Measurement of liver fibrosis

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Measurement of liver fibrosis:

towards a better characterisation of disease stage

Outline

- Liver disease stage and liver tissue collagen quantification is not the same thing
- There is fibrosis variation within each stage score
- There is a relationship between liver morphology and portal hypertension
- Quantification of hepatic collagen reflects HVPG
- Liver biopsy fibrosis measurement at 1 year post transplantation predicts clinical decompensation in HCV patients
- Liver fibrosis measurement can provide clinically useful prognostic information

What do we mean by “liver fibrosis”?

Differences between morphological appearance, description, stage scoring and liver fibrosis measurement

Appearance	Ishak stage: Categorical description	Ishak stage: Categorical assignment	Fibrosis measurement*
	No fibrosis (normal)	0	1.9%
	Fibrous expansion of some portal areas ± short fibrous septa	1	3.0%
	Fibrous expansion of most portal areas ± short fibrous septa	2	3.6%
	Fibrous expansion of most portal areas with occasional portal to portal (P-P) bridging	3	6.5%
	Fibrous expansion of portal areas with marked bridging (portal to portal (P-P) as well as portal to central (P-C))	4	13.7%
	Marked bridging (P-P and/or P-C), with occasional nodules (incomplete cirrhosis)	5	24.3%
	Cirrhosis, probable or definite	6	27.8%

Standish R et al. An appraisal of the histopathological assessment of liver fibrosis. Gut 55;569;2006

Can liver fibrosis be measured as proportion of liver tissue area stained by sirius red in liver biopsies in chronic HCV infection?

Ishak stage	N	CPA % Mean	Median	IQR
0	17	2.8	2.0	1.2.3-4.6
1	37	3.8	3.0	1.9-4.9
2	45	5.8	5.0	3.7-8
3	35	6.4	6.2	3.8-7.7
4	9	9.9	8.4	6.7-14
5	33	14.1	12.5	7.8-19
6	23	21.7	17.3	15.1-29

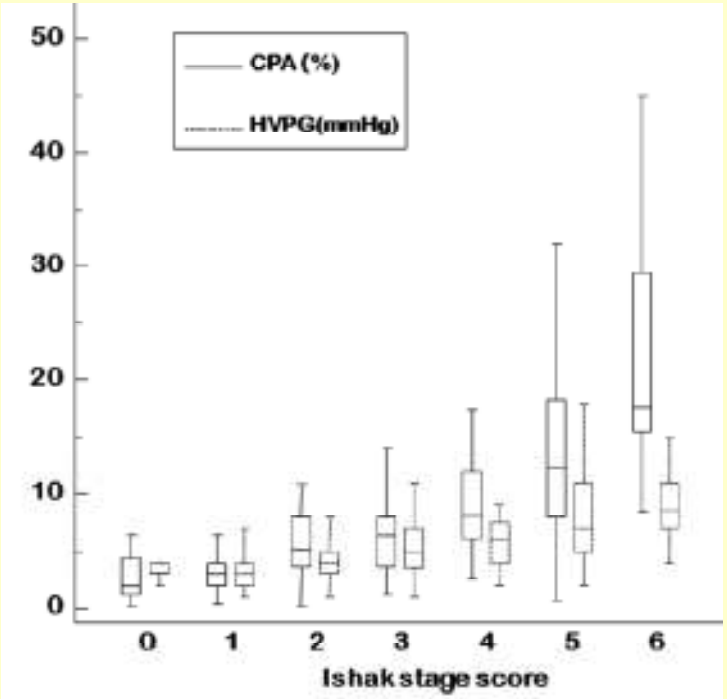
Calvaruso V et al.
Hepatology 49,1236;2009
(TJ liver biopsies)

Table 2. Correspondence Between Area of Fibrosis, as Assessed by Image Analysis (Reference Value) in Whole Sections and METAVIR Stage

METAVIR Stage	Area of Fibrosis by Image Analysis (Mean + SEM)	Range of Fibrosis Area
F0	2 ± 0.14	0.7-2.7
F1	3.4 ± 0.25	2.7-4.6
F2	5.8 ± 0.7	4.6-10.25
F3	14.7 ± 0.77	10.25-19.9
F4	25.1 ± 1.44	19.9-30.2

Bedossa et al.
Hepatology 38,1449;2003
(2x3cm tissue blocks from surgical resections; N=17)

Variability of liver fibrosis within stage scores in chronic hepatitis C



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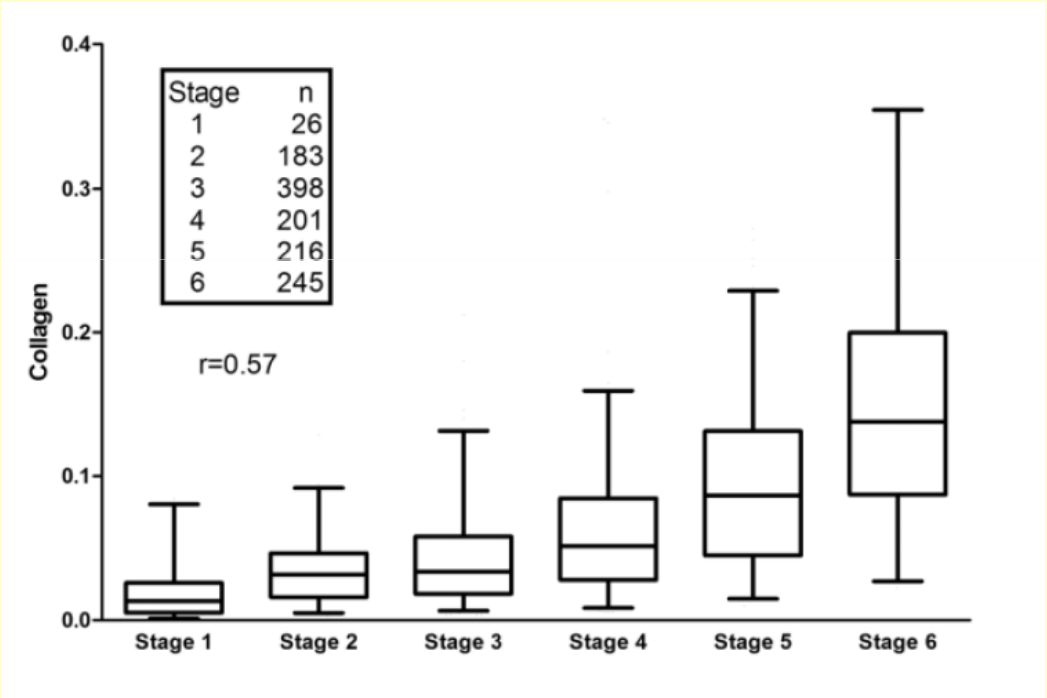


