FROM STEATOSIS TO FIBROSIS AND BEYOND

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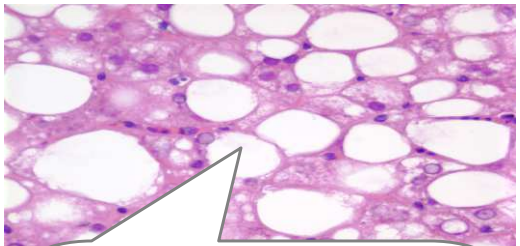
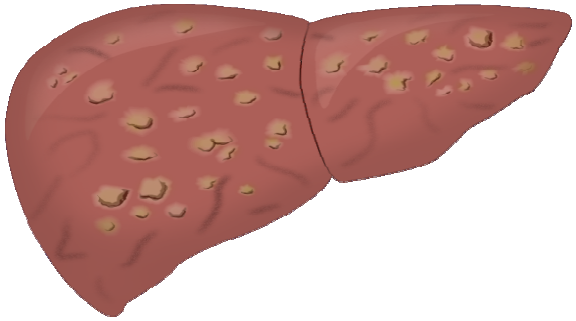
The Spectrum of NAFLD

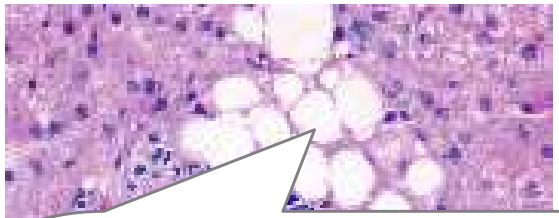
Fatty Liver



Fat infiltration >5% with or without mild inflammation

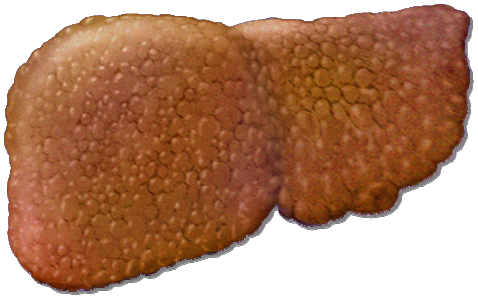
NASH

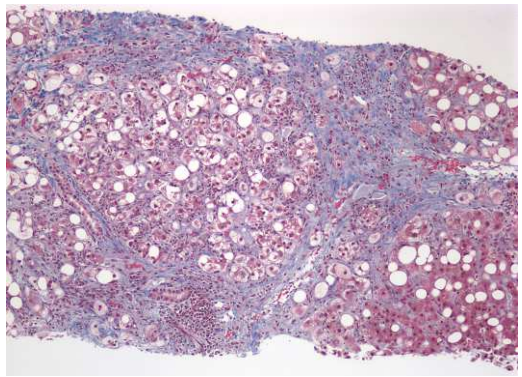




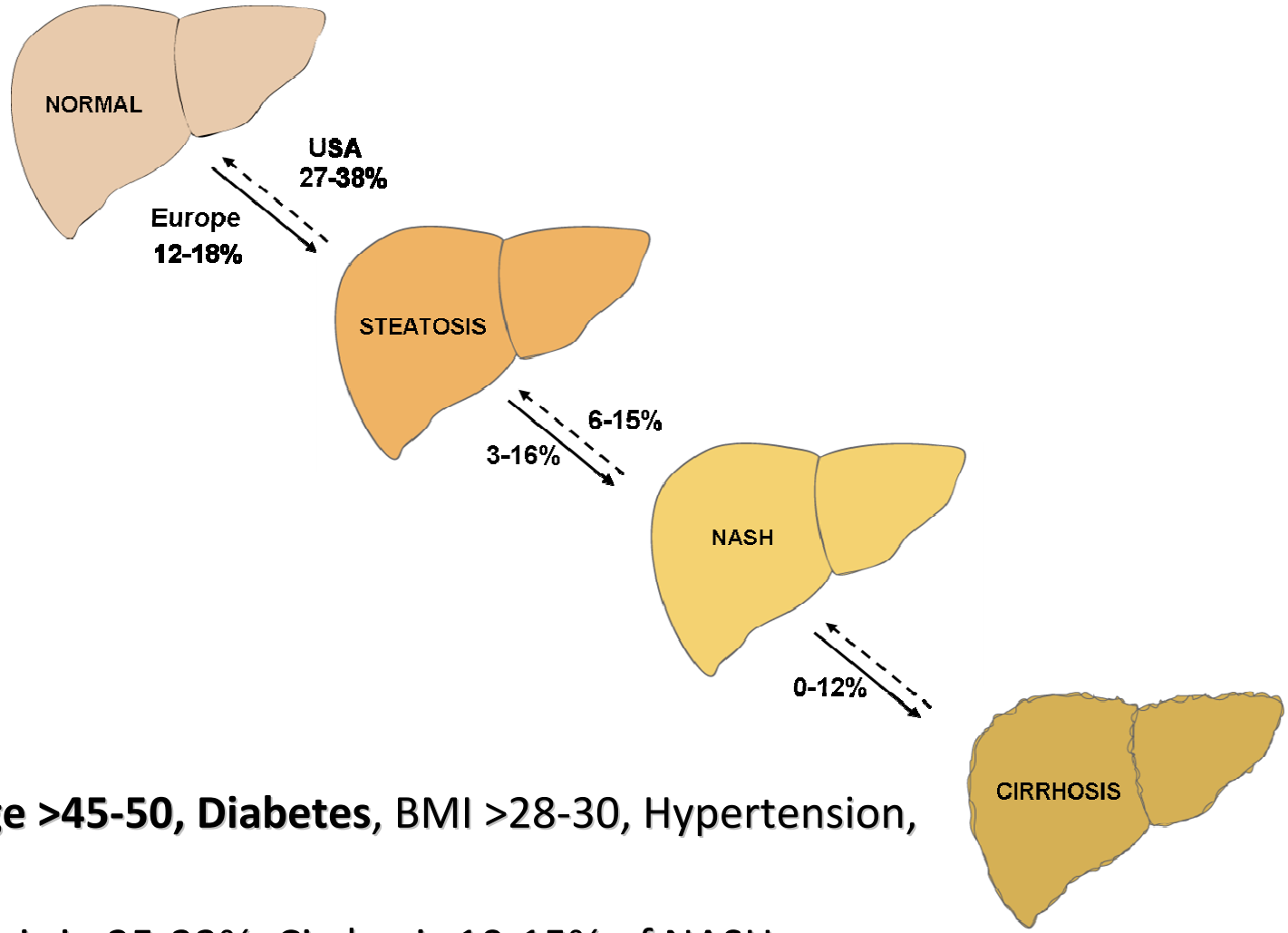
Steatosis + necro-inflammatory changes (ballooning degeneration, Mallory bodies, megamitochondria) and/or fibrosis

Cirrhosis





Risk Stratification



Predictors: **Age >45-50, Diabetes**, BMI >28-30, Hypertension, IR severity.

Bridging Fibrosis in 25-33%, Cirrhosis 10-15% of NASH patients at diagnosis.

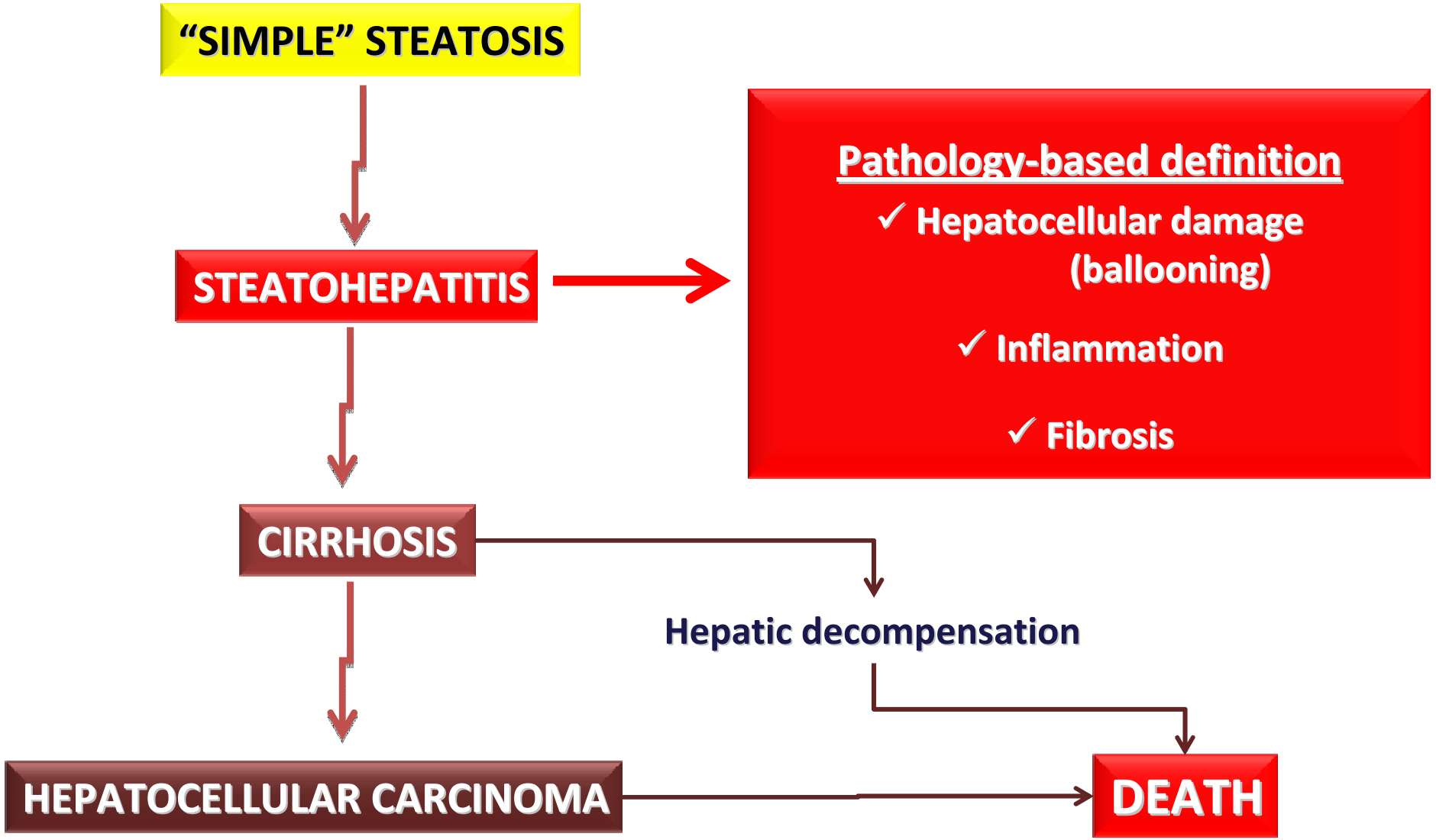
Key Questions

1. What is the significance of steatohepatitis?

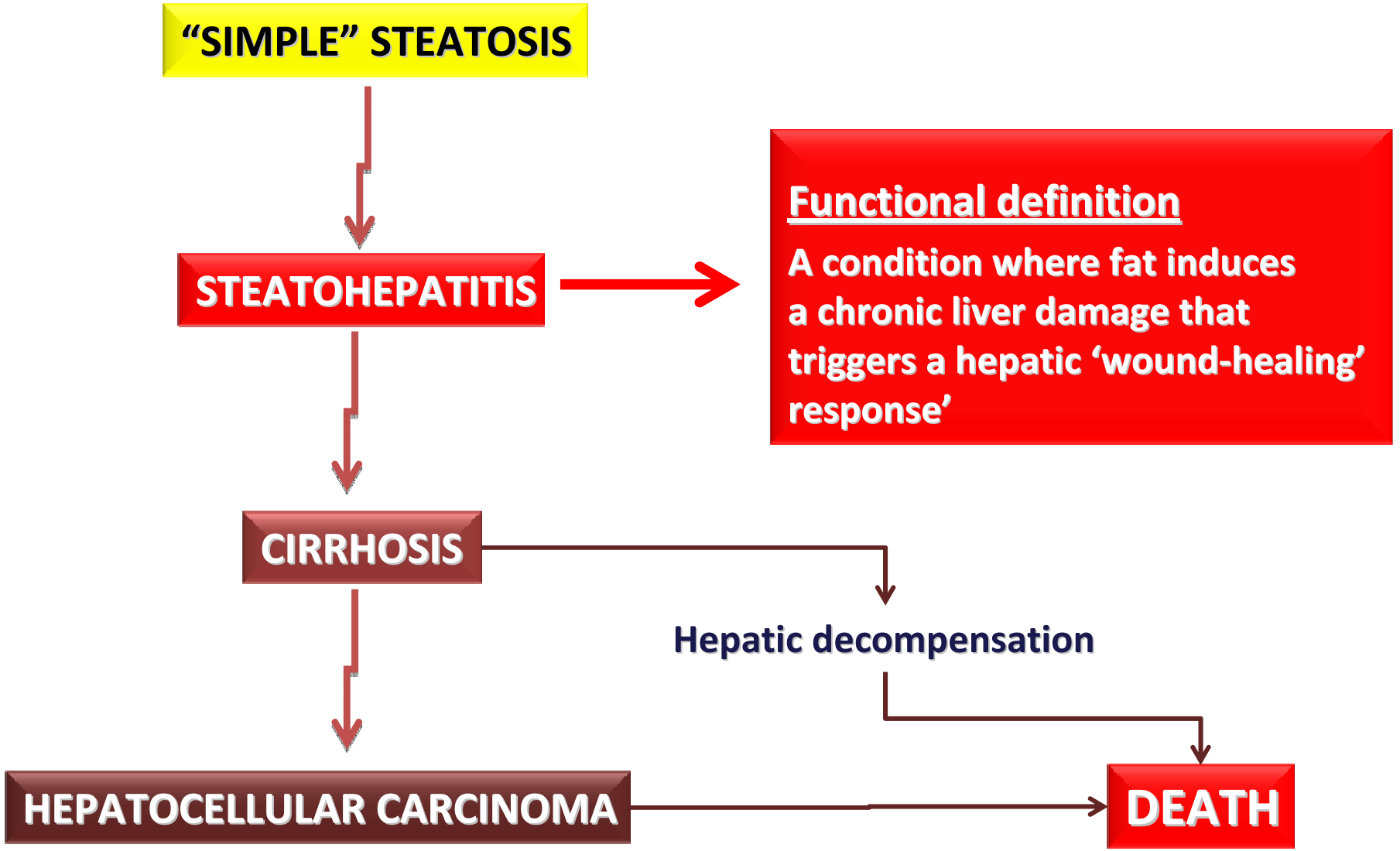
2. How does NASH progress to cirrhosis?

