

# An abdominal pain that you will never forget

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**ESIM**  
**Brighton 2010**



A 60-year-old man was admitted to Internal Medicine Unit complaining of upper abdominal pain and jaundice of some days' duration...

# Past medical history

The patient reported that previously, in concomitance of routine examinations, a **mild cholestatic parameters elevation** (GGT, ALP, bilirubin) was observed. However it was not associated with any clinical manifestation and so additional studies were never performed.

# Past medical history (2)

- His clinical history was unremarkable until December 2004, when he suddenly complained of upper abdominal colicky pain, nausea, and vomiting. On this occasion ultrasonography showed **small gallstones**. These symptoms quickly disappeared spontaneously
- In the following two years the patient referred three other abdominal pain episodes like the previous one
- In March 2006 he experienced a more severe abdominal pain attack: for this reason hospitalization was required and **laparoscopic cholecistectomy** was performed.

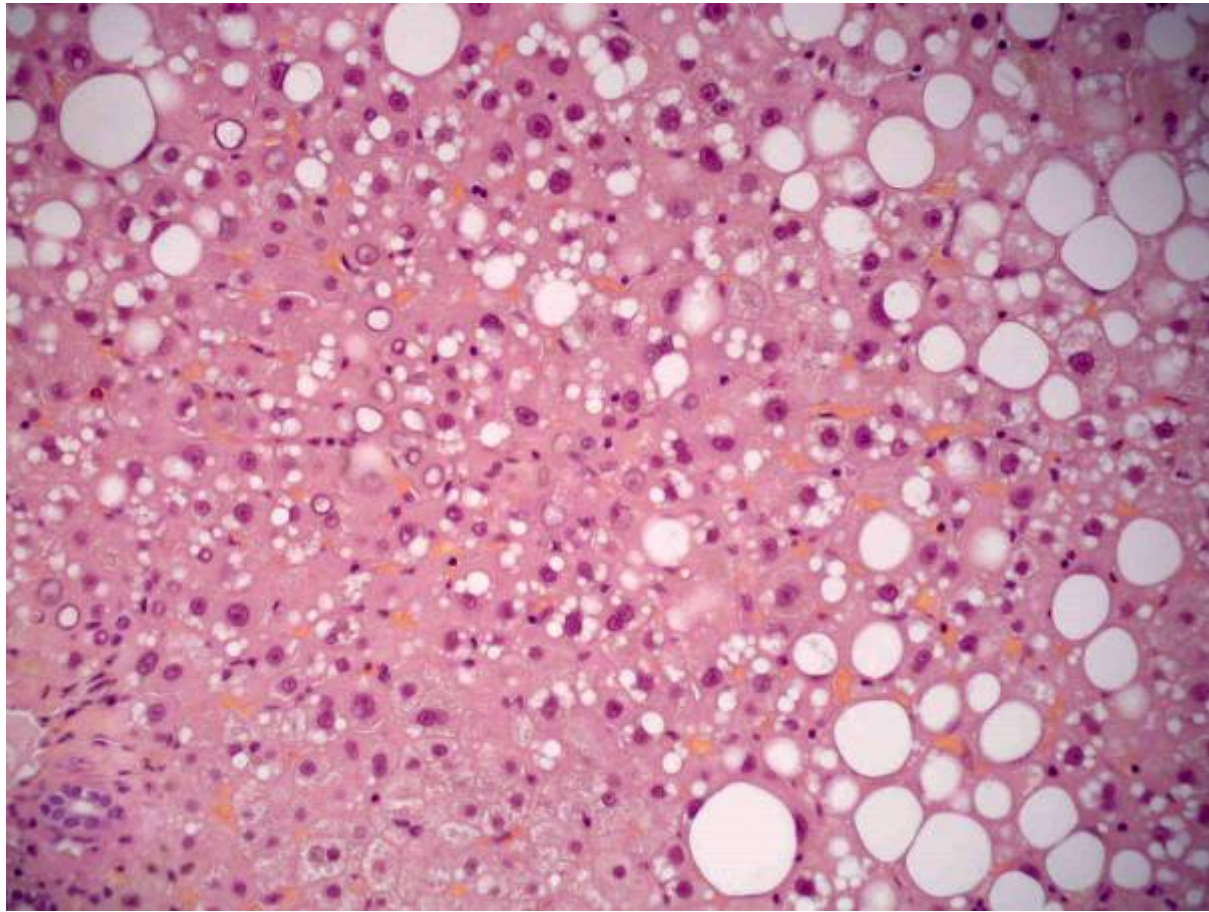
During laparoscopic exploration the surgeon noticed an **abnormal liver appearance**, therefore an intraoperative liver biopsy was performed.



Macroscopic findings of the liver

The liver is enlarged and is dark green in color, with an irregular surface

The histological section showed **micro- and macrovesicular steatosis** with no other pathologic features. The formulated diagnosis was **NASH** (non alcoholic steatohepatitis) so the patient was dismissed and not further investigated.



# Past medical history (3)

- One month before consulting our unit he was admitted to a local hospital for weakness, malaise and jaundice
- **Cholangio-MR** was performed and did not show any abnormality, nevertheless jaundice did not improve and an ERCP was planned
- During the **ERCP procedure cholangiography, biliary brushing** and papillectomy were done
- His hospital course was complicated by iatrogenic pancreatitis secondary to the ERCP
- However clinical and biochemical parameters rapidly normalized with no additional procedures; a residual pancreatic pseudocystis was documented with CT.

- One month after he was admitted to our unit because of marked jaundice and distended abdomen with moderate tenderness and no bowel sounds

# Physical examination

- **Marked jaundice**
- Distended abdomen with moderate tenderness and **no bowel sounds**
- The **liver edge was palpable** 4 cm below the right costal margin at the midclavicular line, with an indistinct margin and firm consistency, whereas the spleen was palpable only during deep inspiration
- **Mild ascites and lower extremities edema were present**
- Heart and lung examinations were inapparent; pulse rate was regular and elevated, up to 120 bpm. Body temperature was normal.

# Initial laboratory investigations

## Peripheral blood

WBC	8990/ $\mu$ l
RBC	$4.4 \times 10^6$ / $\mu$ l
Ht	39.3%
Hb	12.1 g/dl
Plt	$197 \times 10^3$ / $\mu$ l

## Coagulation test

INR	1.20
APTT	43 sec
Fibrinogen	585 mg/dl
XDP	391 ng/ml

**Child-Pugh score  
B9**

## Chemistry

TP	5.3 g/dl
ALB	3.1 g/dl
→ T-Bil	24 mg/dl
→ D-Bil	19 mg/dl
→ GOT	387 IU/l
→ GPT	346 IU/l
LDH	210 IU/l
Ch-E	1799 IU/l

→ ALP	419 IU/l
→ $\gamma$ -GTP	985 IU/l
NH <sub>3</sub>	52 $\mu$ g/dl
BUN	20 mg/dl
Cre	0.9 mg/dl
T-Chol	379 mg/dl
AMY	121 mg/dl
Na	135 mEq/l
K	6.2 mEq/l
Cl	105 mEq/l

ANA, AMA, ASMA and hepatitis B and C serology were all negative, and his serum amylase level was within normal limits.

- Findings on **abdominal X-ray** were consistent with paralytic ileus
- **Ultrasonography** (US) revealed hepatosplenomegaly, a pseudocystis in the pancreas tail, a small amount of free fluid in the Morrison pouch and around the liver, but biliary stones or bile duct dilatation were absent
- **Color Doppler US** showed moderate portal hypertension and portal vein dilatation
- An **abdominal computed tomography (CT) scan** and **cholangio-MR** confirmed the US findings
- Upper **gastroduodenal endoscopy** detected esophageal veins dilatation and gastric congestion

Clinical, laboratory, and  
imaging findings suggested  
primitive paralytic ileus and  
liver failure

# Discussion

- What was the reason of liver failure?
- Which investigations do you suggest during hospitalization in our unit?
- Which kind of treatment do you suggest after admission to our unit?

???

