, Department of Medical and Surgical Sciences*, *[4]Polytechnic University of Marche, Division of Gastroenterology, Ancona, Italy*, *[5]Policlinico Gemelli, Università Cattolica del Sacro Cuore, Internal Medicine and Gastroenterology Unit, Rome, Italy*
Email: alessandro.vitale@unipd.it

Background and Aims:
Metabolic-dysfunction-associated fatty liver disease (MAFLD) represents a new inclusive definition of the whole spectrum of liver diseases associated to metabolic disorders. The main objective of this study was to compare MAFLD and non-MAFLD patients with hepatocellular carcinoma (HCC) included in a nationally representative cohort.

Method:
We analysed 6,882 consecutive HCC patients enrolled from 2002 to 2019 by 23 Italian Liver Cancer centers. We compared three subgroups, pure MAFLD (MAFLD1), combined (metabolic and other) aetiology MAFLD (MAFLD2) and non-MAFLD HCC for epidemiological trends across biennials in the study period and explored future trends using a linear regression model. We also compared these subgroups in terms of baseline characteristics, staging, treatment allocation and death risk using stabilized inverse probability weights and competing risk analyses.

Results:
MAFLD characterized the majority of Italian HCC patients in the study period (68.4%). The proportion of both overall MAFLD and MAFLD1-HCC significantly increased over time (from 50.4% and 3.6% in the first biennium, to 77.3% and 28.9% in 2018-19, respectively, $p<0.001$), driven by type 2 diabetes and obesity epidemics; regression showed that MAFLD1-HCC should overcome MAFLD2-HCC in about ten years in Italy. MAFLD1 HCC patients were older, more frequently males, and had less frequently cirrhosis (74.0% in MAFLD1 vs 90.8% and 92.2% in non-MAFLD and MAFLD2 respectively, $p<0.05$), clinically relevant portal hypertension, and a surveillance-related diagnosis. They were characterized by larger tumours and extra-hepatic metastases compared to the other groups.
After weighting, MAFLD1- and 2-HCC patients showed a significantly lower overall ($p=0.026$, $p=0.004$) and HCC-related ($p<0.001$, $p<0.001$) risk of death, while MAFLD1 had a significantly higher risk for non-HCC-related death vs. non-MAFLD ($p=0.006$).

Conclusion:
The prevalence of MAFLD in Italy is rapidly increasing and involves the majority of HCC patients. Despite a less favourable tumour stage at diagnosis, MAFLD-HCC patients have a lower risk for HCC death, suggesting reduced aggressiveness.

P111 An unexpected side effect of lenvatinib in patients treated for advanced hepatocellular carcinoma: increased hemoglobin level and erythrocytosis

Alina Pascale[1], Laurence Legros[2], yasmina ben merabet[3], Maryse Stang[4], Ayhan Ulusakarya[4], Emma Goldschmidt[4], Maite Lewin[5], Jean-Francois Morere[4], Didier Samuel[3], Olivier Rosmorduc[3]

[1]Paul Brousse Hospital, APHP, Liver Unit, Villejuif, France, *[2]Paul Brousse Hospital, APHP, Hematology and Oncology Unit, Villejuif, France*, *[1]Paul Brousse Hospital, APHP, Liver Unit, Villejuif, France*, *[4]Paul Brousse Hospital, APHP, Oncology Unit, Villejuif, France*, *[5]Paul Brousse Hospital, APHP, Radiology Unit, Villejuif, France*
Email: alina_pascale@yahoo.com

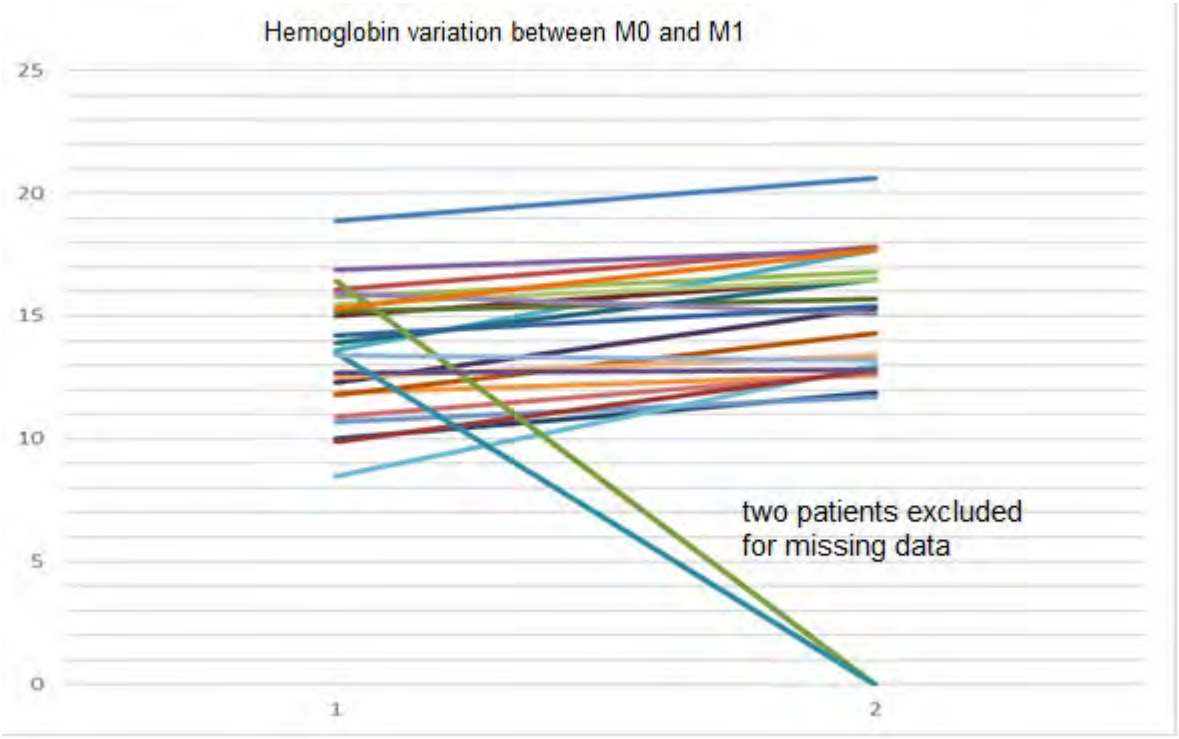
Background and Aims:
Lenvatinib is a tyrosin kinase inhibitor (TKI) approved as a first line treatment for advanced hepatocellular carcinoma (HCC), with well-known side effects: hypertension, proteinuria, fatigue, diarrhea, palmar-plantar erythrodysesthesia syndrome. We report here an unknown side effect of lenvatinib: increased hemoglobin levels and erythrocytosis.

Method:
We included 25 consecutively treated patients in our center since 01/08/2019. Two patients were excluded for missing data. Blood cell counts were performed at treatment initiation, monthly during treatment and another two months after treatment end. Lenvatinib dose was adapted to body weight. Other causes for polycythemia were excluded by testing erythropoietin, B12 vitamin, reticulocyte levels and JAK2 genetic analysis. Side effects were recorded according to Common Criteria Terminology for Adverse Events 4.0. Radiologic response was defined according to RECIST 1.1 criteria. Statistical analysis was performed in SPSS 18.0: t test, Pearson/Spearman correlations and Wilcoxon Ranks Test.

Results:
The median age of patients was 58.3 years. Majority of patients were men (82.6%). All patients had a BCLC B or C HCC, in 60.86% cases on cirrhotic liver; 17.3% had recurrent HCC post liver transplantation; two patients had a fibrolamellar HCC. Lenvatinib was the first line systemic therapy in 87% of patients. The mean of hemoglobin level at base-line (M0), after one (M1) and two months of treatment (M2) were respectively: 13.97 g/dL, 15.38 g/dL and 15.63 g/dL. The mean increase of hemoglobin levels between M0 and M1 was 1.41 g/dL ($p < 0.001$) and then remained sustained during treatment. Ten patients (43.5%), all males, reached levels corresponding to erythrocytosis (> 16.5 g/dL) and were treated with low-dose aspirin for the primary prevention of cardiovascular events. None underwent thrombotic complications and two patients needed a phlebotomy. None of the patients needed to stop treatment due to erythrocytosis. Increased hemoglobin levels were not correlated with: HCC histological type, cirrhotic liver, radiological response, treatment duration, other side effects occurrence ($p > 0.05$). A reversible decrease of hemoglobin level was observed in patients who stopped treatment (56.5%) ($p < 0.05$).

Conclusion:
We report here early and reversible increased hemoglobin values, up to levels corresponding to erythrocytosis, during lenvatinib treatment for HCC. This effect seems to be specific to this setting. It has not been previously described with another TKI in HCC treatment, or in other indication of lenvatinib. We propose a close follow-up of patients, at least monthly by blood cell count and a prophylactic treatment with low-dose aspirin in case of erythrocytosis, allowing to pursuit treatment. A phlebotomy could be considered in selected patients.

Figure:


Hemoglobin variation between M0 and M1
25
20
15
10
5
0
1
2
two patients excluded
for missing data

P112YI Are Radiology-based endpoints robust surrogate outcomes of overall survival in hepatocellular carcinoma patients treated with transarterial chemoembolization?

Ciro Celsa[1], Giuseppe Cabibbo[1], Marco Enea[2], Salvatore Battaglia[3], Giacomo Emanuele Maria Rizzo[1], Anita Busacca[1], Paolo Giuffrida[1], Caterina Stornello[1], Giuseppe BRANCATELLI[4], Roberto Cannella[4], Salvatore Gruttadauria[5], Calogero Camma[1]

[1]University of Palermo, Section of Gastroenterology and Hepatology, PROMISE Department, Palermo, Italy, *[2]University of Palermo, PROMISE*, *[3]University of Palermo, Dipartimento di Scienze Economiche, Aziendali e Statistiche*, *[4]University of Palermo, Dipartimento di Biomedicina, Neuroscienze e Diagnostica avanzata (BIND)*, , *[5]IRCCS-ISMETT (Istituto Mediterraneo per i Trapianti e Terapie ad alta specializzazione), UPMC (University of Pittsburgh Medical Center), , Department for the Treatment and the Study of Abdominal Diseases and Abdominal Transplantation.*
Email: calogero.camma@unipa.it

Background and Aims: Time to progression (TTP) and progression-free survival (PFS) are commonly used as surrogate endpoints in oncology trials. We aimed to assess the surrogacy relationship of TTP and PFS with overall survival (OS) in studies of transarterial chemoembolization (TACE) for unresectable hepatocellular carcinoma (u-HCC) by innovative methods.

Method: A search of databases for studies of TACE for u-HCC reporting both OS and TTP or PFS was performed. Individual patient data were extracted from TTP/PFS and OS Kaplan-Meier curves of TACE arms. Pooled median TTP and OS were obtained from random-effect model. The surrogate relationships of hazard ratios(HRs) and median TTP for OS were evaluated by the coefficient of determination R2.

Results: We identified 13 studies comparing TACE versus systemic therapy or versus TACE plus systemic therapy and including 1932 TACE-treated patients. Pooled median OS was 11.2 months (95% Confidence Interval [95%CI] 7.9-17.8) and pooled median TTP was 5.4 months (95%CI 3.8-8.0). Heterogeneity among studies was highly significant for both outcomes. The correlation between HR TTP and HR OS was moderate (R2 = 0.65. 95%CI 0.08-0.81). R2 value was 0.04 (95%CI 0.00-0.35) between median TTP and median OS.