3. Cirrhosis and beyond

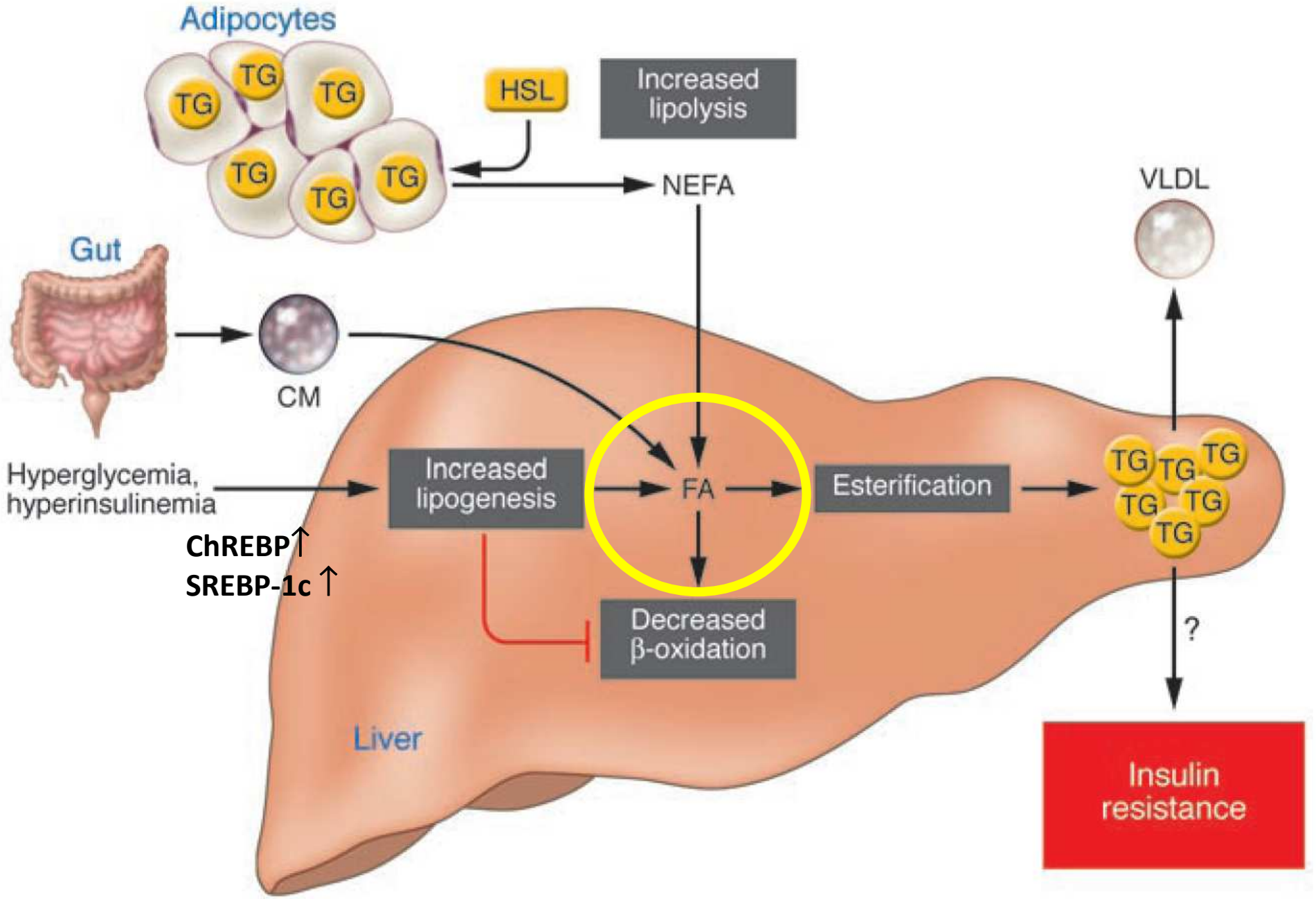
From “Simple Steatosis” to HCC



From “Simple Steatosis” to HCC

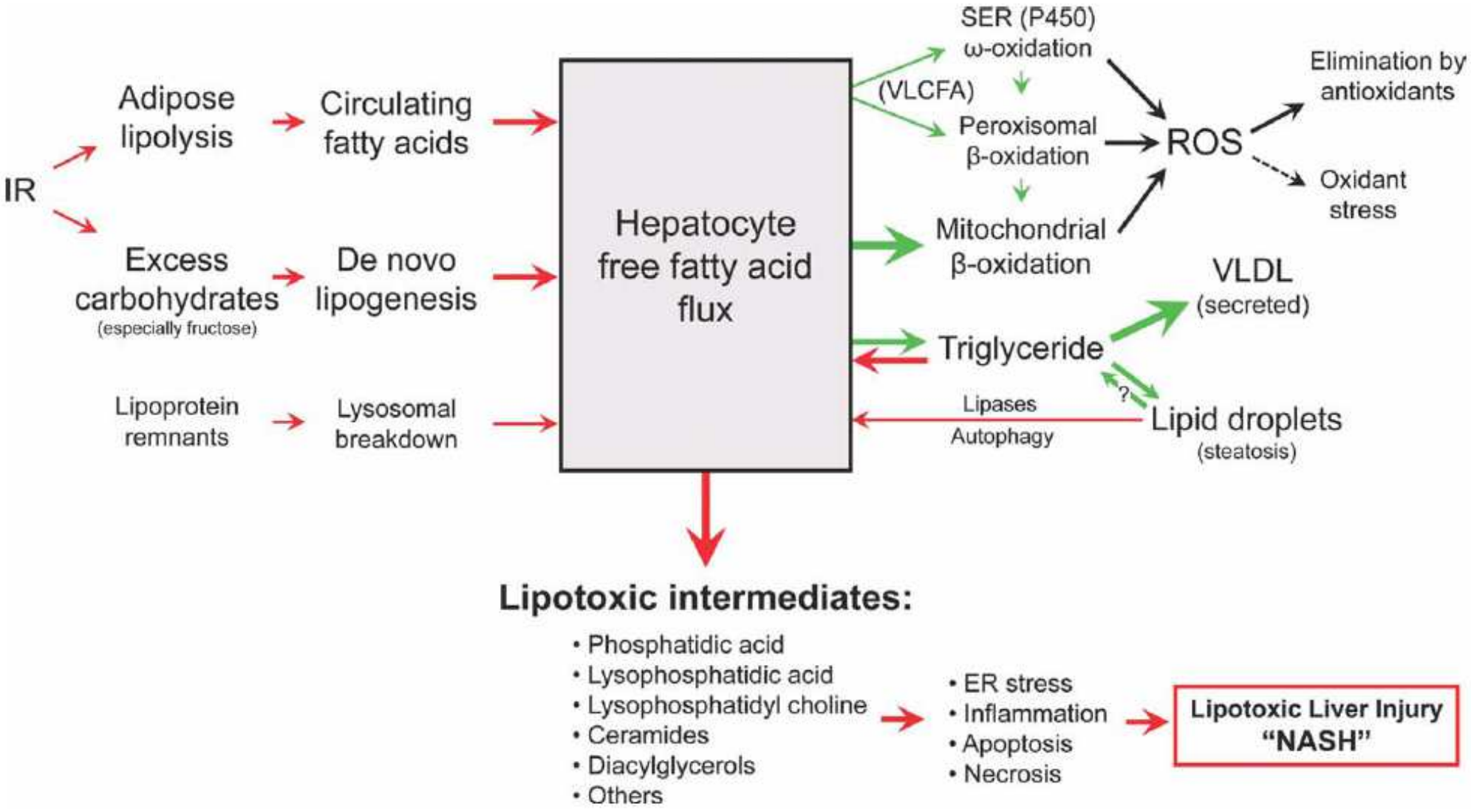


Metabolic Defects Leading to Hepatic Steatosis

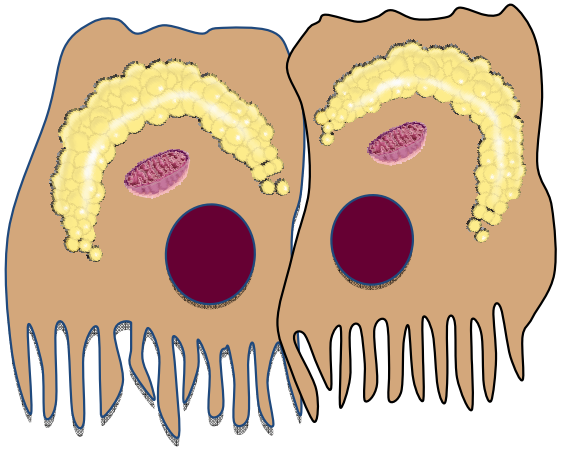


Postic and Girard, J Clin Invest 2008;118:829

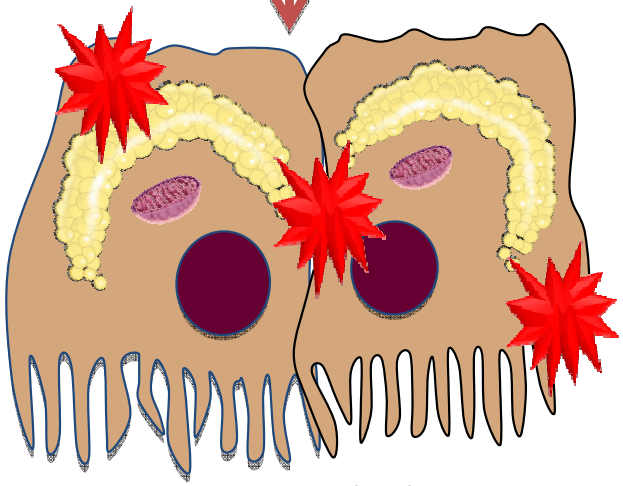
The Pathway of Lipotoxic Liver Injury



Neuschwander-Tetri Hepatology 2010;52:774



Steatosis



Steatosis and damage
(steatohepatitis)