### Urinary porphyrins

Uroporphyrins	116 µg/24 hr
Heptaporphyrins	68 µg/24 hr
Hexaporphyrins	6 µg/24 hr
Pentaporphyrins	8 µg/24 hr
Coproporphyrins I	82 µg/24 hr
Coproporphyrins III	47 µg/24 hr

**Plasma fluorimetric assay (635 nm): positive**  
**Erythrocytic fluorimetric assay (625 nm): positive**  
**Fluorescent erythrocyte percentage: 99.68%**

The obtained data were compatible with erythropoietic protoporphyria (EPP), and the diagnosis was finally confirmed by molecular studies with **ferrochelatase (FECH) gene analysis**.

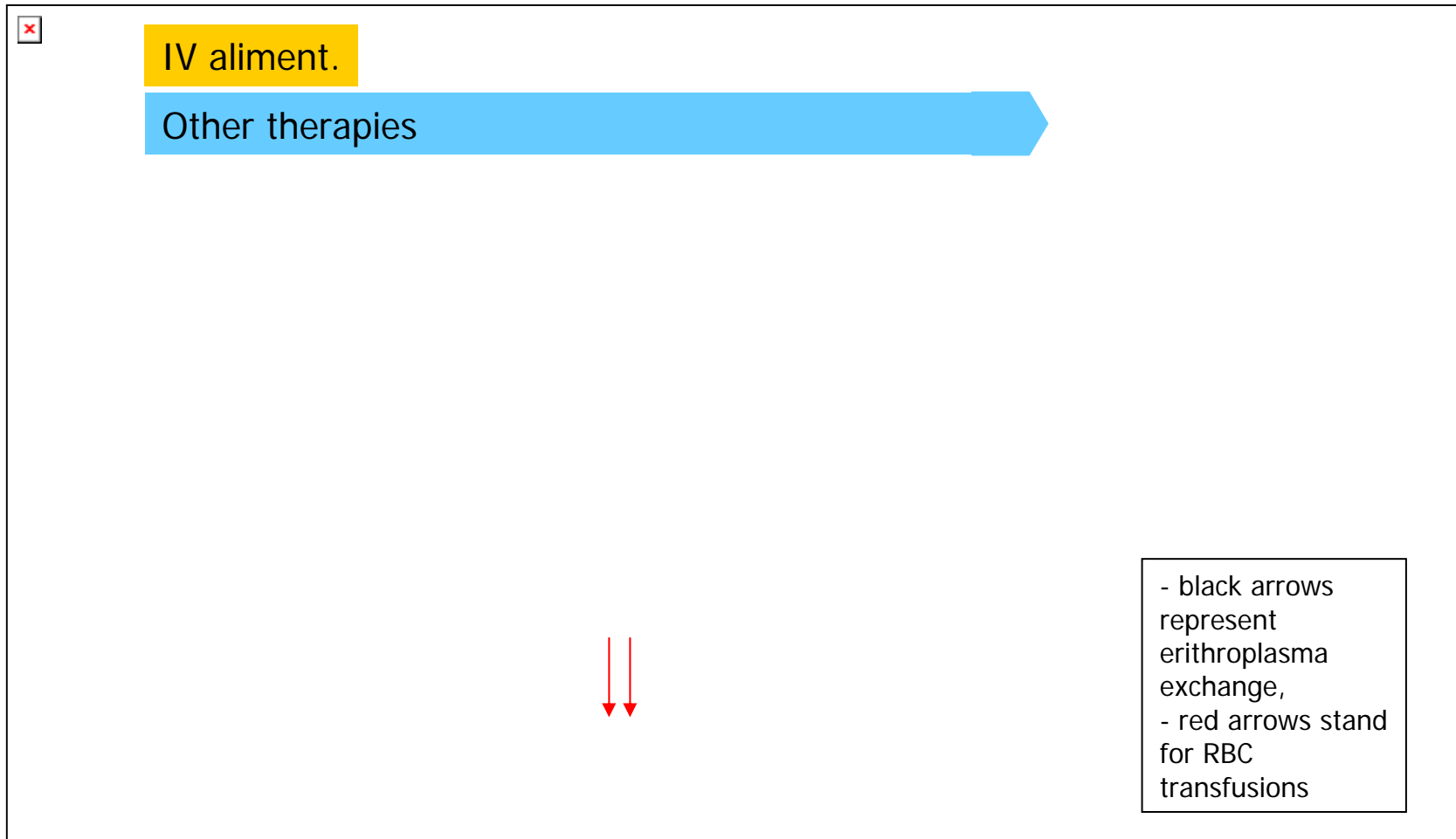
A base-pair insertion (ins T213) in the exon 3 of FECH gene and co-inheritance of a low-expression allele were observed. The low expression allele is the C variant of a single nucleotide polymorphism (SNP; IVS3-48 C/T) in intron 3 of the FECH gene, quite common in general population.

# Therapy

- Intravenous hyperalimentation without allowing oral food intake
- Antibiotic therapy (ciprofloxacin 500 mg/bid) was administered for the initial suspect of acute cholangitis
- Loop diuretics (furosemide 20 mg ev), aldosterone antagonists (canrenoate 100 mg bid) and albumin i.v. (50 ml 20 % bid) were given for liver dysfunction
- Cholestyramine (4 gr tid), activated charcoal (400 mg tid), ursodesoxicholic acid (300 mg qid) and human hemin (200 mg diluted in 100ml of 0.9 per cent sodium chloride in a glass bottle and infused intravenously over at least 30 minutes) were administered

During this period, four **erithroplasma exchanges** were performed (2500 ml of plasma were exchanged with 2500 ml 6% albumin solution together with the exchange of three units of RBC).

In addition, to maintain a Hct level greater than 35 percent, two **RBCs units** were transfused two different times.



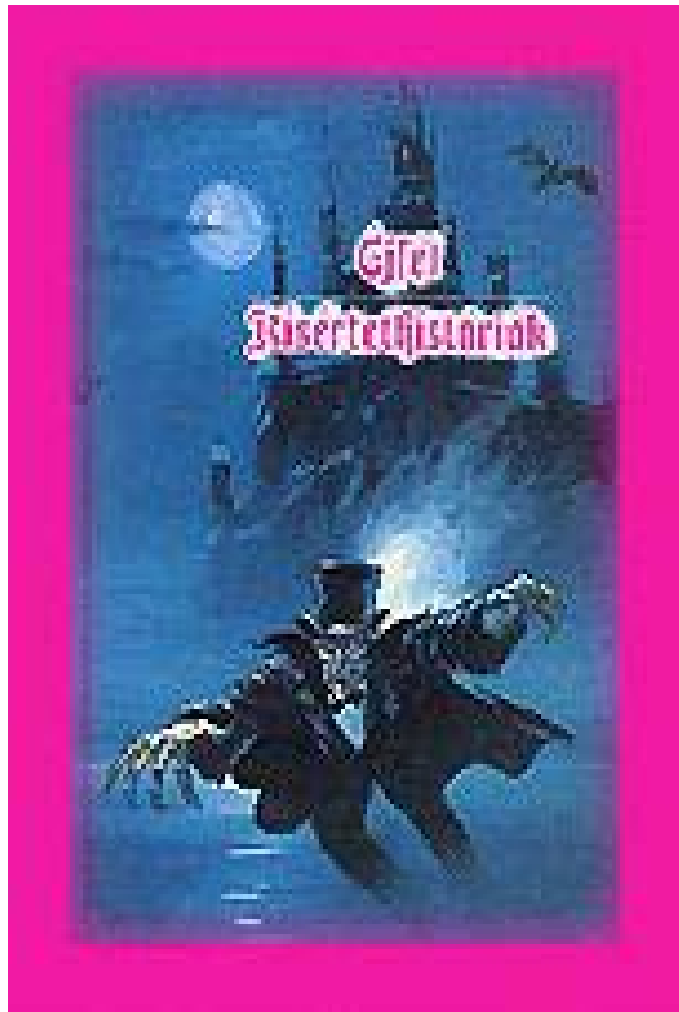
After ten days abdominal pain subsided and bowel sounds were audible. The patient's liver function tests improved as well as his clinical condition. The patient was  
The patient was evaluated for OLT

At day 25 the patient developed delirium, generalized and progressive weakness, and black stool emission. During the night he presented an acute episode of hematemesis, with abundant blood loss. He developed hypovolemic shock and died.

