Inborn Errors of Bile Acid Metabolism

James E. Heubi, M.D.

Professor, Department of Pediatrics

Associate Dean for Clinical and Translational Research

Director of Center for Clinical and Translational Science and Training

University of Cincinnati College of Medicine

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- Consultant to Nordmark, Retrophin, Alnylam

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Outline

- Causes and evaluation for neonatal cholestasis
- Role of bile acids in cholestasis and fat/fat soluble vitamin absorption
- Pathophysiology of inborn errors of BA metabolism
- Diagnosis of inborn errors of BA metabolism
- Commonly identified defects
- Treatment

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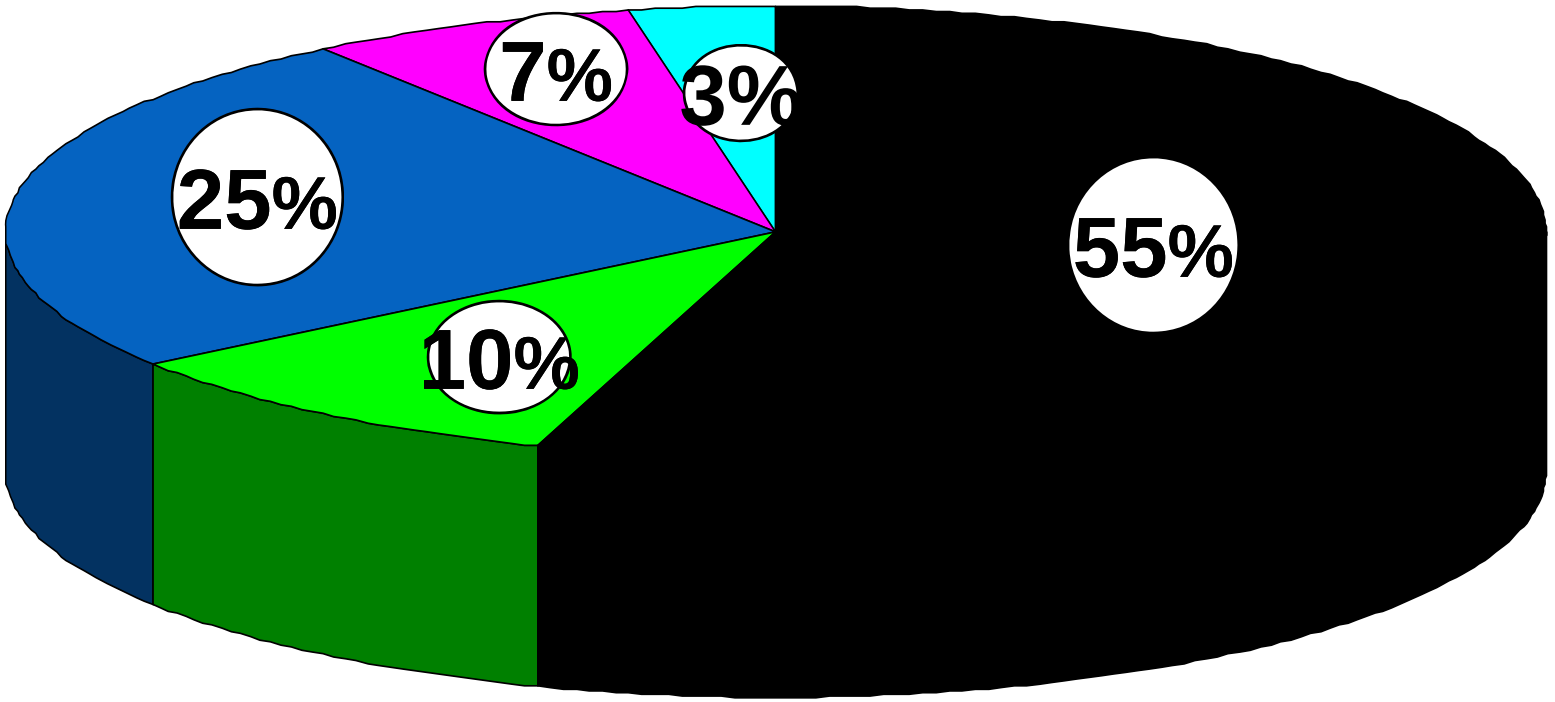
Differential - “Neonatal Cholestasis” 1973



- "Neonatal Hepatitis"
- A-1-AT deficiency
- Biliary Atresia
- Miscellaneous*
- "Viral" (TORCH)

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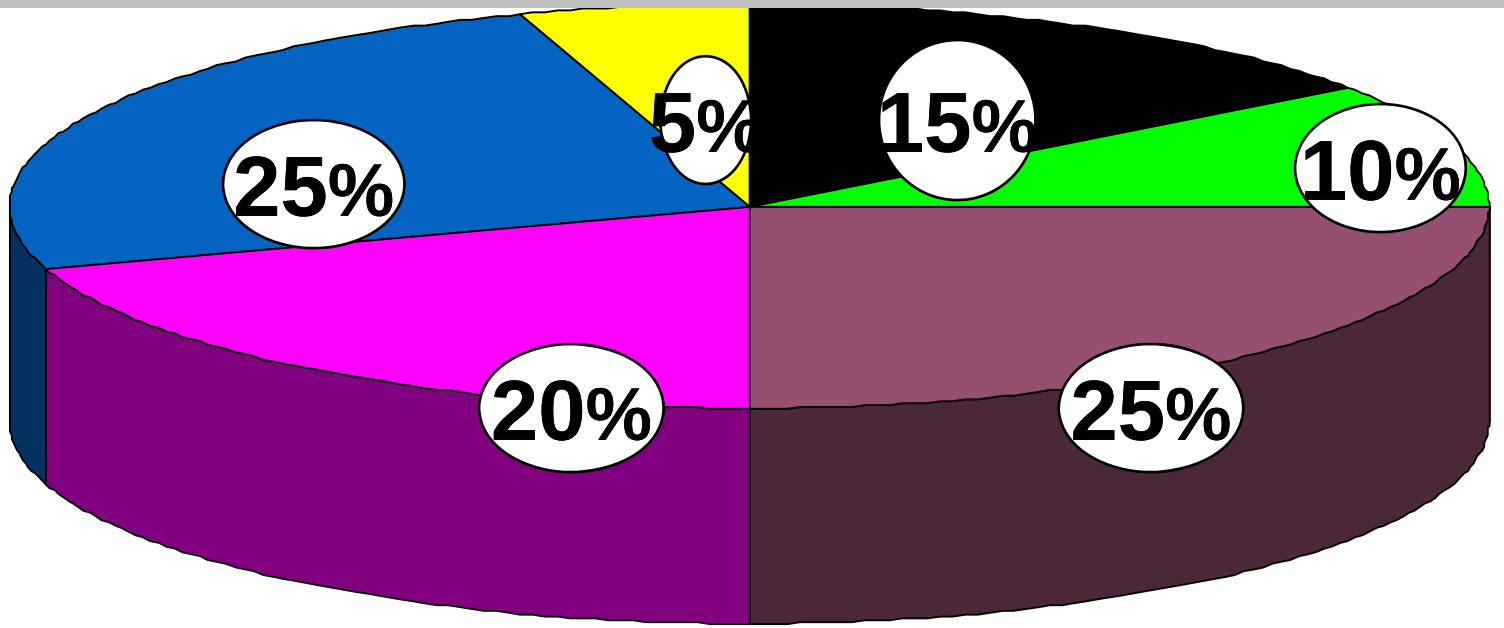
Differential - "Neonatal Cholestasis" 2016



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Role of Bile Acids

- Major metabolic pathway for elimination of cholesterol
- Promote formation/ secretion of bile
- Fat and fat soluble vitamin absorption
- Cathartic action-induce water and electrolyte secretion
- Bacteriostatic properties
- Role in signaling pathways

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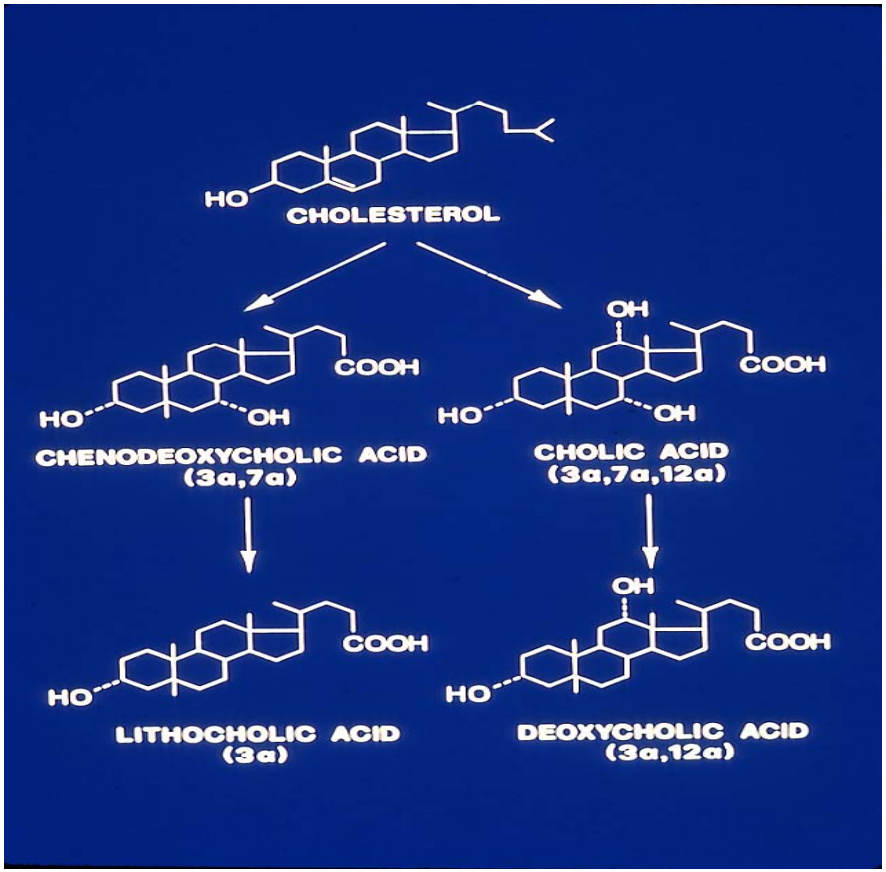
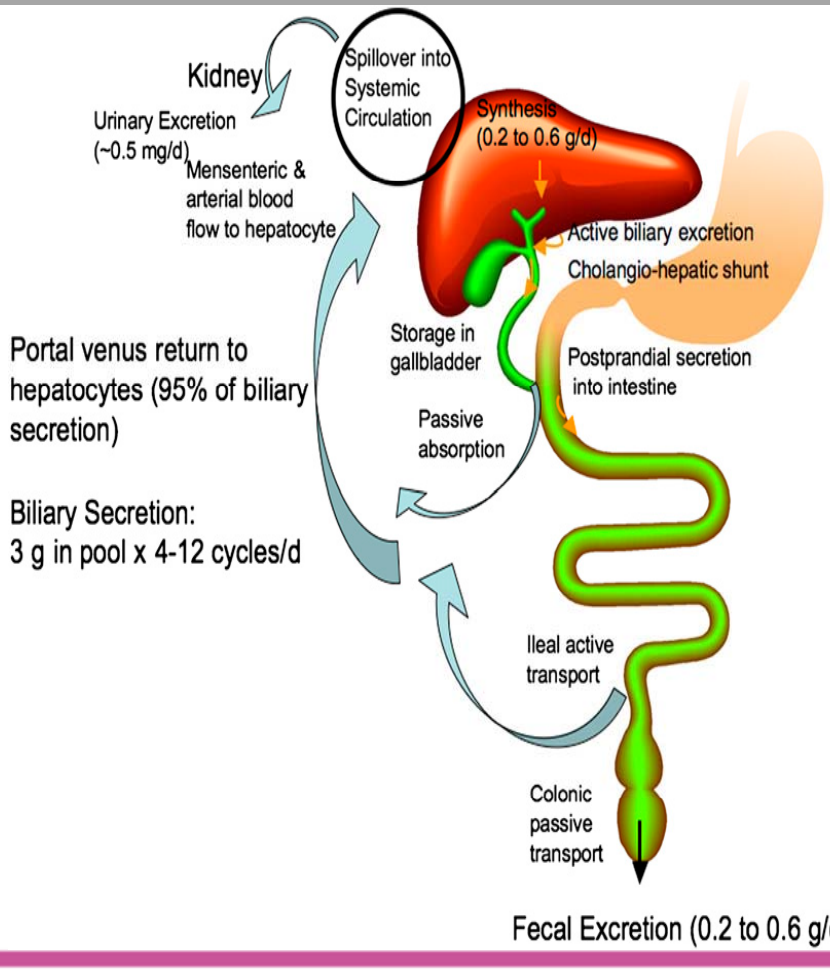
EHC and BA Metabolism 101





Biochemical Anomalies/ Hepatotoxicity of Bile Acid Synthesis Disorders[1-3]

- Lack of, or markedly diminished synthesis of primary bile acids, CA and CDCA→ poor bile flow

- Concomitant production and accumulation of precursors in the pathway proximal to enzyme defect→metabolites may be directly toxic

1. Bove KE, et al. *Pediatr Dev Pathol*. 2000;3(1):1-16.
2. Witzleben CL, et al. *Pediatr Pathol.* 1992;12(2):269-274.
3. Setchell KD, Heubi JE. *J Pediatr Gastroenterol Nutr*. 2006;43(suppl 1):S17-S22.

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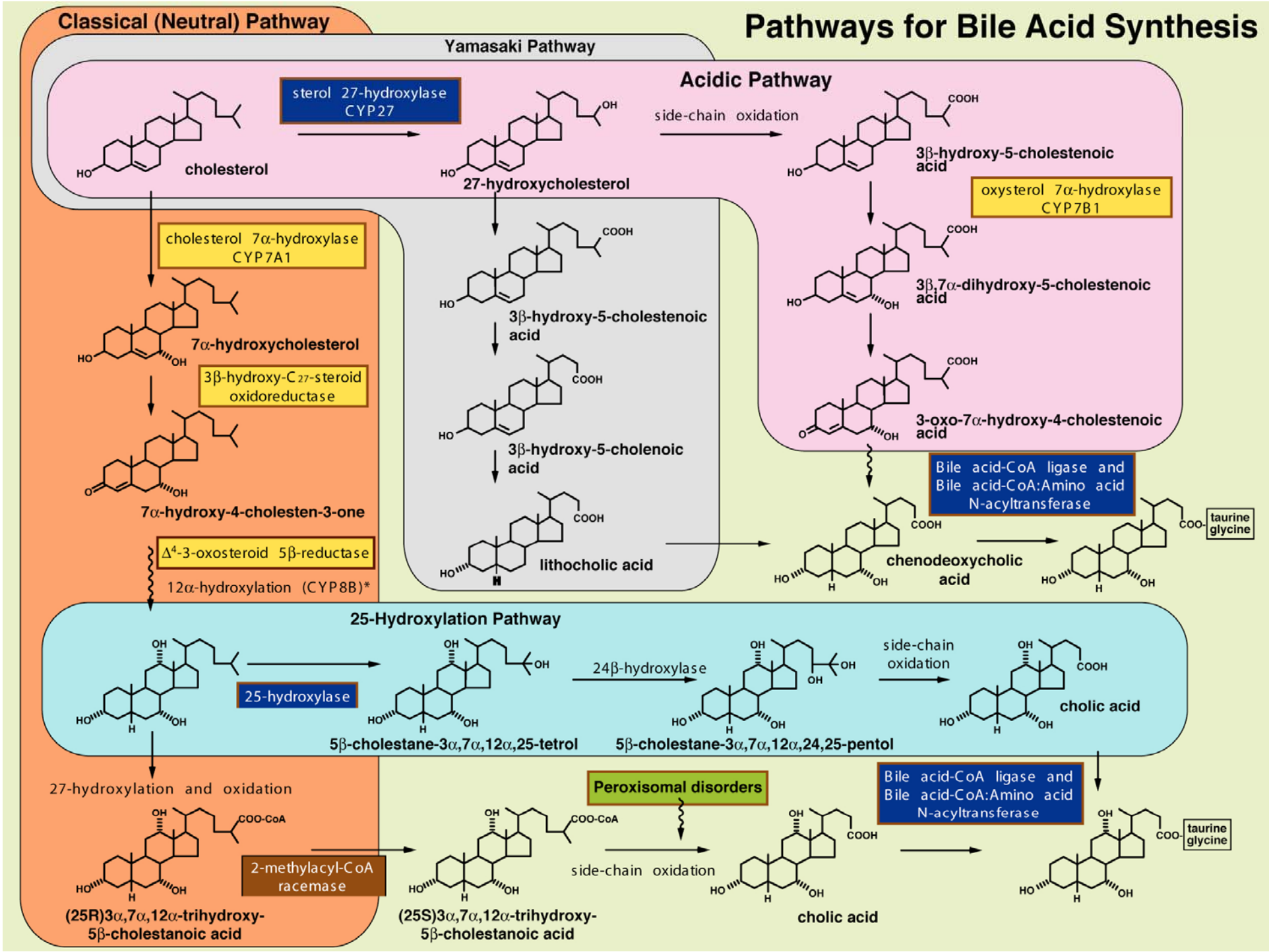
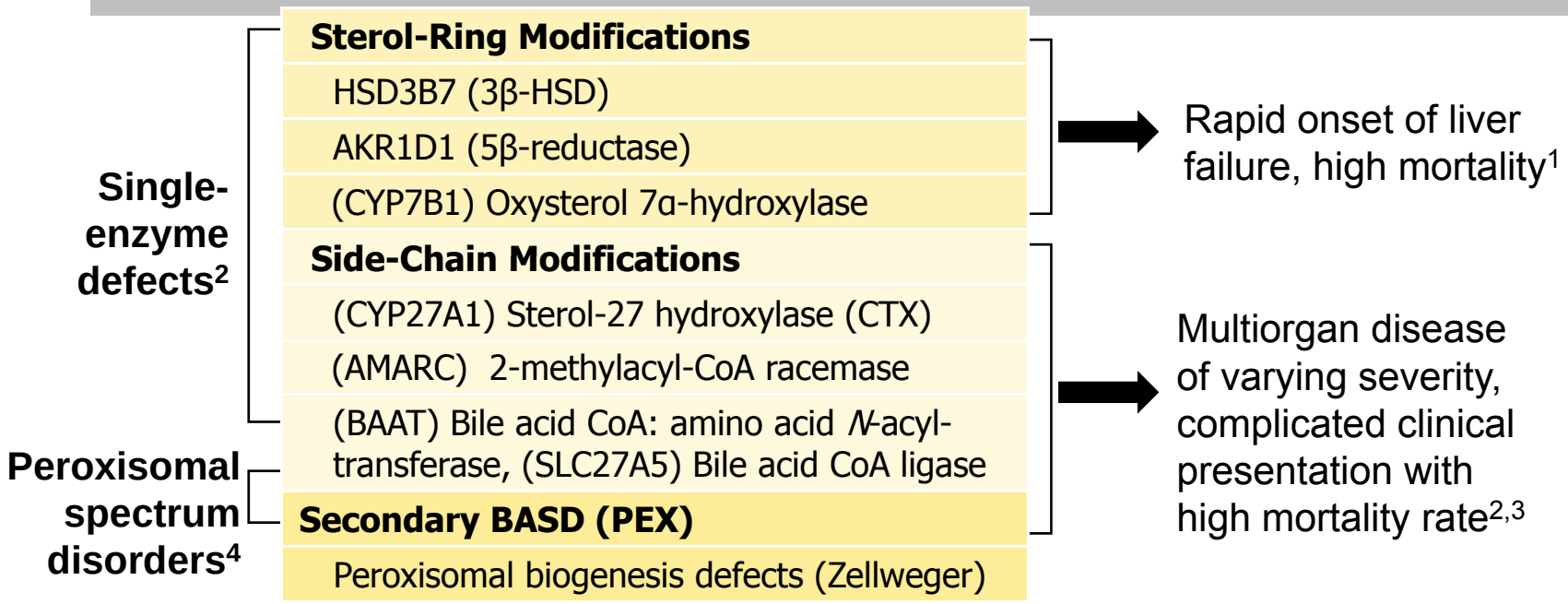