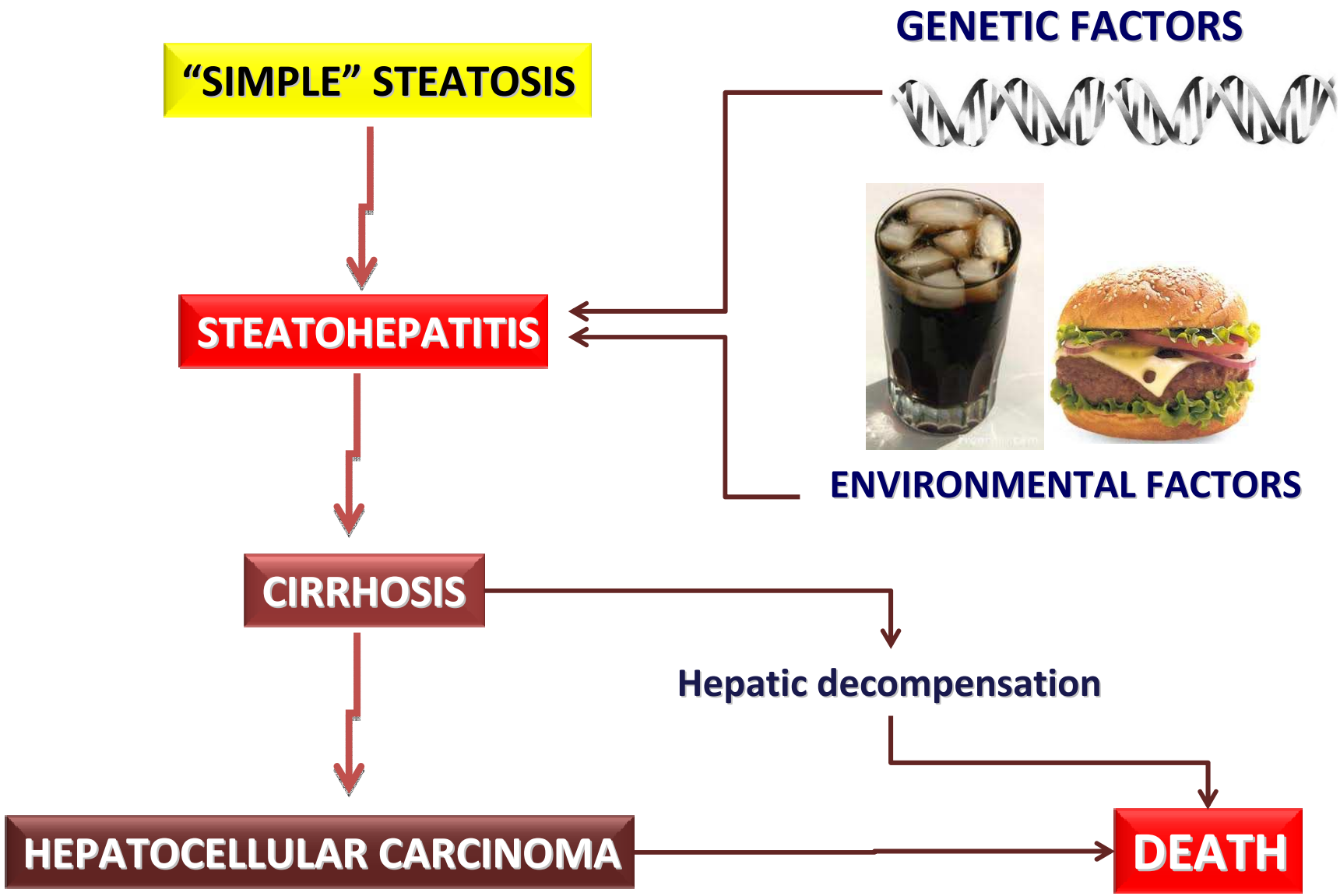
LIPOTOXICITY

Definition of lipotoxicity

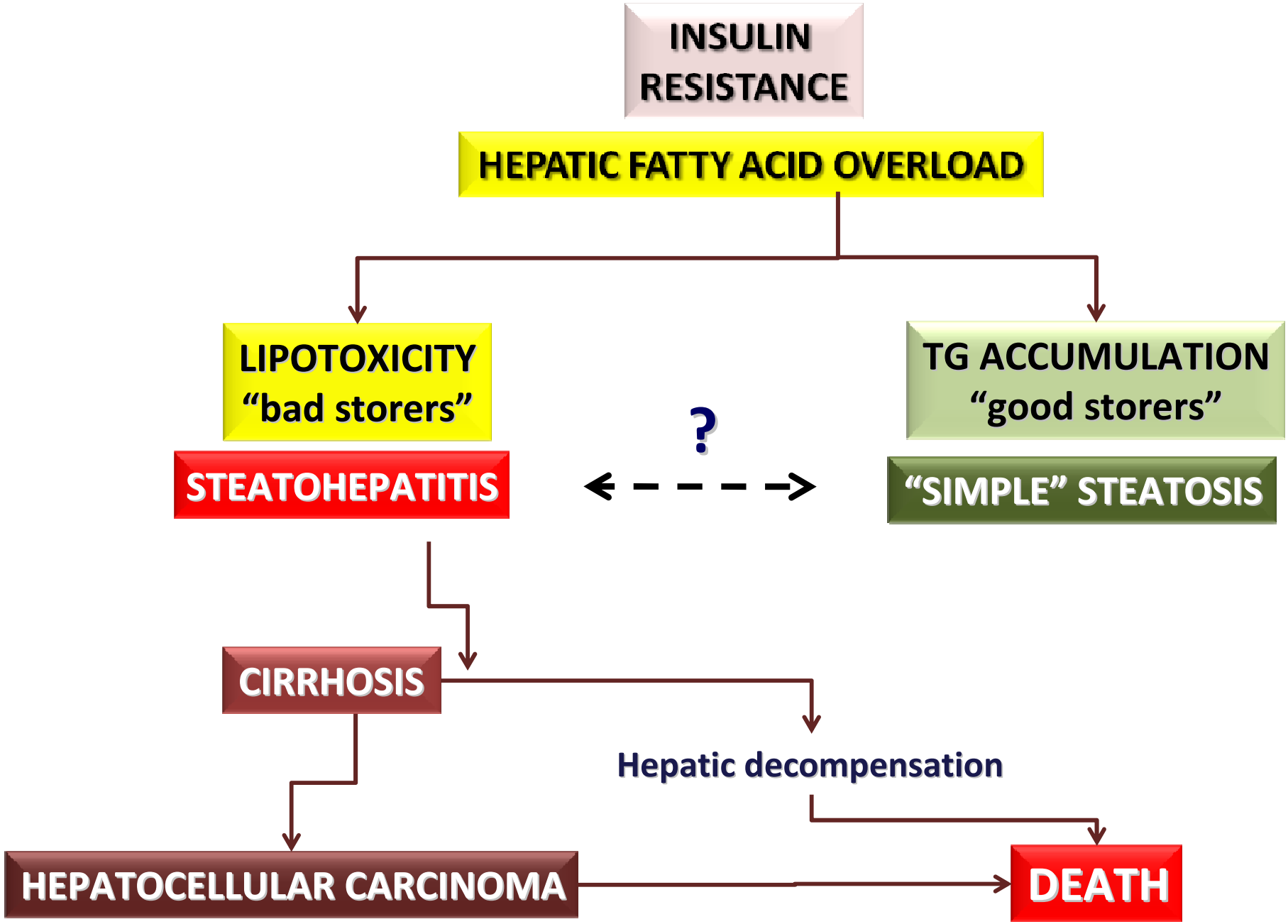
A condition whereby fat accumulation results toxic effects on the cell and causes damage or death

Fatty Acids

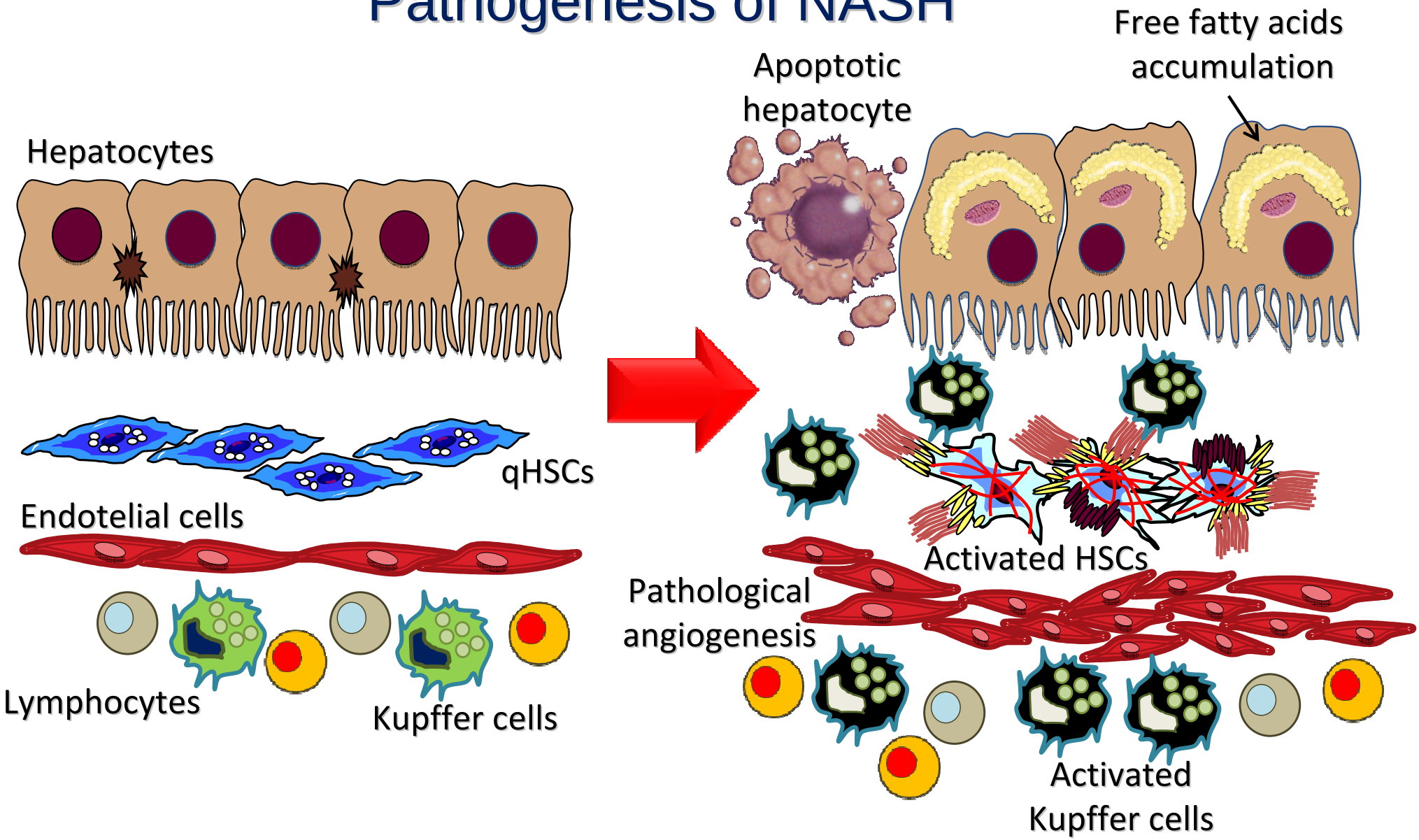
From “Simple Steatosis” to HCC



“Good” and “Bad” Storers



Pathogenesis of NASH



Normal Liver

NAFLD/NASH

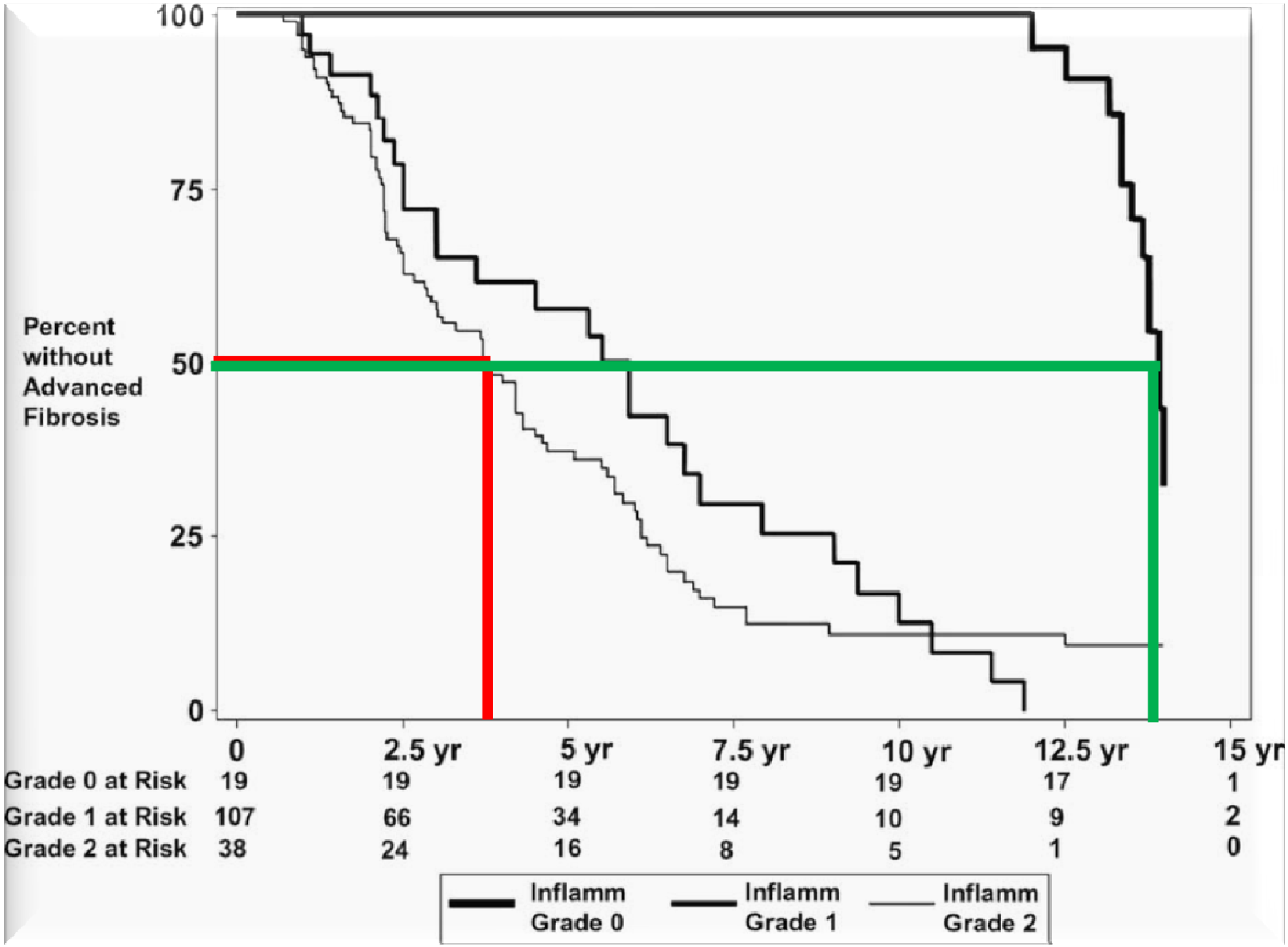
Systematic Review of Risk Factors for Fibrosis Progression in NASH

Statistical significance and adjusted hazard ratios for the components of the multivariate Cox proportional hazards model predicting progression to advanced fibrosis with NASH on initial biopsy.