# Discussion

- ❖ **Differential diagnosis of abdominal pain and paralytic ileus** (choledocholithiasis, acute pancreatitis, acute cholangitis, ... .., **autonomic neurological syndrome**)
- ❖ **Specific therapy** (cholestyramine, activated charcoal, ursodesoxicholic acid, human hemin, erithroplasma exchanges, RBCs transfusion)
- ❖ In this disease **hepatic dysfunction** is a critical outcome determinant -> **OLT**





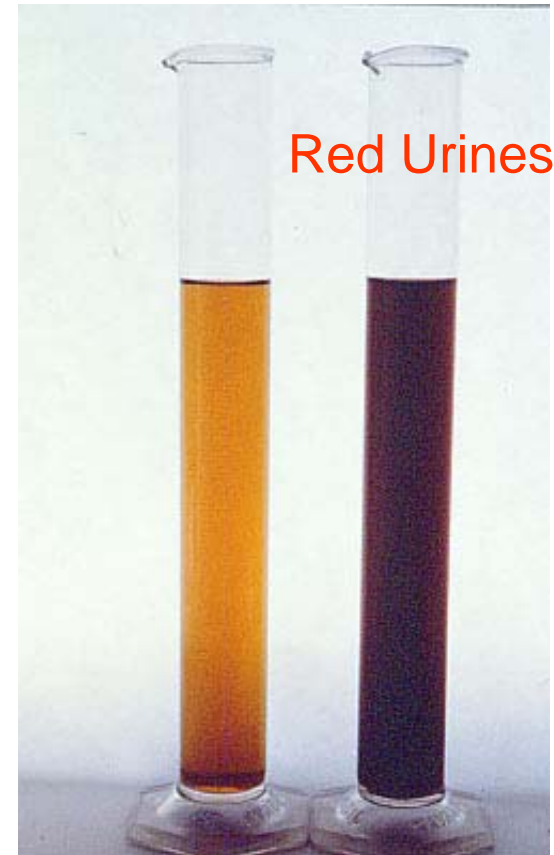
## Human Hereditary Porphyrias: What's new ?



# Human Porphyrias

**“Obscure diseases with confusing names considered only when the need for a diagnosis is desperate”**

*(Antony McDonagh, 1997)*

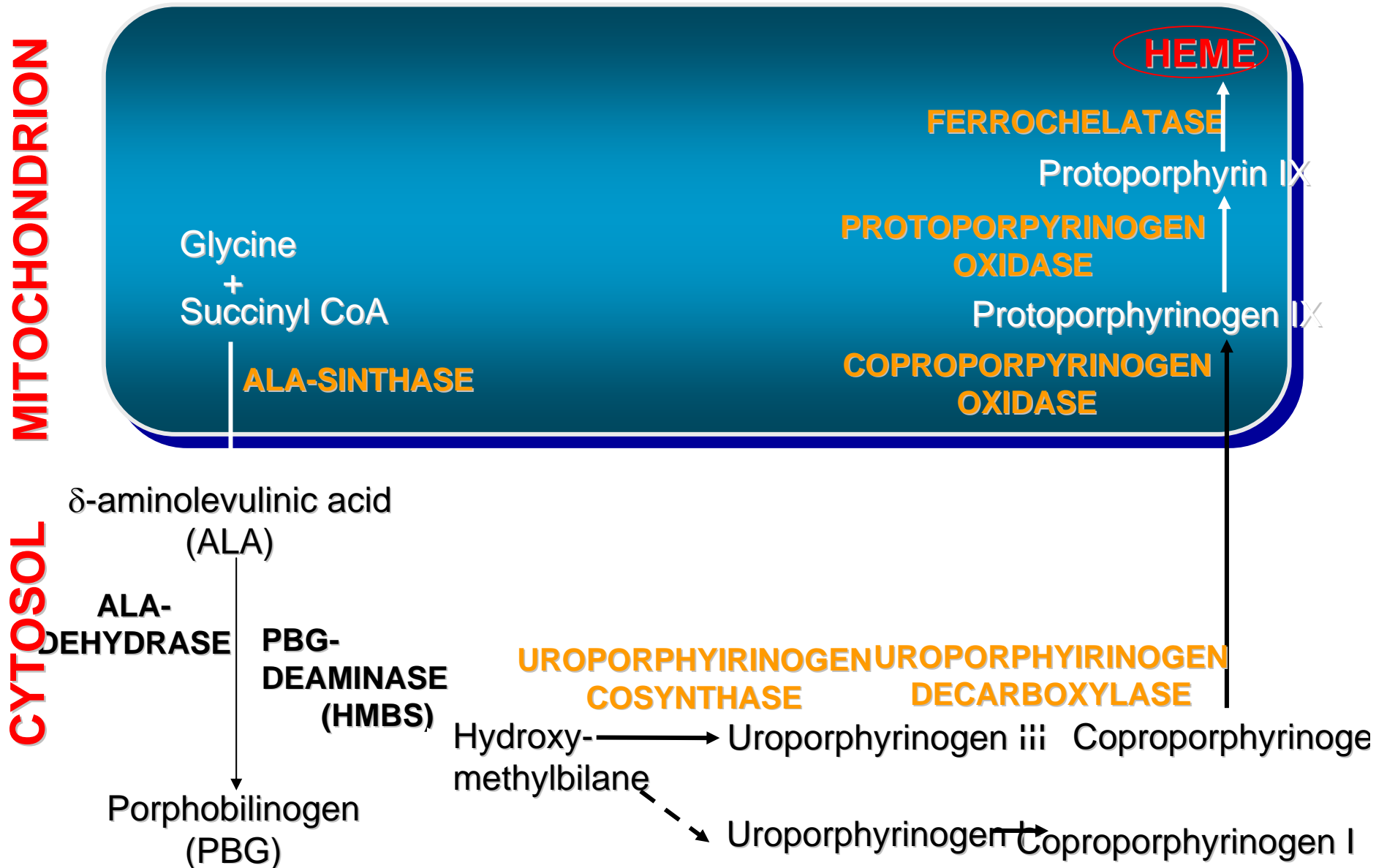


# The Porphyrrias

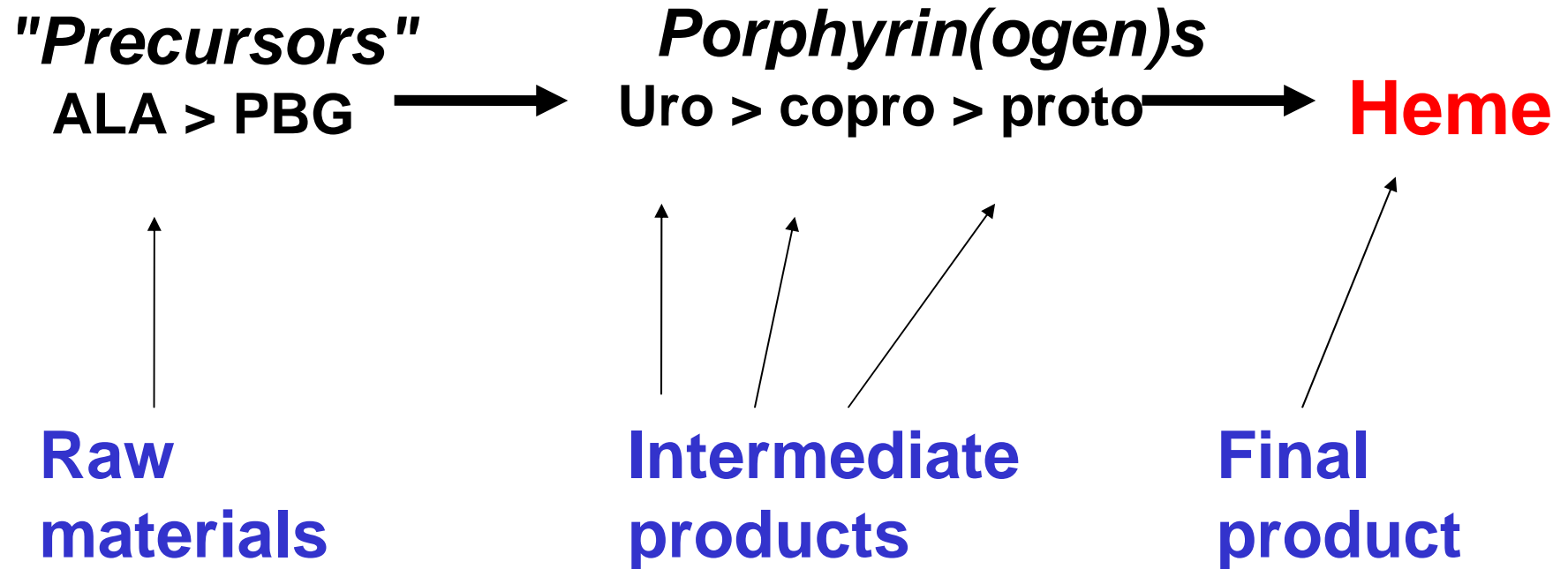
“Porphyrrias” include a wide spectrum of hereditary disorders due to **abnormal heme biosynthesis**