Pathways for Bile Acid Synthesis
Classical (Neutral) Pathway
Yamasaki Pathway
Acidic Pathway
sterol 27-hydroxylase CYP27
cholesterol
27-hydroxycholesterol
side-chain oxidation
3β-hydroxy-5-cholestenoic acid
oxysterol 7α-hydroxylase CYP7B1
3β,7α-dihydroxy-5-cholestenoic acid
3-oxo-7α-hydroxy-4-cholestenoic acid
cholesterol 7α-hydroxylase CYP7A1
7α-hydroxycholesterol
3β-hydroxy-C27-steroid oxidoreductase
7α-hydroxy-4-cholesten-3-one
3β-hydroxy-5-cholenoic acid
lithocholic acid
Bile acid-CoA ligase and Bile acid-CoA:Amino acid N-acyltransferase
chenodeoxycholic acid
taurine
glycine
Δ4-3-oxosteroid 5β-reductase
12α-hydroxylation (CYP8B)*
25-Hydroxylation Pathway
25-hydroxylase
5β-cholestane-3α,7α,12α,25-tetrol
24β-hydroxylase
5β-cholestane-3α,7α,12α,24,25-pentol
side-chain oxidation
cholic acid
27-hydroxylation and oxidation
Peroxisomal disorders
2-methylacyl-CoA racemase
(25R)3α,7α,12α-trihydroxy-5β-cholestanoic acid
(25S)3α,7α,12α-trihydroxy-5β-cholestanoic acid
side-chain oxidation
cholic acid
Bile acid-CoA ligase and Bile acid-CoA:Amino acid N-acyltransferase
taurine
glycine

Clinical Sequelae of BASD: SED vs PD



Clinical phenotype is highly variable — high index of suspicion based on physical examination and laboratory evaluation

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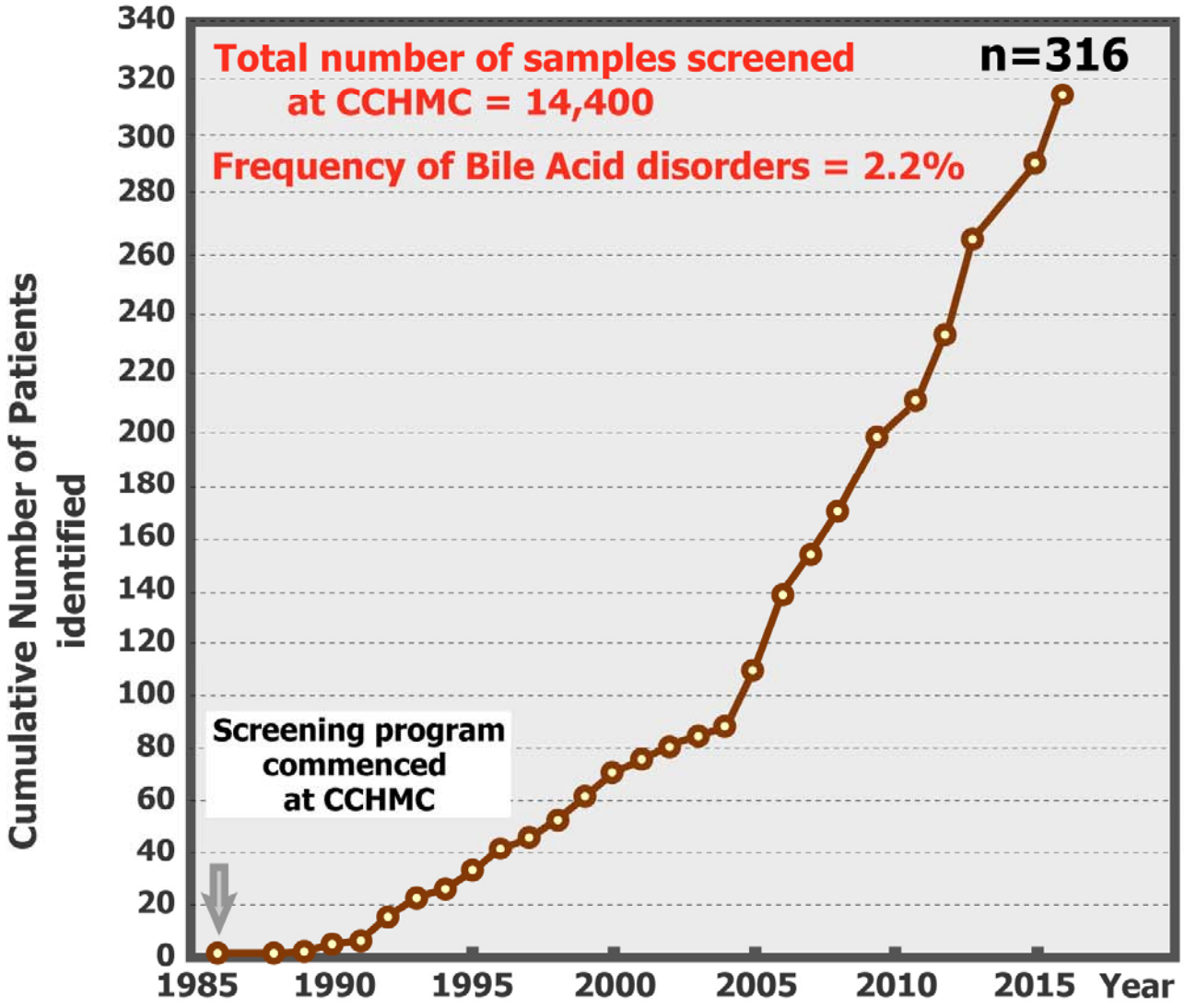
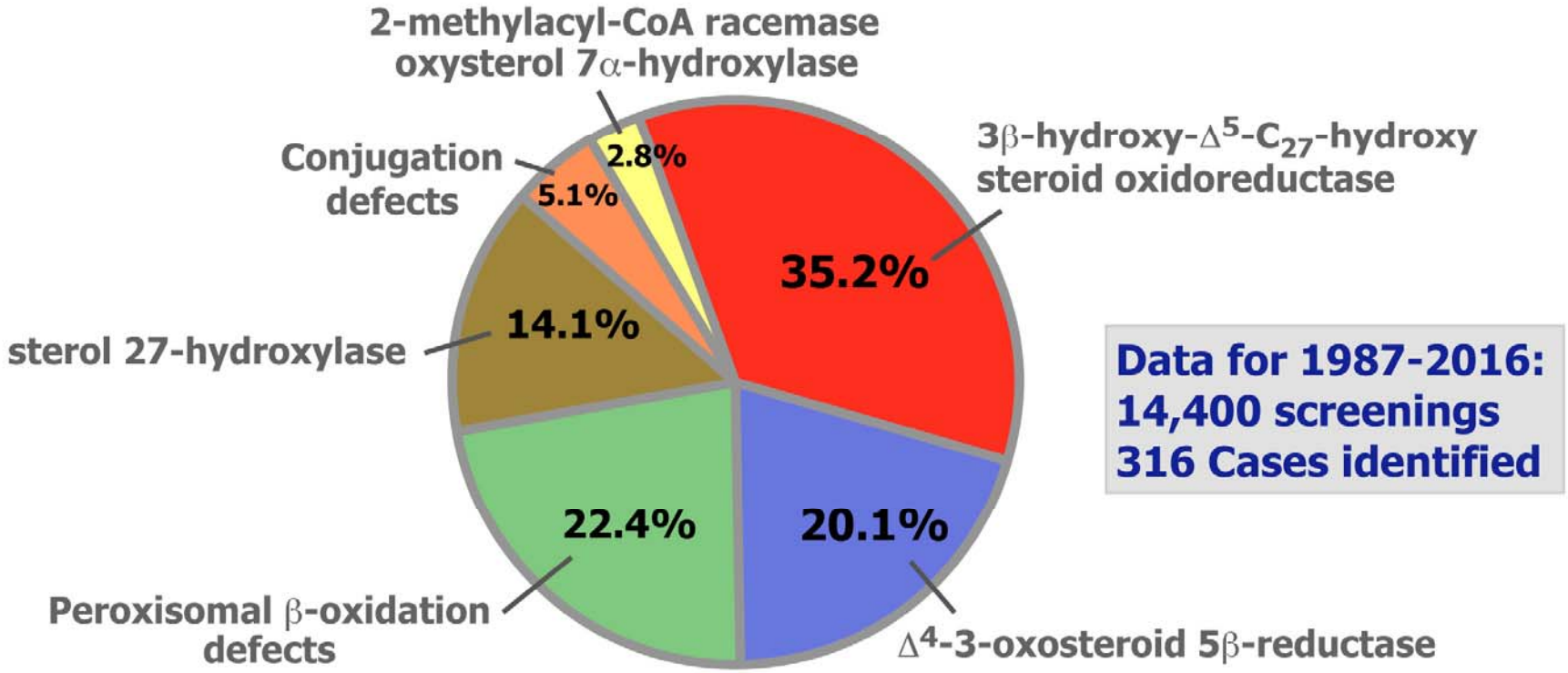
Cumulative Numbers of Patients Diagnosed with Inborn Errors in Bile Acid Synthesis



Defects in Bile Acid Synthesis: 'The Cincinnati Experience'

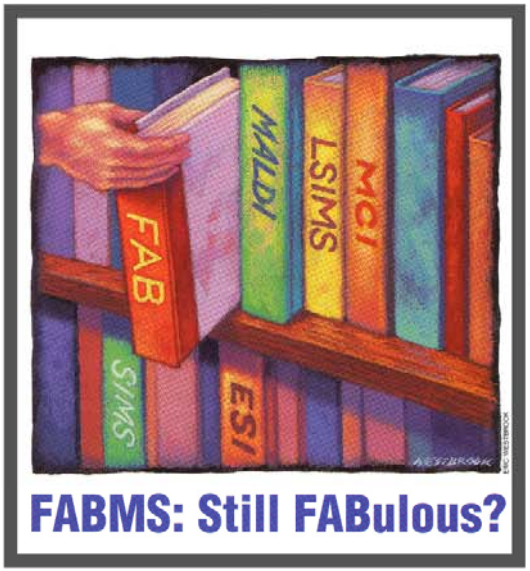


Data for 1987-2016:
14,400 screenings
316 Cases identified

- **Age at diagnosis and clinical presentation is highly variable ranging from early infancy to adulthood - Can be a cause of late-onset chronic cholestasis**

1. Heubi JE, et al. Semin Liver Dis. 2007;27(3):282-294.
2. Bove KE, et al. Pediatr Dev Pathol. 2000;3(1):1-16.
3. Setchell KD, Heubi JE. J Pediatr Gastroenterol Nutr. 2006;43(suppl 1):S17-S22

Fast Atom Bombardment ionization-Mass Spectrometry



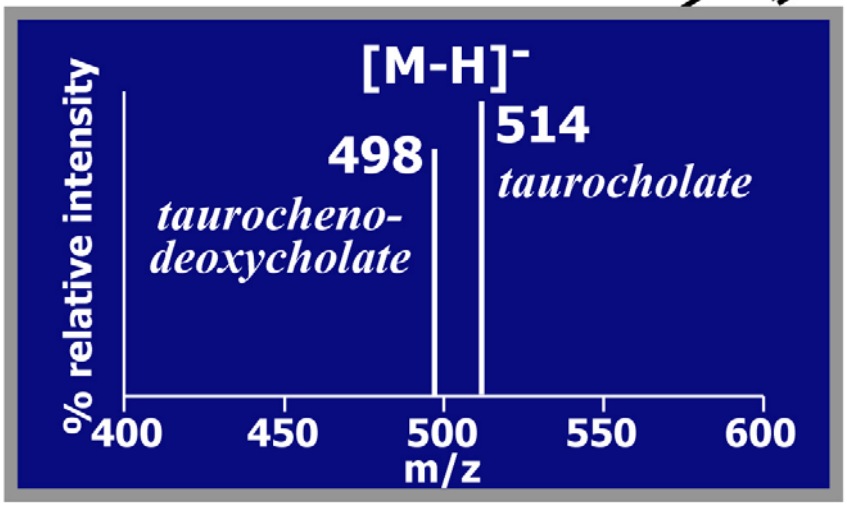
FABMS: Still FABulous?