Variable	Hazard ratio (95% CI)	*p*-Value
Grade 0 inflammation on 1st bx	1.0 [Reference]	Reference
Any grade inflammation on 1st bx	2.5 (1.4–4.3)	0.001*
Grade 1 inflammation on 1st bx	2.5 (1.4–4.3)	0.001*
Grade 2 inflammation on 1st bx	2.4 (1.2–4.8)	0.003*
Grade 3 inflammation on 1st bx	5.7 (0.7–45.0)	0.11
Grade 1 steatosis on 1st bx	1.0 [Reference]	Reference
Grade 2 steatosis on 1st bx	1.0 (0.6–1.6)	0.94
Grade 3 steatosis on 1st bx	1.1 (0.7–1.6)	0.75
Stage 0 fibrosis on 1st bx	1.00 [Reference]	Reference
Stage 1 fibrosis on 1st bx	1.1 (0.7–1.7)	0.82
Stage 2 fibrosis on 1st bx	0.7 (0.4–1.1)	0.13
Age	0.98 (0.96–0.99)	0.009*
Obese (BMI > 30 kg/m^2)	1.1 (0.7–1.6)	0.66
Female gender	0.8 (0.5–1.3)	0.37
Diabetes (clinical history)	0.8 (0.5–1.1)	0.19
Hypertension (clinical history)	1.2 (0.8–1.8)	0.61

Argo CK et al., J Hepatol 2009

Systematic Review of Risk Factors for Fibrosis Progression in NASH



Argo CK et al., J Hepatol 2009

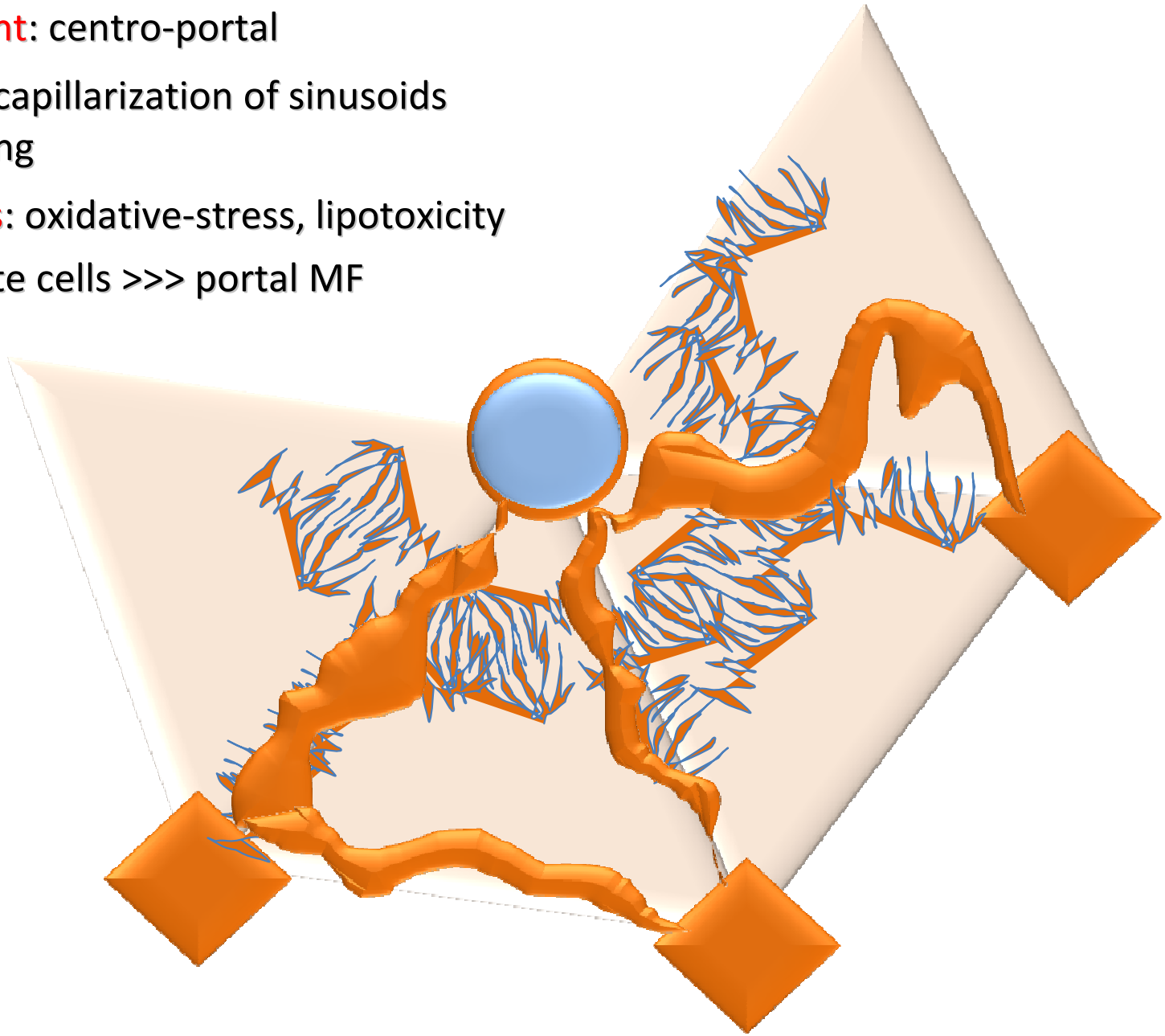
Pericellular Fibrosis and Capillarization of Sinusoids (e.g. ASH/NASH)

Pattern of Development: centro-portal

Key Events: extensive capillarization of sinusoids precedes septal bridging

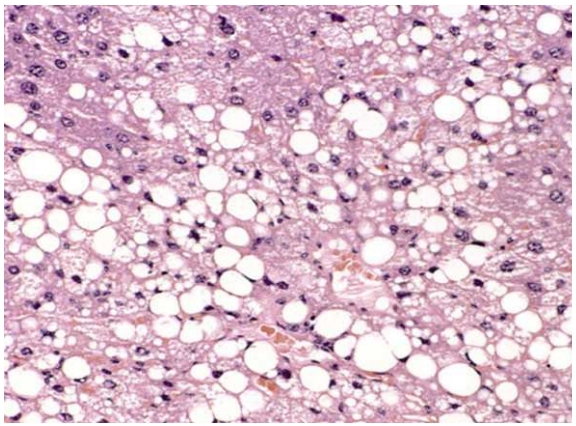
Prevalent Mechanisms: oxidative-stress, lipotoxicity

Fibrogenic cells: stellate cells >>> portal MF

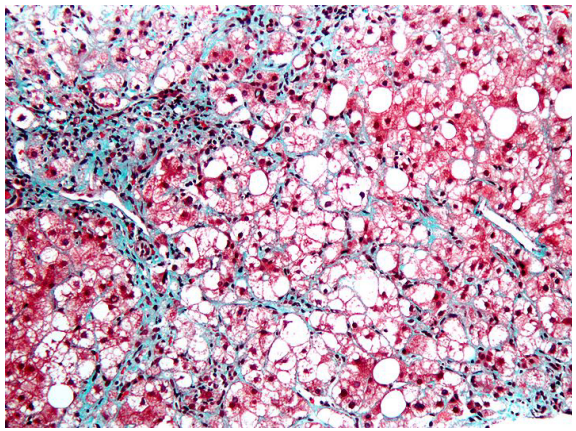


Oxidative Stress-driven Liver Fibrosis (ASH & NASH)

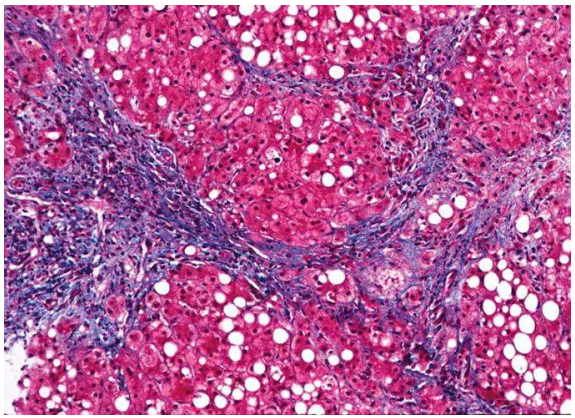
Steatosis (Fatty Liver)

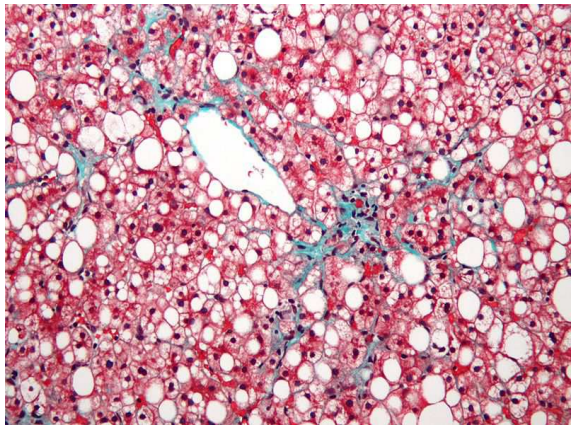


Steatohepatitis with bridging fibrosis



Cirrhosis

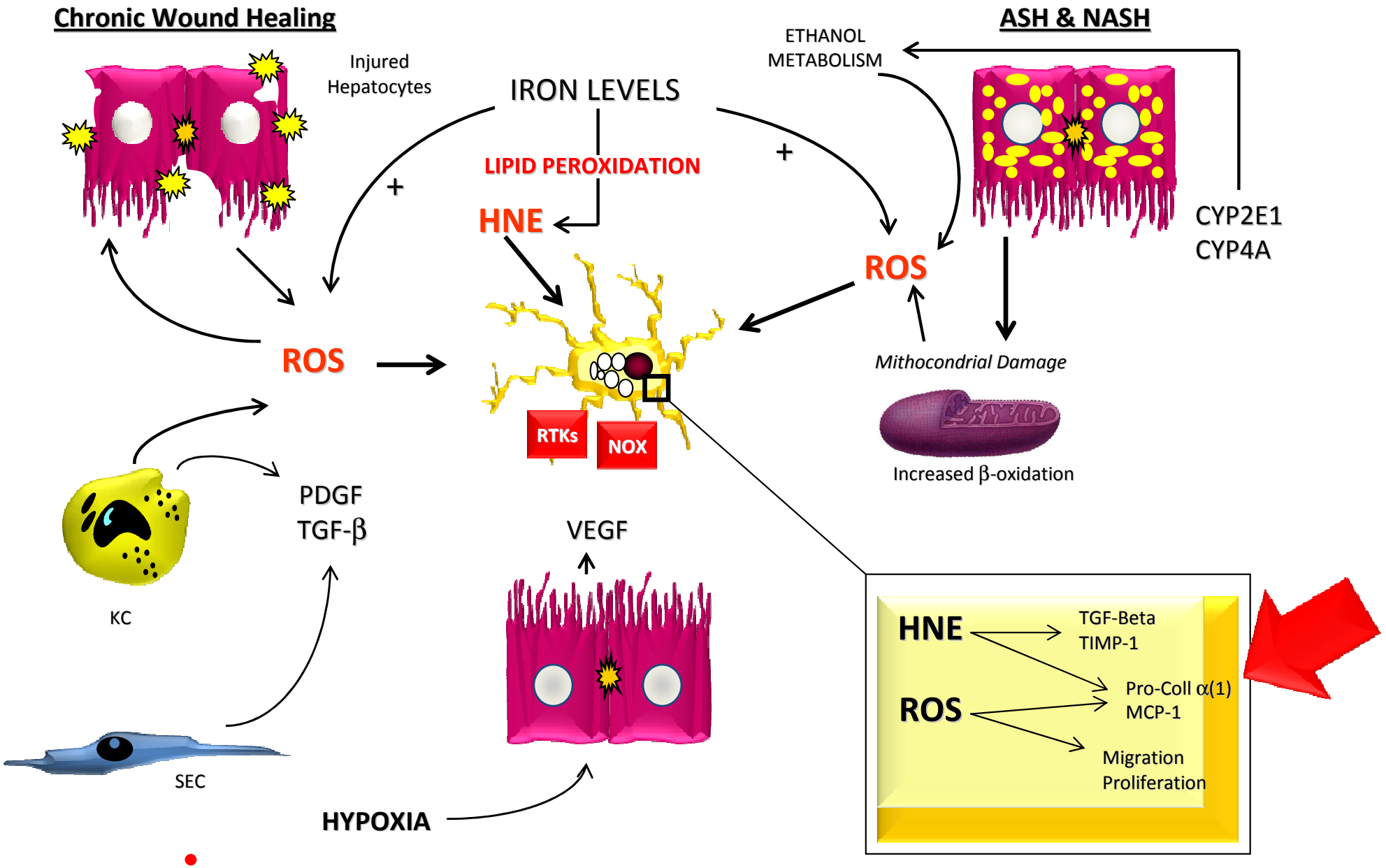




Steatosis with Pericellular Fibrosis

Fibrogenesis in ALD and NAFLD may be partially independent of cell necrosis and inflammation