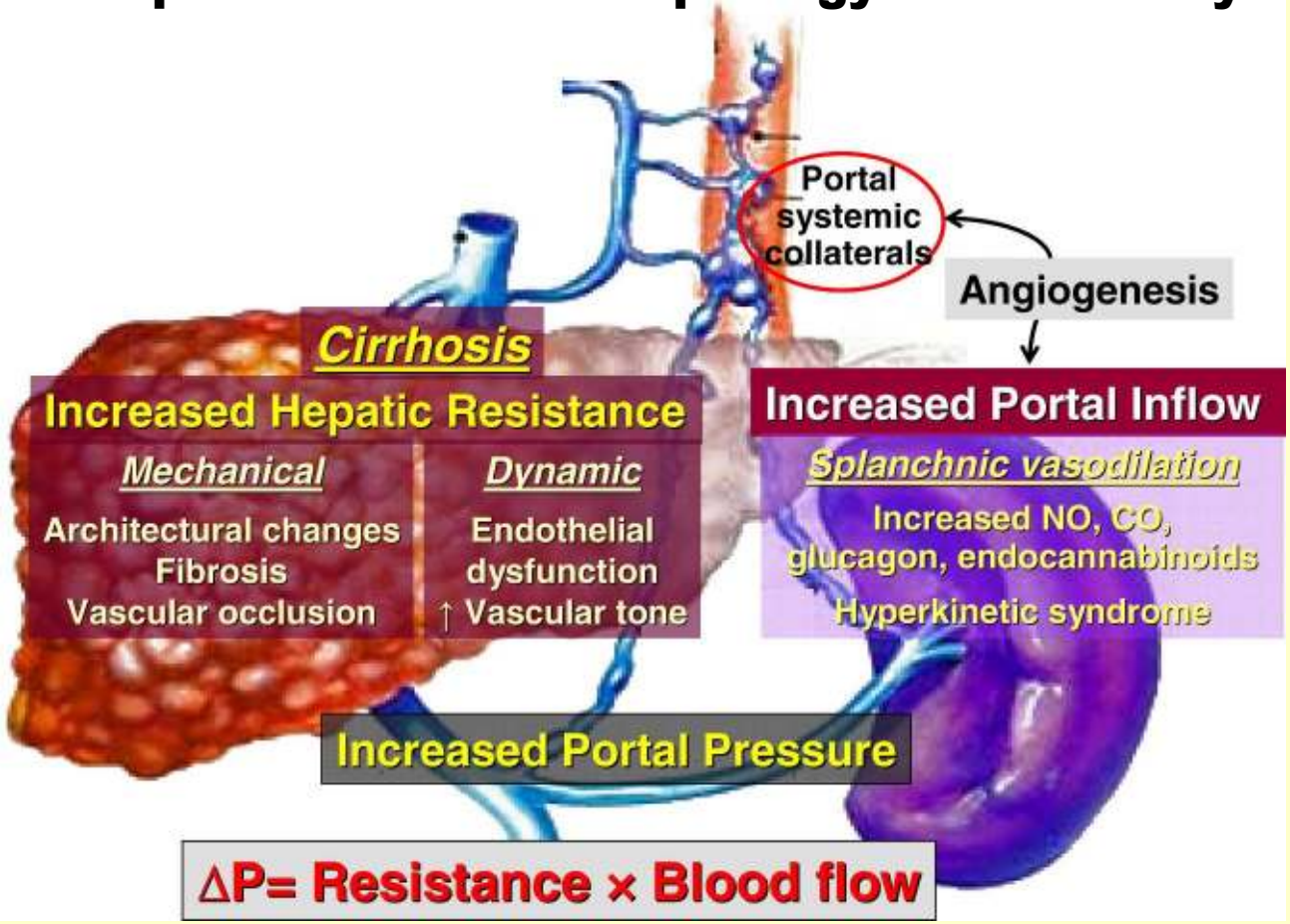
Goodman Z et al.
Hepatology 50,1738-;2009
(liver biopsies from HALT-C trial)

Variability of liver fibrosis

- **Fibrosis variation within stage scores is a biological reality**
- **Amount of liver fibrous tissue is a biological continuum as progressive liver disease develops**
- **It is unreasonable to expect liver fibrous tissue measurements to segregate neatly, with clearly defined cut-offs, into artificial descriptive stage score categories**
- **Intra stage (within any given descriptive stage "score" definition) differences in the amount of fibrosis may have additional prognostic value:**
  - **"vive la difference"**