Urine or Extract and Glycerol matrix

Sample Probe

Atom Gun

Focusing Lens



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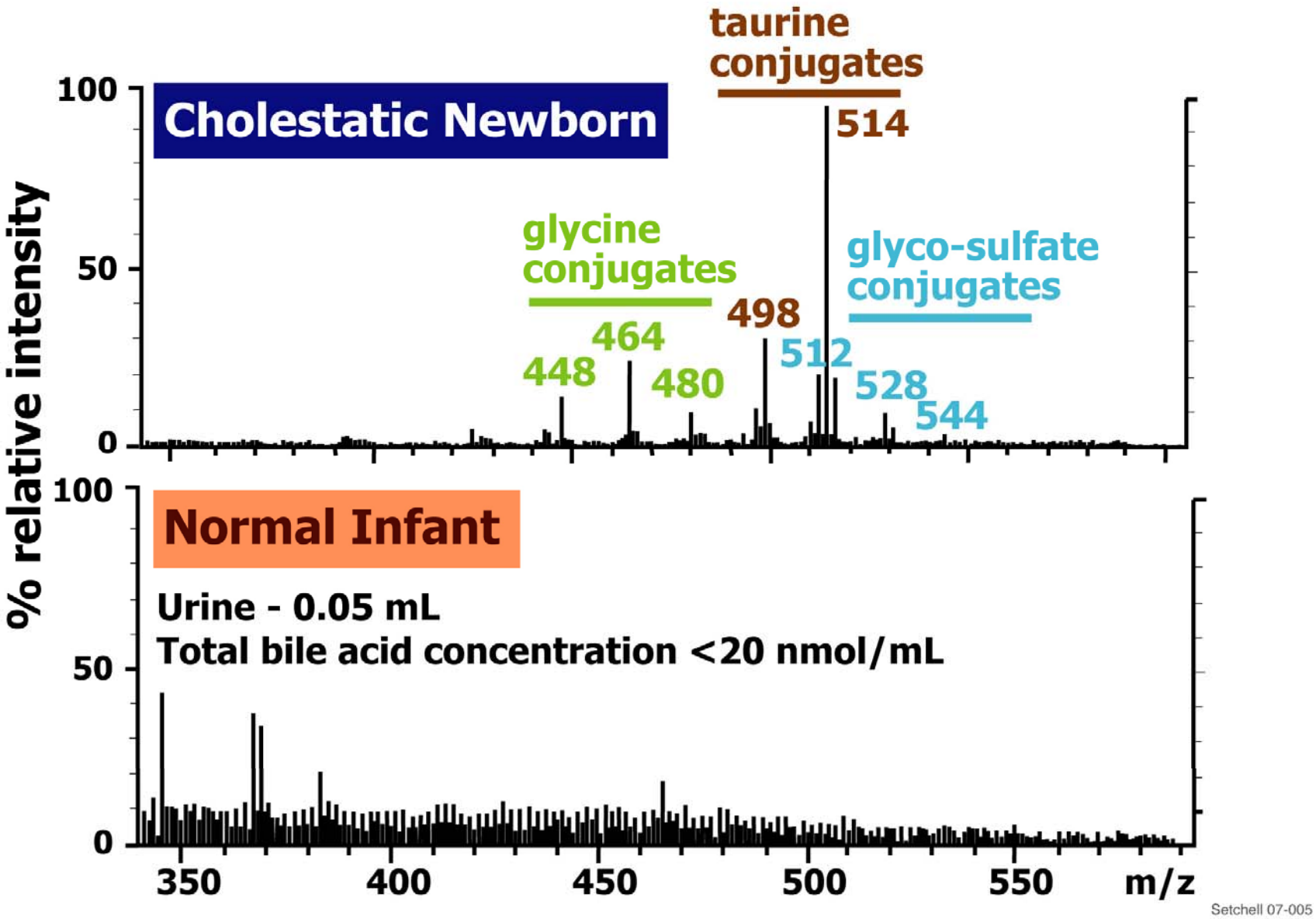
Typical FAB-MS (-ve) spectra comparing urine from a cholestatic newborn with healthy infant



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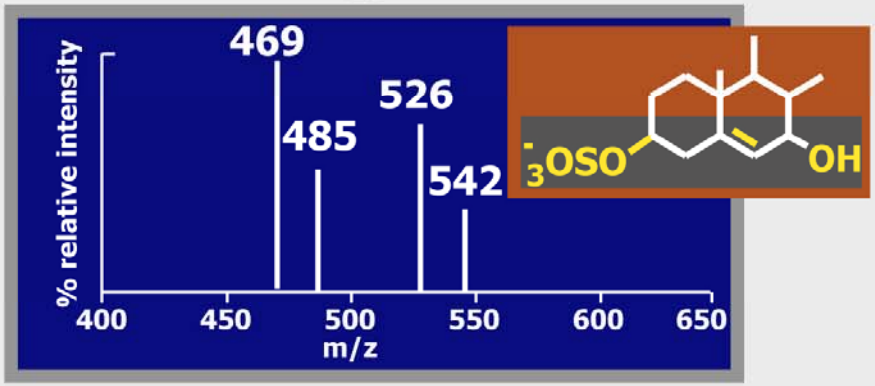
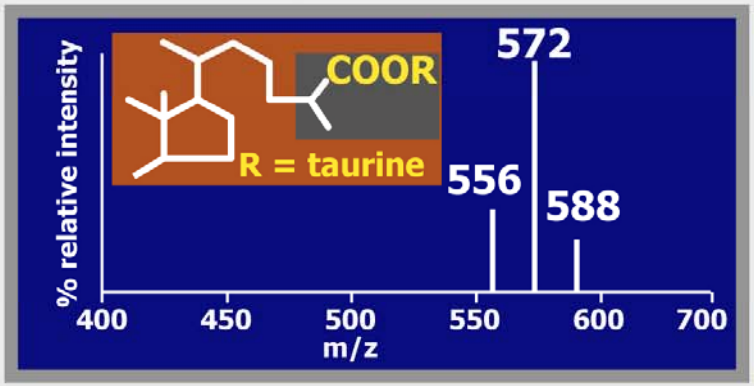
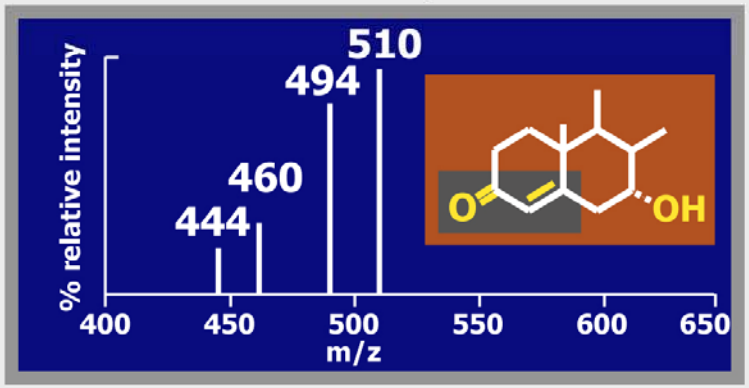
3β-Hydroxy-Δ^5-C_{27}-steroid oxidoreductase



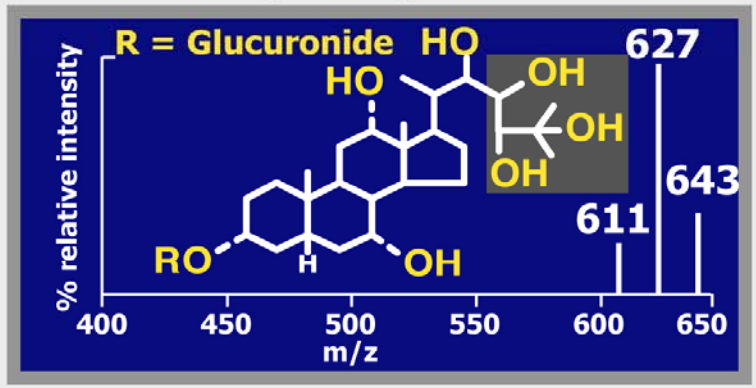
2-Methylacyl-CoA racemase



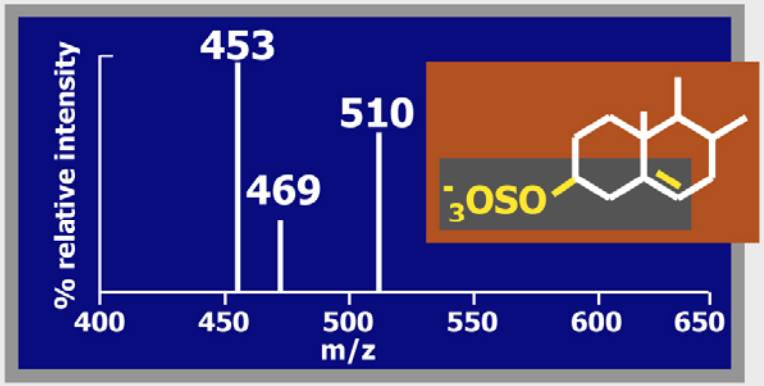
Δ^4-3-Oxosteroid 5β-reductase



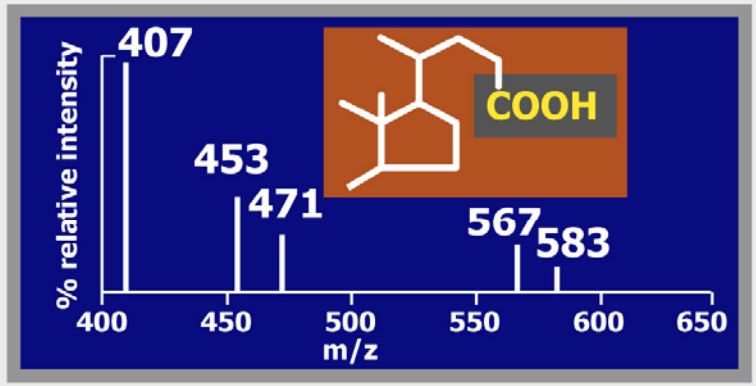
Sterol 27-hydroxylase



Oxysterol 7α-hydroxylase



Bile acid Conjugation defect



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Comparing Intrahepatic Cholestasis in Bile Acid Synthesis and Transporter Disorders

Features	Bile acid defects	PFIC-1, 2, 4 and 5
Age at presentation:	variable, late onset	< 1 year
Liver:	hepatosplenomegaly	hepatosplenomegaly
Pruritus:	absent	severe
Growth failure:	mild/absent	severe
Jaundice:	+/-	mild/severe
Serum transaminases:	elevated	slight to ↑ elevations
Serum GGT:	generally normal	normal
Serum cholesterol:	low	low
Fat-soluble vitamins:	malabsorption	malabsorption
Bile acids:	no primary bile acids	↑ primary bile acids
Treatment:	Bile acid therapy	Transplantation
Prognosis:	excellent	Fatal 1st-2nd decades

Diagnosis of Inborn Errors of Bile Acid Metabolism

- Diagnosis confirmed in Commercial lab
  - www.genetests.org
- CCHMC (formerly jaundice chip)
  - www.cincinnatichildrens.org/service/h/hereditary-liver/tests/
- Emory
  - http://geneticslab.emory.edu/tests/MM340
- Supported by Retrophin
  - http://testcholestasis.com

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3ß-OH steroid dehydrogenase/ isomerase deficiency

- Presents with cholestasis in infancy
- Fat soluble vitamin deficiency (rickets, bleeding)
- Older siblings of affected infants/children
- Low serum gamma GT concentration
- Indolent course→cirrhosis
- May have rapidly progressive course→ transplantation
- May present in later childhood/ adulthood

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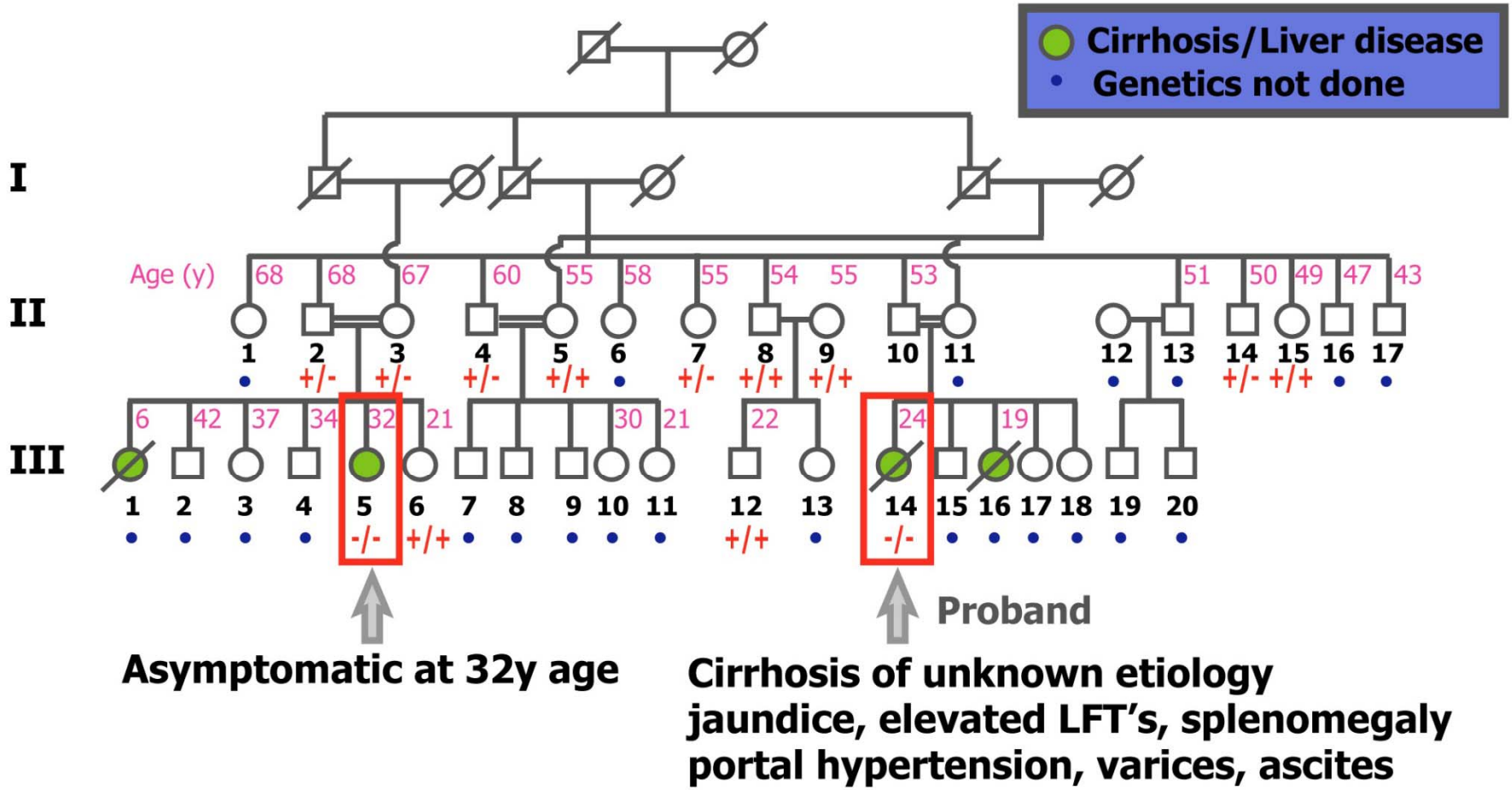
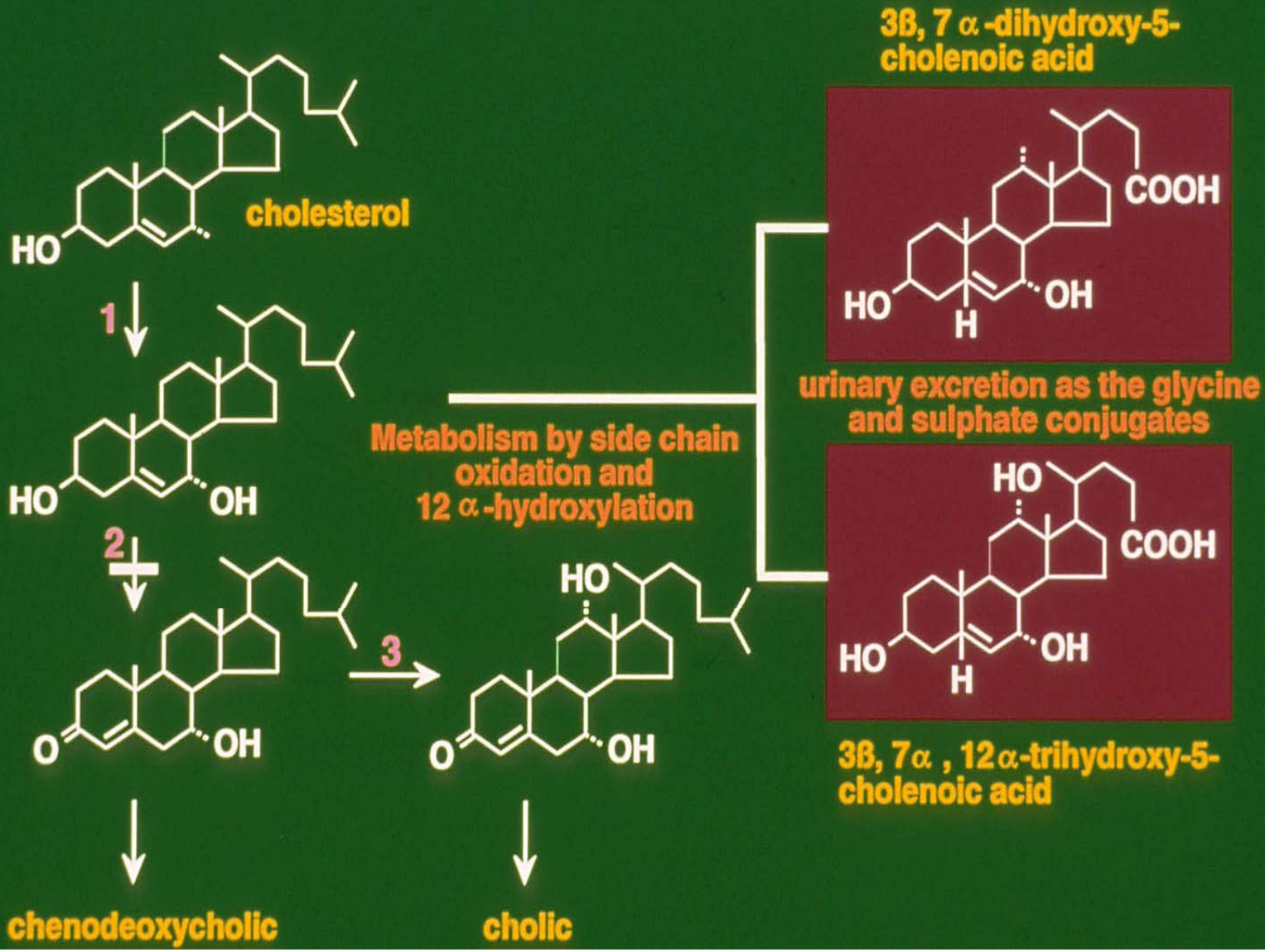
Pedigree of Arab-Iranian Family with 3β-hydroxy-Δ^5-C_{27}-steroid oxidoreductase deficiency