In relation to the enzyme defect along the heme biosynthetic pathway, each type of porphyria is characterized by a specific pattern of overproduction, accumulation and excretion of different heme precursors.

# The heme biosynthetic pathway



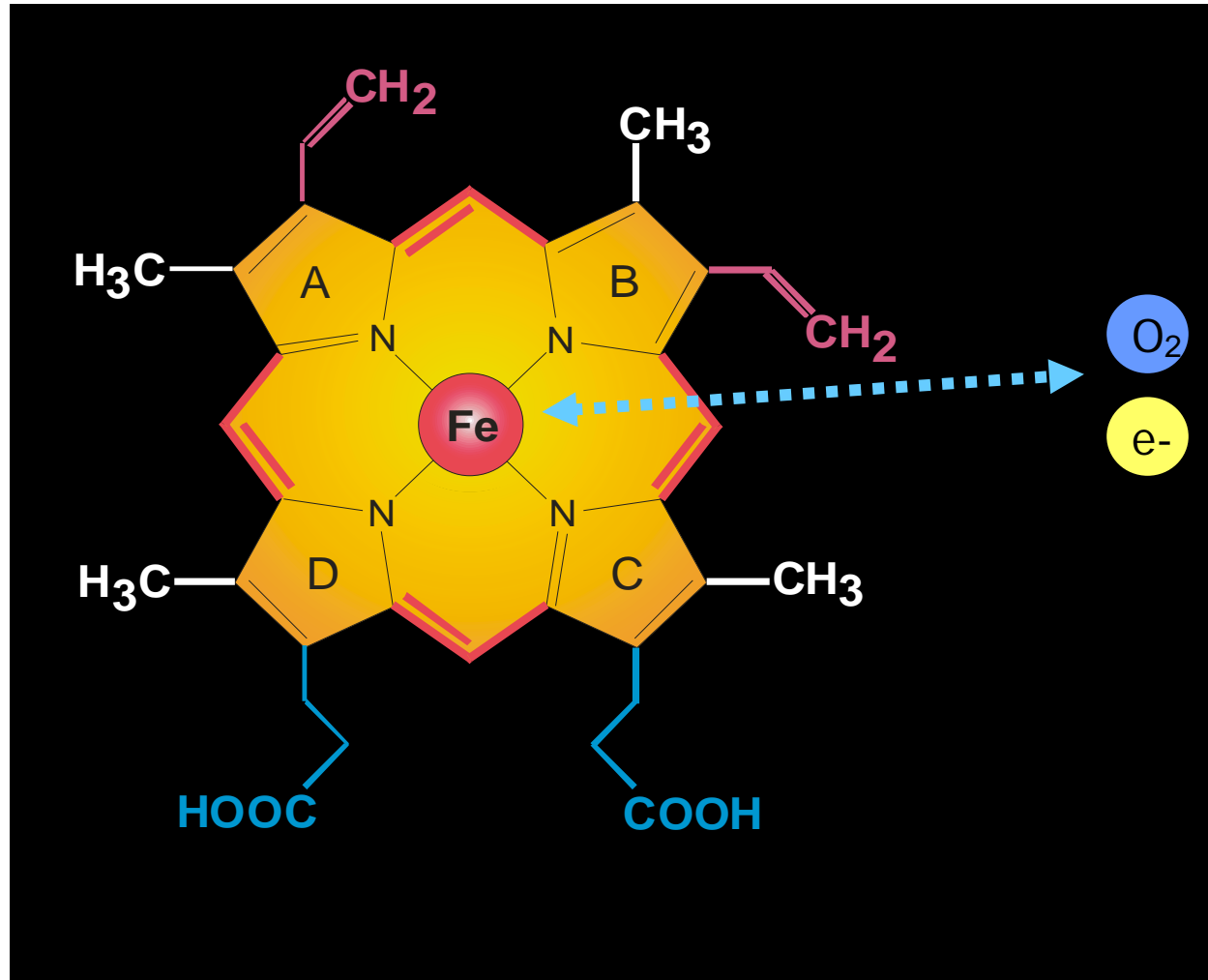
# Heme Biosynthesis



*ALA :  $\delta$  Aminolevulinic acid, PBG : Porphobilinogen*

# Heme

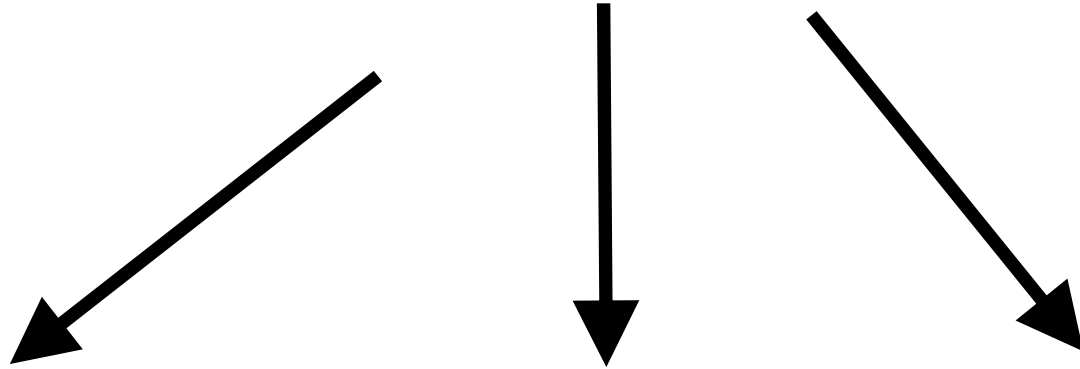
Protoporphyrin IX + Fe<sup>2+</sup>



# Hemoproteins

- **O<sub>2</sub> transport and storage** Hemoglobin / Myoglobin
- **Cellular respiratory chain** Cytochromes
- **Hepatic detoxification** Cytochromes P450
- **H<sub>2</sub>O<sub>2</sub> metabolism** Catalase / peroxydase
- **Fe reduction** Dcytb (duodenal cytochrome b)
- **Tryptophan catabolism** Tryptophan 2,3 dioxigenase
- **Prostaglandins synthesis** Cyclooxygenase
- ...