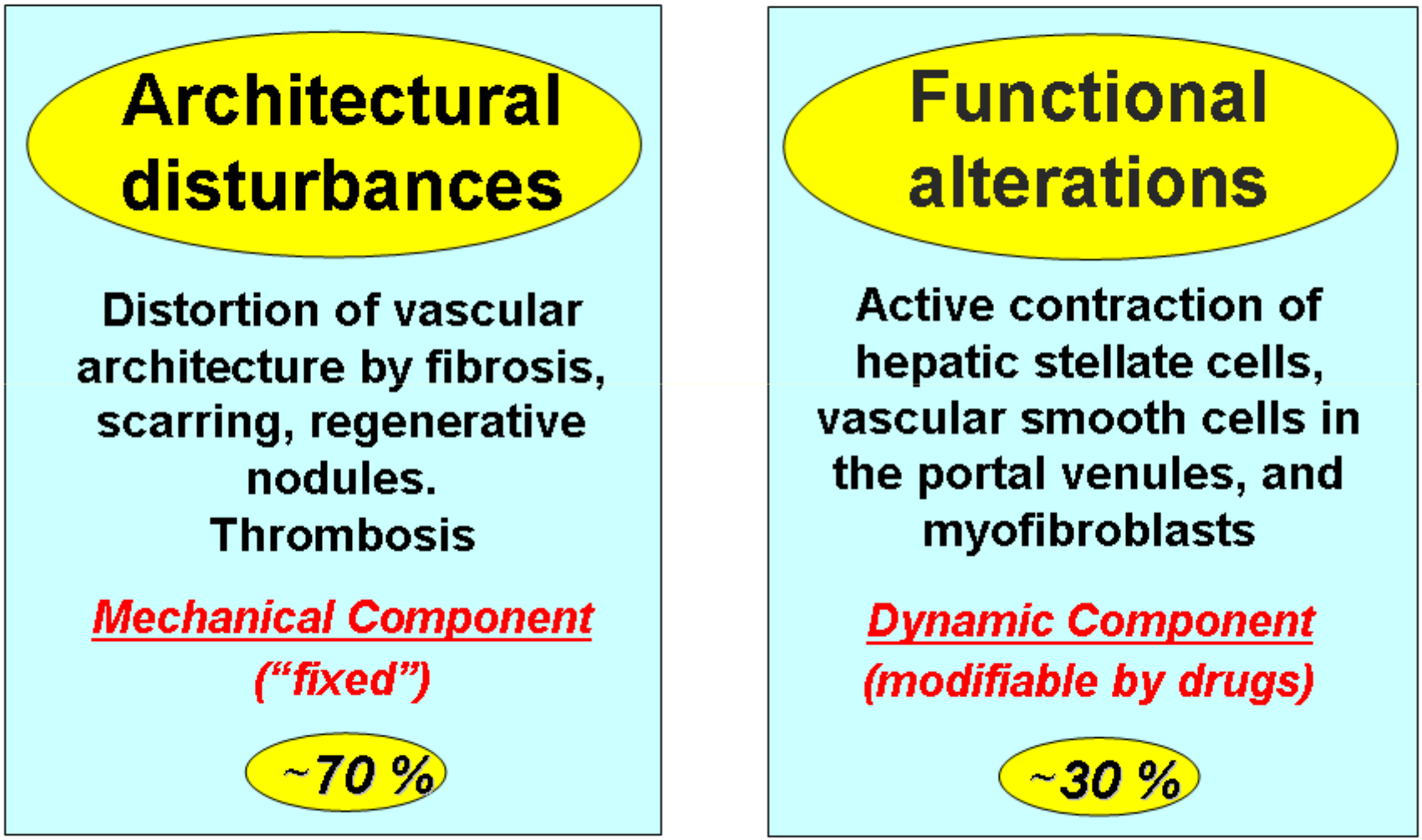
Pathophysiology of portal hypertension

Relationship between liver morphology and haemodynamics



The different contributors to increased portal pressure
Bosch J et al. J Hepatol 48 Suppl1,S68;2008

Increased intrahepatic resistance in cirrhosis



Marra F. Pathophysiology of portal hypertension
EASL Postgraduate course 2008

Relationship between liver disease and haemodynamics

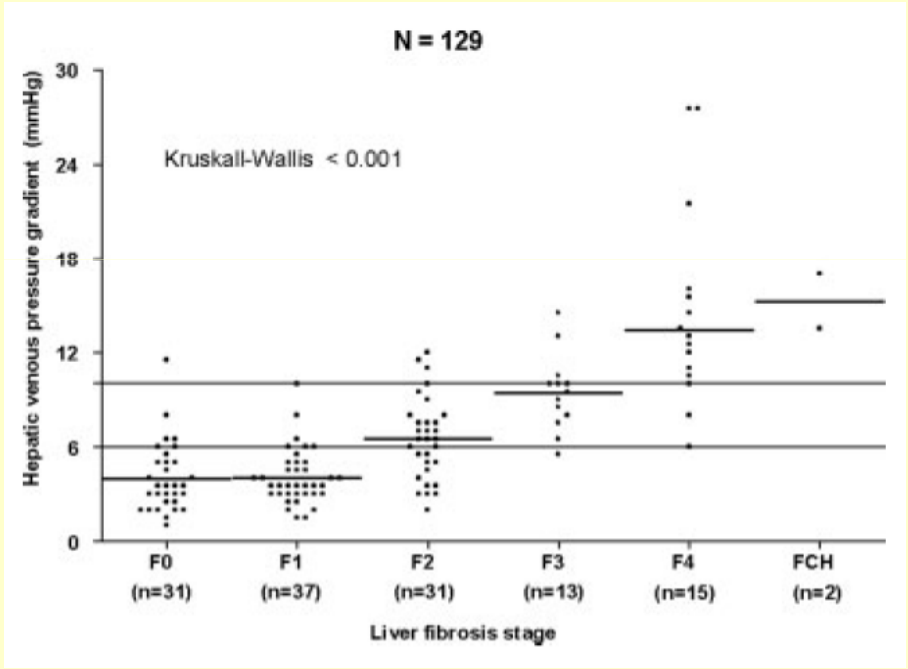
- van Leeuwen D et al. Scand J Gastroenterol. 24,65;1989
- "Wedged hepatic venous pressure (HVWP) recording and venography for the assessment of pre-cirrhotic and cirrhotic liver disease."
  - One of the first papers to explore relationship between liver histopathology and HVWP
- The four main groups and the means of the pressure gradients (WHVP - FHVP) with their 95% confidence limits were
  - Near-normal liver (n = 8), 3.4 mm Hg (2.2-4.6)
  - Chronic active hepatitis (n = 12), 6 mm Hg (4.35-7.65)
  - Chronic hepatitis in transition to cirrhosis (n = 9), 10.3 mm Hg (6.6-14.1)
  - Cirrhosis (n = 8), 15.4 mm Hg (9.4-21.4)
  - A pressure gradient of more than 5 mm Hg was always associated with significant liver disease on liver biopsy