ROS and Related Mediators as Pro-fibrogenic Stimuli



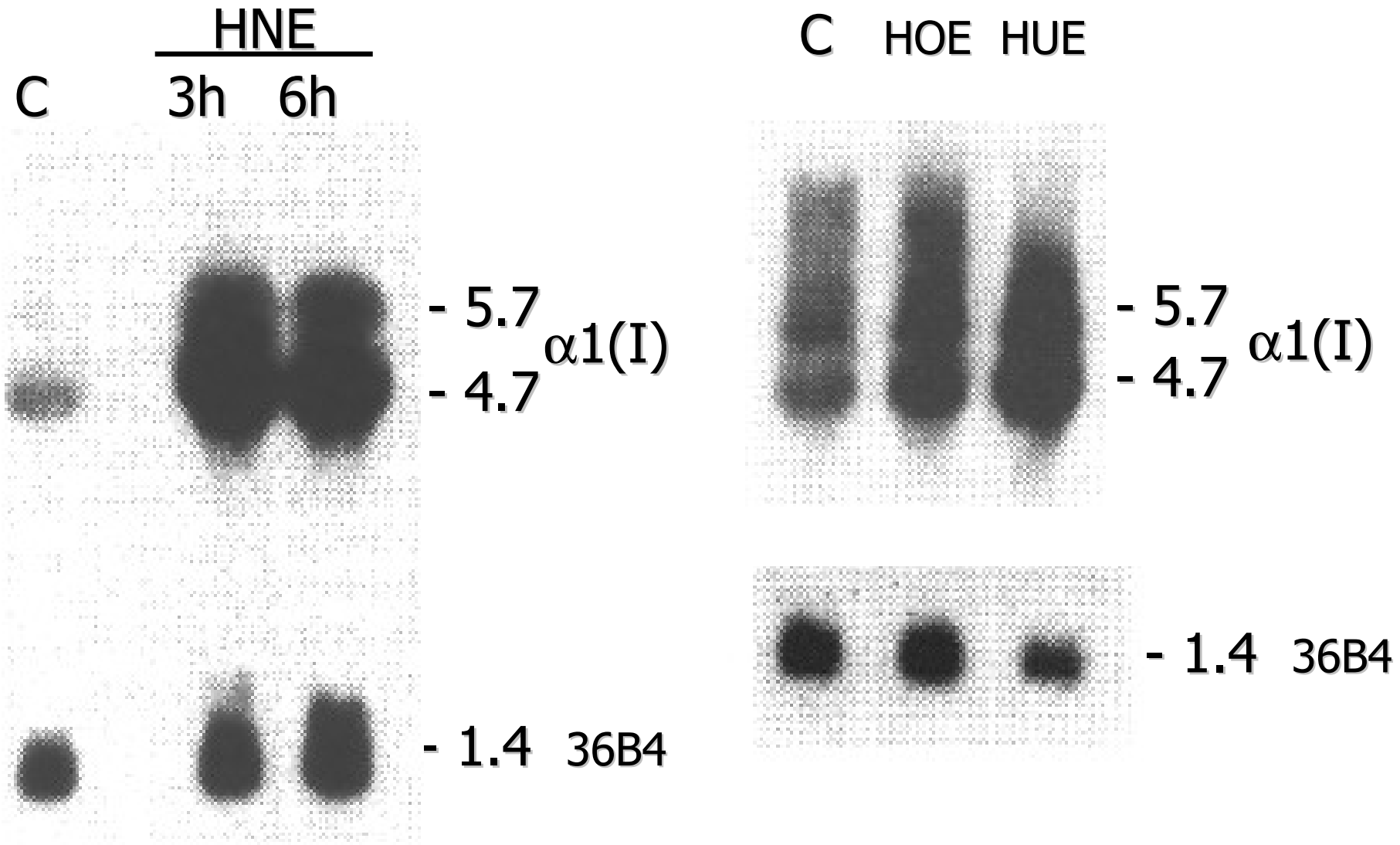
ROS: $H_2O_2, O_2^{\bullet-}$

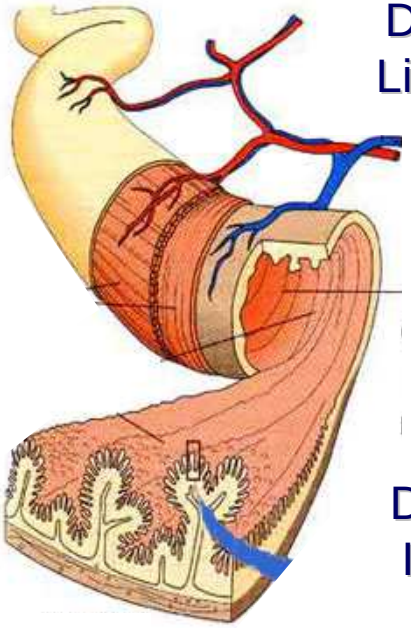
Modified from Novo E & Parola M, Fibrogenesis and Tissue Repair 2008

Reactive Aldehydes Induce Procollagen Type I mRNA Expression in Human HSC

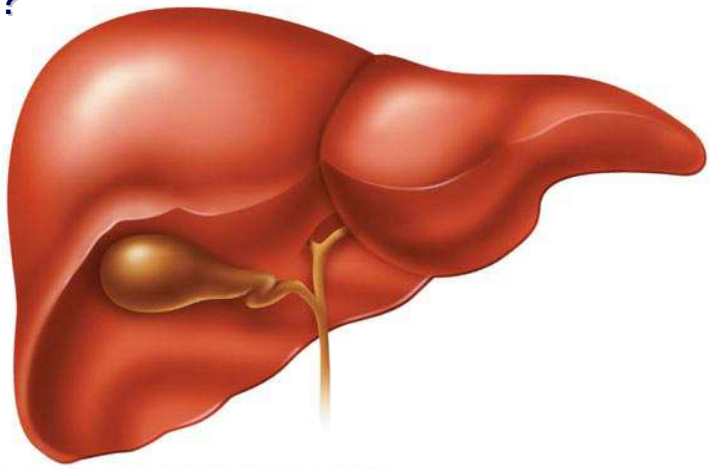
Parola M. et al., Biochem Biophys Res Comm 1996; 222:261-264

Parola M. et al., J Clin Invest 1998; 102:1942-1950





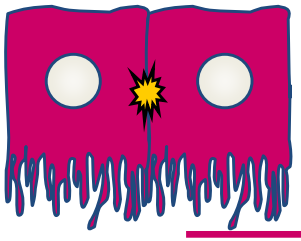
Does the Intestine Contribute to Liver Inflammation and Fibrosis?



Does the Liver Contribute to Intestinal Inflammation and Fibrosis?

The Role of PAMPs and DAMPs in Liver Inflammation and Fibrosis

Damaged /Apoptotic Hepatocytes



Activated Hepatic Stellate Cells/MFs

TLR9

TLR4

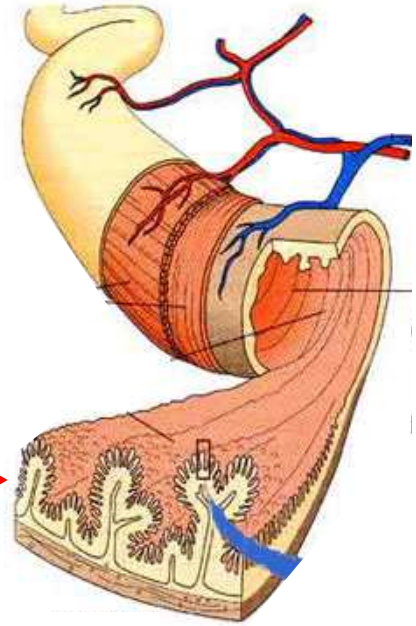
Damage-Associated Molecular Patterns "**DAMPs**" (i.e. apoptotic hepatocyte DNA)

Increased Collagen I synthesis

Increased Pro-fibrogenic (TGF-β) and Pro-inflammatory (IL-6, MCP-1) action

Chronic Liver Disease

- **Changes in Microbiota Bacterial Overgrowth**
- **Decreased Bile Acids**
- **Mucosal Injury (ETOH)**
- **Mesenteric Vein Congestion**
- **Immunosuppression**
- **Obesity/Overweight**
- **High Fat Diet**



Increased *Pathogen-Associated Molecular Patterns* "**PAMPs**" (i.e. LPS) in the portal circulation (proportional to disease stage)

Brun P., et al., Am J Physiol Gastrointest Liver Physiol 2005; 289:G571-G578
Brun P., et al., Am J Physiol Gastrointest Liver Physiol 2007; 292:G518-G525
Seki E and Brenner DA. Hepatology 2008; 48:322-335
Seki E, et al., Nat Med. 2007; 13:1324-1332
Watanabe A, et al. Hepatology 2007; 46:1509–1518

Pinzani M and Macias Barragan J. Expert Rev Gastroenterology and Hepatology 2010

Fructose Consumption is Associated with Fibrosis Severity in Patients with NAFLD

Table 4. Association Between Fructose Consumption and Histologic Feature of NAFLD in the Entire Study Population

	Unadjusted		Adjusted (Model 1)		Adjusted (Model 2)	
	OR[95%CI]	*P* Value	OR[95%CI]	*P* Value	OR[95%CI]	*P* Value
Steatosis						
Fructose consumption						
0 serving	-	-	-	-	-	-
0-7 servings	0.7 [0.4, 1.1]	0.09	0.6 [0.4, 0.9]	0.02	0.7 [0.4, 1.1]	0.10
≥7 servings	0.6 [0.4, 1.0]	0.06	0.4 [0.2, 0.8]	0.007	0.4 [0.2, 0.9]	0.02
Lobular inflammation						
Fructose consumption						
0 serving	-	-	-	-	-	-
0-7 servings	0.8 [0.5, 1.3]	0.30	0.9 [0.5, 1.4]	0.55	0.8 [0.5, 1.4]	0.53
≥7 servings	0.6 [0.4, 1.0]	0.06	0.9 [0.5, 1.8]	0.86	1.1 [0.6, 2.3]	0.70
Ballooning						
Fructose consumption						
0 serving	-	-	-	-	-	-
0-7 servings	0.7 [0.4, 1.1]	0.13	0.9 [0.5, 1.4]	0.62	0.9 [0.5, 1.5]	0.73
≥7 servings	0.7 [0.4, 1.2]	0.25	1.3 [0.7, 2.4]	0.44	1.4 [0.7, 2.7]	0.32
Fibrosis						
Fructose consumption						
0 serving	-	-	-	-	-	
0-7 servings	0.6 [0.4, 0.9]	0.01	0.8 [0.5, 1.3]	0.44	0.9 [0.6, 1.5]	0.78
≥7 servings	0.7 [0.4, 1.2]	0.19	1.7 [1.0, 3.2]	0.07	2.6 [1.4, 5.0]	0.004

Fructose consumption is expressed as reported servings per week. Cumulative odds ratio (OR) and *P*-values were derived from ordinal logistic regression models (Model 1: adjusted for age, sex, BMI, Hispanic ethnicity, and total calorie intake; Model 2: adjusted for age, sex, BMI, Hispanic ethnicity, total calorie intake, triglycerides, HDL-cholesterol, LDL-cholesterol, uric acid, and HOMA-IR).

Abdelmalek et al., Hepatology 2010;51:1961

Fructose as Bad as Alcohol?

1. – Fructose can be metabolized only by the liver and excess leads to insulin-resistance.

2.- Fructose reacts with proteins and polyunsaturated fats generating AGEs (advanced glycation end-products), which amplify oxidative damage.

3. – Chronic excess leads to dyslipidemia and to leptin resistance and obesity.

4. – Fructose causes intestinal bacterial overgrowth and growth of pathogenic bacteria.

5. – Cancer cells favor fructose as energy source .

