Disclosure Information

I hereby declare that I have had business or personal interests in the following industrial enterprises since 1 September 2016:

**Name of the enterprise / Nature of the interest**

<table>
<thead>
<tr>
<th>Enterprise</th>
<th>Interest</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nothing to declare</td>
<td></td>
</tr>
</tbody>
</table>
What’s hot in the liver and biliary tract?

Carolin Lackner
Institute of Pathology
Medical University of Graz
Austria
Overview

• Fibrosis (stage) is a major prognostic factor in alcoholic and non-alcoholic fatty liver diseases

• Neoangiogenesis-related genes are markers of fast-growing hepatocellular carcinoma and poor survival

• Novel insights in the role of IgG4 in the pathogenesis of IgG4-related diseases
Clinical presentation of ALD

Decompensated
- Severely ill
- Jaundiced
- Cirrhotic
- Low 30d survival rate

Early/compensated
- No/mild symptoms
- High 30d survival rate

Predictors of long term-prognosis?
Utility of morphological markers for prediction of long-term outcome in ALD

• Early detection of patients at risk of progressive disease, prevention of cirrhosis, decompensation, eventually liver transplantation

• Diagnosis of cirrhosis and selection of patients for HCC surveillance
Histological parameters and alcohol abstinence determine long-term prognosis in patients with alcoholic liver disease

Carolin Lackner¹*, Walter Spindelboeck², Johannes Haybaeck¹, Philipp Douschan², Florian Rainer², Luigi Terracciano³, Josef Haas⁴, Andrea Berghold⁵, Ramon Bataller⁶, Rudolf E. Stauber²

Journal of Hepatology 2017 vol. 66 | 610–618
Predictors of long-term mortality in ALD

Early/compensated (n=60)  vs  Decompensated (n=132)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Early/Compensated</th>
<th>Decompensated</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pericell fibrosis</td>
<td>Present</td>
<td>Female</td>
<td>0.002</td>
</tr>
<tr>
<td>Sex</td>
<td>0.019</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Bilirubin</td>
<td>0.002</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>INR</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Variables entered:
- Pericellular fibrosis, canalicular cholestasis, ductular cholestasis, ASH, abstinence, sex, bilirubin, INR, albumin, leukocyte count, platelet count, sodium

52% of cases
NAFLD types and disease progression

NAFL 100%

NASH 2-20%

Increased Mortality
Cardiovascular disease
Cancer
Chronic liver disease

Fibrosis progression in NASH: 1 stage/7 y

Morphological key features of NASH & fibrosis stages in NAFLD

CRN stage 0

CRN stage 1

CRN stage 3

CRN stage 4

Burt A, Semin Liver Dis 2015
<table>
<thead>
<tr>
<th>Scoring systems for assessment of NAFLD grade and stage</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CRN NAFLD activity score (NAS)</strong></td>
</tr>
<tr>
<td>NASH diagnosis, NAS score &amp; stage implicated in primary endpoints in clinical trials</td>
</tr>
<tr>
<td>Caveat: Low prognostic relevance of NAS</td>
</tr>
<tr>
<td><strong>Steatosis, Activity, Fibrosis (SAF)</strong></td>
</tr>
<tr>
<td>Inclusion of fibrosis in the definition of disease severity may increase prognostic utility</td>
</tr>
<tr>
<td>Secondary endpoint in clinical trials</td>
</tr>
</tbody>
</table>

*S: Steatosis; I: Lobular Inflammation; B: Ballooning; F: Fibrosis*
SAF score and mortality in NAFLD after up to 41 years of follow-up

Hannes Hagström, Patrik Nasr, Mattias Ekstedt, Stergios Kechagias, Per Stål, Pierre Bedossa & Rolf Hultcrantz

To cite this article: Hannes Hagström, Patrik Nasr, Mattias Ekstedt, Stergios Kechagias, Per Stål, Pierre Bedossa & Rolf Hultcrantz (2017) SAF score and mortality in NAFLD after up to 41 years of follow-up, Scandinavian Journal of Gastroenterology, 52:1, 87-91, DOI: 10.1080/00365521.2016.1230779
Utility of grading and staging systems for prediction of outcome