Relationship between liver morphology and haemodynamics

liver disease stage reflects hepatic venous pressure gradient

(post transplantation HCV liver disease)

- Carrion JA et al. Liv Transpl 12,1791;2006
- 169 liver biopsies (66 percutaneous, 103 transjugular) from 124 HCV-infected liver transplant recipients with determination of hepatic venous pressure gradient (HVPG).
- “Close correlation between liver stiffness measurement and HVPG (Pearson coefficient, 0.84; P < 0.001).”
- “There was a significant positive relationship between the fibrosis stage and HVPG (Kruskal-Wallis P<0.001).”



Relationship between liver morphology and haemodynamics

liver disease stage reflects hepatic venous pressure gradient

(post transplantation HCV liver disease)

- **Samonakis D et al. Liver Transplantation 13,1305;2007**
- **90 consecutive HCV OLT patients underwent 170 hepatic venous pressure gradient (HVPG) measurements concomitant with transjugular liver biopsy**
- **HVPG correlated with Ishak stage (r = 0.73, P < 0.001) for mild (0-3) and severe fibrosis (4-6)**

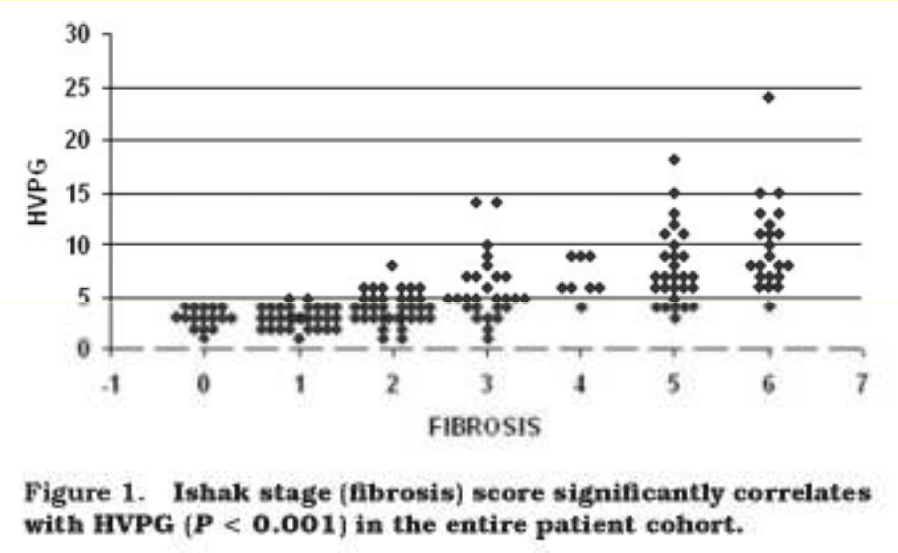


Figure 1. Ishak stage (fibrosis) score significantly correlates with HVPG ($P < 0.001$) in the entire patient cohort.