Homozygous for 2 basepair deletion in exon 1 of *HSD3B7* (c.45_46del AG)
FAB-MS of serum confirmed bile acid synthetic defect

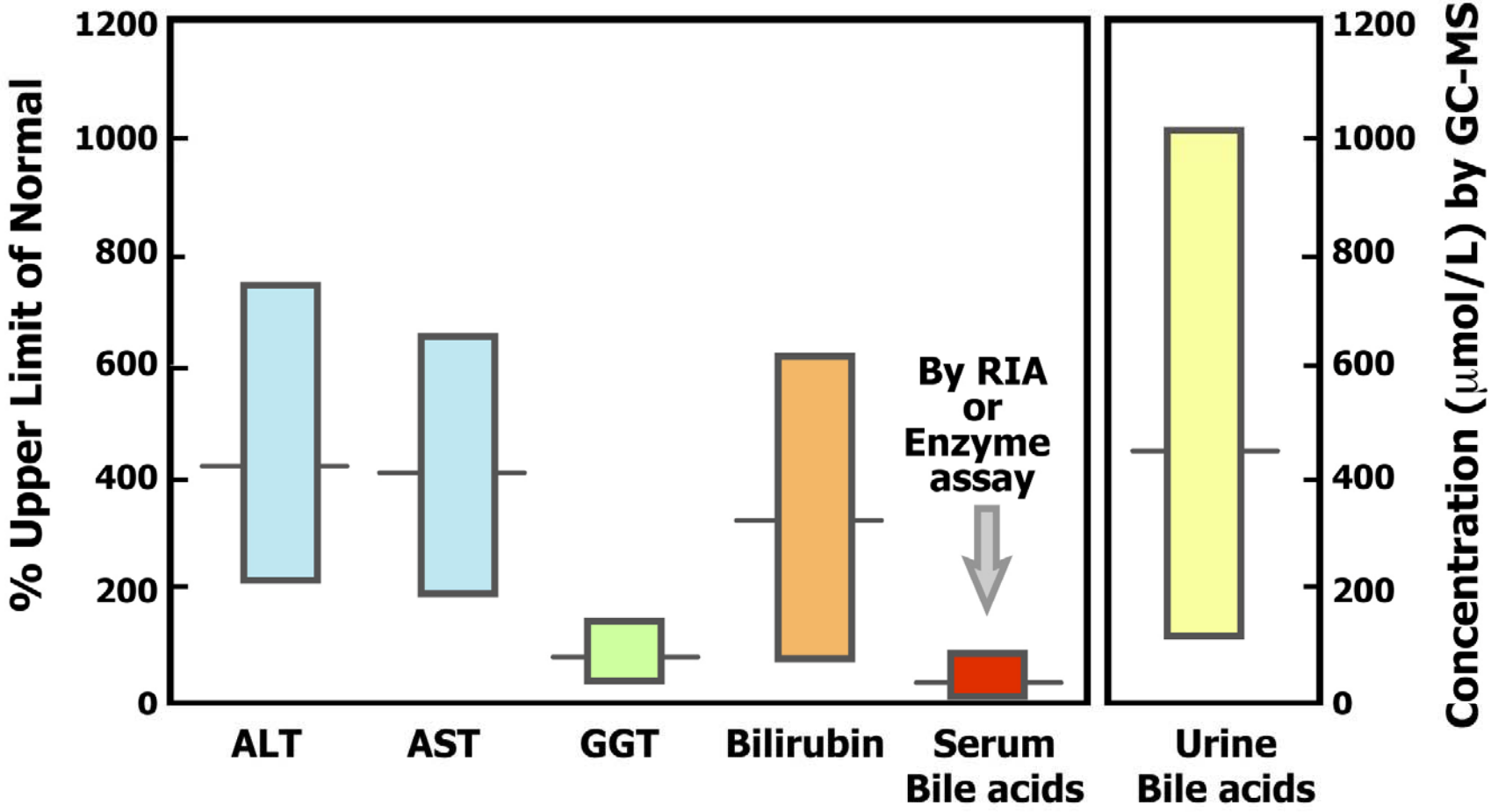
Molho-Pessach, V. et al Hepatology 2012;55:1139-1145

Biochemical defect in 3ß-Hydroxysteroid dehydrogenase/isomerase deficiency

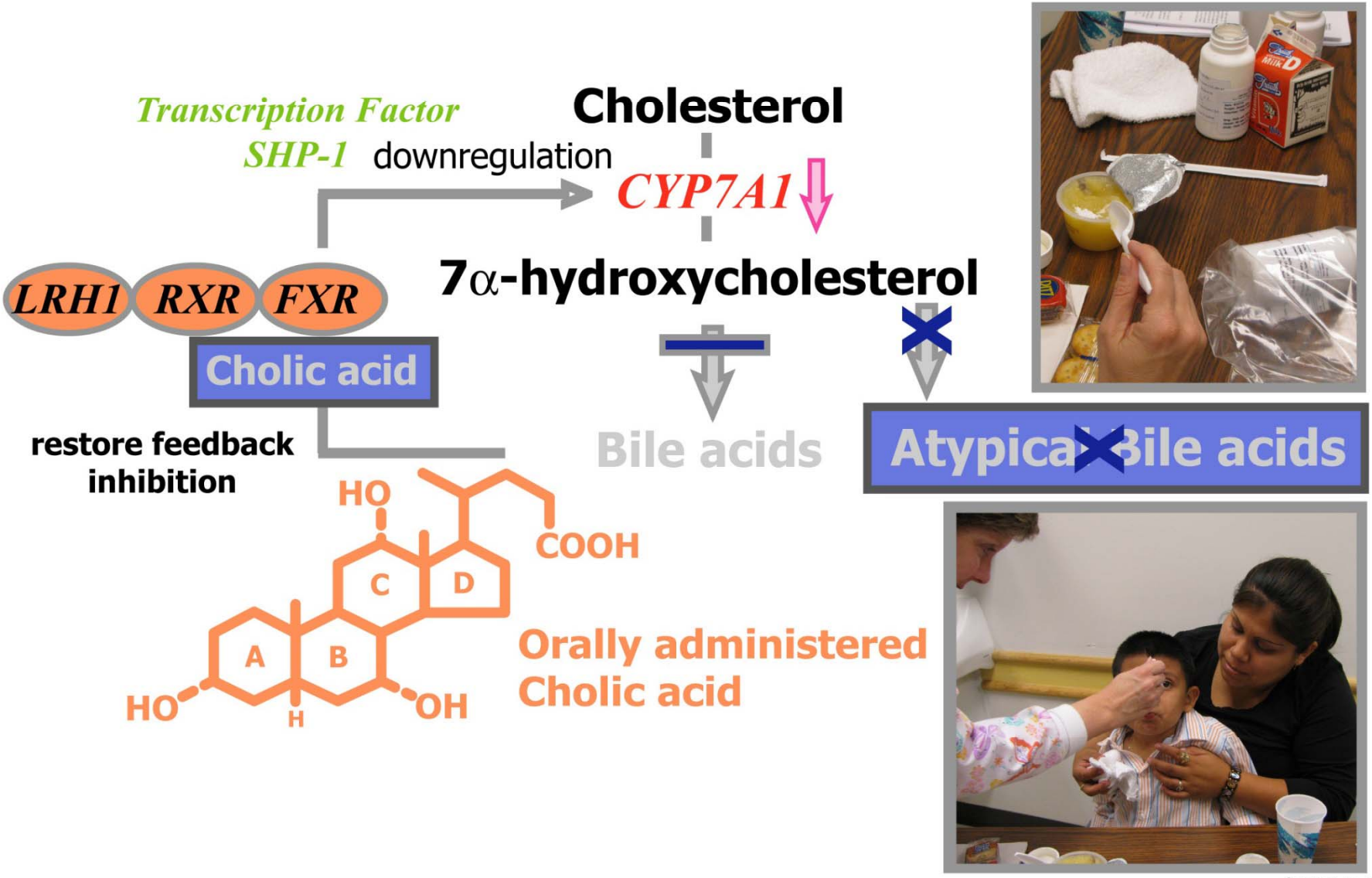


Biochemical Characteristics of Patients with HSD3B7 Deficiency

- Range of baseline values for 19 patients with a biochemically confirmed 3β-hydroxy-Δ^5-C_{27}-steroid oxidoreductase (HSD3B7) deficiency



Metabolic Basis for Cholic Acid Therapy



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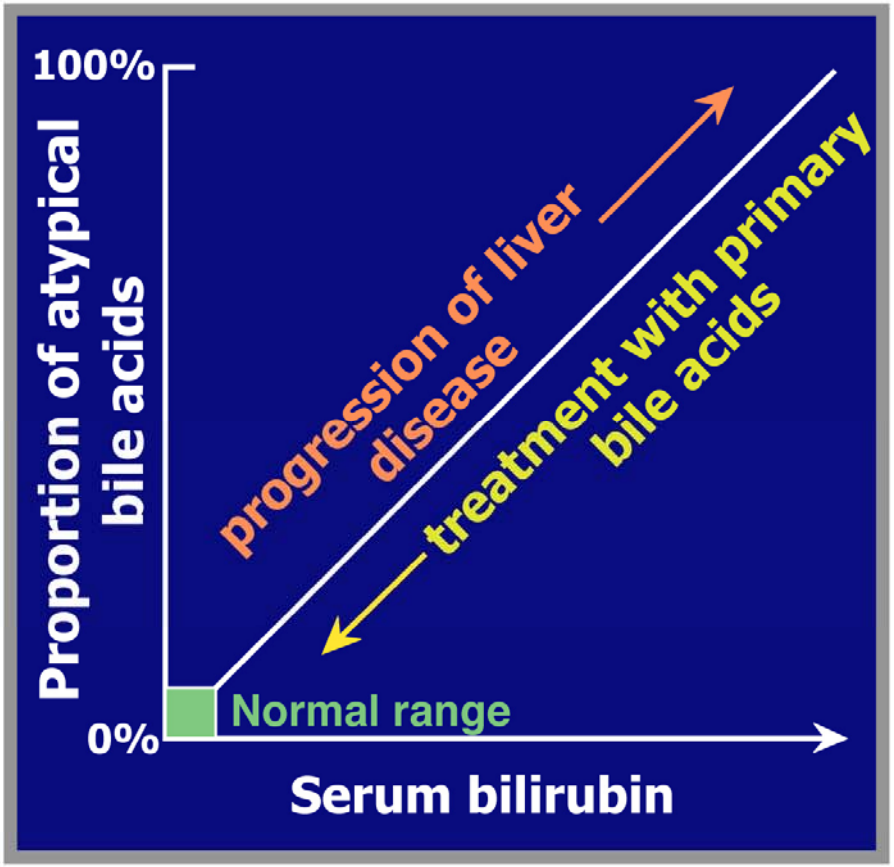
Clinical Manifestation of Inborn Errors in Bile Acid Synthesis and Effect of Primary Bile Acid Therapy



Therapeutic goals:

1. Inhibit endogenous bile acid synthesis - reduce accumulation of atypical bile acids

2. Provide stimulus for bile secretion - generate choleresis

3. Increase intraluminal bile salts - facilitate absorption of fats and fat-soluble vitamins

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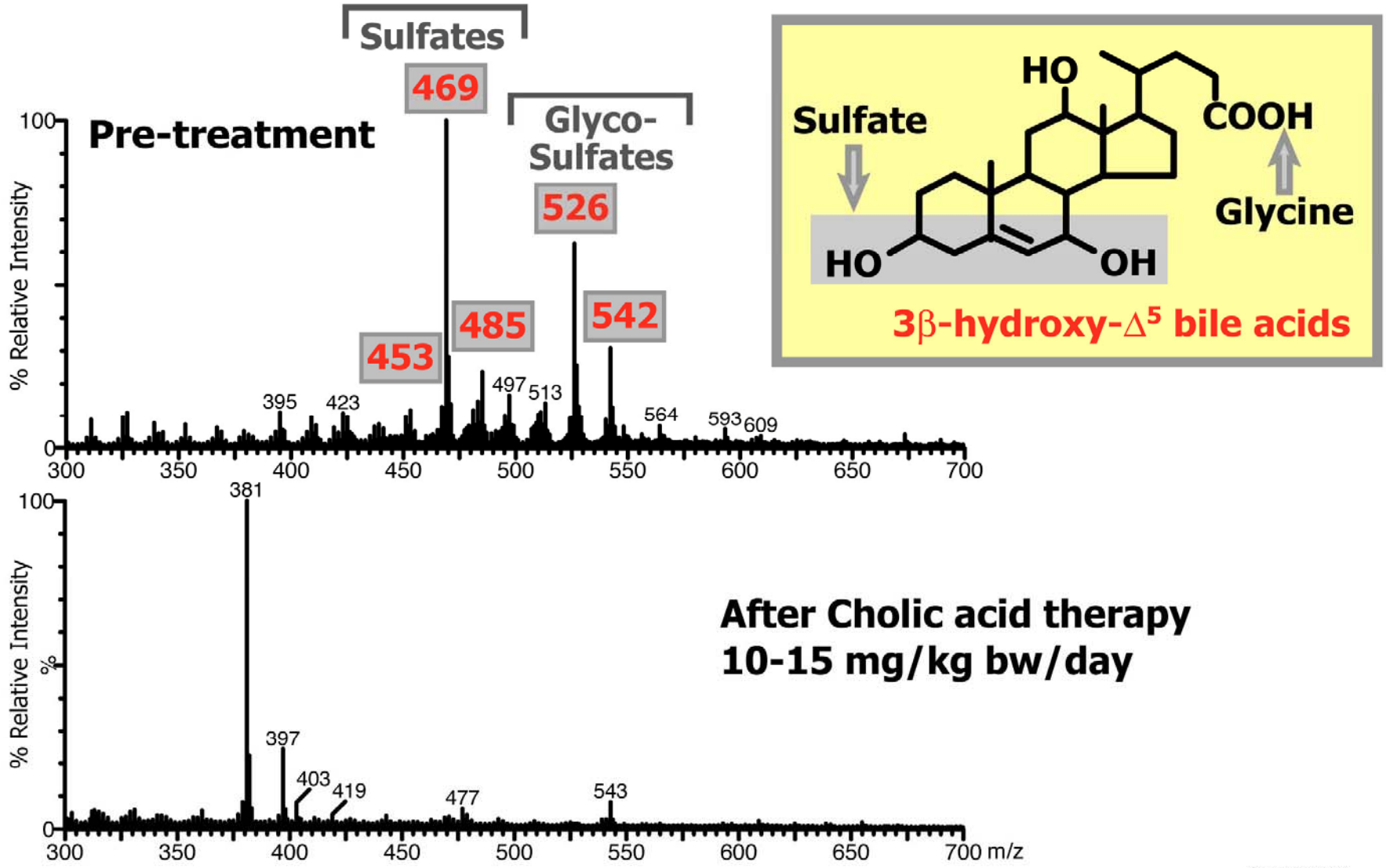
Suppression of Atypical Bile Acids with Cholic Acid Therapy - Urinary FAB-MS profiles