# Heme



**Erythroid  
System  
85%**

**Liver  
14%**

**Other  
Cells  
1%**

# Major Heme Function

- 85%: Oxygen transport  
Hb 98% (**Bone Marrow**), MyoG 2%
- 15%: Oxidation (**Liver** + all cells)

Non-inducible (Mito cyto & cytosolic b5, 15%)  
Inducible (P450, 65%)

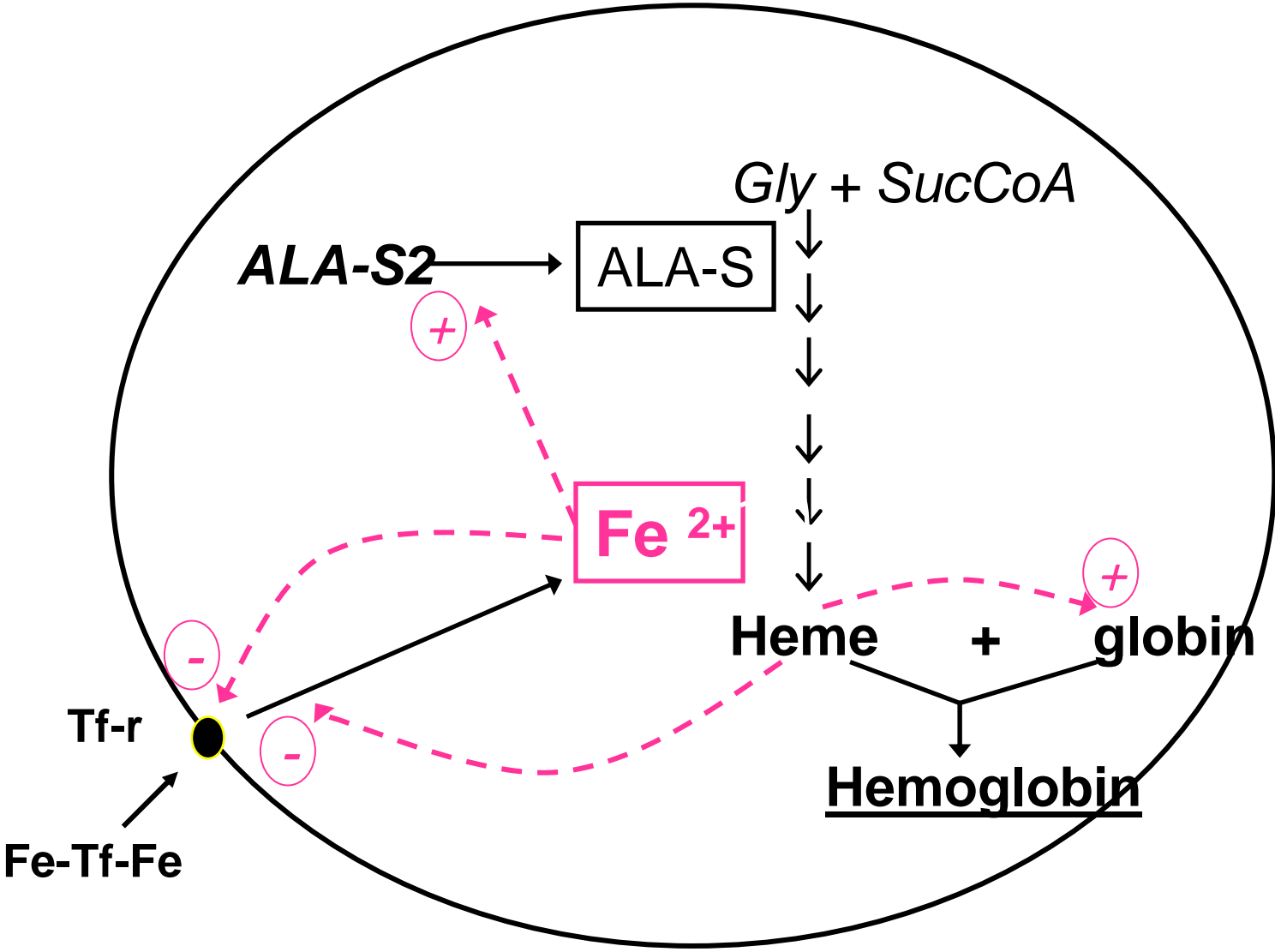
*A single biosynthetic pathway that needs two tissue-specific regulations :*