Alcoholic and Non-alcoholic Fatty Liver in Adolescents: a Worrisome Convergence

Nobili V and Pinzani M, Alcohol and Alcoholism 2011

Metabolic

NAFLD/ NASH

Oxidative stress: reactive oxygen species (ROS) and reactive aldehydes, acetaldhehyde (ETOH)

Insulin resistance, increased FFA synthesis, ER stress (lipotoxicity)

Increased intestinal permeability: PAMPs to the liver and interaction with pro-fibrogenic TLRs

TISSUE DAMAGE, INFLAMMATION, FIBROGENESIS, ANGIOGENESIS

Binge Drinking

ASH

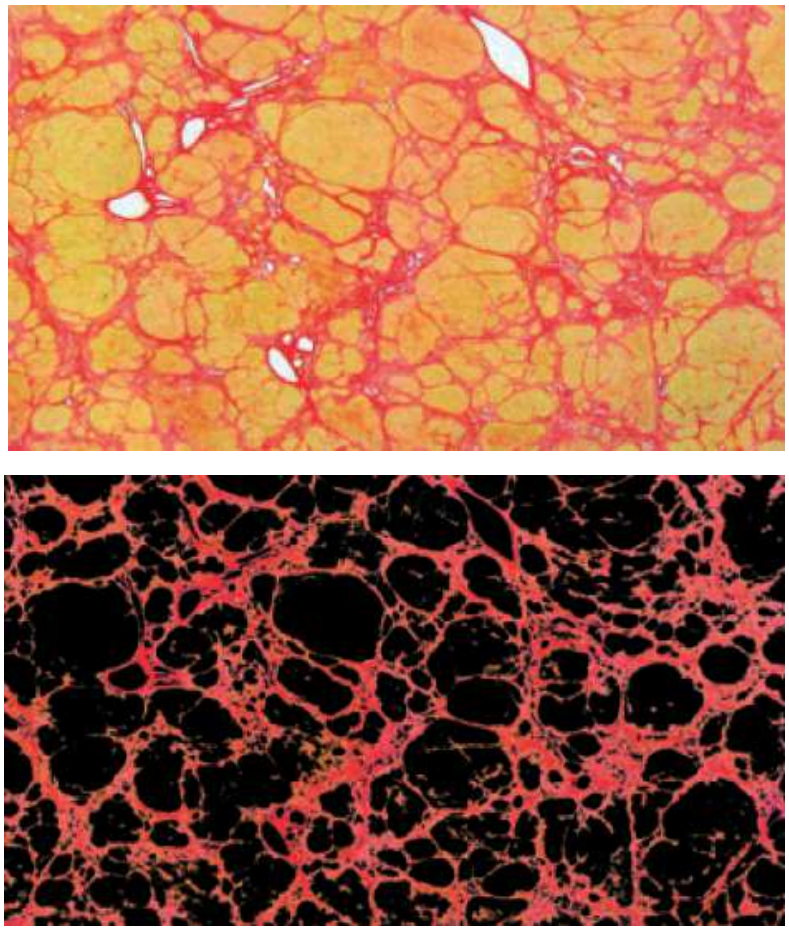
The Diversity of Chronic Liver Diseases

ONE OR MANY CIRRHOSIS ????

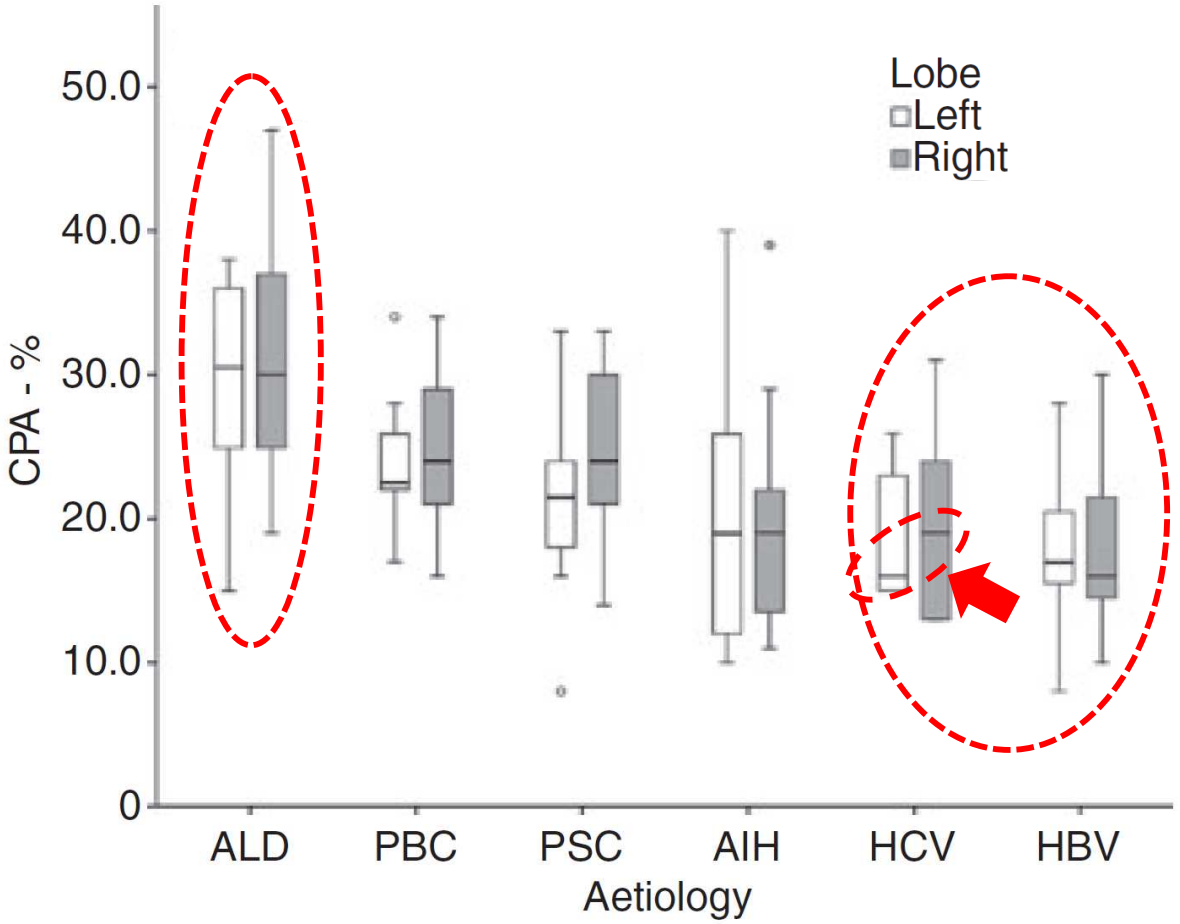
ETIOLOGY !!

Fibrosis Quantity and Distribution in Explanted Cirrhotic Liver Depending on Etiology

Hall A. et al., Histopathology 2012; 60:270-277



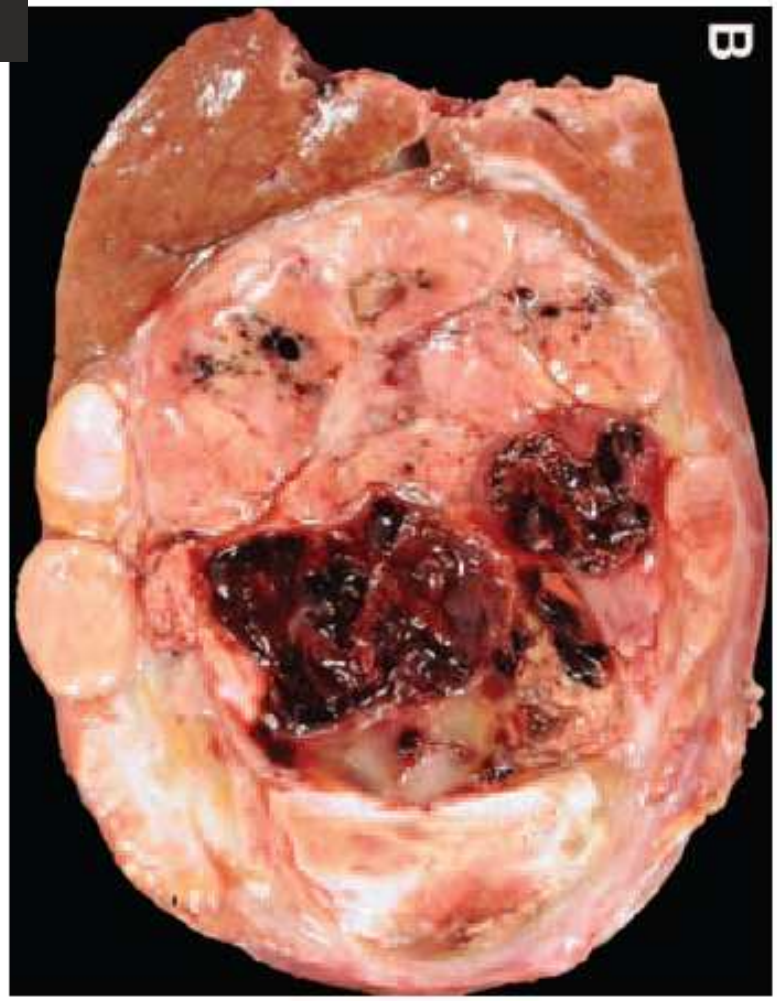
Measurement of Collagen Proportionate Area (CPA)



Hepatocellular Carcinomas in Patients With Metabolic Syndrome Often Develop Without Significant Liver Fibrosis: A Pathological Analysis

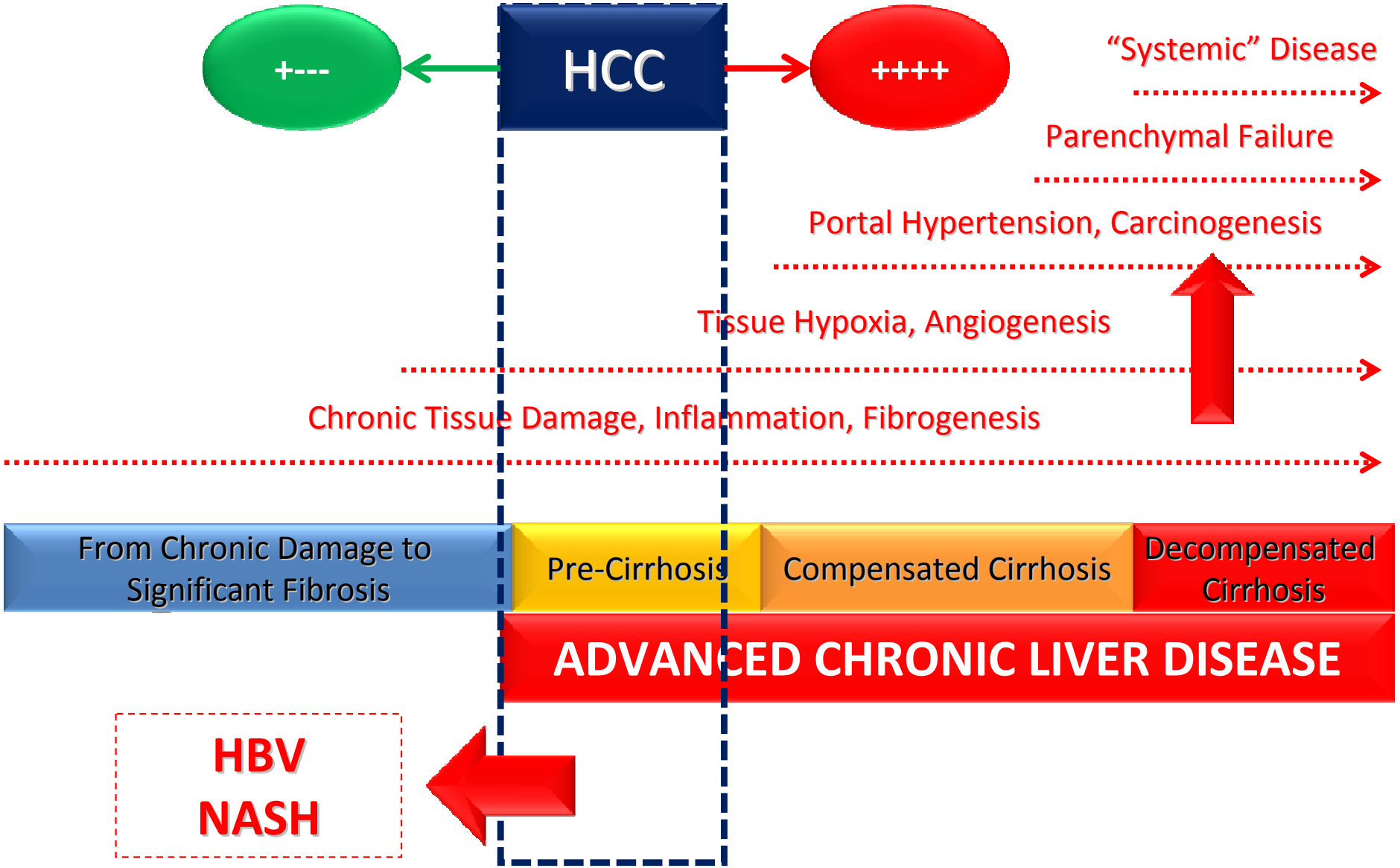
Valérie Paradis,[1,5] Stéphane Zalinski,[2] Emna Chelbi,[1] Nathalie Guedj,[1,5] Françoise Degos,[2] Valérie Vilgrain,[3] Pierre Bedossa,[1,5] and Jacques Belghiti[4]

Hepatology 2009;49:851-859



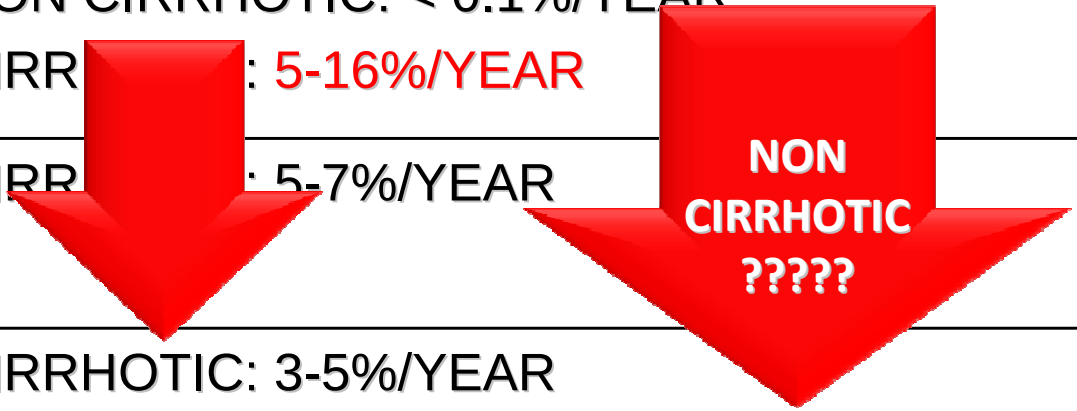
- 31 HCC with metabolic syndrom only
- HCC in liver without significant fibrosis (stage 0-2) is more common than HCC in cirrhosis (65% / 35%) to be compared with viral hepatitis (30% / 70%)
- Malignant degenerescence of liver cell adenomas (telangiectatiac adenomas), 5/31
- Well-differentiated HCC, large size