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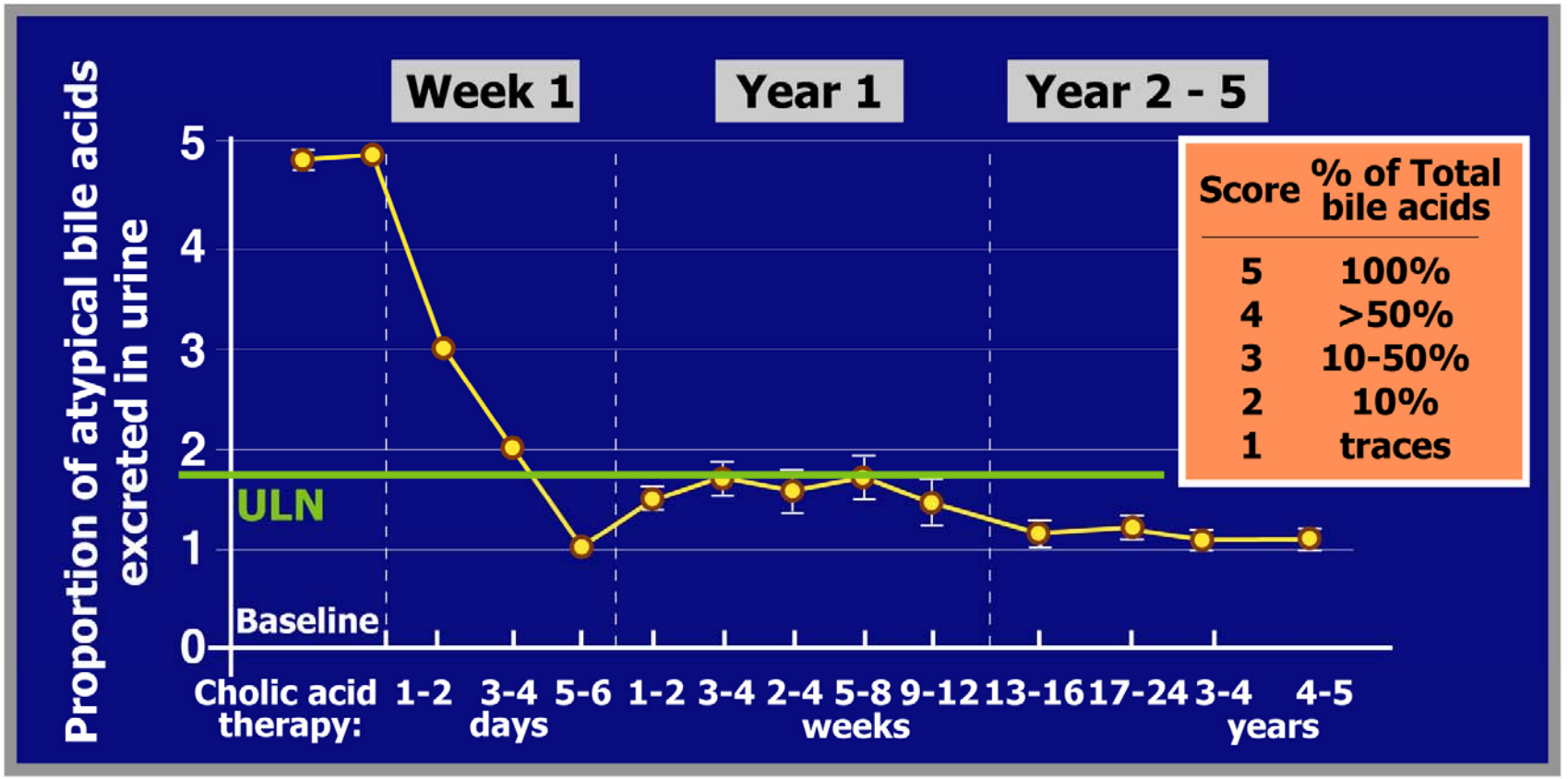
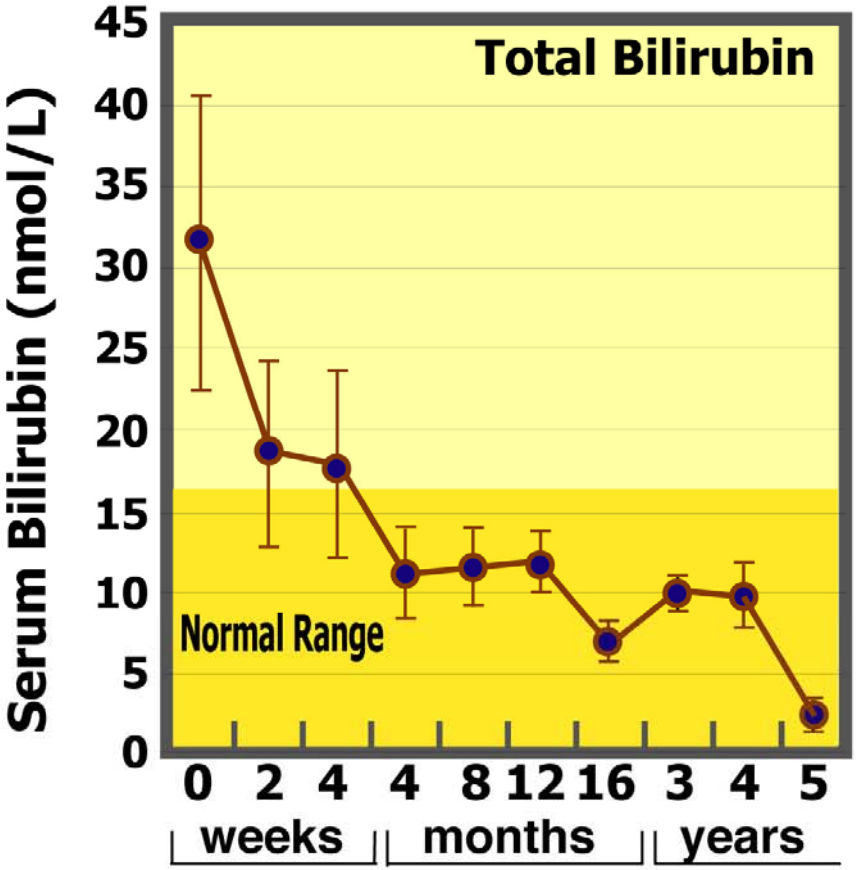
3β-Hydroxy-Δ^5-C_{27}-steroid oxidoreductase deficiency - Effect of Cholic Acid Therapy on Atypical Bile Acids

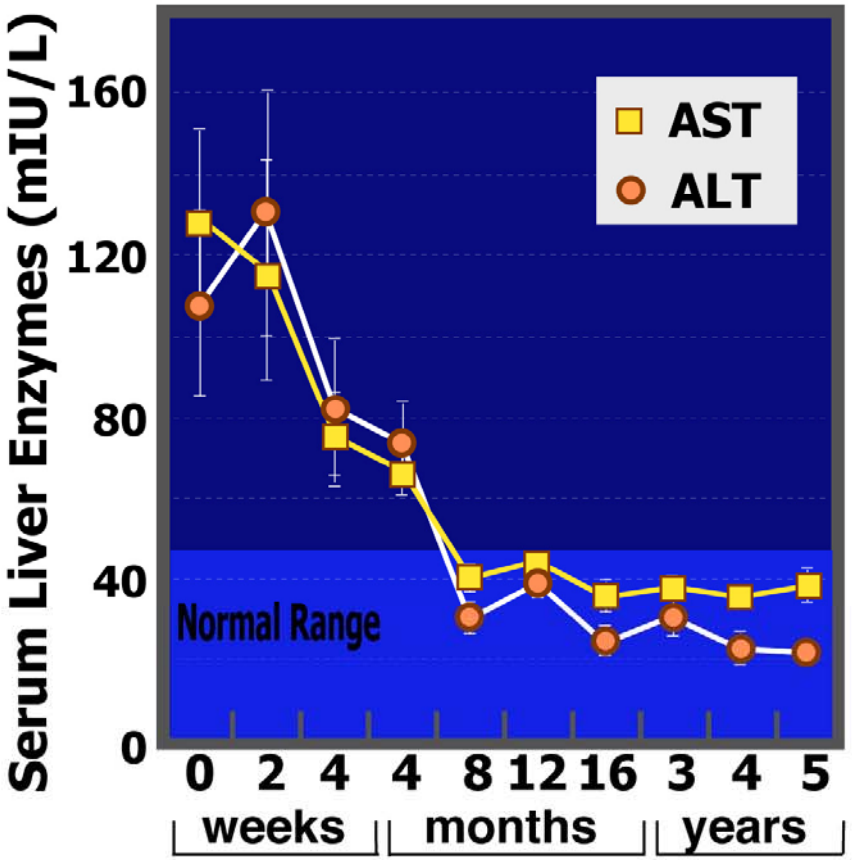
Oral cholic acid therapy 10-15 mg/kg body weight/day
Summary of long-term therapy in 19 patients



Effect of Oral Cholic Acid Therapy on LFTs in Patients with 3β-Hydroxy-Δ^5-C_{27}-steroid oxidoreductase Deficiency

Cholic acid therapy: 10-15 mg/Kg bw/day (19 Patients)





Time after initiating oral cholic acid therapy

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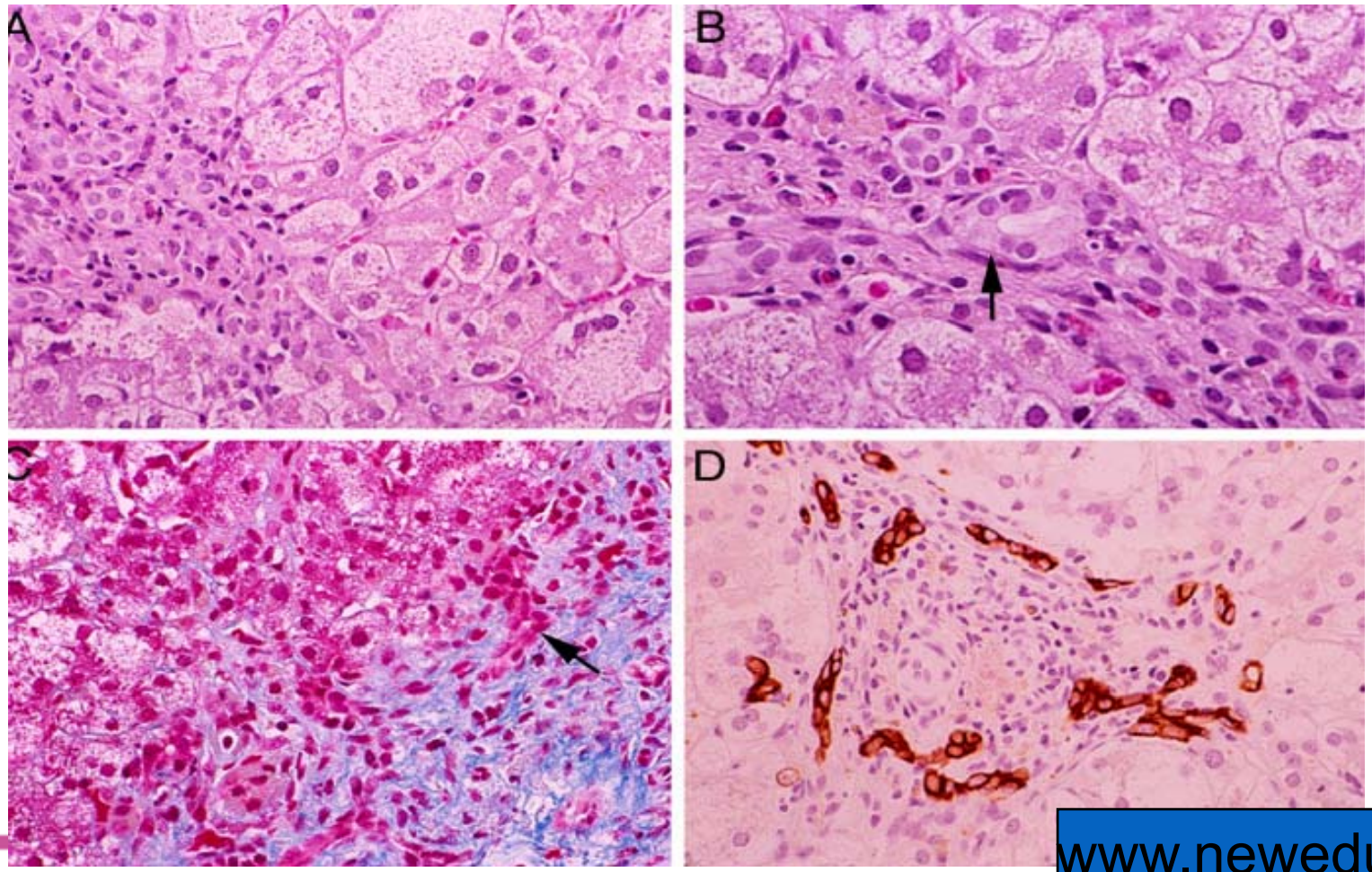
Pathology in 3-HSD



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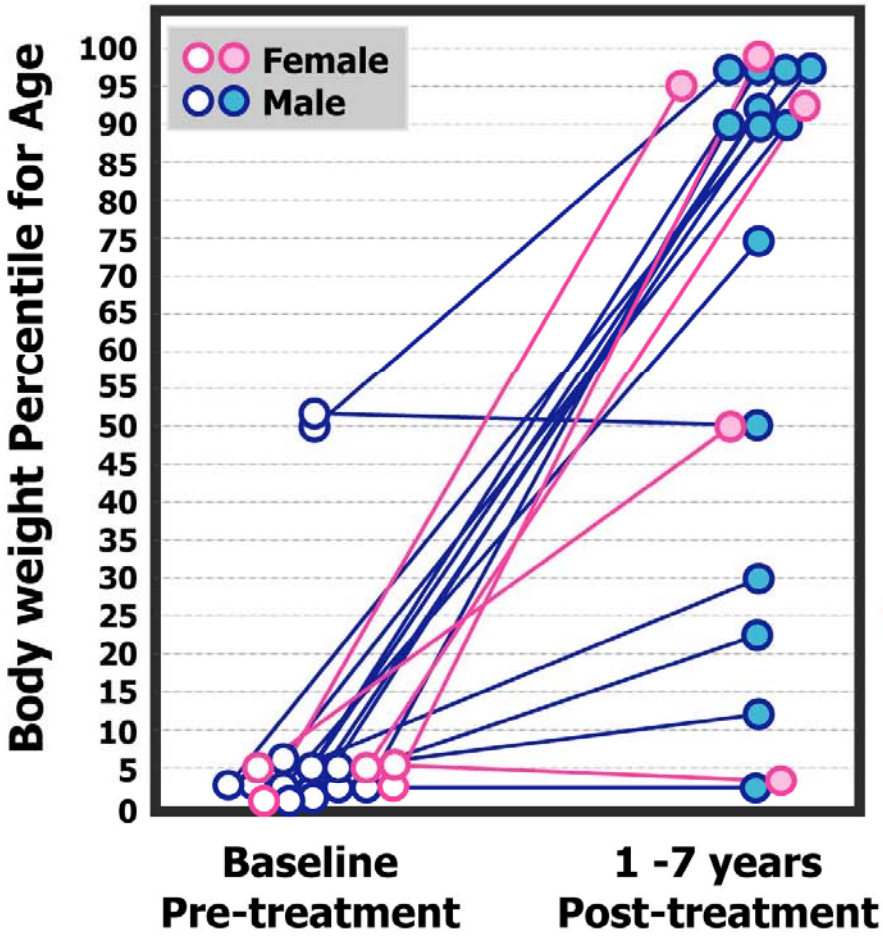
Long-term Effect of Cholic Acid on Growth

3β-Hydroxy-Δ^5-C_{27}-steroid oxidoreductase deficiency (n=19)



Cholic acid therapy
Oral dose 10-15 mg/kg bw/day

- **Long term Cholic acid therapy leads to significant improvement in growth and body weight in most patients**
- **Cholic acid was well tolerated with no significant side-effects reported in >20 yrs of therapy**

Δ^4-3-oxosteroid-5β-reductase deficiency

- Initial description of monochorionic twins presenting with neonatal cholestasis
- Presented with jaundice and varying severity of liver dysfunction
- Rapidly progressive disease leading to cirrhosis in infancy: Previously presumed affected sibling died in infancy

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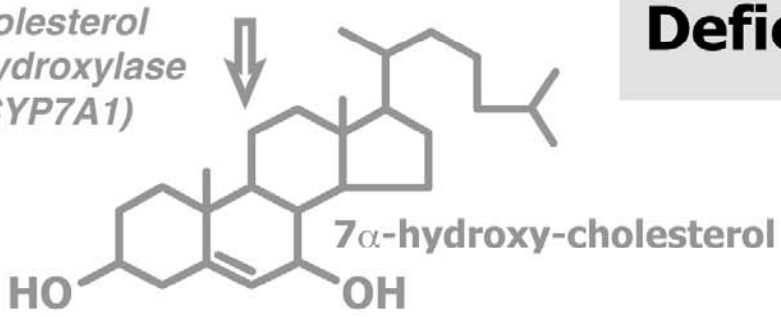
Δ^4-3-Oxosteroid 5β-reductase Deficiency - Biochemistry

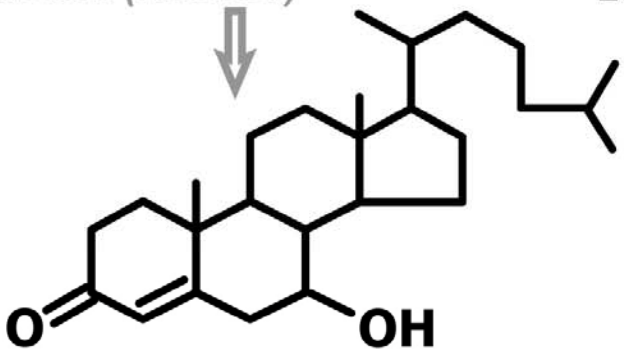
Cholesterol

cholesterol 7α-hydroxylase (CYP7A1)