Conclusion: In studies of TACE for u-HCC, the surrogate relationship of radiology-based endpoints with OS is moderate. Multiple endpoints including hepatic decompensation, macrovascular invasion and extrahepatic spread are needed for future trials comparing systemic therapies or combination of TACE with systemic therapies versus TACE alone.

Figure:

P113 Vimentin expression in hepatocellular carcinoma relates with progenitor cells and epithelial-mesenchymal transition carcinogenesis

Joana Espírito Santo[1], Ana Ladeirinha[2], Ana Alarcão[2], Lia Neves[2], Rui Santos[3], Lina Carvalho[2]

[1]Institute of Anatomical and Molecular Pathology, Faculty of Medicine, University of Coimbra, Adult Liver Transplantation Unit, Coimbra Hospital and Universitary Centre, Portugal, *[2]Institute of Anatomical and Molecular Pathology, Faculty of Medicine, University of Coimbra, Coimbra, Portugal*, *[3]Coimbra Hospital and Universitary Centre, Internal Medicine Department A, Coimbra, Portugal*
Email: jesespsanto@gmail.com

Background and Aims:
Epithelial-mesenchymal transition (EMT) is a cellular reprogramming process during which epithelial cells lose their phenotypic characteristics and acquire mesenchymal cell features. EMT plays a key role in liver inflammation and dependent carcinogenesis and is thought to be associated to hepatic carcinomas increased invasion, proliferation and chemoresistance. Vimentin is a cytoskeletal protein and one of the biomarkers of EMT. Co-expression of vimentin, EpCAM/BerEp4 and CK7/CK19 in hepatic malignant cells indicates clinical aggressive behavior as well as dedifferentiation/stem cells properties.

Method:
A series of 89 hepatocellular carcinomas (HCCs), from 124 liver nodules, were studied concerning 54 consecutive patients undergoing hepatic resection or transplantation (2012-2016). According with 2019 World Health Organization (WHO) histopathological criteria, CK7/CK19, EpCAM/BerEp4 and vimentin immunoexpression in single or clusters of tumour cells were searched.

Results:
From the 89 HCCs studied (18 early and 72 progressed tumours), 39 cases presented vimentin positive cells and 44 cases had relevant vimentin immunoexpression in stroma cells; 18 cases had both immunoexpression in tumour cells and in respective stroma cells and 48 cases showed peritumoural vimentin expression in hepatocytes.
In the series of 39 HCCs expressing vimentin, 6 cases also expressed CK7 /CK19, 15 were CK7+/CK19-, 1 case was CK7-/CK19+; 8 cases of the 39 HCCs expressed EpCAM/BerEp4 where 5 cases were CK7+/CK19+. A significant association ($p<0,05$) was observed between tumour stroma immunoexpression for vimentin and CK7, CK19 and EpCAM/BerEp4 tumour cells immunoexpression.
Twenty-nine HCCs with vimentin positive tumour cells presented pseudoglandular growth pattern, where 13 cases showed that pattern combined with trabecular morphology; 9 cases were G1, 27 cases were G2 and 3 cases were G3; 9 cases displayed evident microvascular invasion and 5 cases had intrahepatic satellite nodules. Out of the 27 patients with HCC tumour cells expressing vimentin, 6 cases had HCC recurrence within 6 years after curative surgery.

Conclusion:
Vimentin expression may reflect either adult stem cells EMT carcinogenesis and/or dedifferentiation of HCC epithelial malignant cells, showing the importance of tumour microenvironment in liver carcinogenesis. The challenging interaction between EMT markers and stemness agents plays a key role in HCC development and emphazises the need for further research in immunotherapy and molecular targeted treatments.

Figure:

P114YI Inducing tissue-resident gamma delta T cells for immunotherapy of hepatocellular carcinoma

Nekisa Zakeri[1], Leo Swadling[1], Laura J Pallett[1], Nathalie Schmidt[1], Mariana Diniz[1], Oliver Amin[1], Stephanie Kucykowicz[1], Amir Gander[2], Massimo Pinzani[3], Brian R Davidson[2], Mala K Maini[1]

[1]Division of Infection & Immunity, University College London, United Kingdom, *[2]Department of Surgery and Liver Transplantation, Royal Free Hospital, University College London, United Kingdom*, *[3]UCL Institute for Liver and Digestive Health, Royal Free Hospital, London, United Kingdom*
Email: nzakeri@doctors.org.uk

Background and Aims:
Gamma delta T cells, in particular Vγ9Vδ2 T cells, are attractive therapeutic targets for cancer immunotherapy due to their potent cytotoxicity, MHC-unrestricted antigen recognition, and capacity for clinical-scale expansion. Early-phase clinical trials have demonstrated that Vγ9Vδ2 T cell-based immunotherapy is safe and well-tolerated, however tumour response rates remain low. We aimed to characterise in detail liver- and tumour- infiltrating Vγ9Vδ2 T cells in patients with hepatocellular carcinoma (HCC), and explore strategies to enhance their therapeutic potential.

Method:
Using multiparameter flow cytometry, we analysed freshly isolated lymphocytes from peripheral blood samples, tumour-free liver tissue, and tumoural tissue resected from patients with HCC or colorectal cancer liver metastases (CRCLM), as well as peripheral blood from healthy controls. Long-lived tissue-sequestered persistence of Vγ9Vδ2 T cells was examined using explants from HLA-mismatched liver transplants. Peripheral blood Vγ9Vδ2 T cells were expanded for 10-days using the aminobisphosphonate Zoledronate and IL-2. Expanded Vγ9Vδ2 T cells, intrahepatic lymphocytes, and tumour-infiltrating lymphocytes, were co-cultured with human hepatoma cell lines (HepG2 and HuH7) which were pre-treated with Zoledronate for 16 hours, promoting tumour cell phosphoantigen accumulation for Vγ9Vδ2 T cell receptor activation.

Results:
Vγ9Vδ2 T cells were reduced in frequency in the blood, liver, and tumours of patients with HCC, in comparison to healthy controls and CRCLM. We demonstrated that a subset of intrahepatic Vγ9Vδ2 T cells with a tissue-resident memory (TRM) phenotype (CD69+CD49a+), had the capacity for long-lived tissue-compartmentalised persistence, and could be replenished from the circulation, an attractive profile to recapitulate with immunotherapy. Zoledronate and IL-2 based expansion of peripheral blood Vγ9Vδ2 T cells induced a *de novo* T_{RM} phenotype (CD69+CD49a+) and higher CXCR3+CXCR6+ expression, promoting liver homing and retention. In addition, Zoledronate pre-treatment of HCC cell lines *in vitro* enhanced the anti-tumour effector function (IFNγ, TNFα, Granzyme B, CD107a) of co-cultured expanded Vγ9Vδ2 T cells, as well as Vγ9Vδ2 T cells directly isolated from HCC livers and tumours, with increased tumour-cell lysis observed.

Conclusion:
A reduction of intrahepatic and intra-tumoural Vγ9Vδ2 T cells in HCC may contribute to tumour immune evasion. We show that Zoledronate can enhance the efficacy of gamma-delta T cell immunotherapy in HCC, via two independent mechanisms: direct delivery to the tumour increases Vγ9Vδ2 T cell recognition, whilst *in vitro* expansion of Vγ9Vδ2 T cells for adoptive cell transfer confers liver-homing tissue residency to achieve long-lived local tumour immunosurveillance.

P115YI Nurse intervention optimizes the care of patients with hepatocellular carcinoma under systemic treatment

Gemma Iserte[1], Neus Llarch[1], nuria granel[1], Víctor Sapena[2], Marta Campos Gomez[2], Marco Sanduzzi Zamparelli[3], Sergio Muñoz Martinez[3], Alejandro Forner[1 2], Jordi Bruix[1 2], María Reig[1 2]

[1] Hospital Clinic Barcelona,, BCLC group, ICMDiM., barcelona, Spain, *[2]CIBERehd, Hospital Clinic, BCLC group, Liver Unit,, barcelona, Spain*, *[3]Fundació Hospital Clinic, BCLC group, Liver Unit,, barcelona, Spain*
Email: giserte@clinic.cat

Background and Aims:
Telephone visits have been part of Nurse Educational Programs for years and are a follow-up tool for liver cancer patients. This tool allows a fluid communication between the patient and the multidisciplinary team, as well as the early identification of treatment side effects, optimizing the management of patients. The aim of this study is to evaluate the impact of advanced practice nurse intervention (APN) managing unscheduled phone visits in patients with advanced hepatocellular carcinoma (HCC) in second and/or third line oral treatment.

Method:
A retrospective, observational and descriptive study of patients who started second and/or third line treatment in a tertiary referral center (BCLC) from 01/2017 to 01/2020, and their phone calls received until 07/2020 or until discontinuation of treatment.

Results:
Forty-one patients with advanced HCC started second or third line treatment; 20 of these patients received treatment with regorafenib and 23 with cabozantinib (2 patients received regorafenib in second line and later cabozantinib in third line). 408 phone calls were registered, 25% of which belonged to confinement period for COVID19 (March 14- June 21, 2020).

In our cohort (36% in regorafenib and 49% in cabozantinib), APNs resolved 100% of administrative consultations and 43% of phone calls related to health problems including potential toxicities of medication or cirrhosis decompensations. In second line cabozantinib patients, APNs resolved up to 73.5% of phone calls related to potential drug toxicity. In only 3.3% of phone calls related to health, patients were referred to the emergency department.

Conclusion:
Telephone consultations managed by the APNs have a positive impact on the outpatient management of patients with HCC in systemic treatment and have been key to maintaining these treatments active during the COVID19 restrictions. The number of patients referred to the emergency department has been low, avoiding unnecessary saturation of the emergency services.

P116 A circulating microRNAs signature identifies DAA-treated HCV-related cirrhosis patients at high risk to develop HCC

Marie-Laure Plissonnier[1], Alessandro Loglio[2], Francesca Guerrieri[1], Floriana Facchetti[2], Claude Caron de Fromentel[1], Mirjam Zeisel[1], Pietro Lampertico[2], Massimo Levrero[1 3 4]

[1]Cancer Research Center of Lyon (CRCL), UMR Inserm U1052-Cnrs 5286 Mixte CLB, Lyon-1 University, Lyon, France, *[2]Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Division of Gastroenterology and Hepatology - "AM and A Migliavacca" Center for Liver Diseases, Milan, Italy*, *[3]University of Rome La Sapienza, Department of Medicine SCIAC, Rome, Italy*, *[4]Hospices Civils de Lyon and Université Lyon1, Department of Hepatology, Hôpital De La Croix Rousse, Lyon, France*
Email: massimo.levrero@inserm.fr

Background and Aims:
Current DAA regimens lead to HCV eradication (SVR - sustained virological response) in the vast majority of treated patients. SVR is associated in HCV cirrhotic patients with the prevention of clinical decompensation and reduction, but not an elimination, of the risk of developing HCC. New diagnostic biomarkers for risk stratification and early diagnosis are urgently needed to increase the access to curative therapeutic options.