Amount of fibrosis within cirrhosis correlates with HVPG

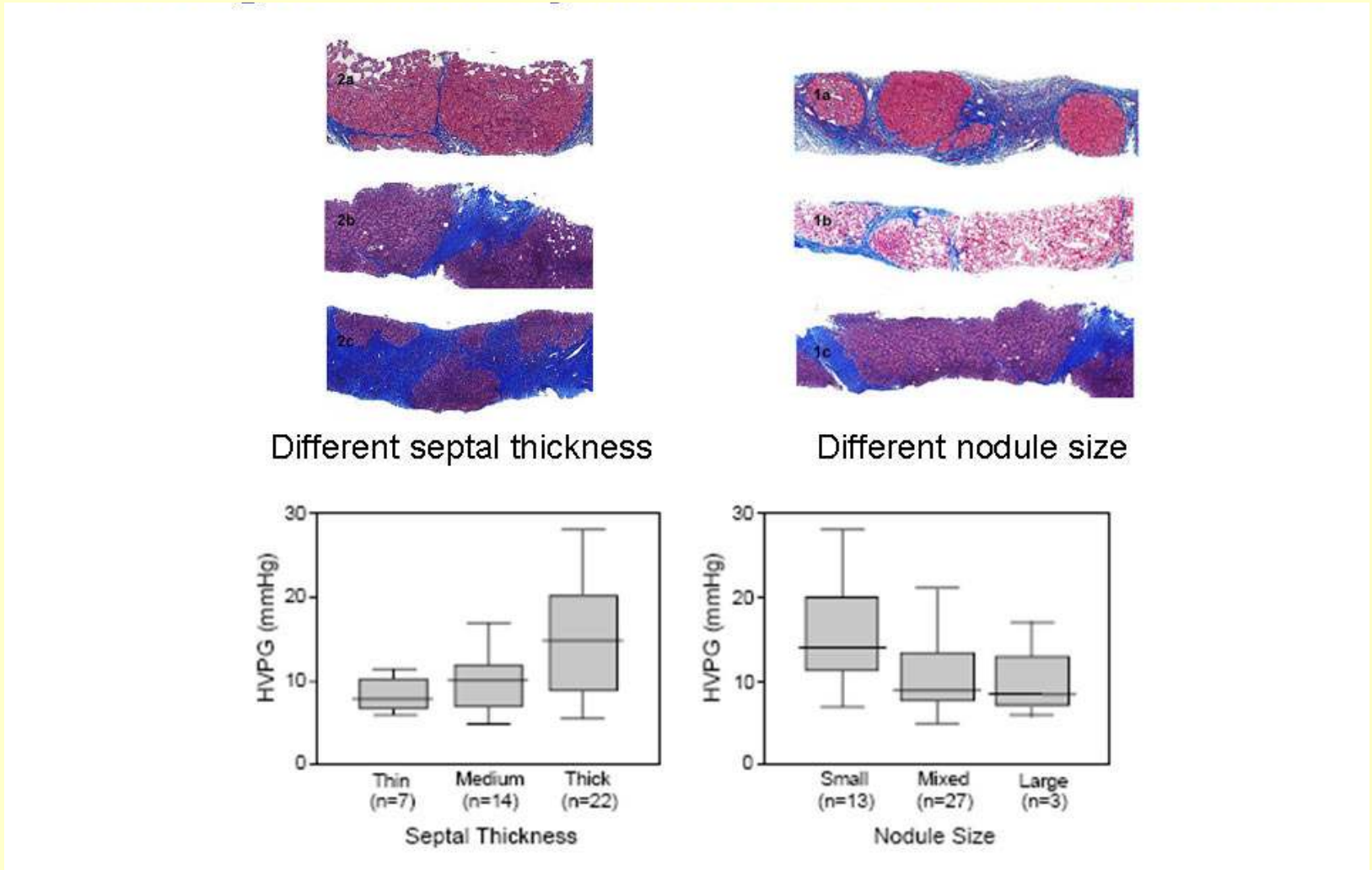
- **"The Laennec grading system for assessment of hepatic fibrosis: validation by correlation with wedged hepatic vein pressure and clinical features"**
  - **Kutami R, Girgrah N, Wanless IR, Sniderman K, Wong FS, Sherman M et al. Hepatol 32, 407A; 2000**
  - **Laennec system ("modified from Metavir system") categorises cirrhosis as mild (thin septa, large nodules), moderate, severe (broad septa, "minute" nodules)**
  - **302 transjugular biopsies (presumably including a range of aetiologies)**
  - **Fibrosis "scores" correlated with HVPG, ascites, INR, bilirubin and inversely with albumin; as well as Child-Pugh class**

Small nodules and thick septa (ie amount of fibrosis) reflects HVPG in cirrhotic patients

(various aetiologies; mainly HCV and alcohol)

- **"Histological-hemodynamic correlation in cirrhosis - a histological classification of the severity of cirrhosis"**
  - **Nagula S et al. J Hepatol 44:111;2006**
  - **43 cirrhotic patients**
  - **(34 transjugular, 9 percutaneous) liver biopsies and hepatic venous pressure gradient (HVPG) measurements were perfomed within 6 mths of each other**
  - **Sinusoidal fibrosis, septal thickness, loss of portal tracts and central veins, nodule size, inflammation, steatosis and iron were analyzed**
  - **Small nodule size (<1.0 mm) and thick septa (>0.4 mm) had the highest independent predictive value for clinically significant portal hypertension (HVPG ≥10 mmHg)**

Small nodules and thick septa (ie amount of fibrosis) indicate higher hepatic venous pressure gradients
Nagula S et al. J Hepatol 44:111;2006



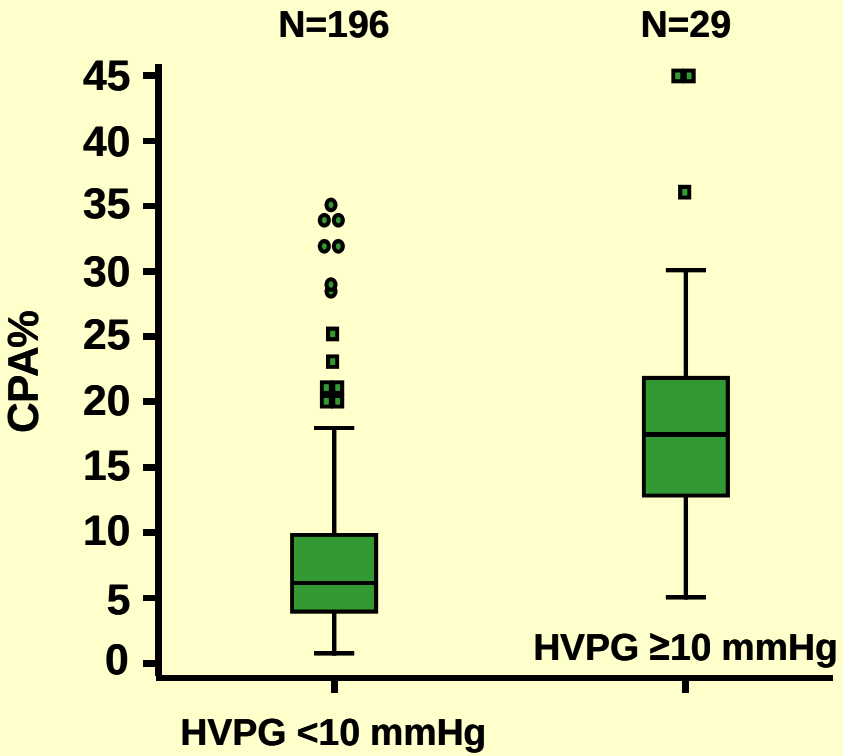
Royal Free computer-assisted image analysis of liver collagen

- Sirius red stained sections
- Grey level thresholding and detection of biopsy area
- Computer assisted RGB detection (green overlay) of sirius red areas
- Structural collagen (eg septal portal tract and large blood vessel, ie unrelated to HCV disease fibrosis) is edited manually and removed from CPA
  - No editing CPA = 12.5%
  - After editing CPA = 5.7%