**BM** : continuous massive erythroid production

**Liver** : rapid, according to local needs

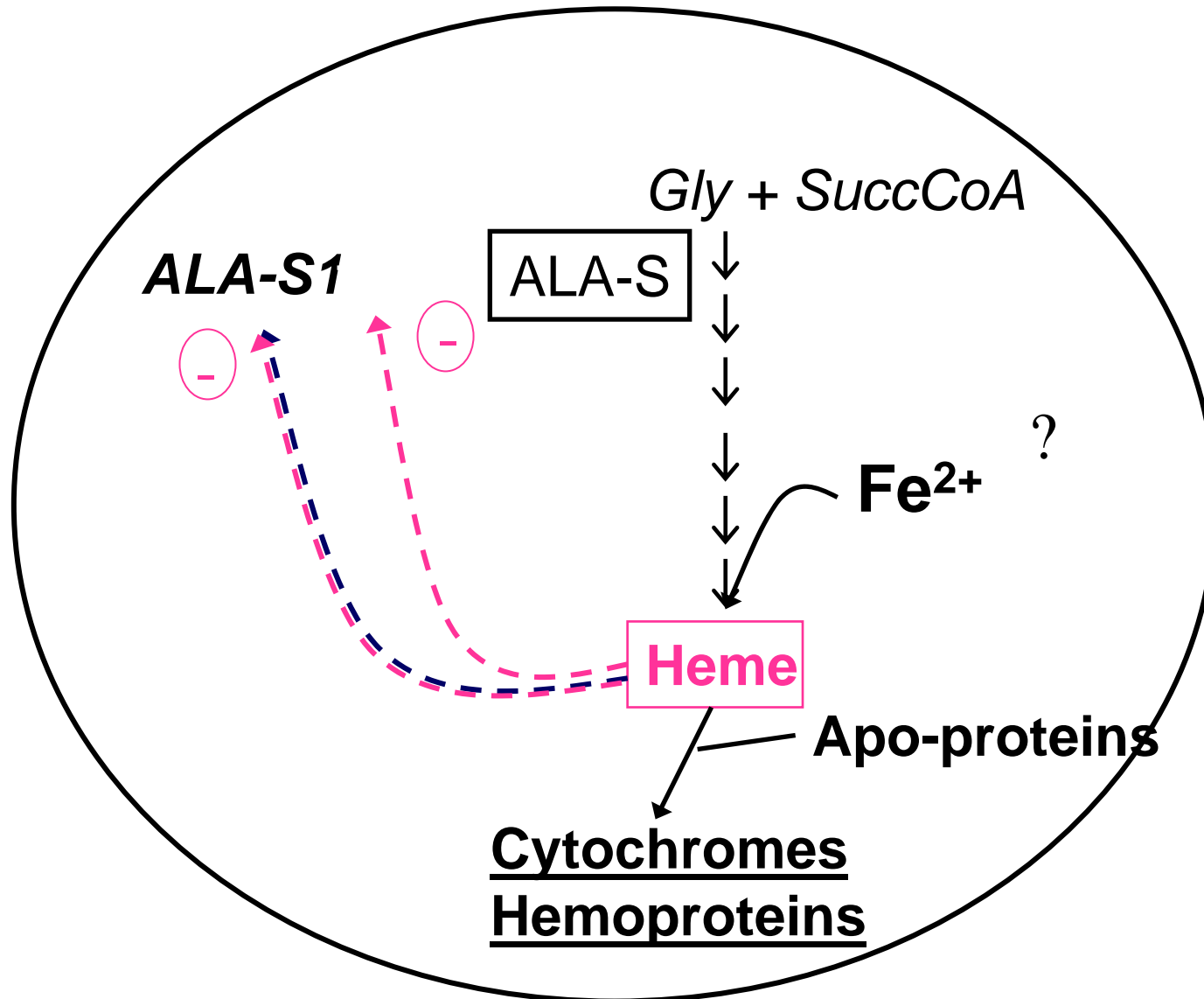
# Erythroid Heme Synthesis Regulation

*B.Marrow : 85% of body heme production*

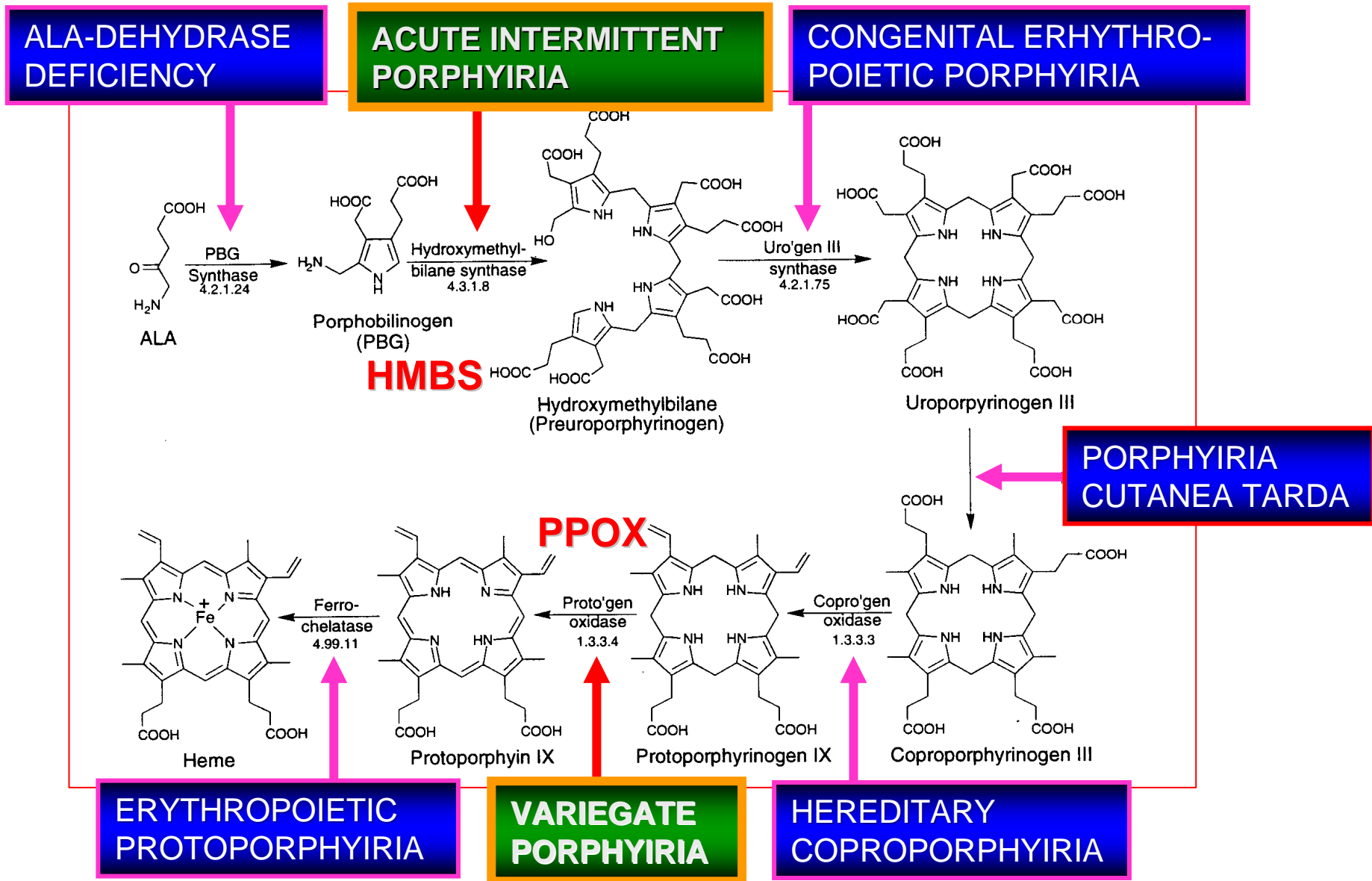


# Hepatic Heme Synthesis Regulation

*Liver : 15% of body heme production*



# Heme biosynthesis and porphyrias



# THE PORPHYRIAS

✓ The porphyrias are classified in **acute or chronic** on the bases of clinical manifestations or in **erythropoietic or hepatic** depending on the site of expression of the enzyme defect.

# CLASSIFICATION OF PORPHYRIAS

## ***ACUTE PORPHYRIAS***

## ***CHRONIC PORPHYRIAS***

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ALAD deficiency porphyria (ADP)  
porphyria (CEP)

Congenital erythropoietic

**Acute intermittent  
Porphyria (AIP)**

**Porphyria cutanea  
tarda (PCT)**

**Erythropoietic  
protoporphyrin (EPP)**

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Hereditary coproporphyrin (HCP)

Variegate porphyria (VP)

# Classification

## Hepatic porphyrias (adult)

- Acute Intermittent Porphyria, AIP
- Hereditary Coproporphyria, HC
- Variegate Porphyria, VP
- Porphyria cutanea, PCT

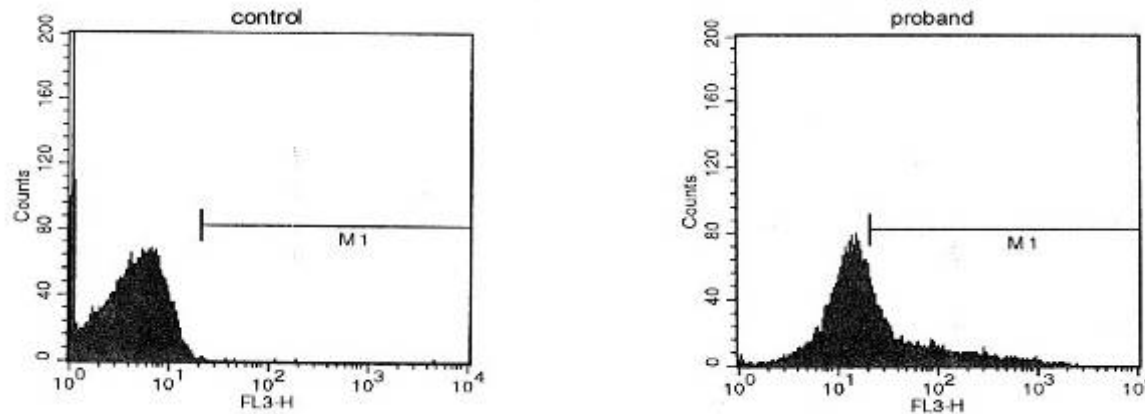
} Acute

## Erythropoietic porphyrias (child)

- Congenital Erythropoietic Porphyria, CEP
- Erythropoietic Protoporphyria, EPP
- X linked Erythropoietic protoporphyria, XLDPP

# Erythropoietic Protoporphyria (EPP)

- Prevalence: 1:75.000-1:200.000
- Accumulation of protoporphyrin IX in erythrocytes, plasma, urine, faeces and skin.



- Early childhood onset of lifelong acute photosensitivity of sun-exposed skin.
- **In about 2% of patients, severe liver disease**

# ERYTHROPOIETIC PROTOPORPHYRIA

Erythropoietic protoporphyria is the most common erythropoietic porphyria and, after *porphyria cutanea tarda* (PCT), the second most common porphyria.

There is phenotypic variation in this disease.

## Clinical Features :

Skin photosensitivity usually begins in childhood. Redness, swelling, burning, and itching can develop within minutes of sun exposure and resemble angioedema (differ from those of other porphyrias).

Liver function is usually normal, but in some patients accumulation of protoporphyrin causes chronic liver disease that can progress to liver failure and death (probably result, in part, from protoporphyrin accumulation in the liver, in fact, protoporphyrin is insoluble, forms crystalline structures in liver cells, and can decrease hepatic bile flow).

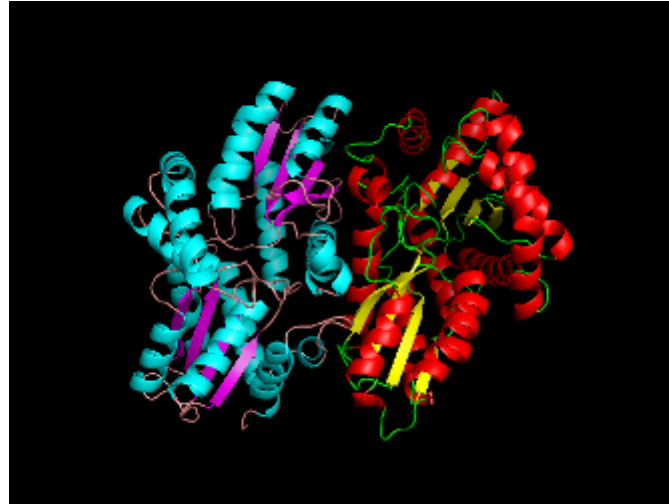
Gallstones composed at least in part of protoporphyrin occur in some patients.