Method:
We performed a retrospective analysis of circulating miRNAs using the NanoString nCounter® technology in selected patients from a prospective longitudinal cohort of 565 HCV cirrhotic patients treated with DAAs in a single clinical center. The study population comprised 12 cirrhotic patients treated with DAA reaching SVR who developed HCC (sera collected before starting DAA treatment (*HCV pre-DAA HCC*) and at HCC diagnosis (*HCV SVR HCC*)) and a case-control cohort of 12 DAA-treated patients who did not develop HCC over a comparable follow-up period after SVR (sera collected before starting DAA treatment (*HCV pre-DAA*) and at SVR (*HCV SVR*)).

Results:
Principal component analysis of > 800 circulating miRNA profiles from the 4 groups showed that DAA-treated patients who develop an HCC after SVR (*HCV SVR HCC*) cluster tightly together and are well separated from the DAA-treated patients who do not develop an HCC after SVR (*HCV SVR*). Unsupervised hierarchical clustering analysis shows that HCV eradication has a strong impact on circulating miRNAs and that HCC development is accompanied by a further reduction of the number of circulating miRNAs detected, supporting the notion that the profile of circulating miRNAs in the HCV SVR HCC patients is the result of both HCV eradication and HCC development. Five miRNAs were specifically detected in *HCV SVR HCC* patients and not in *HCV SVR* patients (*diagnostic signature*). 26 cir-miRNAs are detected in patients who will develop HCC after DAAs treatment and SVR (*HCV pre-DAA HCC*) but not in the patients who will do not develop HCC (*HCV pre-DAA*) (*HCC risk signature* in DAA treated patients).

Conclusion:
Our results show that HCV eradication has a strong impact on cir-miRNAs and identify potential candidate cir-miRNAs for HCC diagnosis and prediction in HCV cirrhosis patients treated successfully with DAAs**.** These miRNAs warrant to be included in new clinical/biological scores for HCC risk stratification in HCV-infected patients.

Figure:

P117 Cancer-associated pathways modulated by O-GlcNAc transferase and Enhancer of Zeste homolog 2 in hepatocellular carcinoma

Margot Thirion[1], Marie-Laure Plissonnier[1], Océane Floriot[1], Francesca Guerrieri[1], Claude Caron de Fromentel[1], Massimo Levrero[1 2], Mirjam Zeisel[1]

[1]Cancer Research Center of Lyon (CRCL), UMR Inserm 1052 CNRS 5286 Mixte CLB, Université Lyon 1 (UCBL1), Lyon, France, *[2]Hospices Civils de Lyon, Service d'Hépato-Gastroentérologie, Lyon, France*
Email: massimo.levrero@inserm.fr

Background and aims:
Despite recent improvements, treatment options for hepatocellular carcinoma (HCC) remain largely unsatisfactory. Epidrugs are currently being evaluated as cancer therapy and a first inhibitor of the histone methyltransferase enhancer of zeste homolog 2 (Ezh2) recently obtained FDA approval. The epigenetic writer Ezh2 is frequently upregulated in HCC tissues and increased Ezh2 expression correlates with the aggressiveness and/or poor prognosis of HCCs. Ezh2 knockdown in HCC cells reverses tumorigenicity in a nude mouse model, suggesting a potential therapeutic value of Ezh2 inhibition in HCC. Ezh2 activity is regulated by post-translational modifications, including glycosylation by O-linked N-acetylglucosamine (O-GlcNAc) transferase (OGT). We have previously shown that OGT expression is increased in tumor tissue from HCC patients. Interestingly, it has been reported that OGT and Ezh2 co-repress a subset of potential tumor suppressor genes in breast cancer MCF-7 cells. The aim of our project is to uncover cancer-associated pathways modulated by OGT and Ezh2 in HCC.

Method:
Liver tissue samples from HCC patients were analyzed by RT-PCR. O-GlcNAcylated proteins were purified using a click chemistry-based protocol and analyzed by Western blot. Multiplex transcriptomic analysis of human hepatoma HepG2 cells was performed using NanoString nCounter® technology. Chromatin immunoprecipitation (ChIP) PCR and Sequencing experiments were done using antibodies directed against OGT, Ezh2 or H3K27m3.

Results:
We showed that in a cohort of 153 HCC patients, OGT and Ezh2 are upregulated in tumor tissue as compared to peri-tumor tissue independently of the HCC etiology. In HepG2 cells, Ezh2 is post-translationally modified by OGT and silencing of OGT and Ezh2 modulates the expression of more than 100 genes belonging to major cancer-associated canonical pathways. ChIP experiments demonstrated that OGT and Ezh2 are recruited to the promoter region of several genes whose expression increased following siRNA-mediated silencing of either OGT or Ezh2 and marked by H3K27m3, suggesting that these genes are targeted for repression by OGT and Ezh2 in transformed liver cells.

Conclusion:
Our data uncovered that OGT and Ezh2 comodulate cancer pathways in transformed liver cells and provide perspectives for epigenetic strategies as potential future anti-HCC therapies.

P118YI Assessment of the impact of COVID-19 pandemic on Liver Cancer Management (CERO-19)

Sergio Munoz-Martínez[1], Víctor Sapena[1], Alejandro Forner[1], Jean Charles Nault[2], Gonzalo Sapisochin[3], Lorenza Rimassa[4], Bruno Sangro[5], Jordi Bruix[1], Marco Sanduzzi Zamparelli[1], Wacław Hołówko[6], Mohammed El-Kassas[7], Tudor Mocan[8], Mohamed Bouattour[9], Philippe Merle[10], Frederik Hoogwater[11], Saleh Alqahtani[12], Helen L. Reeves[13], David J. Pinato[14], Emmanouil Giorgakis[15], Tim Meyer[16], Gerda Elisabeth Villadsen[17], Henning Wege[18], Massimiliano Salati[19], Beatriz Minguez[20], Giovan Giuseppe Di Costanzo[21], Christoph Roderburg[22], Frank Tacke[22], Maria Varela[23], Peter Galle[24], Mario Reis Álvares-da-Silva[25], Jörg Trojan[26], John Bridgewater[27], Giuseppe Cabibbo[28], Christian Toso[29], Anja Lachenmayer[30], Andrea Casadei Gardini[31], Hidenori Toyoda[32], Tom Lüdde[33], Rosanna Villani[34], Ana Matilla[35], Cassia Regina Guedes Leal[36], Monica Ronzoni[37], Manuel Delgado[38], Christie Perelló[39], Sonia Pascual[40], José Luis Lledó[41], Josepmaria Argemi[42], Bristi Basu[43], Leonardo Gomes da Fonseca[44], Juan Acevedo[45], Alexander Siebenhüner[46], Chiara Braconi[47], Brandon M. Meyers[48], Alessandro Granito[49], Margarita Sala[50], Carlos Rodriguez de Lope[51], Lorraine Blaise[2], Manuel Romero Gomez[52], Federico Pinero[53], Dhanny Gomez[54], Vivianne Mello[55], Rogério Alves[56], Alex Vianey Callado França[57], Fernanda Branco[58], Giovanni Brandi[59], Gustavo Pereira[60], Susana Coll[61], Maria Guarino[62], Carlos Benitez[63], Maria Margarita Anders[64], Juan Carlos Bandi[65], Mercedes Vergara Gómez[66], Mariona Calvo[67], Markus Peck-Radosavljevic[68], IGNACIO GARCIA JUAREZ[69], Vicenzo Cardinale[70], María del Mar Lozano[71], Martina Gambato[72], Stefano Okolicsanyi[73], Dalia Morales Arraez[74], Alessandra Elvevi[75], Alberto Muñoz[76], Alberto Lué[77], Massimo Iavarone[78], María Reig[1]

[1]BCLC group, Liver Unit, Hospital Clinic Barcelona, IDIBAPS. CIBERehd. University of Barcelona. Spain., Barcelona, Spain, *[2]Service d'hépatologie, Hôpital Avicenne, Hôpitaux Universitaires Paris-Seine-Saint-Denis, Assistance-Publique Hôpitaux de Paris, Bobigny, France. Unité de Formation et de Recherche Santé Médecine et Biologie Humaine, Université Paris Nord, Paris, France. , France*, *[3]Abdominal Transplant & HPB Surgical Oncology, University Health Network, Toronto General Hospital, University of Toronto, Toronto, Canada., Canada*, *[4]Medical Oncology and Hematology Unit, Humanitas Cancer Center, Humanitas Clinical and Research Center – IRCCS, Rozzano (Milan), Italy. Department of Biomedical Sciences, Humanitas University, 20090 Pieve Emanuele (Milan), Italy, Italy*, *[5]Unidad de Hepatología, Clínica Universidad de Navarra, IDISNA, CIBERehd, Pamplona, Spain., Spain*, *[6]Department of General, Transplant and Liver Surgery, Medical University of Warsaw, Warsaw, Poland., Poland*, *[7]Endemic Medicine Department, Faculty of Medicine, Helwan University, Cairo, Egypt., Egypt*, *[8]3rd Medical Department, "Octavian Fodor" Institute for Gastroenterology and Hepatology, Cluj-Napoca, Romania., Romania*, *[9]AP-HP, Hôpital Beaujon, Department of Digestive Oncology, Clichy, France., France*, *[10]Hepatology, Groupement Hospitalier Lyon Nord, Lyon, France., France*, *[11]Hepato-Pancreato-Biliary Surgery and Liver Transplantation, University Medical Center Groningen, Groningen, The Netherlands., Netherlands*, *[12]Liver Transplant, King Faisal Specialist Hospital & Research Center, Riyadh, Saudi Arabia., Saudi Arabia*, *[13]Liver Unit, Newcastle Hospitals NHS Foundation Trust, Newcastle upon Tyne, UK., United Kingdom*, *[14]Department of Surgery and Cancer, Imperial College London, London, United Kingdom., United Kingdom*, *[15]Division of Transplantation, Department of Surgery, UAMS Medical Center, Winthrop P. Rockefeller Cancer Institute, Little Rock, United States., United States*, *[16]Oncology, Royal Free Hospital, London, UK., United Kingdom*, *[17]Hepatology and Gastroenterology, Aarhus University Hospital, Aarhus, Denmark., Denmark*, *[18]Department of Internal Medicine, Gastroenterology and Hepatology, University Medical Center Hamburg-Eppendorf, Hamburg, Germany., Germany*, *[19]PhD Program, Clinical and Experimental Medicine, University Hospital of Modena and Reggio Emilia, Modena, Emilia-Romagna, Italy., Italy*, *[20]Liver Unit, Hospital Universitari Vall d´Hebron, Liver Diseases Research Group, Vall d´Hebron Institut of Research (VHIR), Vall d´Hebron Barcelona Hospital Campus. Universitat Autònoma de Barcelona, Barcelona, Spain., Spain*, *[21]Liver Unit, A Cardarelli Hospital, Naples, Italy., Italy*, *[22]Department of Hepatology and Gastroenterology, Charité- University medicine Berlin, Berlin, Germany., Germany*, *[23]Department of Gastroenterology and Hepatology. Hospital Universitario Central de Asturias. IUOPA. ISPA. Universidad de Oviedo. Oviedo, Spain., Spain*, *[24]I. Dept. of Internal Medicine, University Medical Center Mainz, Mainz, Deutschland., Germany*, *[25]GI/Liver Unit, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil., Brazil*,