Relationship between liver morphology and haemodynamics

Liver biopsy fibrosis measurement reflects hepatic venous pressure gradient

- 225 liver biopsies from 115 patients
- Hepatic venous pressure gradient (HVPG) measured at the time of transjugular liver biopsy
- Liver collagen was expressed as a proportion of liver biopsy tissue area (collagen proportionate area, CPA) and compared with the HVPG
- Significant correlation between CPA and HVPG (Spearman's r=0.61; p<0.001)
  - Calvaruso V et al. Hepatology 49,1236;2009



Collagen proportionate area reflects hepatic venous pressure gradient (HVPG) better than Ishak stage (or grade) Calvaruso V et al. Hepatology 49,1236;2009

Predictors of portal hypertension (ie HVPG ≥ 6mmHg)

Predictor	Multivariate Analysis OR (95% CI)	P value
Ishak grading score	1.214 (0.940 – 1.567)	0.138
Ishak staging score	1.372 (0.979 – 1.923)	0.067
Collagen Proportionate Area(%)	1.206 (1.094 – 1.331)	**< 0.001**

Predictors of "clinically significant" portal hypertension (ie HVPG ≥ 10mmHg)

Predictor	Multivariate Analysis OR (95% CI)	P value
Ishak grading score	1.126 (0.812 – 1.561)	0.477
Ishak staging score	1.577 (1.000 – 2.482)	0.05
Collagen Proportionate Area(%)	1.105 (1.026 – 1.191)	**0.009**

CPA adds value to the liver biopsy assessment of liver disease stage

- Measurement of CPA (as a HVPG surrogate) improves the histological description of liver disease stage
- A relationship between the amount of liver collagen and portal pressure could be useful to stratify prognostic groups
- Histological disease stage, CPA and HVPG are surrogates for patient outcome measures
  - Correlation with prognosis and treatment outcomes is necessary

Computer-assisted image analysis of liver collagen at one year biopsy can predict clinical outcome in HCV post liver transplant patients

Manousou P et al. Hepatology 50(S4),302A;2009

- **Biopsies : one year post LT (12-15m); >15mm in length)**
- **Decompensation : the first occurrence of**
  - **ascites, hydrothorax**
  - **variceal bleeding**
  - **jaundice (≥3mg/dl – in the absence of other causes)**
  - **encephalopathy**

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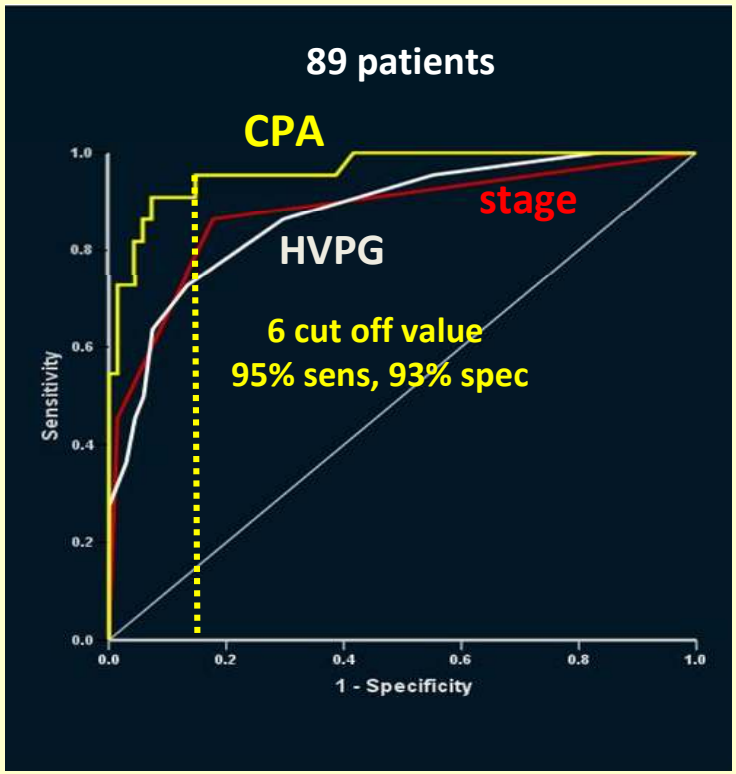
- **Variables analysed (associated with HCV fibrosis in the literature):**
  - **Age/gender recipient/donor**
  - **HCC / ALD as associated aetiologies**
  - **Cold/warm ischemic time**
  - **Blood group compatibility and HLA disparity**
  - **Histological acute hepatitis**
  - **Rejection episodes and treatment**
  - **CMV viraemia, bacterial infections/antibiotics**
  - **Diabetes mellitus pre/post-transplant**
  - **Genotype , viral load pre/post-Tx, antiviral treatment/SVR**
  - **Initial/1 year – immunosuppression,**
  - **HVPG, Ishak stage and CPA at one year biopsy**

Computer-assisted image analysis of liver collagen at one year biopsy can predict clinical outcome in HCV post liver transplant patients

Manousou P et al. Hepatology 50(S4),302A;2009

- **ROC curve of CPA, HVPG and stage at 12m post LT for the prediction of clinical decompensation**
  - **CPA AUROC 0.962**
    - **95% CI: 0.921 – 1.000**
  - **Stage AUROC 0.877**
    - **95% CI: 0.781 – 0.972**
  - **HVPG AUROC 0.874**
    - **95% CI: 0.790 – 0.959**
- **In Cox regression analysis, only CPA could predict clinical decompensation (p=0.0001, Exp(B)=1.158, 95%CI 1.102 – 1.217)**