# Erythropoietic Protoporphyria (EPP)



**light/sun  
sensitivity**

# FERROCHELATASE



- Enzyme localized on mitochondrial membrane
- Active as omodimere

# Autosomal-dominant EPP (dEPP)

Inherited disorder caused by deficiency of Ferrochelatase (FECH)

*FECH* mutation → abolishment of FECH activity

and

Low expressed *FECH* allele

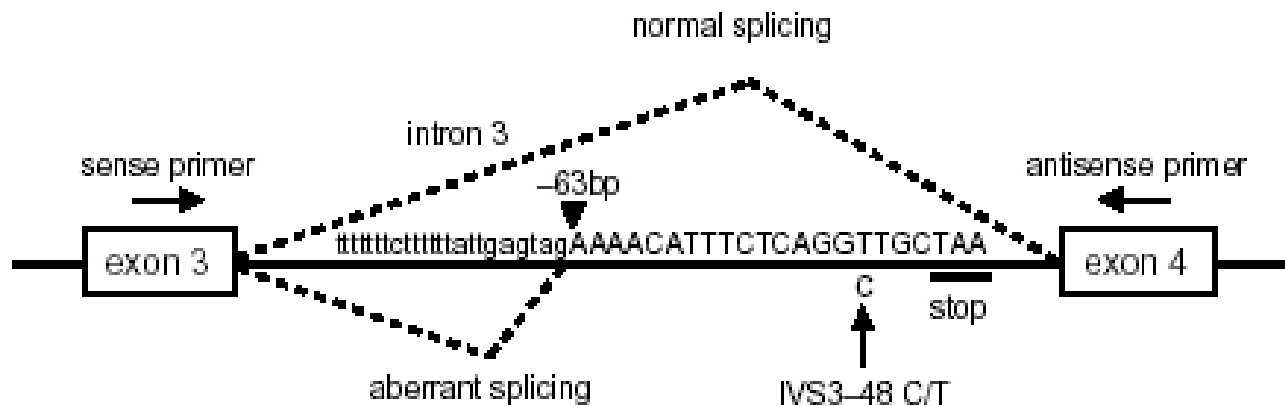
a) IVS3~48 T/C

b) Haplotype:

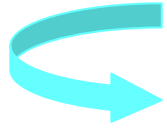
-251 A/G

IVS1~23 C/T

IVS3~48 T/C

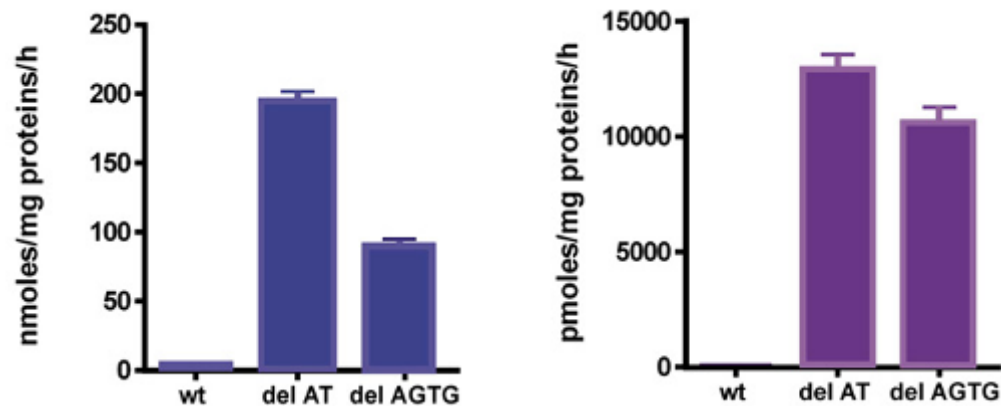


## Whatley et. Al (2008) : X-linked Erythropoietic Protophoryria



c.1706–1709 delAGTG and c.1699–1700delAT in exon 11 *ALAS2*

Expression studies in E.coli showed that both deletions increase ALAS2 activity



**Expression of wild-type and mutant ALAS2 enzymes:  
Rates of formation of ALA and porphyrin**

**Increased ALAS2 activity could explain the over-production of protoporphyrin IX, despite of normal FECH activity often observed in some patients**



**7 Italian unrelated *FECH*-negative EPP families (19 subjects)**



**No mutation on: *ALAS2* IRE**

**Mitoferrin (SLC25A37)**

**Interacting region of ABC7 with the iron sulfur cluster of FECH**

# TREATMENT OF PROTOPORPHYRIA

- Oral  $\beta$ -carotene
- Cholestyramine and other porphyrin absorbents such as activated charcoal
- Caloric restriction and drugs or hormones that may induce the heme pathway or impair hepatic excretory function should be avoided
- Transfusions or intravenous heme therapy may suppress erythroid and hepatic protoporphyrin production and are sometimes beneficial
- **Liver transplantation has been carried out in some patients with severe liver complications**

## Skin lesions in Variegate Porphyria





# Skin lesion treatment

## ■ PCT :

- recurrent phlebotomies
- Low dose chloroquine
  - ➔ works very well

## ■ VP :

- *Neither phlebotomies nor chloroquine are efficacious*

***Porphyrins vs porphyrinogens accumulation ?***

# Acute Porphyrias

# Clinical manifestations in acute porphyrias

<b>Visceral symptoms</b>	<b>Iperactivity SNS</b>	<b>CNS</b>	<b>Peripheral nervous system neuropathy</b>
Abdominal pain Nausea Vomiting Constipation Diarrhea (rare) Fever	Tachycardia Hypertension Ortostatic Hypotension (rare) Tremors Urinary retention Excessive sweating Electrolytic	Depression Confusion Agitation Instabilità emotiva Hallucinations Anxiety Insomnia Delirium Convulsions Epilepsy	Motor weakness Paraplegia Tetraplegia Optic nerves atrophy Disfagia Oftalmoplegia Paresthesia

# Common case report of acute attacks in AHP

- **Woman <35, recurrent periods of severe abdominal and back pain with nausea, vomiting, constipation, insomnia, irritability... frequently before the menses. Important loss of weight and weakness after weeks, and sometimes dark or red urines...**
- **All physical examination normal. Opiates only active on pain**
- **No diagnosis at ED...but after recurrent venue :**
  - Surgeon or psychiatrist or both consulted...
  - ...Often severe consequences with increased pain, extensive paralysis, mental disturbances
- **Diagnosis usually made in ICU...on red urine, differential diagnosis of polyradiculonevritis**

## **Acute Neurovisceral Attack :** ***General Clinical Features***

- **Women (80 %), men (20 %)**
- **Mean age 20-45, rare before puberty**
- **Severe abdominal pain +++**
- **Risk of neuropathy (paralysis)**
- **Precipitating factors (hormones, fasting, infection, drugs, alcohol, stress...)**

# Acute Porphyric Attack :

## *Incidence of Symptoms*

■ <b>Abdominal and back pain</b>	<b>99 %</b>
■ Muscle weakness	90
■ Vomiting, constipation	72
■ Tachycardia	62
■ Insomnia, anxiety, agitation	60
■ Hypertension	45
■ Convulsions	15
■ Paralysis	10
■ <i>Colored/red urines</i>	<i>90</i>