[26]Medical Clinic 1, Goethe University Hospital, Frankfurt, Germany., Germany, [27]Oncology, University College of London, London, United Kingdom., United Kingdom, [28]Section of Gastroenterology and Hepatology, PROMISE, University of Palermo, Palermo, Italy., Italy, [29]Department of Surgery, Geneva University Hospitals, Geneva, Switzerland., Switzerland, [30]Department of Visceral Surgery and Medicine, Inselspital, Bern University Hospital, Bern, Switzerland., Switzerland, [31]Medical Oncology, University of Modena and Reggio Emilia, Modena, Italy., Italy, [32]Department of Gastroenterology, Ogaki Municipal Hospital, Ogaki, Japan., Japan, [33]Clinic for Gastroenterology, Hepatology and Infectious Disease, University Hospital Düsseldorf, Düsseldorf, Germany., Germany, [34]Liver Unit, Department of Surgical and Medical Sciences, University of Foggia, Foggia, Italy., Italy, [35]Gastroenterology and Hepatology, H.G.U. Gregorio Marañon, CIBERehd, Madrid, Spain., Spain, [36]Gastroenterology, Hospital Federal dos Servidores do Estado, Rio de Janeiro, Brazil., Brazil, [37]Medical Oncology Unit, IRCCS Ospedale San Raffaele, Milan, Italy., Italy, [38]Digestive Disease, University Hospital La Coruña, La Coruña, Spain., Spain, [39]Gastroenterology and Hepatology, University Hospital Puerta de Hierro, Majadahonda, Spain., Spain, [40]Liver Unit, HGU Alicante. CIBERehd. Alicante, Spain., Spain, [41]Gastroenterología y Hepatología, Hospital Universitario Ramón y Cajal, Madrid, Spain., Spain, [42]Internal Medicine - Liver Unit, Clinica Universidad de Navarra, Pamplona, Spain., Spain, [43]Department of Oncology, University of Cambridge and Cambridge University Hospitals NHS Foundation Trust, Cambridge, United Kingdom., United Kingdom, [44]Clinical Oncology, Sao Paulo Clinicas Liver Cancer gourp. Insitituto do Cancer do Estado de Sao Paulo. University of Sao Paulo, Brazil., Brazil, [45]South West Liver Unit, University Hospitals Plymouth NHS Trust, Plymouth, United Kingdom., United Kingdom, [46]Department of Medical Oncology and Hematology, University Hospital Zurich and University of Zurich, Rämistrasse 100, CH-8091, Zurich, Switzerland., Switzerland, [47]Medical Oncology, Beatson West of Scotland Cancer Centre /. University of Glasgow, Glasgow, UK., United Kingdom, [48]Department of Oncology, Juravinski Cancer Centre, McMaster University, Hamilton, Canada., Canada, [49]Division of Internal Medicine, Azienda Ospedaliero-Universitaria di Bologna. Department of Medical and Surgical Sciences, University of Bologna, via Albertoni 15, Bologna, Italia., Italy, [50]Gastroenterology, Hepatology Unit, Hospital Doctor Josep Trueta, CIBERehd, Girona, Spain., Spain, [51]Servicio de Aparato Digestivo, Hospital Universitario Marqués de Valdecilla, IDIVAL, Santander, Spain., Spain, [52]SeLiver group. UGC de Enfermedades Digestivas, Instituto de Biomedicina de Sevilla- Hospital Virgen del Rocío-CIBERehd, Seville, Spain., Spain, [53]Liver Unit, Hospital Universitario Austral, Pilar, Argentina., Argentina, [54]HPB Surgery and Hepatology, Nottingham University Hospitals NHS Trust, Nottingham, UK., United Kingdom, [55]Oncology, AMO, Salvador, Brazil., Brazil, [56]Gastroenterology, Hospital do Servidor Publico Estadual de São Paulo, Sao Paulo, Brazil., Brazil, [57]Medicine, Federal University of Sergipe, Aracaju, Brazil., Brazil, [58]Hepatology, Clinionco, Porto Alegre, Brazil., Brazil, [59]Division of Oncology - Department of Experimental, Diagnostic and Specialty Medicine, S.Orsola-Malpighi Hospital, Bologna, Italy., Italy, [60]Gastroenterology and Hepatology Unit, Hospital Federal de Bonsucesso, Rio de Janeiro, Brazil., Brazil, [61]Hepatology section, Gastroenterology Department, Hospital del Mar. IMIM (Hospital del Mar Medical Research Institute). Barcelona, Spain., Spain, [62]Dpt of Clinical Medicine and Surgery, University of Naples Federico II, Napoli, Italy., Italy, [63]Gastroenterology, Pontificia Universidad Católica de Chile, Santiago, Chile., Chile, [64]Sección hepatología, Hospital Aleman, Buenos Aires, Argentina., Argentina, [65]Hepatology, Hospital Italiano, Buenos Aires, Argentina., Argentina, [66]Unitat d'Hepatologia. Servei d'Aparell Digestiu. Parc Taulí Sabadell Hospital Universitari. Institut d'Investigació i Innovació I3PT. Universitat Autònoma de Barcelona. Sabadell. Barcelona, Spain, [67]Oncología Médica, Institut Català d'Oncologia, L'Hospitalet del Llobregat, Spain., Spain, [68]Innere Medizin & Gastroenterologie, Klinikum Klagenfurt am Wörthersee, Klagenfurt am Wörthersee, Austria., Austria, [69]Gastroenterology Department, National Institute of Medical Sciences and Nutrition Salvador Zubirán, Mexico City, Mexico., Mexico, [70]Department of Medico-Surgical Sciences and Biotechnologies, Sapienza University of Rome, Polo Pontino, Latina, Italy., Italy, [71]Aparato Digestivo, Hospital Universitario Infanta Leonor, Madrid, Spain., Spain, [72]Department of Surgery, Oncology and Gastroenterology Padua University Hospital, Padua, Italy, Multivisceral Transplant Unit, Gastroenterology, Padua, Italy., Italy, [73]Department of Surgical Disciplines, Gastroenterology and Digestive Endoscopy, Umberto Parini Hospital, Aosta, Italy., Italy, [74]Gastroenterology and Hepatology, Hospital Universitario de Canarias, La Laguna, Spain., Spain, [75]Division Gastroenterology and Center for Autoimmune Liver Diseases San Gerardo Hospital University of Milano - Bicocca School of Medicine Monza, Italy., Italy, [76]Sección Hepatología, Hospital Dr. Carlos Bonorino Udaondo, Ciudad Autónoma de Buenos Aires, Argentina., Argentina, [77]Gastroenterology, Hepatology and Nutrition Unit, San Jorge General Hospital, Huesca, Spain., Spain, [78]Foundation IRCCS Ca' Granada Ospedale Maggiore Policlinico – Division of Gastroenterology and Hepatology – CRC "A.M. and A. Migliavacca" Center for Liver Disease, Milan, Italy., Italy

Email: mreig1@clinic.cat

Abstract P118YI is under embargo until 5 February 2021. This abstract has been selected to be highlighted during official EASL Press Office activities or in official EASL Press Office materials that will be made publicly available on the Summit website on 5 February 2021 on the day of their presentation at the Summit.

Industry must not issue press releases – even under embargo – covering the data contained in abstracts selected to be highlighted during official EASL Press Office activities or in official EASL Press Office materials until the individual embargo for each data set lifts.

Media must not issue coverage of the data contained in abstracts selected to be highlighted during official EASL Press Office activities or in official EASL Press Office materials until the individual embargo for each data set lifts.

Journalists, industry, investigators and/or study sponsors must abide by the embargo times set by EASL. Violation of the embargo will be taken seriously. Individuals and/or sponsors who violate EASL's embargo policy may face sanctions relating to current and future abstract submissions, presentations and visibility at EASL Congresses. The EASL Governing Board is at liberty to ban attendance and/or retract data.

P119YI Modified Page B Score Performance in Predicting HCC Risk in patients with chronic hepatitis B on antiviral therapy.

Fatma Ben Farhat[1], bochra bouchanou[1], Ennaifer Rym[1], Myriam Ayari[1], houda ben nejma[1]

[1]*Mongi Slim Hospital, Gastroenterology, Tunisia*
Email: fatmabf8@gmail.com

Background and Aims:
Hepatocellular carcinoma (HCC) is frequently associated with chronic liver disease, particularly chronic hepatitis B. Several scores are currently available to predict the incidence of HCC in patients monitored for chronic hepatitis B on antiviral therapy. The most widely used is the Page B score. The purpose of our study was to evaluate the performance of a new modified Asian Page B score (mPage B) in predicting the risk of HCC in these patients.

Method:
We enrolled all patients with chronic hepatitis B on antiviral therapy hospitalized or outpatient follow-up at Mongi slim hospital, department of hepatogastroenterology over a period of eleven years from 2006 to 2017. We excluded patients who had an inaugural HCC. The Page B score and mPage B score were calculated in all patients. The mPage B score is calculated based on age, gender, platelet count and albumin level. The risk was then staged as low: Group 1 (mPage B score<=8 and Page B score <=9), intermediate: Group 2 (mPage B score between 9 and 12 and Page B score between 10 and 17) and high: Group 3 (mPage B score >=13 and Page B score>=18). Statistical study was conducted using SPSS 22.0 software.

Results:
Sixty patients were included in our study. The mean age was 46.6 years [+/-11]. We noted a large masculine predominance. Indeed, the sex ratio was 2,3. Cirrhosis was noted in 32 patients. Twenty patients (33%) developed HCC within the first 5 years of follow-up (mean time to progression was 21 months). The mPage B and Page B scores were calculated in all patients. According to the mPage B score, 25% of patients had a low risk of HCC, 36.7% had an intermediate risk and 38% had a high risk. Of the patients who were at low risk, no one developed HCC. The prevalence of HCC for groups 2 and 3 was 22% and 65% respectively. According to the Page B score, low risk was noted in 26.6%, intermediate risk in 50% and high risk in 23.3%. The prevalence of HCC in groups 1, 2 and 3 was 12.5%, 37.5% and 50% respectively. Correlation between Page B and mPage B was strong and positive ($\sigma = 0.871$, $p < 0{,}001$). The area under the mPage B score curve was 0.88 and the area under the Page B score curve was 0.82.

Conclusion:
Our study showed that the mPage B score was better than the Page B score in predicting the risk of HCC in chronic hepatitis B especially for those at low HCC risk. Based on our results, we propose to adapt the monitoring according to the mPage B score and to reduce the number of abdominal ultrasounds for patients at low risk of HCC.

P120 Circulating microRNAs predict hepatocellular carcinoma in NUC-suppressed HBV cirrhotic patients

Marie-Laure Plissonnier[1], Francesca Guerrieri[1], Alessandro Loglio[2], Floriana Facchetti[2], Claude Caron de Fromentel[1], Pietro Lampertico[2], Mirjam Zeisel[1], Massimo Levrero[1 3 4]

[1]Cancer Research Center of Lyon (CRCL), UMR Inserm U1052-Cnrs 5286 Mixte CLB, Lyon-1 University, Lyon, France, *[2]Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Division of Gastroenterology and Hepatology - "AM and A Migliavacca" Center for Liver Diseases, Milan, Italy*, *[3]University of Rome La Sapienza, Department of Medicine SCIAC, Rome, Italy*, *[4]Hospices Civils de Lyon and Université Lyon1, Department of Hepatology, Hôpital de La Croix Rousse, Lyon, France*
Email: massimo.levrero@inserm.fr

Background and Aims:
CHB infection (257 million HBV chronic carriers, 886 000 deaths yearly) is the first cause of HCC worldwide (up to 50%). Current therapies suppress but do not eradicate the virus (persistence of HBV cccDNA and integrated sequences). HBV suppression by nucleot(s)ide analogues (NUCs) reduces but does not eliminate the risk of HCC (1.4-2.8% per year in the first 5 to 7 years of treatment). The identification of patients at higher risk of developing HCC among HBV-related cirrhosis patients treated with a supprerssive NUC therapy represents an important medical need.