Hagström H, Scand J Gastroenterol

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Statistical model</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Univariate</td>
<td>Multivariate</td>
<td>Multivariate Adjusted for stage</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>p-value</td>
<td></td>
</tr>
<tr>
<td>Disease severity by SAF</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moderate vs. mild</td>
<td>0.1</td>
<td>0.64</td>
<td>0.89</td>
<td></td>
</tr>
<tr>
<td>Severe vs. Mild</td>
<td>&lt;0.001</td>
<td>0.017</td>
<td>0.18</td>
<td></td>
</tr>
<tr>
<td>FLIP diagnostic algorithm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NASH vs. NAFL</td>
<td>0.07</td>
<td>0.67</td>
<td>0.72</td>
<td></td>
</tr>
<tr>
<td>Fibrosis stage 3-4 vs. 0-2</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAS 0-8</td>
<td>0.036</td>
<td>0.85</td>
<td>0.3</td>
<td></td>
</tr>
<tr>
<td>SAF activity score 0-4</td>
<td>&lt;0.001</td>
<td>0.036</td>
<td>0.32</td>
<td></td>
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</table>
Stage as the main predictor of outcome

<table>
<thead>
<tr>
<th>Study</th>
<th>Type</th>
<th>Design</th>
<th>NAFD Type</th>
<th>Number</th>
<th>Follow-up Years</th>
</tr>
</thead>
<tbody>
<tr>
<td>Younossi ZM</td>
<td>R</td>
<td>NAFL 78; NASH 132</td>
<td>Median: ~12</td>
<td>CRN 4</td>
<td></td>
</tr>
<tr>
<td>Ekstedt M</td>
<td>P</td>
<td>NAFLD 229</td>
<td>Mean: ~26</td>
<td>CRN ≥3</td>
<td></td>
</tr>
<tr>
<td>Angulo P</td>
<td>R</td>
<td>NAFL 335; NASH 284</td>
<td>Median: ~13</td>
<td>CRN ≥1</td>
<td></td>
</tr>
<tr>
<td>Dulai PS</td>
<td>M</td>
<td>NAFLD 1495</td>
<td>Median: ~17</td>
<td>CRN ≥1</td>
<td></td>
</tr>
</tbody>
</table>

Liver-related mortality risk increases exponentially with increase in fibrosis stage

R...Retrospective; P...Prospective; M...Meta-analysis
BCLC Staging System and Treatment Strategy
EASL Clinical Practice Guideline on the Management of HCC  J Hepatol 2012

Prognostic markers reflecting tumor biology for improved patient selection for OLT?

Milan Criteria:
1 Tumor ≤ 5cm
Up to 3 tumors, none >3cm

HCC

Stage 0
PST 0, Child-Pugh A

Stage A-C
PST 0-2, Child-Pugh A-B

Stage D
PST >2, Child-Pugh C*

Terminal stage (D)

Very early stage (0)
Single <2 cm, Carcinoma in situ

Early stage (A)
Single or 3 nodules ≤3 cm, PS 0

Rate of recurrence 10-20%

Resection
Liver transplantation
TACE
Sorafenib
Best supportive care

Curative treatments (40-40%)
Median OS >60 mo, 5-y survival: 40-70%

Target: 20% OS: 20 mo (45-14)
Target: 40% OS: 11 mo (5-14)
Target: 10% OS: <3 mo
HCC tissue markers of adverse prognosis

• Low differentiation grade, venous invasion, positive resection margin
• High microvessel density; high expression of VEGF, Hif, MMPs, cyclins & CDKs, Ki67; loss of p16,18,27,57 expression;
• Angiogenesis-related mRNA signatures
• MiRNA-signatures
  Down regulation of miR-99a,-124,-139,-145, -199b
  Up-regulation of miR-222,-135a,-155,-182,-10b,-17-5p,-221,-21
• Proliferation subclass
  Enrichment in signaling pathways related to proliferation & tumor recurrence or stem cell features
• High frequency of LOH

Sia D, Gastroenterology 2017
Berretta M, Oncotarget 2017
Nault J, Gastroenterology 2013
Obstacles to clinical implementation

Experimentally derived marker/algorithm

Validation by independent group(s)/collectives
Consensus finding

Transfer to clinical applicability
- Minimize complexity
- Adjust to clinical material
- Define assay conditions
- Determine clinically relevant applications