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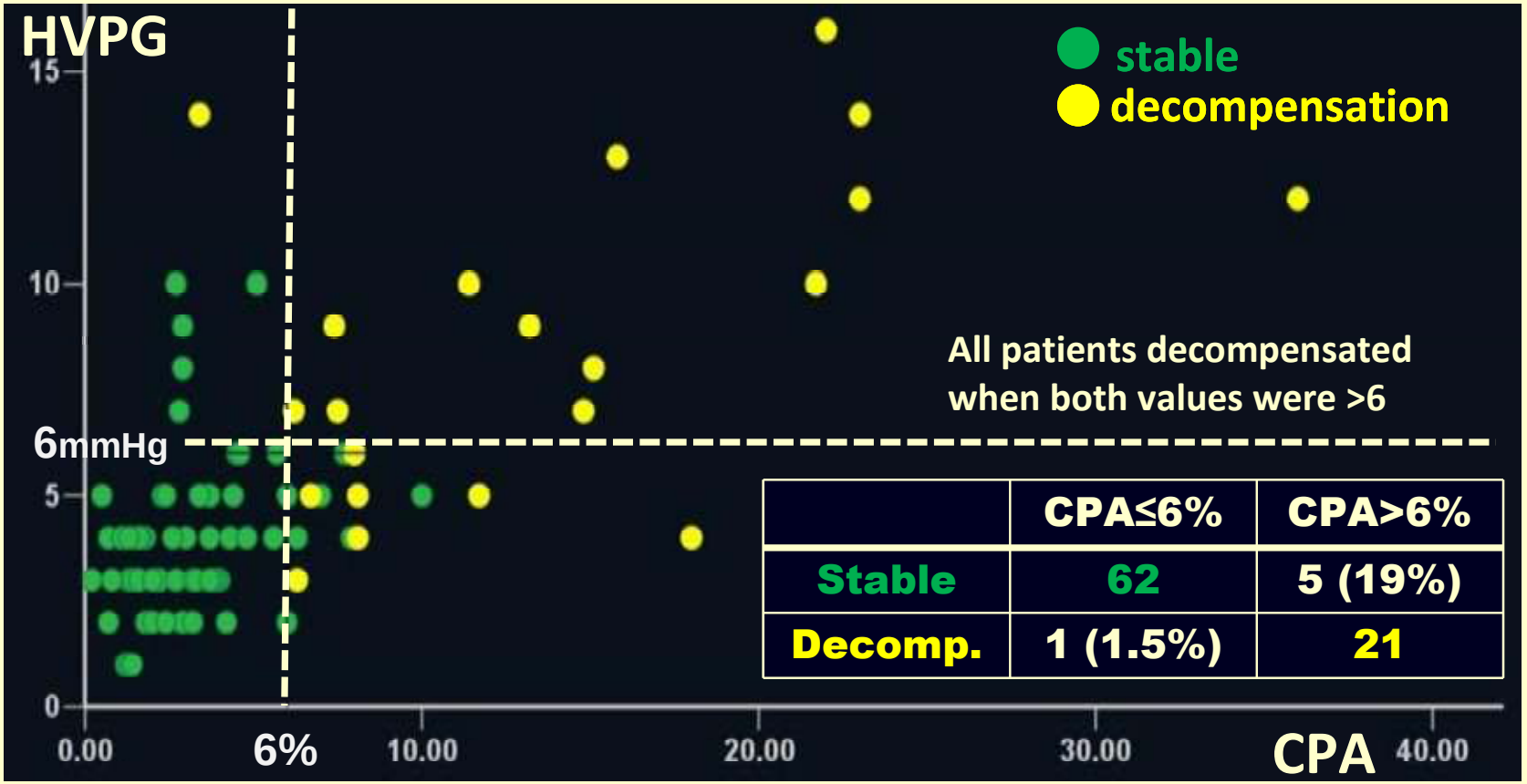

	CPA≤6%	CPA>6%
Stable	62	5 (19%)
Decomp.	1 (1.5%)	21

CPA and HVPG (89 patients)

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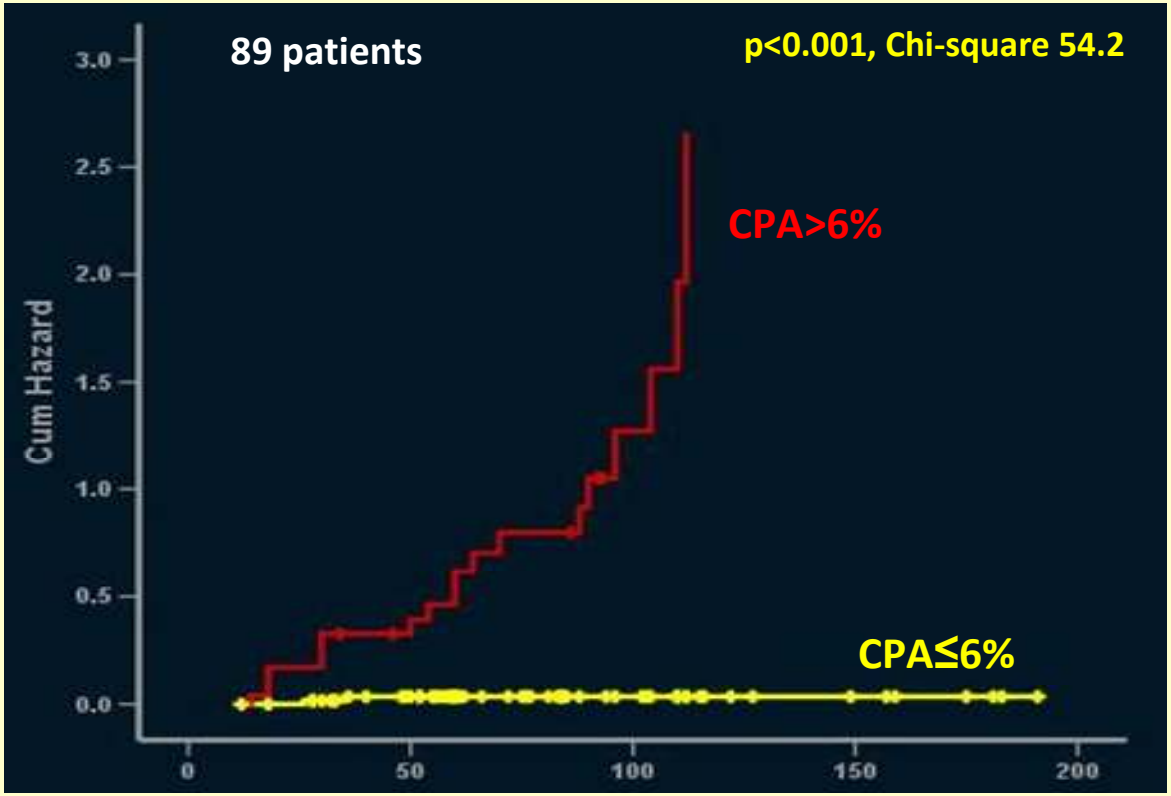