Method:
We performed a retrospective analysis of > 800 circulating miRNAs using the NanoString nCounter® technology in patients selected from a prospective longitudinal cohort of 258 HBV cirrhotic patients treated with ETV or TDF. The study population comprised 12 NUC-suppressed cirrhotic patients with who developed HCC (sera collected at HCC diagnosis (*HBV HCC*) and 12 months before HCC (*HBV pre-HCC*)) and a matched case-control cohort of 12 NUC-suppressed cirrhotic patients who did not develop HCC over a comparable follow-up period (*NUC controls*).

Results:
Principal component analysis (PCA) of circulating miRNAs (631/826) detected in all patients shows that: a) *HBV pre-HCC* cir-miRNAs profiles cluster together and are separated from the *NUC control* group before HCC development; b) HBV patients before HCC development cluster tightly together; c) cir-miRNAs profiles in HBV HCC patients are well separated from the pre-HCC group. HCC development is accompanied by an upregulation of cir-miRNAs. 170/631 miRNAs were significantly differentially expressed (DE). The unsupervised hierarchical clustering analysis of the 170 DE cir-miRNAs shows in HCC patients 2 clusters of cir-miRNAs each detected in about half of patients and additional small clusters specific to the pre-HCC group. A 38 cir-miRNAs signature ($p<0,01$) predicts HCC with 5 five cir-miRNAs are significantly upregulated in the pre-HCC group. 12 cir-miRNAs are specifically detected in the *HBV pre-HCC* group.

Conclusion:
Our study identifies cir-miRNAs that are associated with a higher risk of developing HCC among HBV-related cirrhosis patients with sustained viral suppression.

P121YI Liver decompensation as late complication in HCC patients with long term response following selective internal radiation therapy.

Diederick van Doorn[1], Pim Hendriks[2], Mark Burgmans[2], Daphne Rietbergen[3], Minneke Coenraad[4], Otto van Delden[5], Roel Bennink[5], Tim Labeur[1], heinz-josef klümpen[6], Ferry Eskens[7], Adriaan Moelker[8], Erik Vegt[8], Dave SPRENGERS[9], Nahid Mostafavi[1], Jan IJzermans[10], Bart Takkenberg[1]

[1]Amsterdam UMC, University of Amsterdam, Gastroenterology and Hepatology, Amsterdam, Netherlands, *[2]Leiden University Medical Center (LUMC), Radiology, Leiden, Netherlands*, *[3]Leiden University Medical Center (LUMC), Radiology and Nuclear Medicine, Leiden, Netherlands*, *[4]Leiden University Medical Center (LUMC), Gastroenterology and Hepatology, Leiden, Netherlands*, *[5]Amsterdam UMC, University of Amsterdam, Radiology and Nuclear Medicine, Amsterdam, Netherlands*, *[6]Amsterdam UMC, University of Amsterdam, Medical Oncology, Amsterdam, Netherlands*, *[7]Erasmus MC University Medical Center, Medical Oncology, Rotterdam, Netherlands*, *[8]Erasmus MC University Medical Center, Radiology and Nuclear Medicine, Rotterdam, Netherlands*, *[9]Erasmus MC University Medical Center, Gastroenterology and Hepatology, Rotterdam, Netherlands*, *[10]Erasmus MC University Medical Center, Surgery, Rotterdam, Netherlands*
Email: d.j.vandoorn@amsterdamumc.nl

Background and Aims:
Selective internal radiation therapy (SIRT) is used for the treatment of intermediate and advanced stage hepatocellular carcinoma (HCC). The aim of this study was to assess the long-term liver-related complications of SIRT in patients with HCC who had not developed Radioembolization Induced Liver Disease (REILD).

Method:
In this multicenter retrospective study, patients who had undergone SIRT between 2011 and 2019 were included. A predefined subgroup consisting of patients who had not developed REILD was specifically explored. Biochemical, radiological and performance status data were analyzed. Primary outcomes were clinical and/or biochemical signs of liver decompensation 6 months or later after SIRT, defined as a Child Pugh (CP) score ≥B7. Secondary outcomes were overall survival (OS), tumor response and time to progression (TTP). Data were compared with a matched cohort of patients treated with sorafenib.

Results:
Eighty-five (total cohort) patients were included in this analysis, of whom 16 (14%) developed REILD. Of the remaining 69 patients, 38 (55%) developed liver decompensation CP≥B7 after 6 months or later, that were compensated CP A at baseline. Thirty patients from these patients (79%) developed clinically relevant ascites. Median OS of all patients analyzed was 18 months (95% CI 14-22). In the group of patients without REILD, median OS in patients with CP≥B7 was significantly shorter than that of patients without CP≥B7; 16 (95% CI 11-21) vs 31 months (95% CI 19-43); p=0.001. In the case-matched analysis, median OS was significantly longer in patients treated with SIRT; 17 (95% CI 12-21) vs 11 months (95% CI 8-14) for patients treated with sorafenib; p=0.027. Liver decompensation occurred significantly more often in the SIRT cohort (62% versus 22%, p<0.001). ALBI-score was an independent predictor for liver decompensation (OR 0.07; 95% CI 0.01-0.48; p=0.006) and OS (HR 2.82; 95% CI 1.43-5.60; p=0.003).

Conclusion:
Liver decompensation often develops as late complication of SIRT in HCC patients who have not developed REILD after SIRT and is associated with shorter OS. The ALBI score was predictive for the development of liver decompensation and OS and may be a valuable marker for patient selection for SIRT.

P122YI Prognostic performance of Toronto HCC risk index and the alpha-fetoprotein rate in patients with Hepatocellular carcinoma

Nouha Trad[1], mohamed ghanem[1], baha ben slimen[1], khouloud boughoula[1], sondes bizid[1], hatem ben abdalah[1], riadh bouali[1], mohamed nabil abdelli[1]

[1]military hospital of tunis, Tunisia
Email: nouha1med7@gmail.com

Background and Aims:
Hepatocellular carcinoma (HCC) is a life-threatening complication of cirrhosis. The alpha fetoprotein (AFP) conventionally used for HHC screening and the Toronto HHC risk index (Toronto index) which is a simple score recently proposed for the prediction of HHC, could have prognostic value.
Our objective was to assess the prognostic performance of the Toronto index and AFP rate at the time of diagnosis of HCC on the prediction of overall one-year survival.

Method:
This is a retrospective study including consecutive cirrhotic patients with HCC followed in our department, between January 2010 and December 2019. Overall survival was assessed by Kaplan-Meier survival analysis using log-rank. Demographic, clinical, and paraclinical data were collected.

Results:
A total of 219 cirrhotic patients were included. Sixty-one (27,8%) of them had HCC with a mean age of 64,3 ± 10,1 years and a sex ratio of 3,35. The patients were classified according to the BCLC classification: 3,2% stage (0), 33,8% stage (A), 28.9% stage (B), 19,1% stage (C) and 15 % stage (D). Toronto index was statistically associated with BCLC classification (p=0,011) but not with one-year survival (p=0,136). A significant correlation was noted between the AFP rate and the stage of the BCLC classification (p=0,04). Twenty-one patients (classified as stage BCLC 0 and A) underwent curative radiofrequency treatment (34,4%) and two patients underwent surgical resection (3,2%). Thirteen patients classified as stage B underwent chemoembolization (21,3%) and three patients treated with sorafenib (4,9%). One-year overall survival was 42,3% in patients with AFP below 14 ng/ml and 15,3% in patients with AFP above 14ng/ml (p = 0.032). The area under the ROC curve for the AFP rate in the one-year survival prediction was 0.664 [95% CI: 0.502-0.826].

Conclusion:
At a cutoff of 14 ng/ml, the AFP rate had a good prognostic value in one-year survival prediction, unlike the Toronto index. This suggested the interest of its inclusion in the prognostic scores of the HCC.

P123YI Safety and benefit of the combination of regorafenib and nivolumab in patients with hepatocellular carcinoma (HCC) in progression beyond two lines of oral chemotherapy

yasmina ben merabet[1], Maite Lewin[2], Alina Pascale[1], Catherine Guettier[3], Edoardo Poli[1], Emma Goldschmidt[4], Jean-Charles Duclos-Vallée[1], Jean-Francois Morere[4], Didier Samuel[1], Audrey COILLY[1], Olivier Rosmorduc[1]

[1]AP-HP Hôpital Paul Brousse, , Centre Hépato-Biliaire, Villejuif, France, *[2]AP-HP Hôpital Paul Brousse, , Radiology, Villejuif, France*, *[3]AP-HP Hôpital Bicêtre , Anatomopathology, Villejuif, France*, *[4]AP-HP Hôpital Paul Brousse, , Cancerology, Villejuif, France*
Email: yasmina.benmerabet@aphp.fr

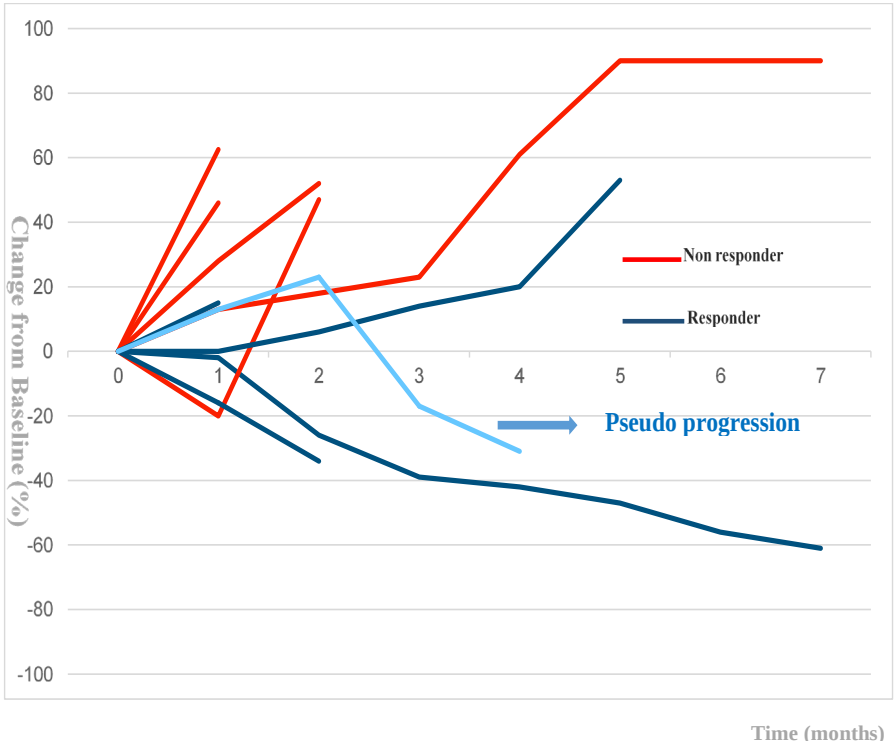
Background and Aims:
Outcome of patients with HCC in progression after more than two lines of oral chemotherapy is very poor. Anti-angiogenic could have an immunomodulatory activity in HCC by enhancing the effects of anti-PD1. In the present study, we assessed the safety and efficacy of the combination of regorafenib with nivolumab for patients with advanced HCC in this setting.