Integration and prospective testing in clinical trials
Consensus/guidelines

Diagnostic application

Open questions from application
(bedside-bench research)

Costs Refunding
Quality management
Technology changes
Therapeutic strategies
Community acceptance

Liver Biopsy !!
Prognostic markers may be assessed in HCC tumor biopsies:

Neoangiogenesis-related genes are hallmarks of fast-growing hepatocellular carcinomas and worst survival. Results from a prospective study

Erica Villa,1 Rosina Critelli,1 Barbara Lei,1 Guido Marzocchi,2 Calogero Cammà,3 Gianluigi Giannelli,4 Patrizia Pontisso,5 Giuseppe Cabibbo,3 Marco Enea,6 Stefano Colopi,2 Cristian Caporali,2 Teresa Pollicino,7 Fabiola Milosa,1 Aimilia Karampatou,1 Paola Todesca,1 Elena Bertolini,1 Livia Maccio,8 Maria Luz Martinez-Chantar,9 Elena Turola,1 Mariagrazia Del Buono,1 Nicola De Maria,1 Stefano Ballestri,10 Filippo Schepis,1 Paola Loria,10 Giorgio Enrico Gerunda,11 Luisa Losi,8 Umberto Cillo12

Neoangiogenesis-related genes identified from tumor biopsy predict fast growth rate of HCC and outcome  

*Villa E, Gut 2015*

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Table 4: Cox regression analysis of baseline factors associated with mortality in the training cohort

<table>
<thead>
<tr>
<th>Variables</th>
<th>Univariate analysis</th>
<th>Multivariate analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HR (95% CI)</td>
<td>p Value</td>
</tr>
<tr>
<td>Gender*</td>
<td>0.687 (0.302 to 1.564)</td>
<td>0.371</td>
</tr>
<tr>
<td>Age, years</td>
<td>0.989 (0.956 to 1.024)</td>
<td>0.540</td>
</tr>
<tr>
<td>Aetiology</td>
<td>1.587 (0.845 to 2.980)</td>
<td>0.151</td>
</tr>
<tr>
<td>Treatment (yes/no)†</td>
<td>0.919 (0.844 to 1.000)</td>
<td>0.051</td>
</tr>
<tr>
<td>Edmondson–Steiner grading</td>
<td>1.443 (1.571 to 2.398)</td>
<td>0.036</td>
</tr>
<tr>
<td>Macrovascular invasion</td>
<td>4.818 (2.140 to 10.846)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Tumour volume at baseline</td>
<td>1.003 (0.518 to 1.944)</td>
<td>0.993</td>
</tr>
<tr>
<td>Platelets (×10^3/mm^3)</td>
<td>0.586 (0.271 to 1.268)</td>
<td>0.586</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>0.492 (0.253 to 0.882)</td>
<td>0.017</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>1.111 (0.515 to 2.397)</td>
<td>0.788</td>
</tr>
<tr>
<td>Bilirubin (mg/dL)</td>
<td>1.069 (0.964 to 1.185)</td>
<td>0.207</td>
</tr>
<tr>
<td>AFP (ng/mL)</td>
<td>1.153 (0.776 to 1.712)</td>
<td>0.481</td>
</tr>
<tr>
<td>Five-gene risk signature</td>
<td>1.548 (1.296 to 1.849)</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>
IgG4-related sclerosing cholangitis (ISC) key features

Associated with type 1 autoimmune pancreatitis in >95% of cases

Reviewed in Zen Y, J Gastroenterol 2016
Annexin A11 is targeted by IgG4 and IgG1 autoantibodies in IgG4-related disease

Lowiek M Hubers,1 Harmjan Vos,2 Alex R Schuurman,1 Robin Erken,1 Ronald P Oude Elferink,1 Boudewijn Burgering,2 Stan F J van de Graaf,1 Ulrich Beuers1

IgG1 and IgG4 antibodies bind to annexin

Hubers LM, Gut 2017
IgG4 antibodies block binding of IgG1 to annexin

- Annexin is a novel autoantigen targeted by IgG1&4 ab in the serum of patients with IgG4-RD but not in disease mimickers like PSC or pancreatobiliary carcinoma.

- IgG4 ab block binding of IgG1 abs to annexin supporting an anti-inflammatory role of IgG4.

Hubers LM, Gut 2017