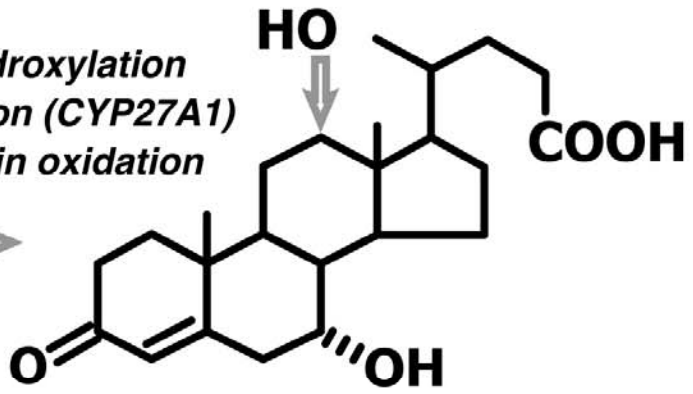
7α-hydroxy-cholesterol

3β-hydroxy-Δ^5-C_{27}-steroid oxidoreductase (HSD3B7)



7α-hydroxy-4-cholen-3-one (sterol-C4)

(+/-)12α-hydroxylation
27-hydroxylation (CYP27A1)
and side-chain oxidation



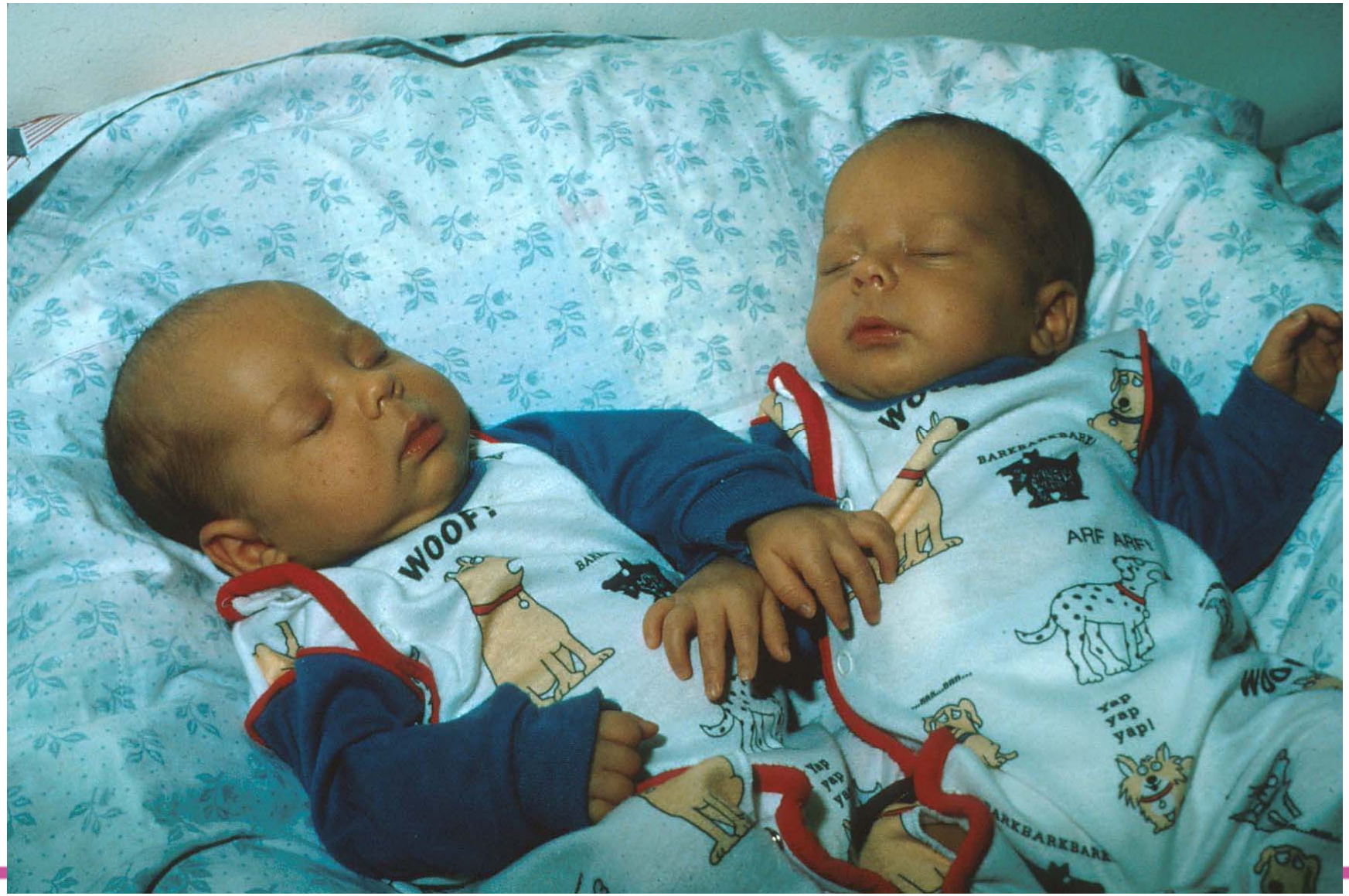
3-oxo-7α-hydroxy-4-cholenoic acid
and
3-oxo–7α,12α-dihydroxy-4-cholenoic acid

Δ^4-3-oxosteroid 5β-reductase (AKR1D1)

primary bile acids
[Not Detected]

Conjugation with taurine and glycine

cholestasis, liver injury



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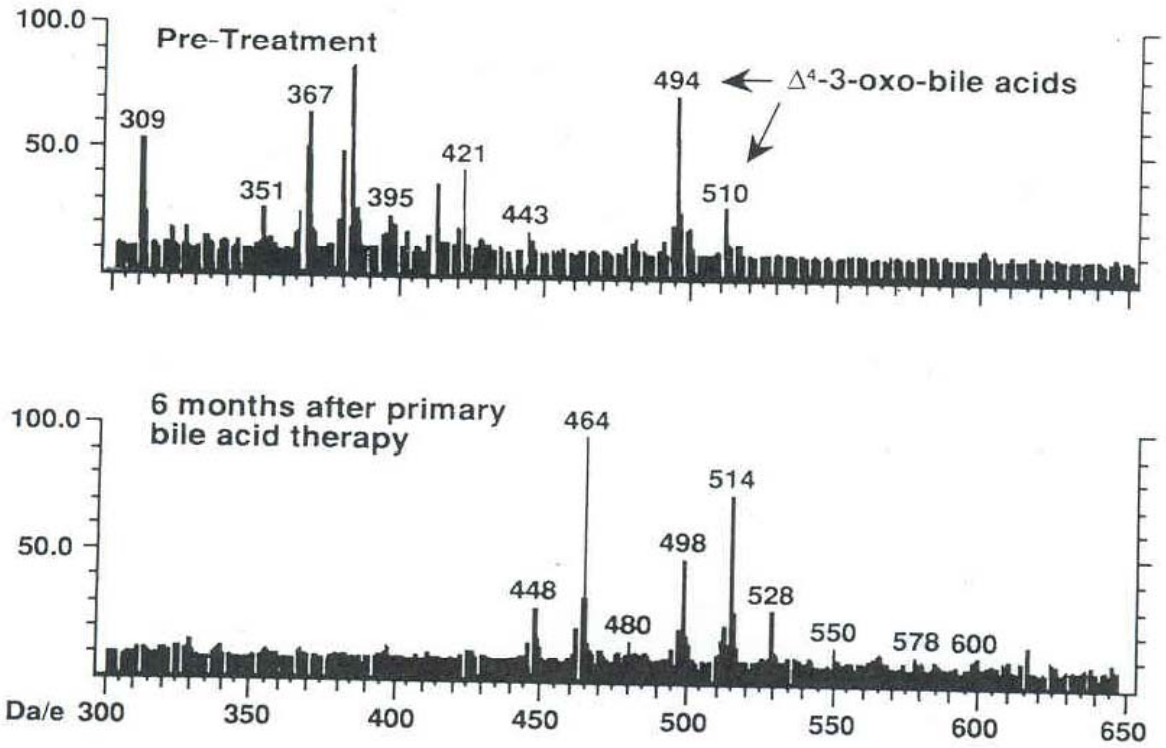
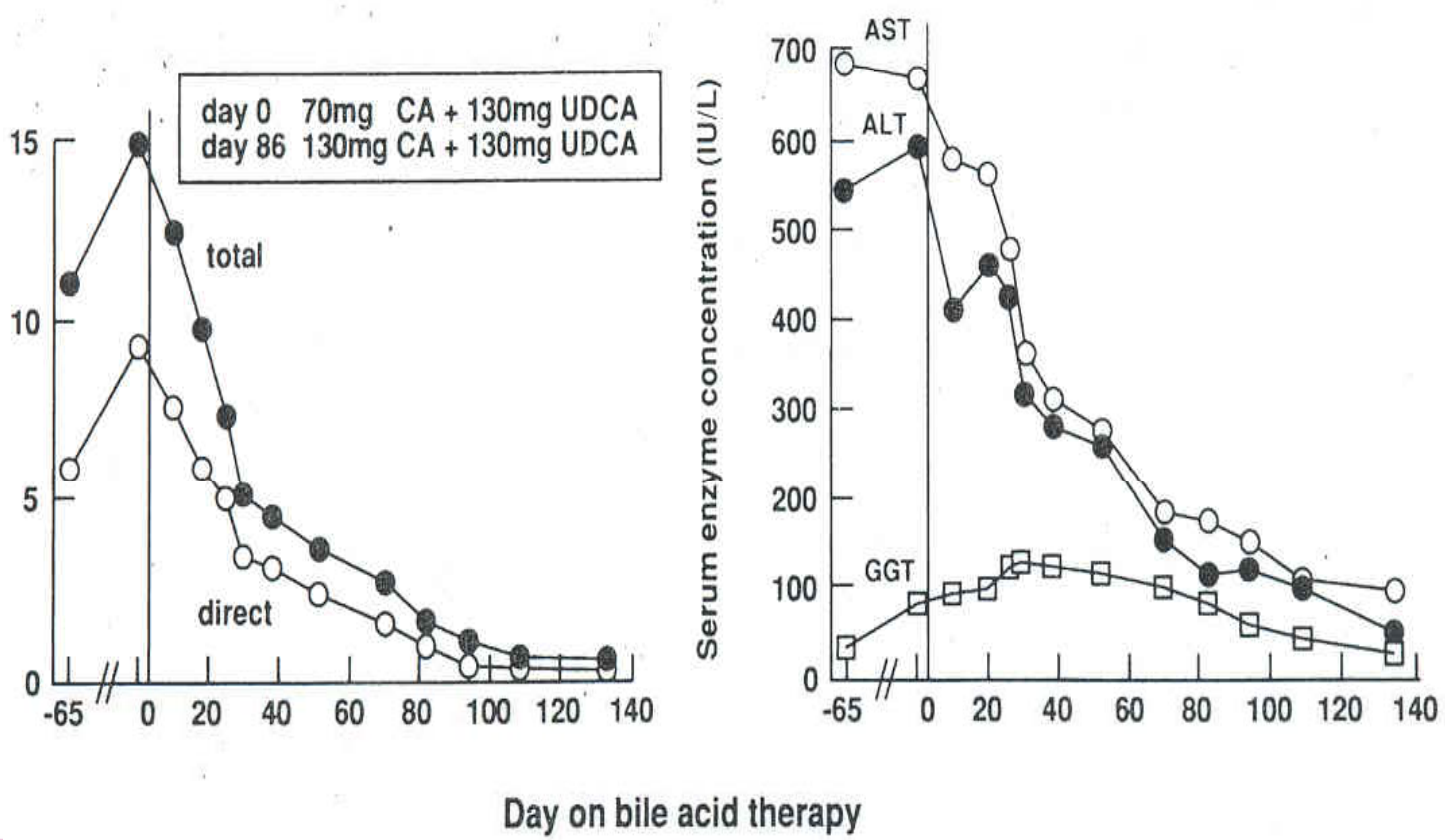
Effect of Therapy on Bile Acid Excretion



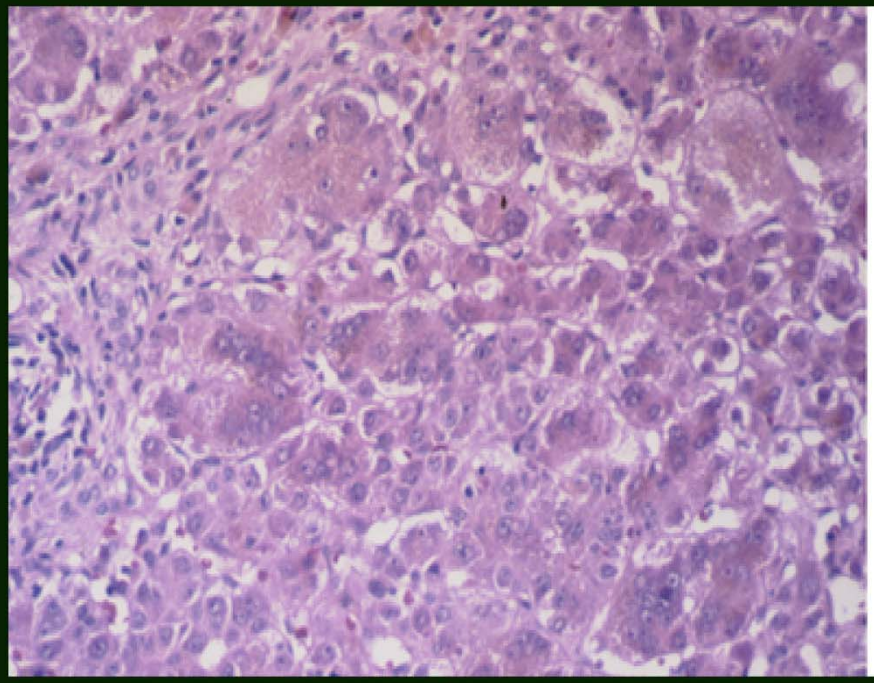
Biochemical Response to Therapy



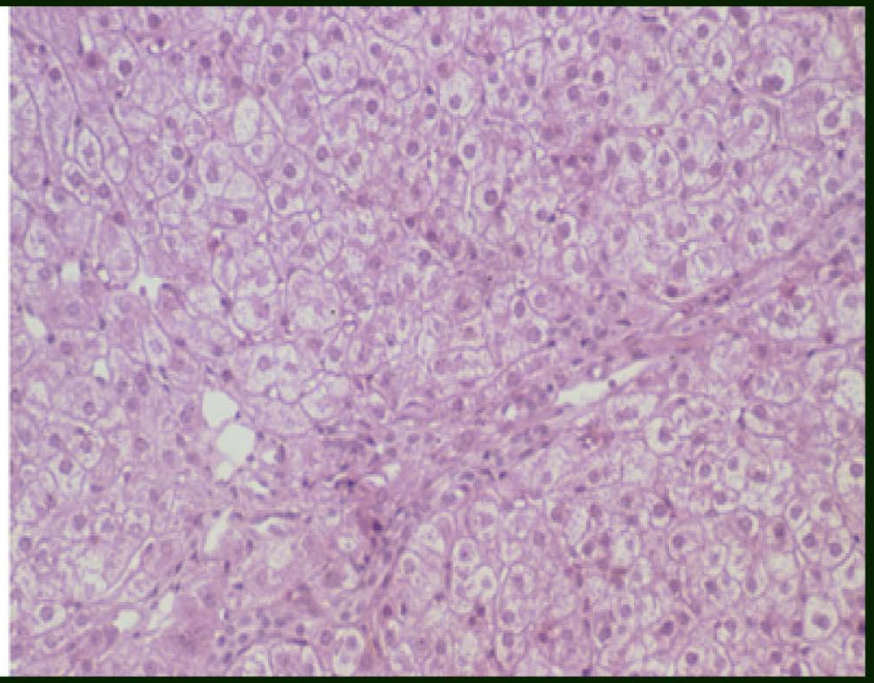
Effect of cholic acid therapy on liver histology: Patient with a bile acid synthetic defect

Δ^4-3-Oxosteroid 5β-reductase deficiency

Cholic acid: 15 mg/kg/day



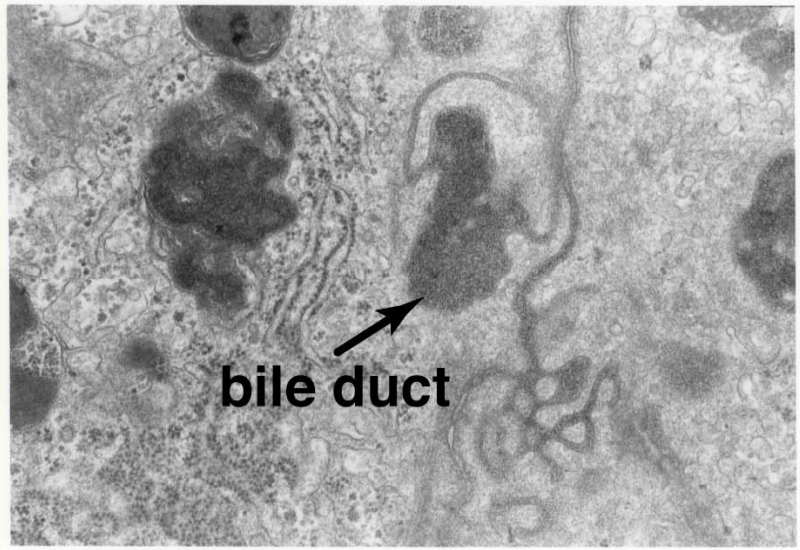
Liver: Age 3 months
(Before treatment)