Method:
Successive patients presenting with an advanced or metastatic HCC in progression beyond a second line of oral chemotherapy, a well-preserved hepatic function (i.e. Child-Pugh score < 7) and an ECOG score ≤ 2 were included in this single-center proof of concept study. They received a combination of regorafenib (80mg daily; 3 out of 4 weeks) and nivolumab (3mg/kg intravenously, every 14 days) until progression or unacceptable toxicity. The primary endpoint was the safety based on Common Criteria Terminology for Adverse Events version 4.0 (NCI CTC). The secondary endpoints were the objective response rate (ORR), the disease control rate (DCR), and the time to response (TTR).

Results:
Ten successive patients with progressive HCC after 2 or 3 lines of oral chemotherapy were prospectively included between 01.2019 and 01.2020. Median age was 69.5 years (IQR [62; 73]). Eighty % patients were in the BCLC-C score (including 2 patients with a tumoral portal thrombosis). Median follow-up was 7 months (IQR [6; 8]) and median duration of treatment was 4 months (IQR [4 ;5]). The main adverse events (AEs) were of grade 1 - 2 and were related to the regorafenib treatment. Two patients discontinued the anti-PD1 treatment: one for a grade 3 cytolysis and one for a grade 2 rhizomelic pseudo-polyarthritis that required corticosteroids. One patient discontinued regorafenib after a grade 2 reversible heart failure. The response rate was 30% (with no complete response) with a median time to response of 2.5 months (IQR [2,1 ;3.3]) and a median duration of control disease of 5 months (IQR [3; 5]). The disease control rate was 50 % in this population. All responders started to decrease AFP only after 2 cycles of nivolumab and response (or stable disease) was assured by CT-Scan only after 4 cycles. Predictive Molecular and histological factors of response have also been studied.

Conclusion:
Regorafenib and nivolumab may offer an unexpected control disease rate of 50 % with an acceptable safety in majority of patients with HCC in progression beyond 2 lines of oral chemotherapy, not eligible for the combination atezolumab + bevacizumab.

Figure: Percentage change from baseline in sums of target lesions by RECIST 1.1 per investigator imaging review



P124YI Incidence of Hepatocellular Carcinoma in patients with Non-Alcoholic Fatty Liver Disease. A Systematic Review Meta-Analysis and Meta-Regression

Marco Sanduzzi Zamparelli[1], Lorenzo A Orci[2 3], Berta Caballol[1], Víctor Sapena[1], Nicola Colucci[2 4], Ferran Torres[5 6], Jordi Bruix[1], Christian Toso[2 3], María Reig[1]

[1]Hospital Clinic of Barcelona. IDIBAPS.CIBERehd., BCLC group. Liver Unit. , *[2]Geneva University Hospitals and Faculty of Medicine, Abdominal and Transplantation Surgery, Department of Surgery*, *[3]Geneva University Hospitals and Faculty of Medicine, Hepato-pancreato-biliary Center*, *[4]University of Pavia*, *[5]Hospital Clinic of Barcelona. IDIBAPS.CIBERehd., Medical Statistics core facility*, *[6]Universitat Autònoma de Barcelona, Biostatistics Unit, Faculty of Medicine*
Email: mreig1@clinic.cat

Background and Aims:
Nonalcoholic fatty liver disease (NAFLD) may been identified as a risk factor for hepatocellular carcinoma (HCC), but the extent of this association still needs to be properly addressed. The aim of the present study is to estimate by a meta-analysis the pooled-incidence rates of HCC across the disease spectrum of NAFLD.

Method:
In this systematic review, we searched Web of Science, Embase, Pubmed, and the Cochrane library from January 1st, 1950 through July 30th, 2020. We included studies reporting on HCC incidence in patients with NAFLD. The main outcomes were pooled HCC incidences in patients with NAFLD at distinct severity stages. Sensitivity analyses and meta-regression analyses were carried out to address heterogeneity. The protocol for this review was registered in Prospero (CRD42018092861).

Results:
We identified 10,263 studies and 18 of those involving 470,404 patients were finally included. Heterogeneity in cirrhotic patients was of 81% and of 98% in non-cirrhotic. In patients with NAFLD without established cirrhosis, HCC incidence was 0.03 per 100 person-years (PYs) (95% confidence interval 0.01-0.07, I^2=98%). When considering studies that only included patients with cirrhosis the rate was of 3.78 per 100PYs (2.47 - 5.78, I^2=81%). Among the latter patients, those undergoing regular HCC screening displayed an incidence of 4.62 per 100PYs (2.77-7.72, I^2= 77%). Sensitivity analyses and meta-regression analyses did not significantly improve heterogeneity.

Conclusion:
The high heterogeneity of the studies hampers robust conclusions. However, patients with NAFLD-related cirrhosis have a risk of developing HCC similar to that reported for patients with cirrhosis from other aetiologies. Data documenting the risk in patients with NASH or steatosis are limited, but HCC incidence in these populations may lie below thresholds used to recommend HCC screening. Considering the high heterogeneity and the absence of informative screening programs in non-cirrhotic patients, well-designed prospective studies are needed in such subgroup of patients to properly assess the risk.

Figure:

P125YI Performance of ten non-invasive liver function tests in predicting one-year mortality in patients with hepatocellular carcinoma

Nouha Trad[1], mohamed ghanem[1], baha ben slimen[1], khouloud boughoula[1], sondes bizid[1], hatem ben abdalah[1], riadh bouali[1], mohamed nabil abdelli[1]

[1]military hospital of tunis, gastroenterology, Tunisia
Email: nouha1med7@gmail.com

Background and Aims:
Hepatocellular carcinoma (HCC) remains nowadays cancer with a poor prognosis and one of the main causes of mortality in cirrhotic patients. Its management is often based on BCLC staging , however, this classification has gray areas, especially in the intermediate stage.

Our objective was to evaluate the performance of ten non-invasive liver function tests in predicting one-year mortality in cirrhotic patients with HCC.

Method:
We performed a retrospective analysis of data from consecutive cirrhotic patients with HCC recruited from January 2010 to December 2019. In addition to the CHILD score, the following scores were calculated when HHC was diagnosed: MELD, albumin-bilirubin grade(ALBI), platelet-albumin-bilirubin grade(PALBI), fibrosis-index based on 4factors(FIB-4), aspartate-aminotransferase-to-platelet ratio(APRI), Lok index, cirrhosis discriminant index(CDS), King's score, Goteborg-University Cirrhosis Index(GUCI), and aspartate-aminotransferase to alanine-aminotransferase ratio(AAR).

Results:
In total, 219 patients were included. Sixty-one patients had HHC (27,85%).The mean age was 64,38 ± 10,17 and the sex-ratio was 3,35. Viral origin (80,32%) was the predominant etiologies of cirrhosis. The one-year mortality rate was 70.5%. The following scores have been statistically associated with one-year mortality: FIB-4 (p=0,012); APRI (p=0,038) and AAR (p=0,037). FIB-4 had the best area under the curve ROC (AUROC) in predicting one-year mortality (AUROC=0,703 [95%CI: 0,569-0,837]) followed by AAR (AUROC=0,682 [95%CI: 0,538-0,825]) and APRI (AUROC=0,657 [95%CI: 0,513-0,801]. At the cut-off of 5,26, FIB-4 had a sensibility and specificity of 80,1% and 56,4% respectively in the prediction of one-year mortality. At the cut-off of 1,49, AAR had a sensibility and specificity of 72,5% and 64,1% respectively. At the cut-off of 1,48, APRI had a sensibility and specificity of 70,1% and 45,6% respectively in the prediction of one-year mortality.

Conclusion:
In our study, FIB-4, AAR and APRI had a good prognostic value in predicting one-year mortality. In addition to BCLC staging, these simple, non-invasive scores could be useful in current practice to classify patients with HHC and guide management.

P126 Impact of Hepatocellular Carcinoma screening in patients with Non-Achoholic Fatty Liver Disease and survival analyzis: a Brazilian Cross-Section Study

Regiane Saraiva de Souza Melo Alencar[1 2], Aline Lopes Chagas[2 3], Leonardo Gomes da Fonseca[2 4], Claudia Maccali[2 3], Cláudia Megumi Tani[2 3], Lisa Rodrigues da Cunha Saud[2 3], Mariana Pinheiro Xerfan[2 3], Claudia Oliveira[2 5], Venâncio Avancini Ferreira Alves[2 6], Luiz Augusto Carneiro D'Albuquerque[2 7], Paulo Herman[2 7], Flair Jose Carrilho[2 8]

[1]Insituto do Câncer do Estado de São Paulo, São Paulo, Hospital das Clínicas , University of São Paulo School of Medicine, São Paulo, Brazil, Service of Hepatology, Sao Paulo, Brazil, *[2]São Paulo Clínicas Liver Cancer Group, University of São Paulo School of Medicine, São Paulo, Brazil*, *[1]Insituto do Câncer do Estado de São Paulo, São Paulo, Hospital das Clínicas, University of São Paulo School of Medicine, São Paulo, Brazil, Service of Hepatology, Sao Paulo, Brazil*, *[4]Insituto do Câncer do Estado de São Paulo, São Paulo, Hospital das Clínicas , University of São Paulo School of Medicine, São Paulo, Brazil, Service of Oncology, Sao Paulo, Brazil*, *[5]Division of Clinical Gastroenterology and Hepatology, Hospital das Clínicas, Department of Gastroenterology, University of São Paulo School of Medicine, São Paulo, Brazil., Gastroenterology, São Paulo, Brazil*, *[6]Department of Pathology, University of São Paulo School of Medicine, São Paulo, Brazil., Pathology, São Paulo, Brazil*, *[7]Department of Gastroenterology, University of São Paulo School of Medicine, São Paulo, Brazil., Gastroenterology, São Paulo, Brazil*, *[5]Division of Clinical Gastroenterology and Hepatology, Hospital das Clínicas, Department of Gastroenterology, University of São Paulo School of Medicine, São Paulo, Brazil., Gastroenterology, São Paulo, Brazil*
Email: rssmalencar@gmail.com

Background/Aim:
The natural history of Non-alcoholic fatty liver disease (NAFLD) has been a subject of interest during the last decade. However, the evaluation of hepatocellular carcinoma (HCC) development, the role of screening, treatment and outcomes in this group is still controversial. The aim of the present study was to evaluate the clinical features, HCC screening, treatment modalities and overall survival in NAFLD-HCC related patients.

Methods: This was a cross-sectional study at the Instituto do Câncer do Estado de São Paulo,University of Sao Paulo School of Medicine with approval of the local research ethics committee. Patients with HCC diagnostic, from May 2010 to May 2019, with NAFLD or cryptogenic disease were included. Clinical characteristics, comorbidities and therapeutic modalities were collected. Survival analyzis were calculated using Kaplan-Meier.