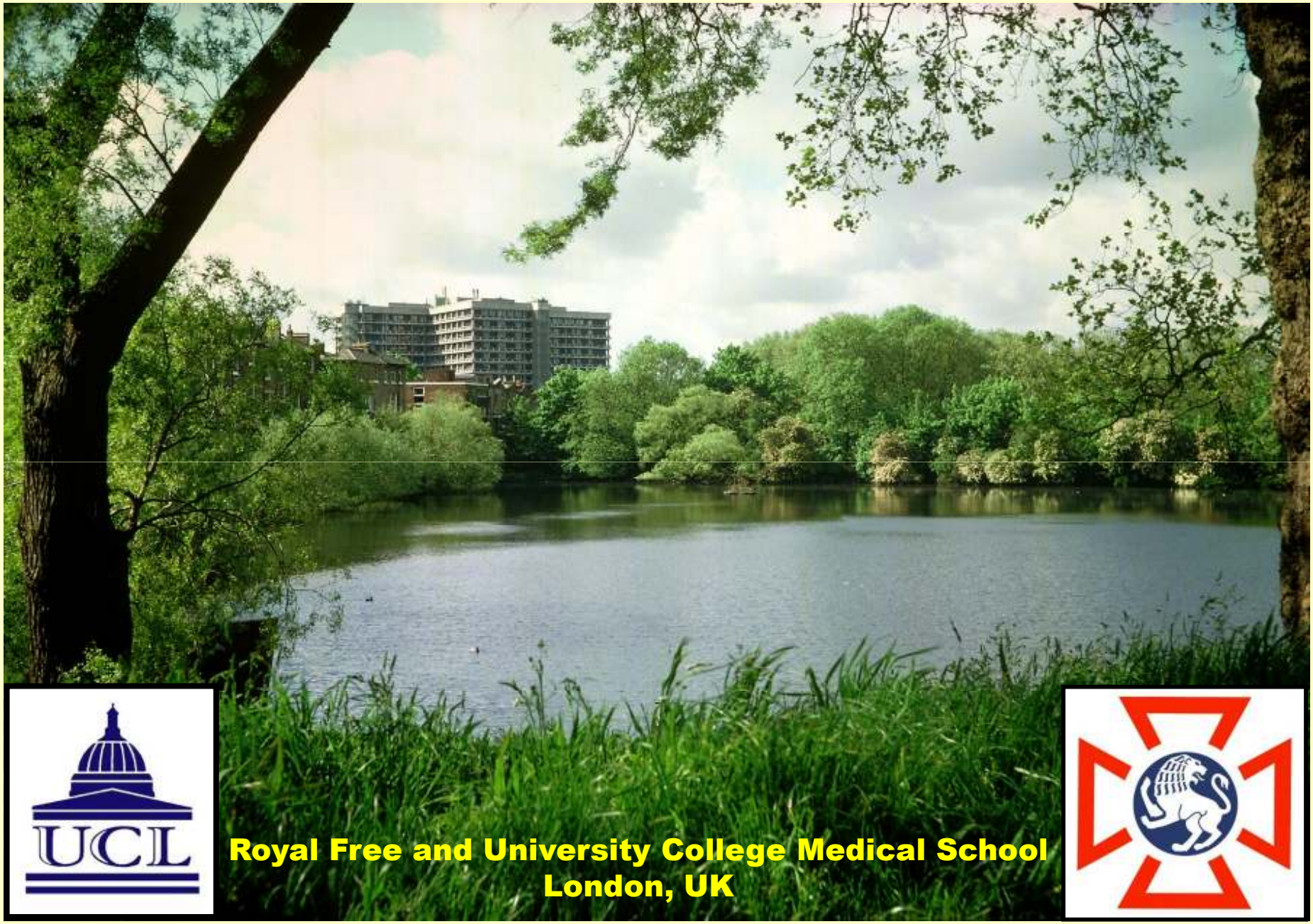
Time (mths) to first episode of decompensation with a CPA cut off value of 6%

Computer-assisted image analysis of liver collagen at one year biopsy can predict clinical outcome in HCV post liver transplant patients
Manousou P et al. Hepatology 50(S4),302A;2009

- **CPA measurement at 1 year post LT was predictive of clinical decompensation in HCV patients:**
  - **CPA had good sensitivity and specificity**
  - **CPA was a better predictor than Ishak stage and HVPG**
  - **CPA combined with HVPG had better precision**

Measurement of liver fibrosis:
towards a better characterisation of disease stage
Conclusion

- The morphological definitions of liver disease stages have been useful
- Other clinically relevant aspects such as portal hypertension should be reconsidered from the histopathological perspective
- Liver biopsy fibrosis measurement reflects HVPG and improves prognostic evaluation
- Liver biopsy fibrosis measurement at one year post transplantation predicts clinical decompensation in HCV patients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