Progression of CLD: Key Pathophysiological Points



Incidence of HCC According to Different Chronic Fibrogenic Liver Diseases

CHRONIC HBV INFECTION	NON CIRRHOTIC: 0.4-0.6%/YEAR CIRRHOTIC: 5-19%/YEAR
CHRONIC HCV INFECTION	NON CIRRHOTIC: < 0.1%/YEAR CIRR : 5-16%/YEAR
GENETIC HEMOCHROMATOSIS	CIRR : 5-7%/YEAR
NAFLD/NASH	CIRRHOTIC: 3-5%/YEAR >> OBESITY AND DIABETES INDEPENDENTLY ASSOCIATED WITH INCREASED RISK OF CANCER
ALCOHOLIC LIVER DISEASE	CIRRHOTIC: 5-8%/YEAR >> ALCOHOLISM INDEPENDENTLY ASSOCIATED WITH INCREASED RISK OF CANCER
AUTOIMMUNE HEP., PBC, PSC, WILSON DISEASE	LOW INCIDENCE OF HCC (< 0.5 %/YEAR)



Obesity and Cancer Mortality

Calle E *et al*., NEJM 2003; 348:1625-1638

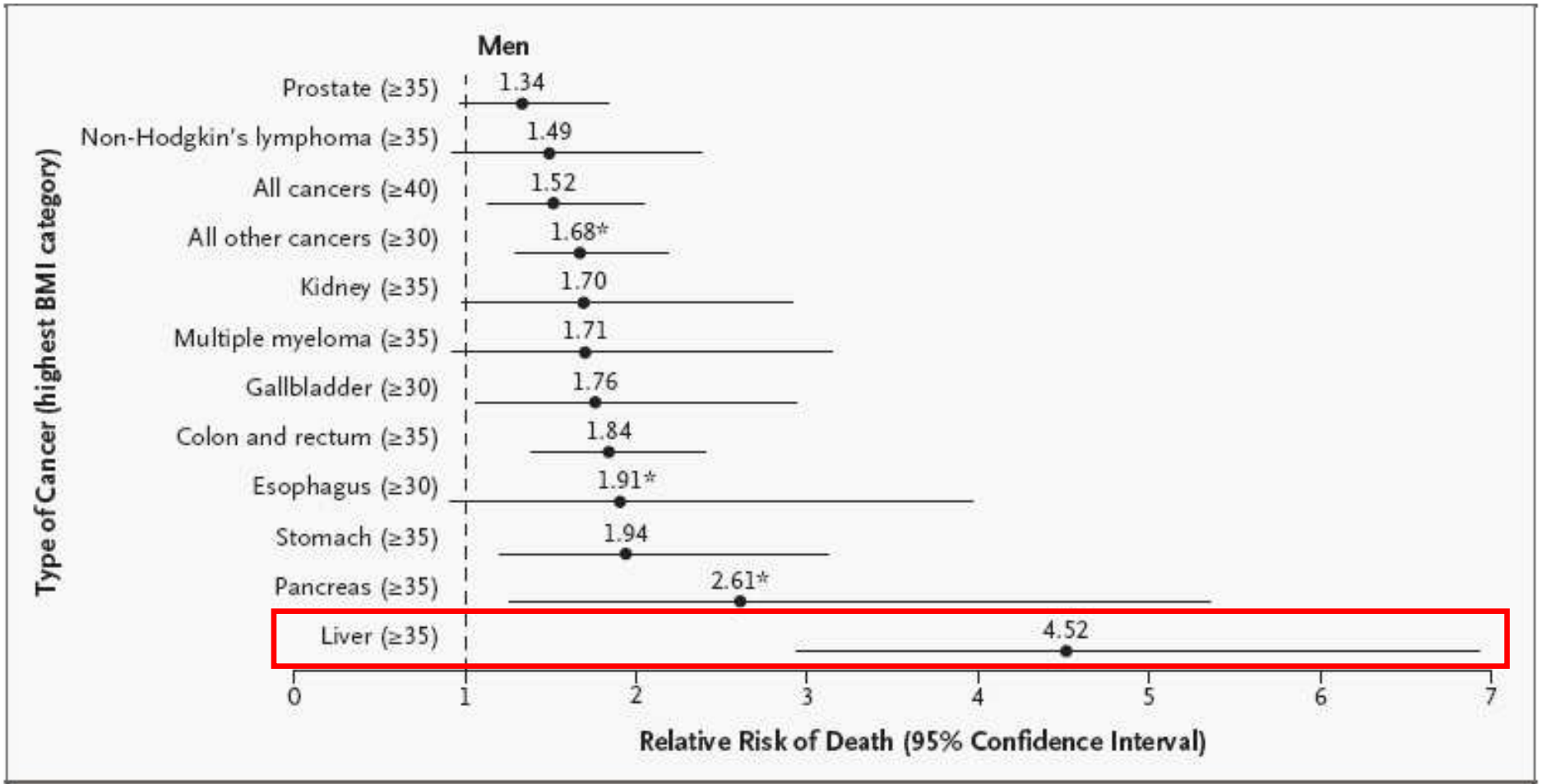


Figure 1. Summary of Mortality from Cancer According to Body-Mass Index for U.S. Men in the Cancer Prevention Study II, 1982 through 1998.

For each relative risk, the comparison was between men in the highest body-mass-index (BMI) category (indicated in parentheses) and men in the reference category (body-mass index, 18.5 to 24.9). Asterisks indicate relative risks for men who never smoked. Results of the linear test for trend were significant (P≤0.05) for all cancer sites.

Overweight, Obesity and Risk of HCC: a Meta-Analysis of Cohort Studies

Larsson SC and Wolk A, British J Cancer 2007; 97:1005-1008

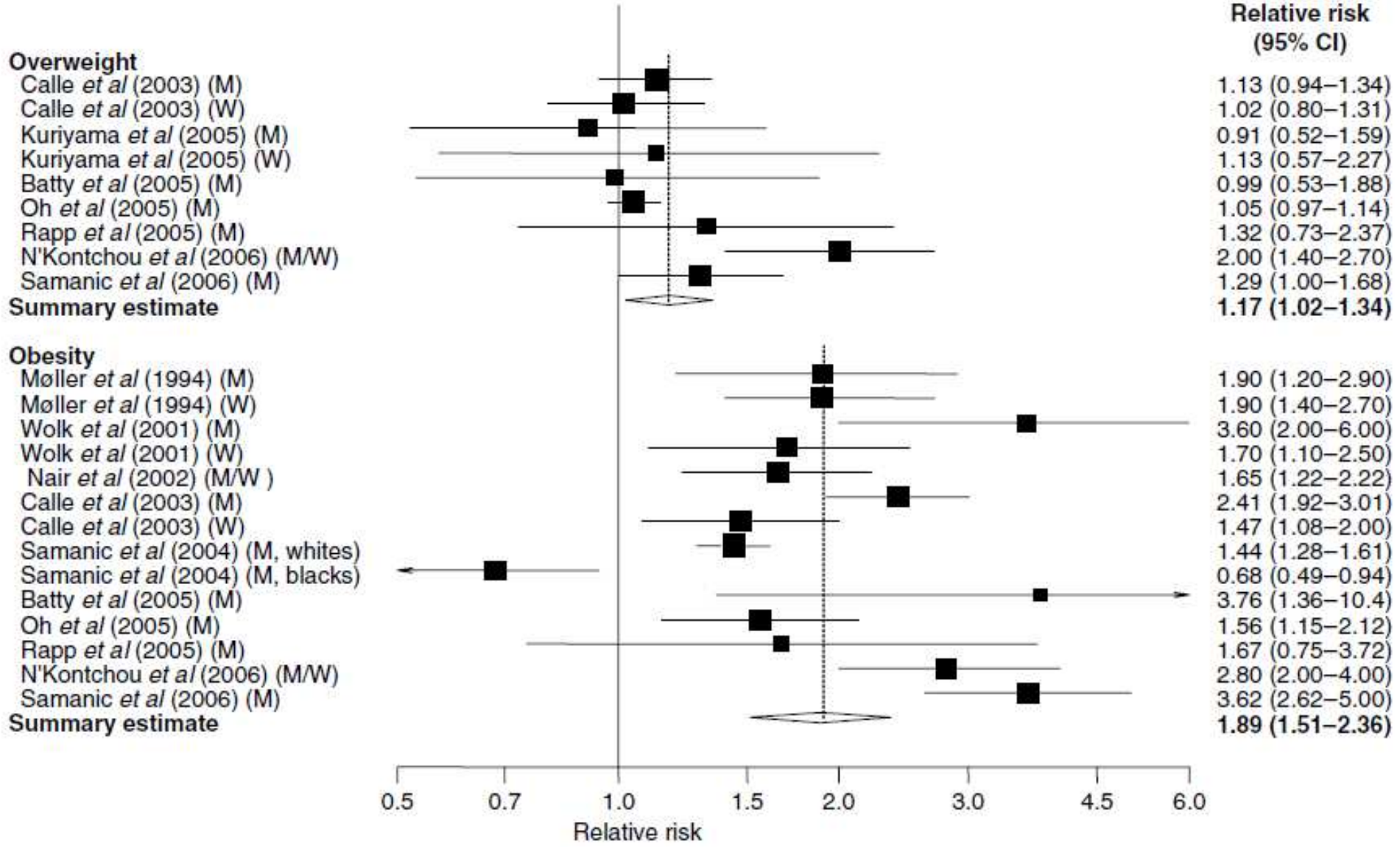


Figure 1 Relative risks of liver cancer associated with overweight and obesity. Relative risk estimates are for overweight and obese persons compared with normal weight persons. Tests for heterogeneity: overweight, $Q = 16.83$, $P = 0.03$; $I^2 = 52.5\%$; obesity, $Q = 88.03$, $P < 0.001$; $I^2 = 86.4\%$. M = men; W = women.

GASTROENTEROLOGY 2008;135:111–121

Metabolic Factors and Risk of Hepatocellular Carcinoma by Chronic Hepatitis B/C Infection: A Follow-up Study in Taiwan

CHI–LING CHEN,[*] HWAI–I YANG,[‡] WEI–SHIUNG YANG,[*,§] CHUN–JEN LIU,[*,§,‖] PEI–JER CHEN,[*,§,‖] SAN–LIN YOU,[‡] LI–YU WANG,[¶] CHIEN–AN SUN,[#] SHENG–NAN LU,[**] DING–SHIN CHEN,[*,§,‖] and CHIEN–JEN CHEN,[‡,‡‡]

	RR HCC (95% IC)
Controls	1
Diabetes HCV-/HBV-	3.49 (1.08-11.3)
Diabetes HCV+	60.3 (23.6-153.6)
Diabetes HBV+	43.5 (20.5-92.3)
Obesity-Diabetes-HCV +	134.5 (17.5-1035)
Obesity-Diabetes-HBV +	264.7 (35.2-1993)

23.820 patients followed for 14 yrs

Primary Prevention: EASL-EORTC Guidelines

Geographical Distribution and Main Risk Factors for HCC Worldwide*

Geographic area	AAIR M/F	Risk factors HCV (%)	Risk factors HBV (%)	Alcohol (%)	Others (%)
Europe	6.7/2.3	60-70	10-15	20	10
Southern	10.5/3.3				
Northern	4.1/1.8				
North America	6.8/2.3	50-60	20	20	10 (NASH)
Asia and Africa		20	70	10	10 (Aflatoxin)
Asia	21.6/8.2				
China	23/9.6				
Japan	20.5/7.8	70	10-20	10	10
Africa	1.6/5.3				
WORLD	16/6	31	54	15	

*Updated from Llovet *et al.* [99], according to IARC data [4]. AAIR, age-adjusted incidence rate.