Results: A total of 131 patients were included, 60.3% male, 80.9% caucasian, with mean age of 66.1±9.7 years and BMI 29. Cirrhosis was present in 90.1% of patients, 51.9% were Child-Pugh A and 46.5% presented liver decompensation before HCC diagnosis, with ascites being the most common complication (80.3%). Risk factors for NAFLD were present in 94.6% of the patients: hypertension 77.4%; Type II Diabetes 68.5%; Obesity 40.3%; Dyslipidemia 40.3%; Overweight 39.5%; Glucose intolerance 7.2%. Tobacco consumption was present in 45% of patients. Thirty percent of the cases had cancer related symptoms at HCC diagnosis, with abdominal pain in 75%. Only 29% of patients were in HCC screening program before diagnosis, with ultrasound and alpha-fetoprotein every 6 months. In the majority of patients HCC diagnosis was made by imaging (85.5%), being computed tomography the main method (87%). Most patients presented 1 nodule (57.2%) at diagnosis, with the largest nodule diameter average of 54.6mm. Mean alpha-fetoprotein was 15,118ng/ml. Regarding tumor staging at diagnosis, 40.4% were within Milan Criteria and according the Barcelona Staging System (BCLC), the patients were distributed: 0 – 5.3%; A – 42.7%; B - 25.2%; C - 16% and D – 10.7%. HCC treatment was performed in 84.7% of patients. Regarding the first treatment, the most common modalities were: Transarterial chemoembolization in 47.7%; Radiofrequency ablation in 17.1%; Sorafenib in 16.2% and surgical resection in 9.9%. Of these, 40.5% of patients underwent more than one treatment during the follow up. Liver transplantation was performed in 16.2% of cases. Median overall survival (OS) was 26.2 months (95% CI: 19.3-45.7 months). Concerning HCC screening, patients that were screened for HCC presented better survival (48.3 x 21.0 months, p=0.03).

Conclusions:
In the preliminary results of our study, only one third of patients with NAFLD and HCC were performing HCC screening and were these patients had better survival. This highlights the importance of screening NAFLD patients for HCC.

P127YI Whole-transcriptome profiling of biopsies from unresectable intrahepatic cholangiocarcinoma (iCCA) reveals a prognostic signature with treatment implications.

Massimiliano Salati[1], Colm O'Rourke[2], Colin Rae[3], Guido Carpino[4], Maria Giuseppina Prete[3 5], Luca Reggiani bonetti[6], Francesco Amato[3], Rosie Rosie Upstill-Goddard[3], Stefano Cascinu[7], Andrea Spallanzani[8], Fabio Gelsomino[8], Francesco Caputo[8], Gabriele Luppi[8], Massimo Dominici[8], Jesper B. Andersen[9], Chiara Braconi[3]

[1]Modena Cancer Centre, University Hospital of Modena, Modena, Italy, Department of Oncology, , Modena, Italy, *[2]Biotech Research and Innovation Centre (BRIC), Department of Health and Medical Sciences, University of Copenhagen, Ole Maaløes Vej 5, 2200, Copenhagen N, Denmark., Denmark*, *[3]Institute of Cancer Sciences, University of Glasgow.*, *[4]Department of Movement, Human and Health Sciences, Division of Health Sciences, University of Rome "Foro Italico,", Rome, Italy*, *[5]Humanitas Clinical and Research Center – IRCCS, Humanitas Cancer Center, Department of Medical Oncology and Hematology, via Manzoni 56, 20089 Rozzano, Milan, Italy*, *[6]Department of Diagnostic, Clinic and Public Health Medicine, University of Modena and Reggio Emilia, Modena, Italy*, *[7]Department of Medical Oncology, IRCCS San Raffaele Hospital, Milan, Italy.*, *[8]Division of Oncology, Department of Oncology and Hematology, University Hospital of Modena, Modena, Italy.*, *[9]Biotech Research and Innovation Centre (BRIC), Department of Health and Medical Sciences, University of Copenhagen, Ole Maaløes Vej 5, 2200, Copenhagen N, Denmark.*
Email:

Background and Aims:
Transcriptome profiling provides useful insights for therapeutics. However, only data for resected disease are available to date, limiting the clinical implications for unresectable advanced iCCA (iCCA). Here, we applied transcriptomics to derive a prognostic signature in advanced iCCA, using a clinically-annotated cohort of Rapid Progressor (RP) and Long Survivor (LS) patients undergoing first-line chemotherapy.

Method:
RP and LS were defined according to overall survival (OS): RP ≤ 6 months, LS ≥24 months. Transcriptome profiling was performed with TempO-Seq targeted-sequencing on pretreatment liver biopsies. An RP-LS signature was developed from differentially expressed genes (DEGs; fold-change≥2, p<0.05). This signature was individually evaluated in four resected cohorts, including survival analyses (Kaplan-Meier, Cox proportional hazards) and correlation analyses (signaling pathways, hepatobiliary subtypes, immune functionality). Potential responsiveness of samples to checkpoint inhibitors was estimated using TIDE.

Results:
In total, 13 biopsied iCCA (7 RP, 6 LS) were included. Each group was well-balanced regarding clinico-pathologic features (age, sex, ECOG p=0.26; disease status p=0.99). Median OS was 3 versus 24 months in RP and LS, respectively. Overall, 504 genes were found to be differentially expressed between the groups. These included 310 genes categorized as LS-high (significantly over-represented in Hedgehog signaling and mismatch repair), and 194 genes categorized as RP-high (over-represented in NOTCH, IL-17, TNF signaling pathways) (p<0.05). RP cases showed higher inflammatory pathological scores by H&E assessment, and higher TIDE scores (P<0.0001), suggesting a likelihood of response to immune checkpoint inhibitors. Application of the RP-LS signature to four resected iCCA cohorts (n=403 cases) showed a higher RP-LS score consistently associated with shorter OS in Kaplan-Meier analyses. The signature was found to be a stage-independent predictor of OS in multivariate Cox analyses. The RP-LS signature reproducibly correlated with oncogenic pathway-activity (VEGF, P53), metabolic reprogramming (including glycolysis), previously-reported molecular subtypes of hepatobiliary cancers, depleted M2 tumor-associated macrophages and microsatellite instability (MSI) score.

Conclusion:
We showed feasibility of whole-transcriptome profiling of biopsies from advanced iCCA and identified a gene-signature with prognostic implications. The RP-LS signature was reiterated in tumours from resected patients classified as resectable with predominant good prognosis. Based on our signature, it is possible to speculate that patients rapidly progressing on chemotherapy may benefit from immunotherapy checkpoint or VEGF inhibitors.

Figure:

P128 Complete response after biological downstaging in patients with hepatocellular carcinoma: XXL-like prioritization for liver transplantation or “waiting and see” strategy?

Alessandro Vitale[1], Paolo Angeli[1], Patrizia Burra[1], Umberto Cillo[1]

[1]*University of Padua, Padova, Italy*
Email: alessandro.vitale@unipd.it

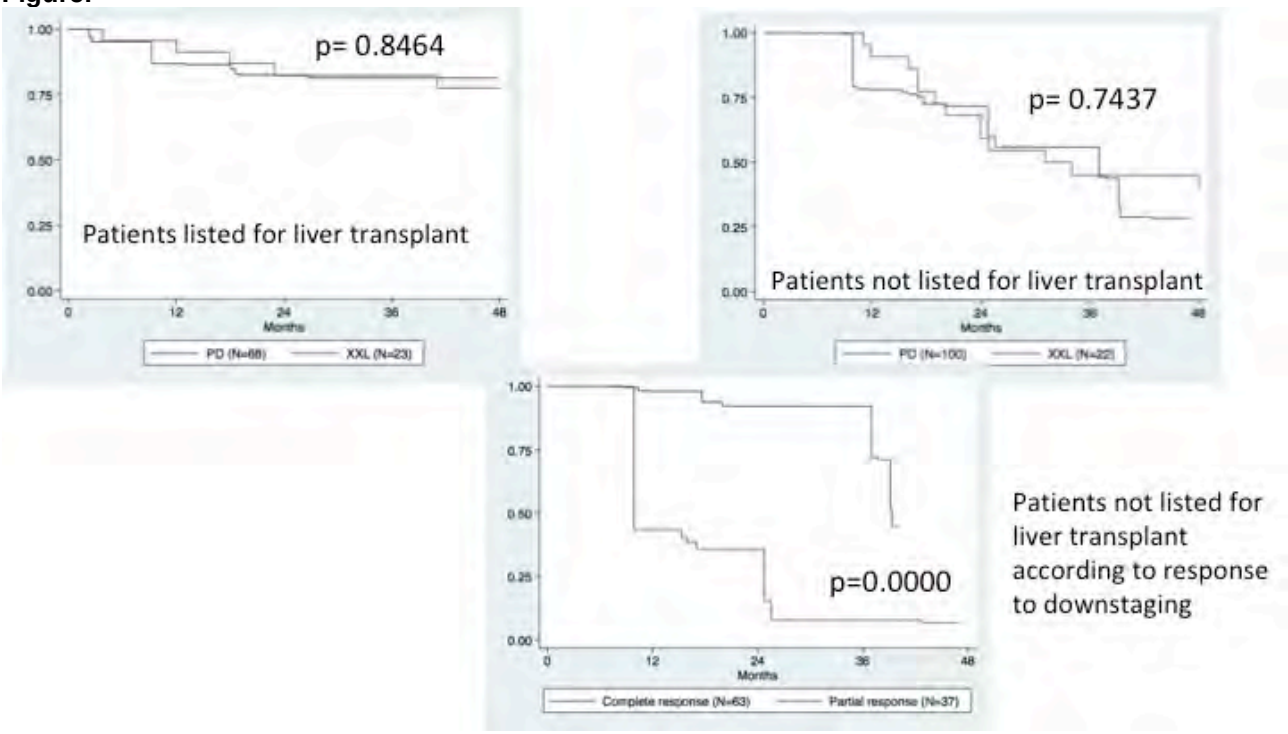
Background and Aims:
Although some methodological and ethical issues, the recently published XXL trial represents the first prospective validation of “biological downstaging” in liver transplantation for hepatocellular carcinoma (i.e. selecting tumors with a good biology irrespective of morphological criteria). The aim of this study is to compare our downstaging protocol with the XXL protocol in terms of downstaging failure rates and patient outcome.

Method:
A total of 191 patients undergoing surgical downstaging and potentially eligible for transplantation from 2012 to 2018 at our Center, were selected according to the XXL trial enrolment criteria. Differently from the XXL trial we used an aggressive surgical downstaging protocol, and patients with a complete response to downstaging did not receive any prioritization to transplant. Downstaging failure was defined as progressive disease or post treatment mortality. The statistical method "matching-adjusted indirect comparison” was used to match the study group to the XXL population and to compare the proportion of downstaging failures. The software Engauge digitizer was used to allow a statistical comparison between Kaplan Meier survival curves.

Results:
Downstaging failure rate was significantly lower in our cohort than in the XXL trial (12% vs. 39%). Patients with partial response to downstaging had a much greater probability of being included in the waiting list than patients with a complete response (OR 20.4, 95% CI 6.9-69.9, p=0.0001). Although patients with complete response were not prioritized to transplant, the survival curves of our cohort overlapped with that of the XXL protocol (p> 0.05). Survival curves of non-transplant candidates with a complete response to downstaging were similar to that of transplanted patients (p> 0.05).