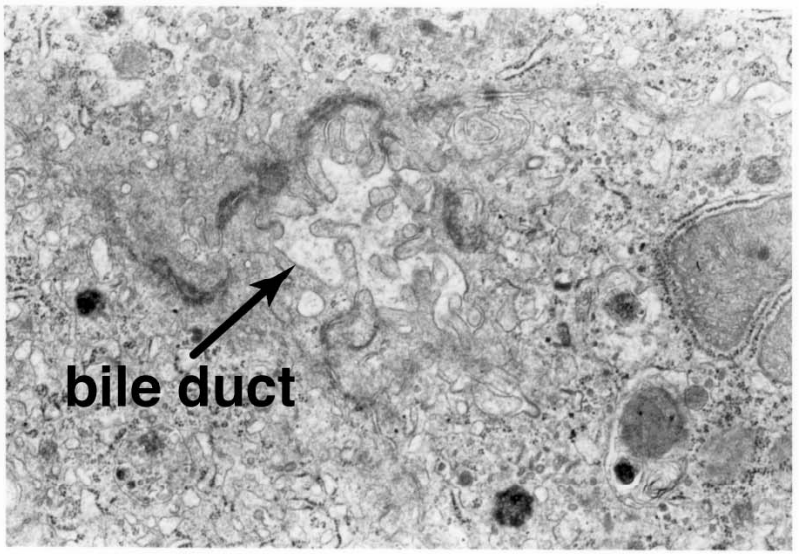
Liver after 16 months
of cholic acid therapy

EM of liver of a patient (SG) with Δ^4-3-oxosteroid 5β-reductase deficiency

Bile ducts poorly developed, lacking normal microvillus structure and filled with electron-dense material



Before treatment



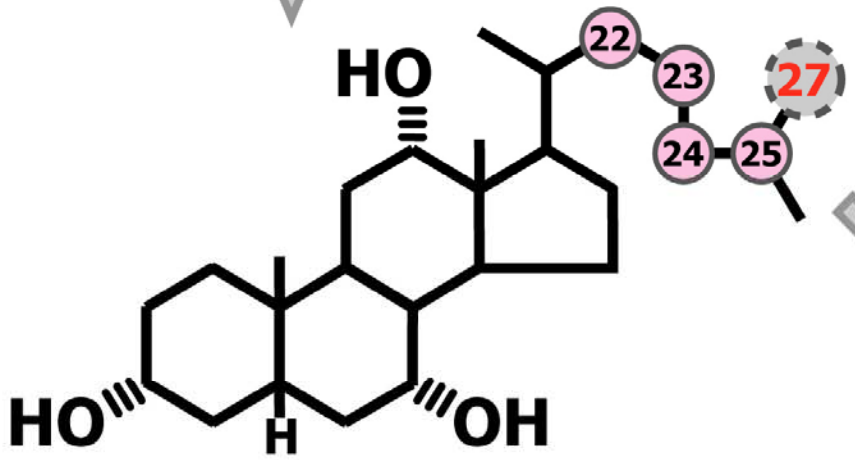
After 6 months of bile acid therapy

Sterol 27-hydroxylase Deficiency (CTX) - Metabolic Defect

cholesterol

sterol intermediates

27-hydroxylation (CYP27) and side-chain β-oxidation



5β-cholestane-3α,7α,12α-triol

Cholic acid [Chenodeoxycholic]

developmental pathway

microsomal 25-hydroxylation

24β-hydroxylation

Cholic acid

bile alcohols (22-,23-,24-,25-hydroxylated)

Urine/Bile/Feces

Setchell 12-061

Clinical History-Index Patient

- Asian male (parents 1st cousins), full term pregnancy, BW 2.6 kg
- At 8 weeks, prolonged jaundice, pale stools
  - Serum bilirubin 4.9 mg/dl, albumin 3.9 gm/dl, AST 275 u/L, ALP 1281 U/L
  - Cholic acid started
- Age 3 months, poor growth, weight 3%ile, hepatosplenomegaly
  - Serum AST 260 U/L, ALT 212 U/L, GGT 95 U/L, ALP 2555 U/L

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Clinical History Index Patient

- Impression: Neonatal hepatitis
- Age 4 months: jaundice ↓, pruritus
- Age 5 months: resolved jaundice, pruritus improved, BW at 50th%ile
- Age 8 months: LFT's normal
- Age 13 months: seizures

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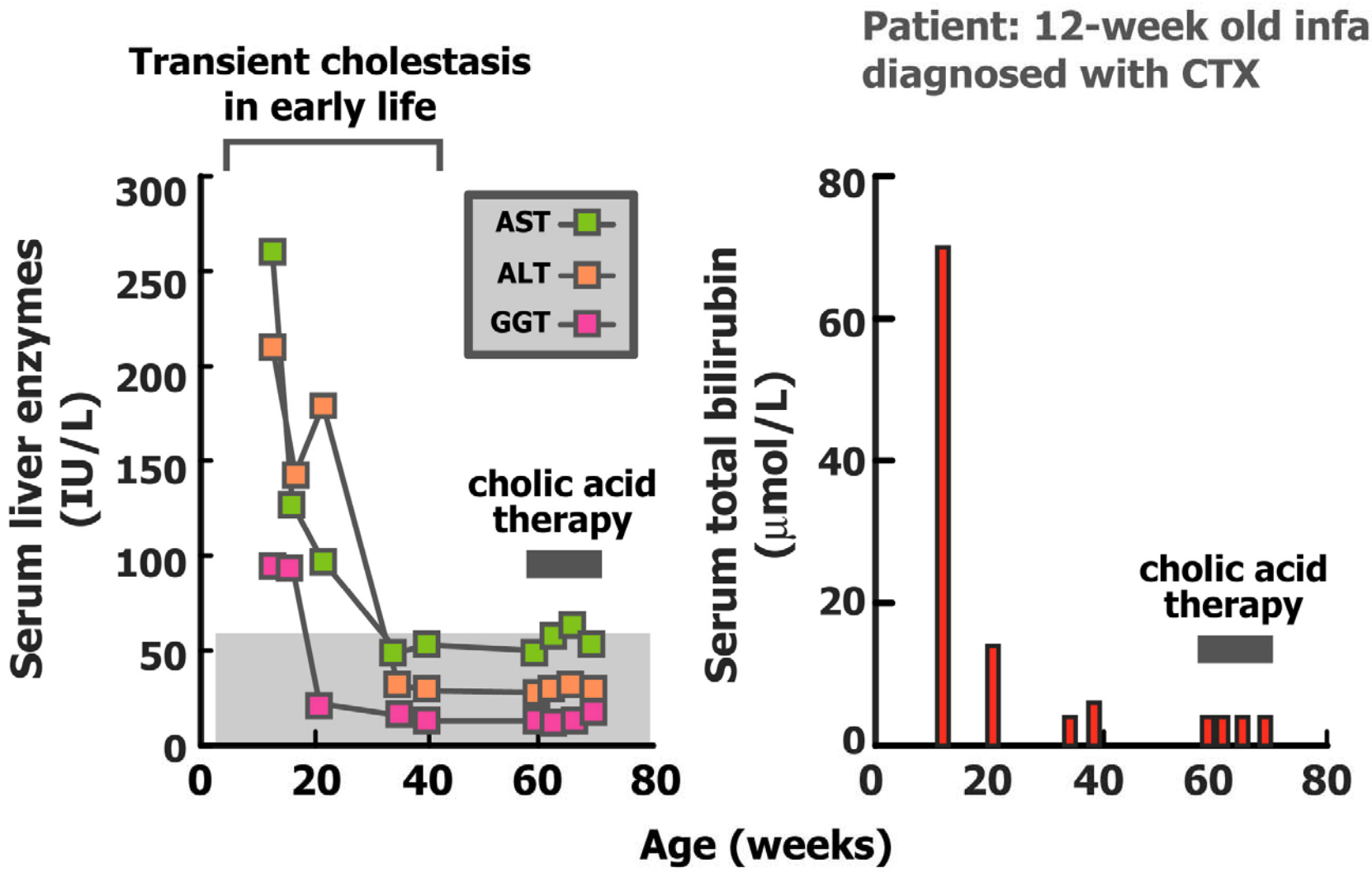
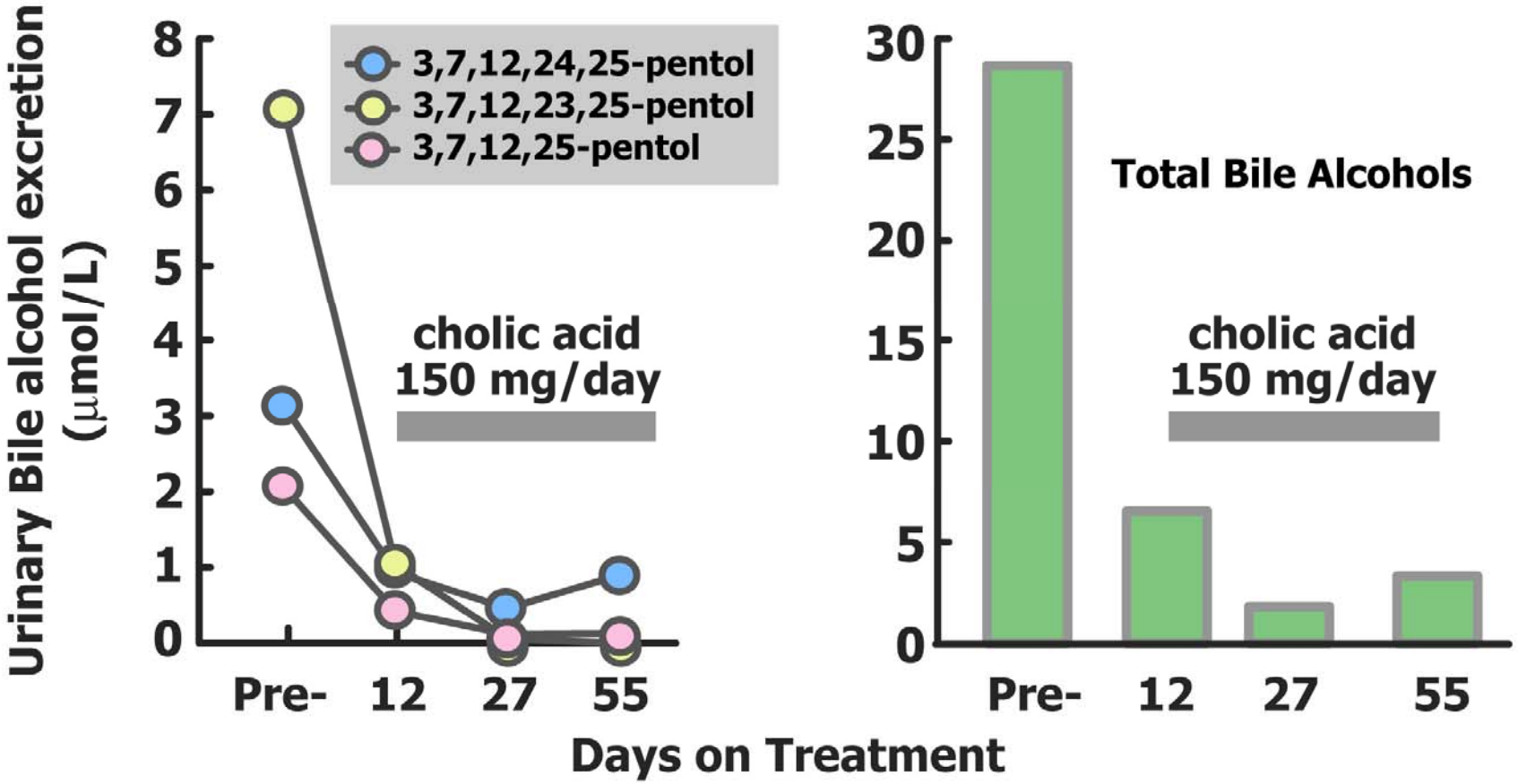
Liver Function Tests in an Infant with Sterol 27-hydroxylase Deficiency



Setchell 12-067

Changes in Urinary Bile Alcohol Excretion with Cholic Acid Therapy

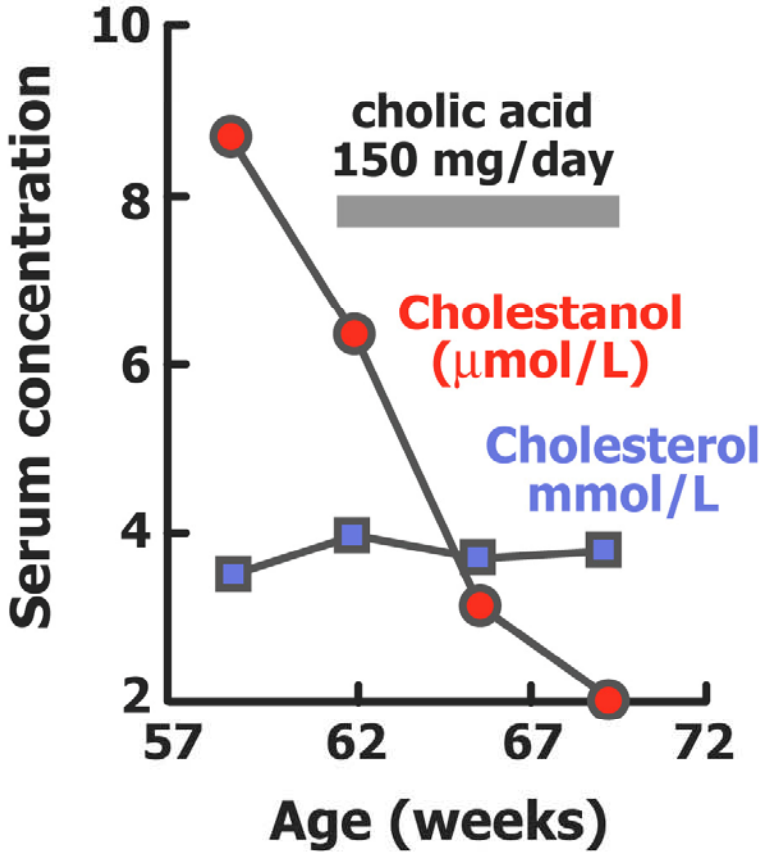
Patient : MM - Sterol 27-hydroxylase deficiency
Oral cholic acid 150 mg/day

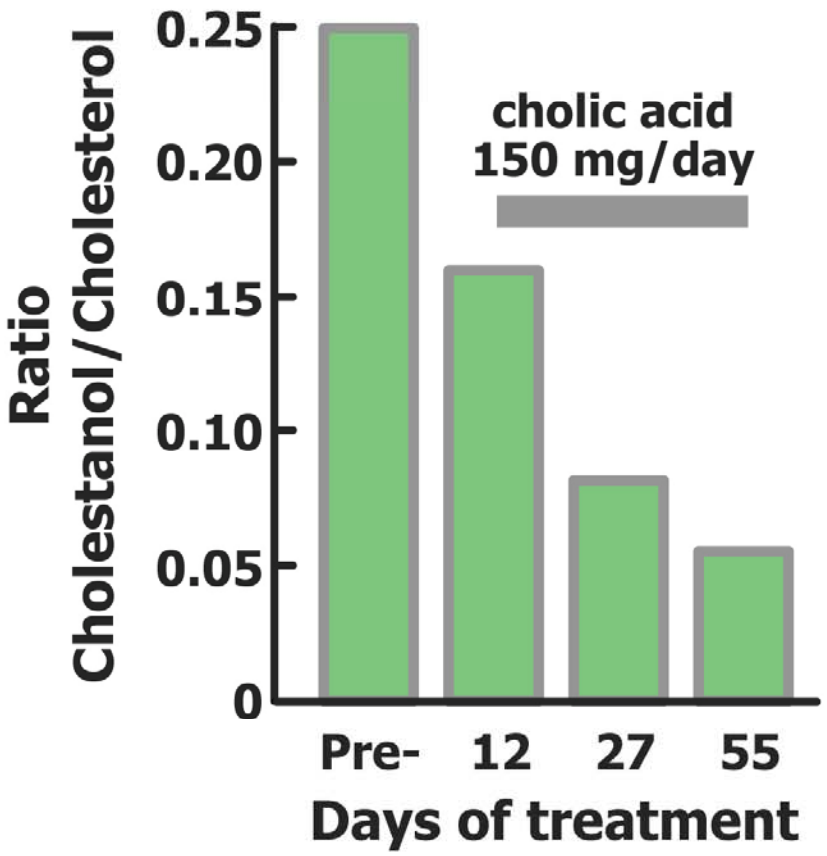


Setchell 12-065

Cholestanol/Cholesterol Changes in Response to Cholic Acid Therapy

Patient : MM - Sterol 27-hydroxylase deficiency
Oral cholic acid 150 mg/day





Setchell 12-062

Disorders of Peroxisomal Function

- Zellweger syndrome, Neonatal Adrenoleukodystrophy, Refsum syndrome
- Profound muscular weakness, liver disease, fatal early in life
- Characterized by absent peroxisomes or defects of enzymes in peroxisomes
- BA and defects of FA metabolism:
  - Accumulation of VLCFA, Pipecolic Acid, Phytanic, Pristanic Acids
  - Di- and Trihydroxycholestanoic acid