Categories of Adult Patients in Whom Surveillance is Recommended

1. Cirrhotic patients, Child-Pugh stage A and B*
2. Cirrhotic patients, Child-Pugh stage C awaiting liver transplantation**
3. Non-cirrhotic HBV carriers with active hepatitis or family history of HCC***
4. Non-cirrhotic patients with chronic hepatitis C and advanced liver fibrosis F3****

*Evidence 3A; strength B1;

**evidence 3D; strength B1;

***evidence 1B; strength A1 for Asian patients; evidence 3D; strength C1 for Western patients;

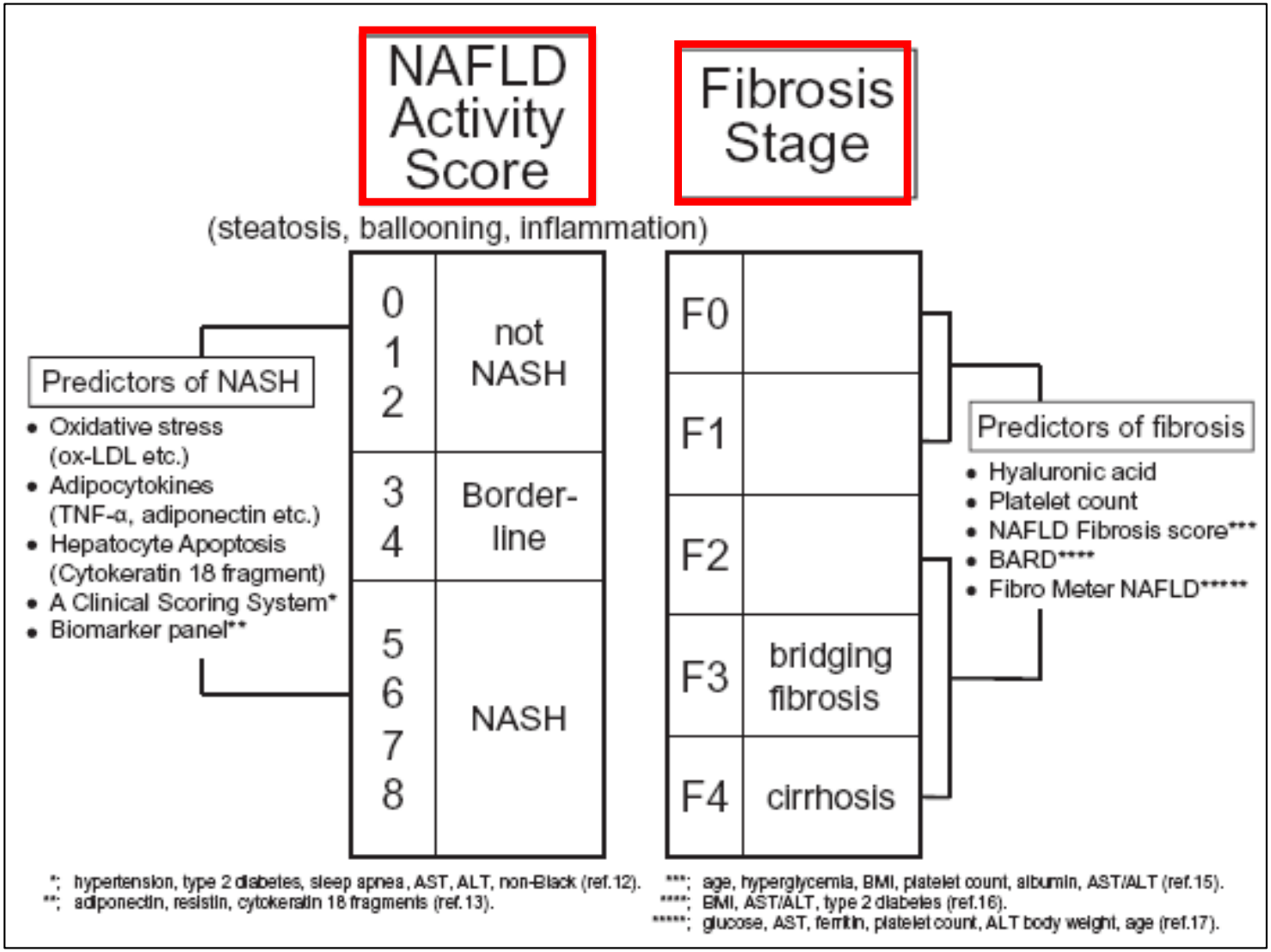
****evidence 3D; strength B1 for Asian patients; evidence 3D; strength B2 for Western patients.

EASL–EORTC Clinical Practice Guidelines: Management of hepatocellular carcinoma

European Association for the Study of the Liver*,
European Organisation for Research and Treatment of Cancer

Journal of Hepatology **2012** vol. 56 | 908–943

Serum and Clinical Predictors of NASH in NAFLD



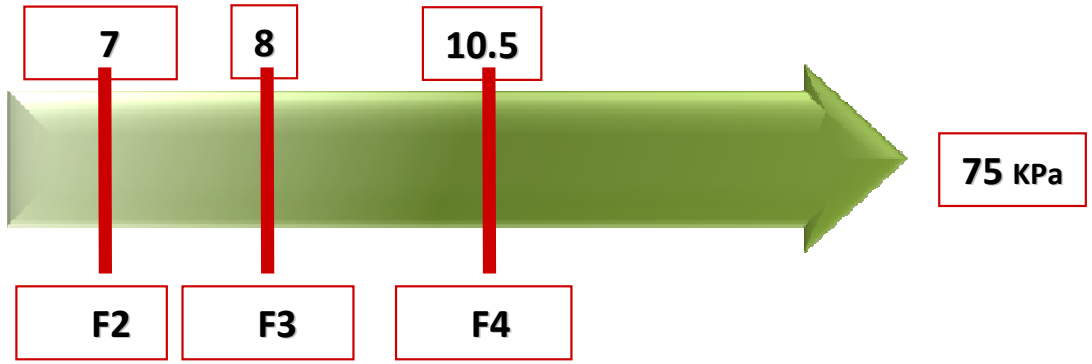
Transient Elastography in NASH

Fibroscan®

- assessment of fibrosis measuring liver stiffness
- initial promising results in NAFLD *Wong, Hepatology 2010*

BUT

- further validation needed
- Failure Rate → 25.5% if BMI≥ 30 and 2.6% if BMI < 30
- special probe for obese patients *De Ledinghen, J Hepatol 2009*
- caution in NAFLD => results may be influenced by steatosis *Gaia, J Hepatol 2011*

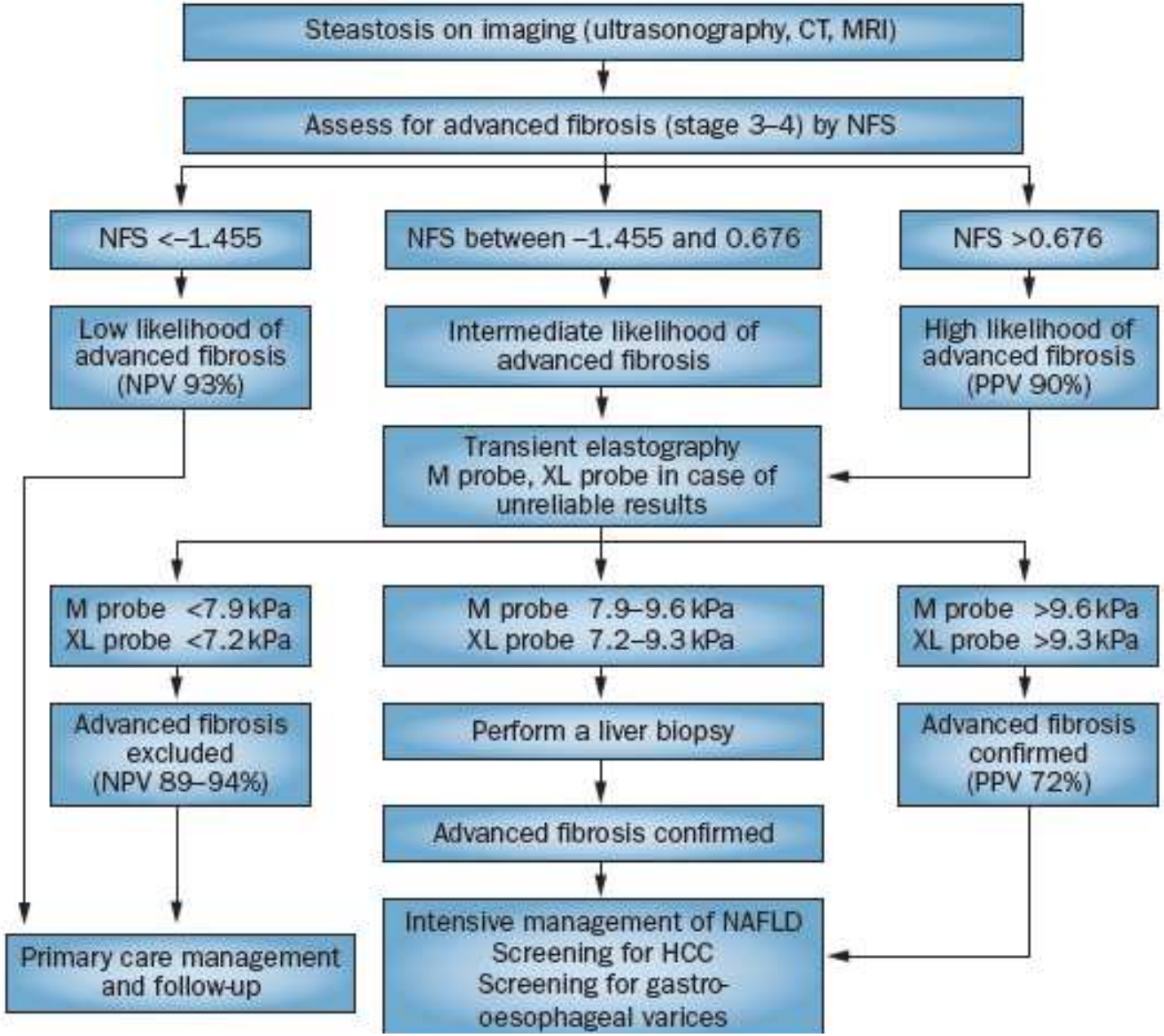


Stiffness: 7.2 Kpa
(IQR 2.5, SR 70%)

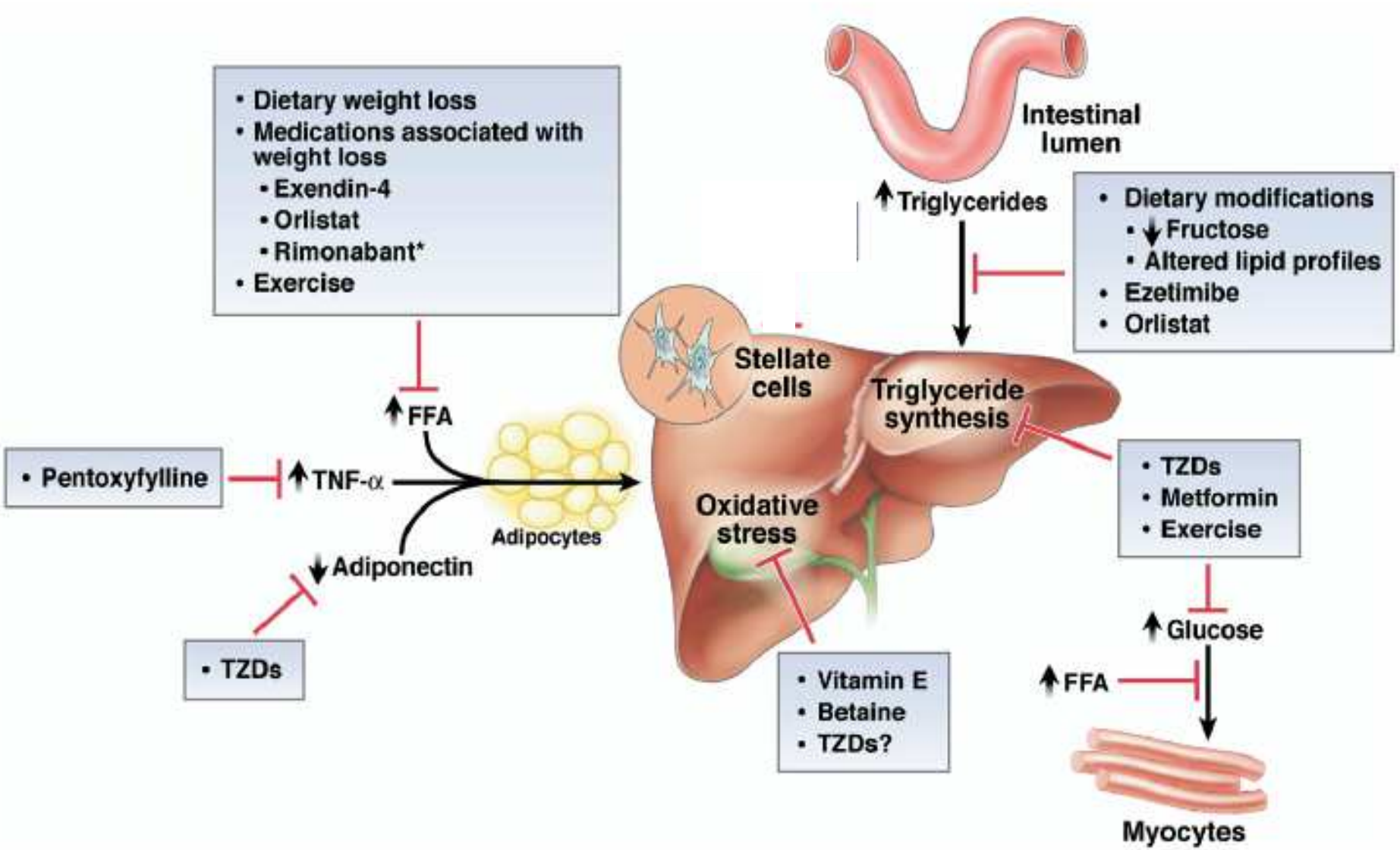
NAFLD fibrosis score: –1,017 (“grey area”)

Noninvasive evaluation of NAFLD

Laurent Castera, Valérie Vilgrain and Paul Angulo



Targets for Therapy



Torres & Harrison, Gastroenterology 2008

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www.ucl.ac.uk/medicine/liver-and-digestive-health