Conclusion:
Our study represents a validation of the current Italian policy of denying any prioritization to patients with complete response to downstaging. Such a policy seems to spare organs without worsening patient outcome.

Figure:


p= 0.8464
Patients listed for liver transplant
1.00
0.75
0.50
0.25
0.00
0
12
24
36
48
Months
PD (N=68)
XXL (N=23)
p= 0.7437
Patients not listed for liver transplant
PD (N=100)
XXL (N=22)
p=0.0000
Complete response (N=63)
Partial response (N=37)
Patients not listed for liver transplant according to response to downstaging

P129YI PSMA PET/CT imaging has equivalent performance to MRI for characterising hepatocellular carcinoma

Veronica Wong[1], Joshua Yip[1], Vincenzo Fragomeli[1], Martin Weltman[1], Han Loh[1], Robert Mansberg[1]

[1]*The University of Sydney, School of Medicine and Health Sciences, Australia*
Email: veronica.wong@health.nsw.gov.au

Background and Aims:
Prostate specific membrane antigen (PSMA) is known to be expressed by hepatocellular carcinoma (HCC). This is an exploratory study aimed at comparing Ga-68 HBED-CC PSMA positron emission tomography/computed tomography (PET/CT) to conventional imaging with magnetic resonance imaging (MRI) and triple phase CT in patients with a history of suspected or treated HCC.

Method:
In this small cohort study, patients were prospectively recruited from a tertiary hospital outpatient clinic with suspected or treated HCC. In addition to routine surveillance as recommended by the multidisciplinary liver cancer team, a PSMA PET/CT was performed. Imaging and clinical characteristics were compared over a follow-up period of up to 12 months.

Results:
49 individual lesions were assessed in 19 patients, of which 25 had previously been treated with either resection, microwave ablation, radiotherapy or chemoembolisation. Median follow-up period was 204 days. PSMA PET/CT had similar efficacy to MRI for the detection of HCC, with a sensitivity of 90% and a specificity of 70% and sensitivity of 87% and a specificity of 73% for PSMA PET/CT and MRI respectively. PSMA PET/CT had a higher negative predictive value of 90%. PSMA PET/CT also detected 4 lesions which were not found on MRI and CT, of which one of these was biopsy-proven to be HCC, leading to significant earlier management in 4/19 patients.

Conclusion:
PSMA PET/CT has equivalent performance to MRI for HCC detection and has potential for evaluating equivocal lesions.

Figure:

Comparison of screening modalities				
(n)	**PSMA (49)**	**MRI (30)**	**CT (49)**	**AFP (19)**
True negative	19	11	27	7
True positive	20	13	7	5
False negative	2	2	15	6
False positive	8	4	0	1
Sensitivity	90	87	32	45
Specificity	70	73	100	88
PPV	71	76	100	83
NPV	90	85	64	54

P130YI Stellate cell activation in liver cancer treatment

Xiaowen Liang[1 2], Qi Ruan[1 2], Haolu Wang[1 2], Leslie Burke[2], Lu Cao[2], Kim Bridle[2], Michael Roberts[1], Darrell Crawford[2]

[1]The University of Queensland Diamantina Institute, The University of Queensland, Faculty of Medicine, Queensland, Australia, *[2]Gallipoli Medical Research Institute, Greenslopes Private Hospital, Greenslopes , Australia*
Email: d.crawford@uq.edu.au

Background and Aims:
Liver cancer patients are candidates for chemotherapy such as transarterial chemoembolisation for hepatocellular carcinoma (HCC) and systemic chemotherapy for cholangiocarcinoma (CC). Chemotherapeutics may activate hepatic stellate cells (HSCs), the key components of the liver cancer microenvironment with important roles in tumour growth, chemoresistance and metastasis. However, the molecular mechanisms underlying the chemotherapy-induced HSC activation have not been well determined.

Method:
We first investigated the chemotherapy-induced HSC activation in liver cancer using three *in vitro* models. The human HSC line, LX-2, was co-cultured with cisplatin-treated human HCC cell lines (Huh7 or PLC/PRF/5) or human CC cell line (Hucct1) in a transwell system or 3D mixed-cell spheroids. Then LX-2 cells were cultured in conditioned media (CM) from cisplatin/doxorubicin-treated Huh7 cells. The activation markers of HSCs, including platelet-derived growth factor receptor β (PDGFRβ) and α-smooth muscle actin (α-SMA) were assessed. The human cytokine array was used to identify cytokines released from cancer cells after chemotherapy. Reactive oxygen species (ROS) were assessed in tumor cells and HSCs in CM model. HSC activation was assessed in orthotopic mouse HCC model and human biopsy samples before and after chemotherapy. Finally, HSC activation markers and correlation to the prognosis were investigated in human HCC samples using TCGA data.

Results:
We confirmed that HSCs can be activated through the paracrine factors of chemotherapy stimulated liver cancer cells, as evidenced by significantly increased expressions of α-SMA, PDGFRβ (Fig. 1A&B) and collagen type I alpha 1 chain (COL1A1). CXCL5 was obviously increased in doxorubicin treated Huh7 CM but no significant changes of cytokine or chemokine in cisplatin treated huh7 CM. Cancer cells generated the high level of ROS after chemotherapy and induced PDGFRβ activation in HSCs. The HSC activation further increased the level of ROS in cancer cells. The expression of PDGFRβ was increased in mouse (Fig. 1C&D) and human HCC after chemotherapy. Our results were validated using TCGA database, where gene expression of HSC activation markers increased in advanced HCC patients after chemotherapy.

Conclusion:
This study identifies, for the first time, that ROS/PDGFRβ loop contributes to platinum-drugs induced HSC activation via amplification of oxidative stress and downstream signaling. Doxorubicin may activate HSCs through stimulating cancer cells to release chemokines (CXCL5). Understanding this mechanism could provide insights into the development of potential therapeutic targets for targeting activated HSCs to improve the efficacy of liver cancer chemotherapy.

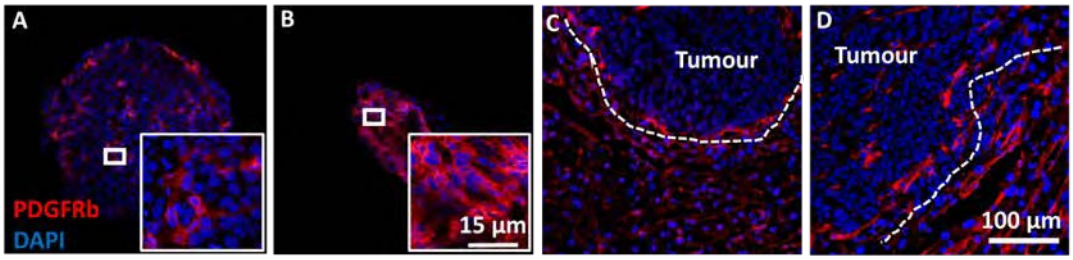


Fig. 1 PDGFRβ expression. A&B. Untreated or cisplatin-treated mixed-cell spheroids; C&D. Untreated or cisplatin-treated mouse tumours.

P131 Efficacy and safety of pemigatinib in European patients with previously treated locally advanced or metastatic cholangiocarcinoma: a FIGHT-202 subgroup analysis

Antoine Hollebecque[1], Davide Melisi[2], Arndt Vogel[3], Mairéad Geraldine McNamara[4], Teresa Mercade Macarulla[5], Eric Van Cutsem[6], Huiling Zhen[7], Luis Feliz[8], Ghassan Abou-Alfa[9]

[1]Gustave Roussy, Department of Adult Medicine, Villejuif, France, *[2]Università degli studi di Verona, Digestive Molecular Clinical Oncology Research Unit, Verona, Italy*, *[3]Hannover Medical School, Department of Gastroenterology, Hepatology and Endocrinology, Hannover, Germany*, *[4]University of Manchester, The Christie NHS Foundation Trust, Manchester, United Kingdom*, *[5]Vall d'Hebron University Hospital, Vall d'Hebron Institute of Oncology, Barcelona, Spain*, *[6]University Hospitals Leuven and KU Leuven, Department of Digestive Oncology, Leuven, Belgium*, *[7]Incyte Corporation, Wilmington, United States*, *[8]Incyte Biosciences International Sàrl, Morges, Switzerland*, *[9]Memorial Sloan Kettering Cancer Center , Weill Medical College at Cornell University, New York, United States*
Email: antoine.hollebecque@gustaveroussy.fr

Background and Aims:
FGF/*FGFR* genomic alterations (GAs) are present in 10-16% of patients (pts) with intrahepatic cholangiocarcinoma (CCA). The potent and selective oral fibroblast growth factor receptor (FGFR) 1-3 inhibitor pemigatinib is approved in the US for pts with previously treated advanced CCA with *FGFR2* fusions/rearrangement (RE), based on the phase 2 FIGHT-202 study (NCT02924376) showing an objective response rate (ORR) of 35.5%, a disease control rate (DCR) of 82%, and median duration of response (mDOR), progression-free survival (mPFS), and estimated overall survival (mOS, immature) of 7.5, 6.9, and 21.1 months (mo), respectively [Abou-Alfa GK, et al. *Lancet Oncol.* 2020;21(5):671-684]. We conducted a post-hoc subgroup analysis in pts from Europe (EU: Belgium, France, Germany, Italy, Spain, UK).

Method:
Pts with advanced disease and progression after >=1 prior treatment were included and, based on confirmed *FGF/FGFR* status, assigned to cohort A (*FGFR2* RE), B (other *FGF/FGFR* GA), or C (no *FGF/FGFR* GA). Pts received pemigatinib 13.5 mg QD (21-day cycle; 2 weeks on, 1 week off) until disease progression or unacceptable toxicity. The primary endpoint was centrally confirmed ORR in cohort A.

Results:
The data cutoff (Mar 22, 2019) used for this analysis was identical to that of the published primary analysis. Of 147 pts enrolled, 35 (24%) were from EU (10 France, 8 Italy, 17 other countries); of those, 32, 3, and 0 were enrolled in cohorts A, B, and C, respectively. Median (range) age was 59 (34-77) years; 66% were female; 46% had >=2 prior therapies. At data cut-off, 25 pts had discontinued treatment, (21 for disease progression); 10 pts (all cohort A) were continuing treatment. In cohort A, ORR (95% confidence interval) was 40.6% (23.7-59.4) with 1 complete response (**Figure**); mDOR was 7.5 mo (5.5-7.5), DCR was 87.5% (71.0-96.5), mPFS was 6.9 mo (4.8-9.1), and mOS was 14.7 mo (11.7-not reached). In cohort B, 2 of 3 pts had stable disease (PFS, 2.1 and 4.0 mo). The most common AEs were diarrhoea (80%; grade 3/4, 6%), alopecia (66%; 0%), and hyperphosphataemia (60%; 0%); 14% of pts had hypophosphataemia, 60% had nail toxicities, and 1 pt (3%) had retinal detachment. AEs led to dose reductions and interruptions in 20% and 57% of pts, respectively.

Conclusion:
Efficacy and tolerability of pemigatinib in EU pts were similar to the published findings in the total study population.

Figure: Best percentage change in target lesion size (cohort A)