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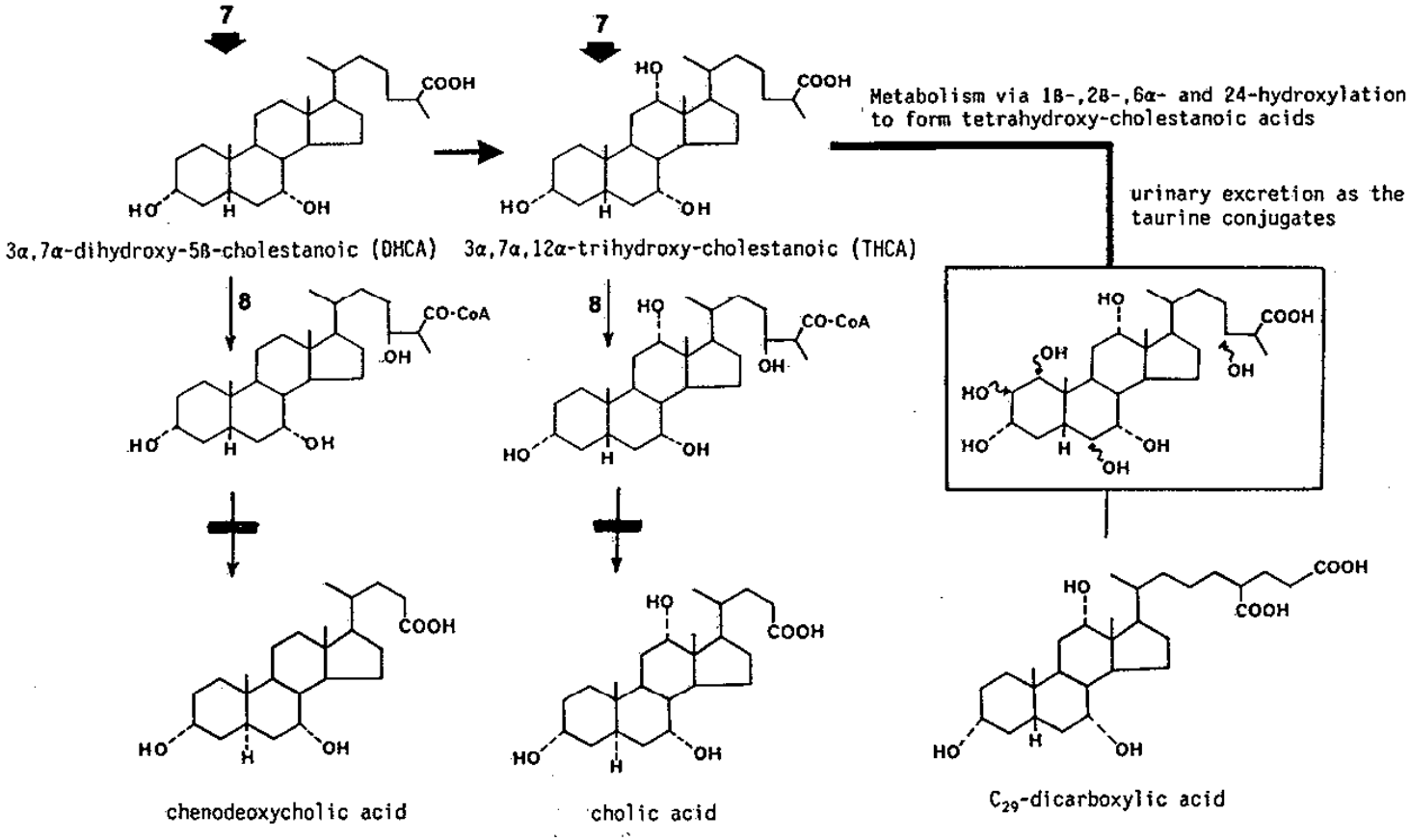
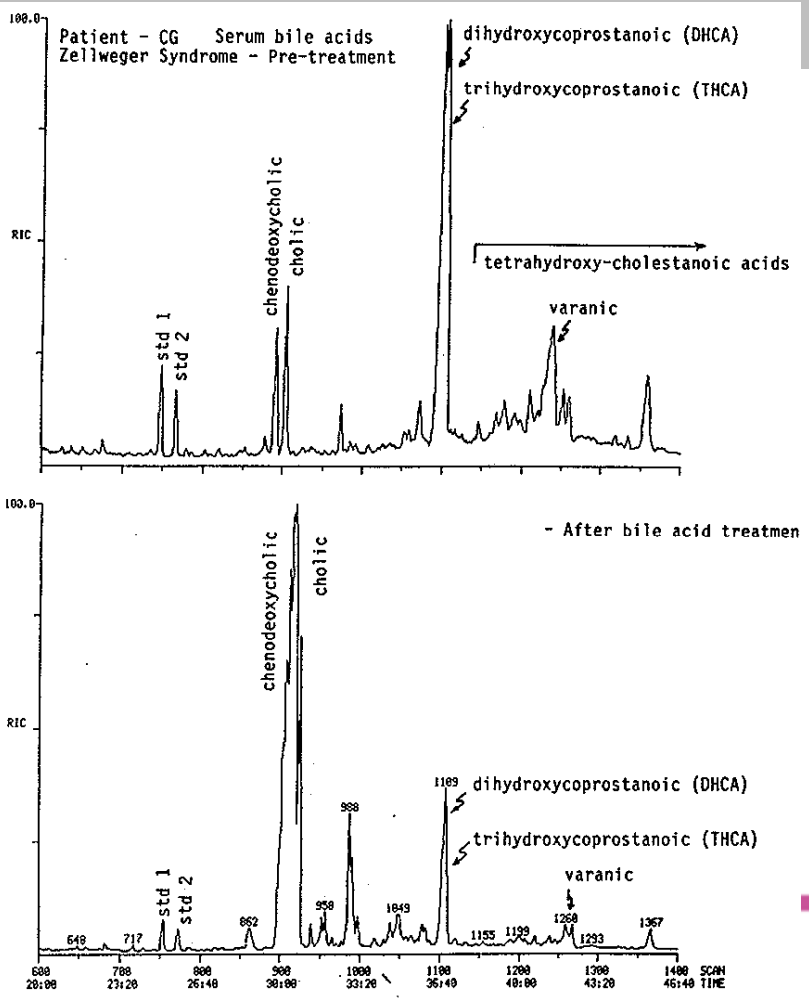
Effect of peroxisomal disorders on BA synthetic pathway



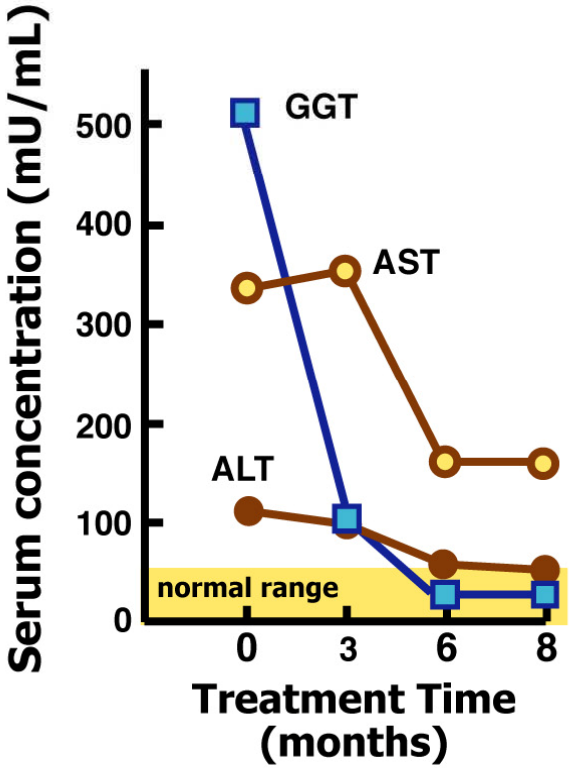
Effect of CA in Peroxismal Disorders

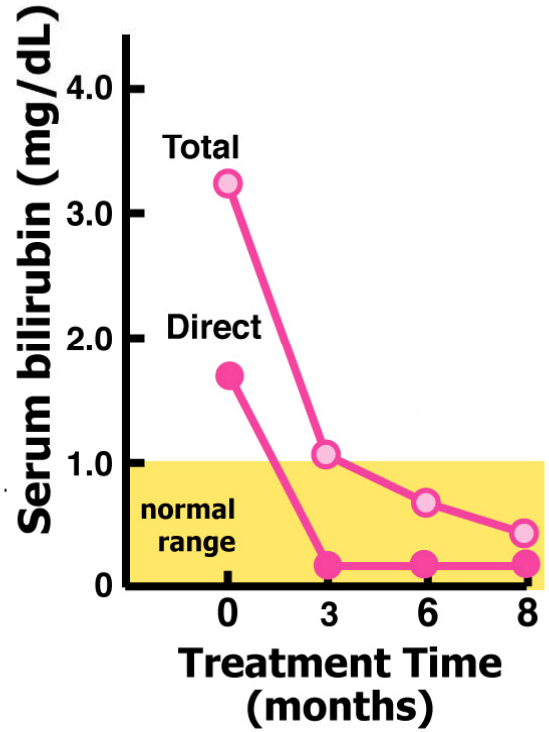


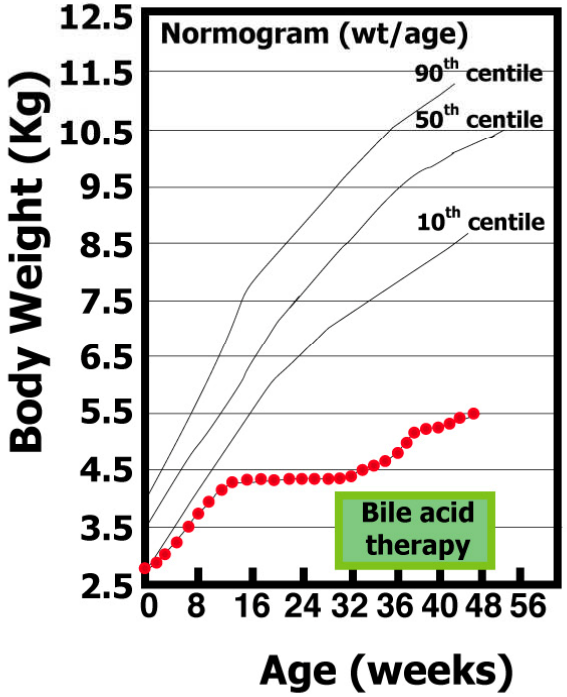
Effect of Oral Primary Bile Acid Treatment on LFT's and Growth in a Patient with Zellwegers

Index Case:

Cholic and chenodeoxycholic acid (15 mg/kg bw/day)

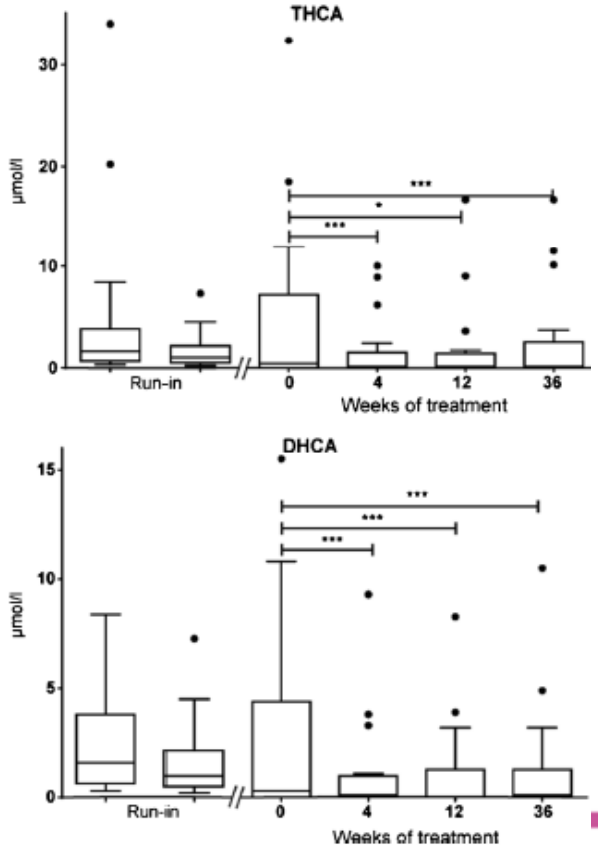






Reference: Setchell et al., Hepatology 1992; 15:198-207

Response to Cholic Acid



Berendse K, et al. J Inherit Metab Dis DOI 10.1007/s10545-016-9962-9

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Safety/Efficacy of Cholic Acid

- Bile acid therapy singled out as curative for BA defects by IOM*
- No drug-related adverse events in > 20 year use
- Impressive clinical response
  - Normalization of liver chemistries
  - Resolution of histologic abnormalities
  - Improved growth
- Treatment failures only in advanced ESLD
- Exceptions (Cholic acid does not work!)
  - Oxysterol 7α-hydroxylase deficiency
  - Conjugation defects (treated with glycocholic acid)

*IOM Report on Rare Diseases and Orphan Products 2010

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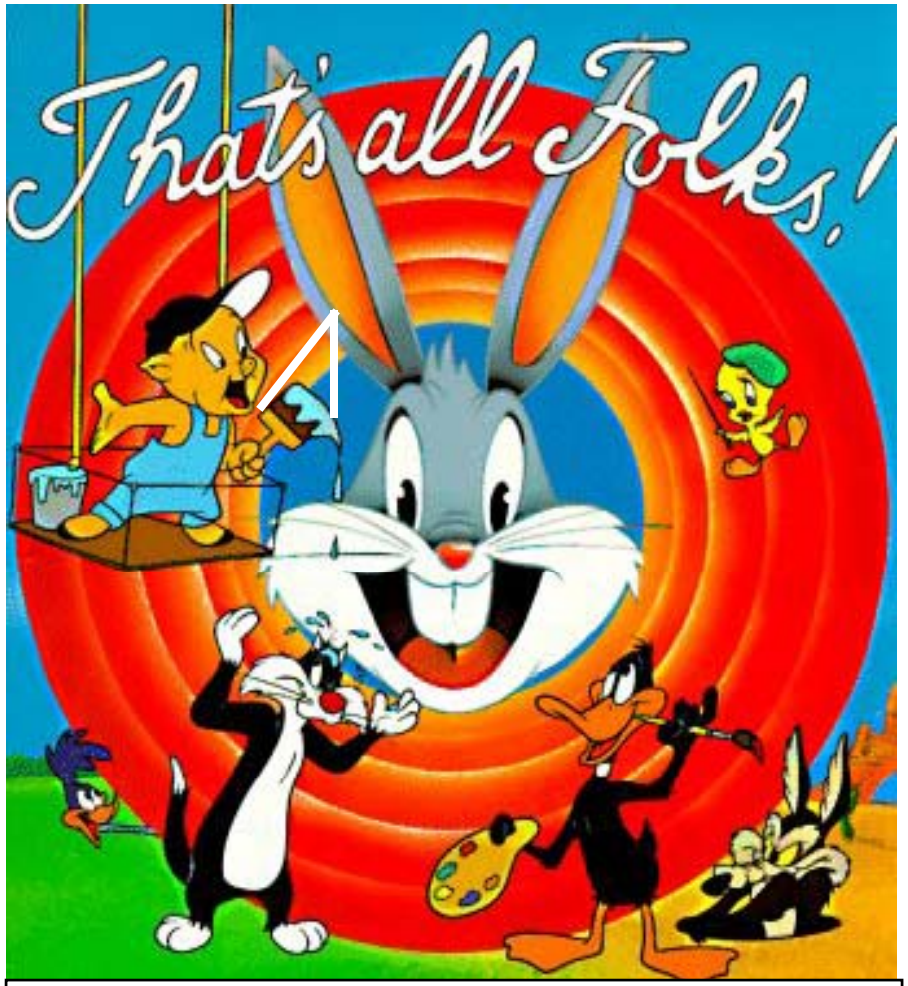

Gracias

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