# Neuropathy in acute attacks

## Autonomic

**Abdominal pain**

**Constipation**

**Vomiting**

**Hypertension**

**Tachycardia**

## Peripheral

**Motor neuropathy**

**Extremity pain**

## CNS

**Anxiety**

**Hallucinations**

**Agitation**

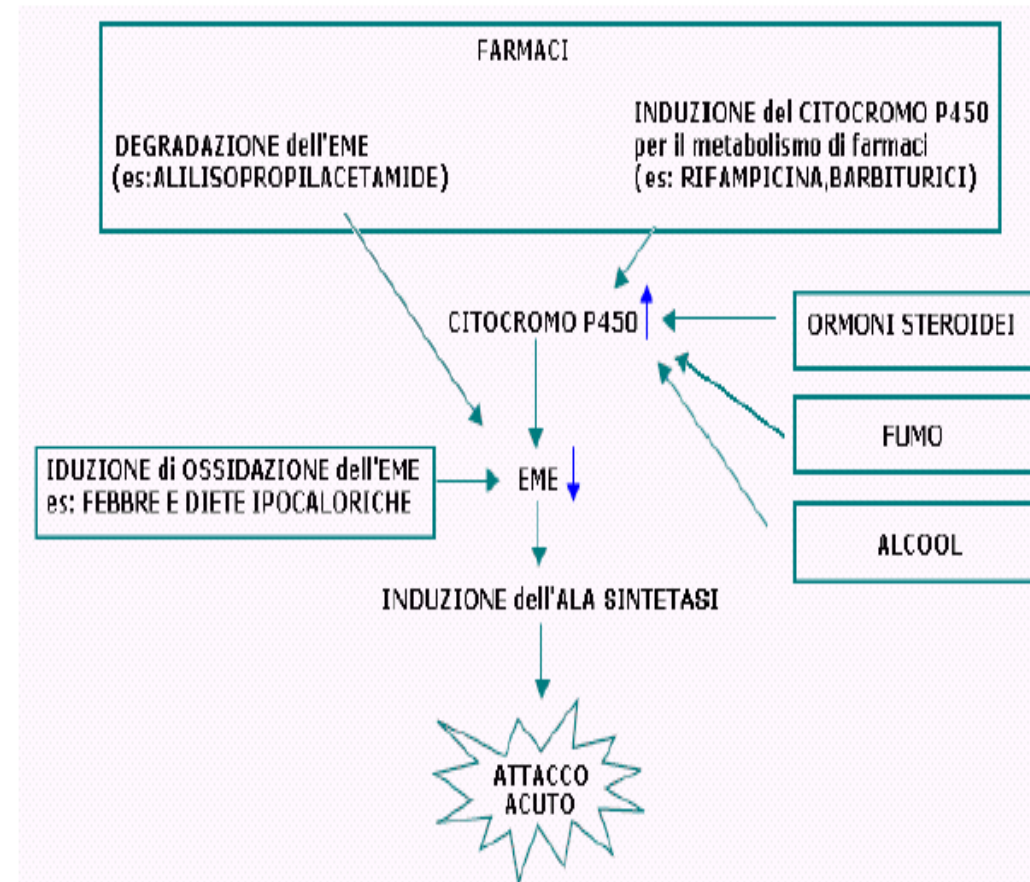
**Epilepsy**

# Hypotheses of the pathogenesis of nervous system dysfunction in acute porphyrias

- ALA, PBG or porphyrins overproduced and accumulating in liver or nervous system are neurotoxic
- A relative heme deficiency in the liver and/or nervous system leads to decreased hemeprotein function in neural tissues
- Abnormal products derived from ALA or PBG are neurotoxic (free radicals, hydroxyhemopyrroline, porphobilin)
- Depletion of essential substrates or cofactors resulting from the disturbance of heme synthesis cause the symptoms (depletion of pyridoxal phosphate, zinc or glycine)

# Trigger factors in acute porphyrias

Drugs  
Estrogens  
Oral contraceptives  
Pregnancy  
Menses  
Alcohol  
Smoke (?)  
Infections  
Stress  
Hypocaloric intake



# Biological diagnosis

## *Symptomatic Patient*

- ALA, **PBG** in urine : 10 to 50 X N  
→ **Acute attack of hepatic porphyria**  
*Sensibility and specificity: 100%*
- ALA, PBG & porphyrins in urine, faeces and plasma  
→ **Type of acute hepatic porphyria**  
→ **orientation for enzymatic diagnosis**

# I step: biochemical analysis

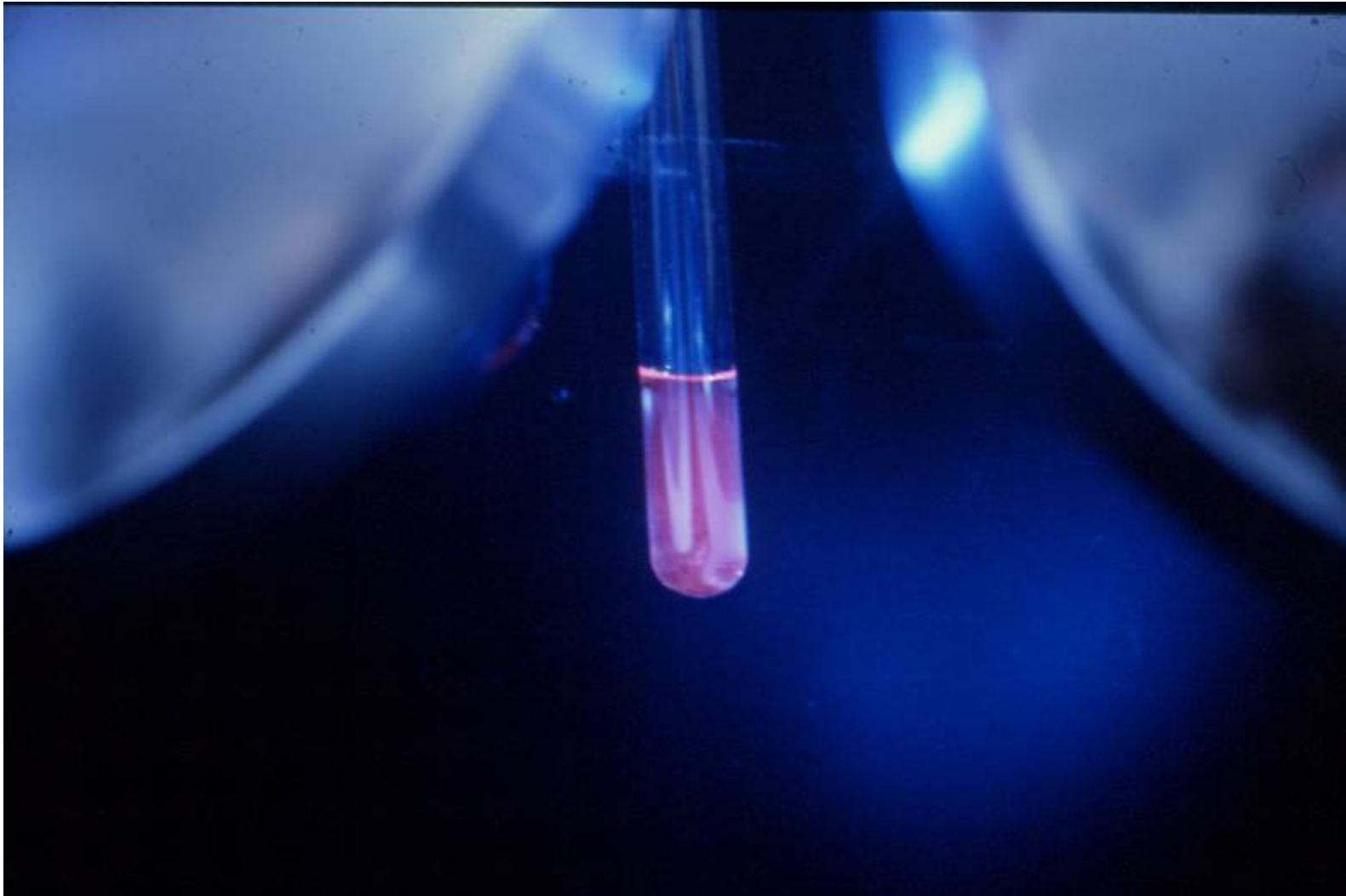
a) Plasma: Fluorescent pick

b) Erythrocytes: Fluorocytes

c) Urine: ALA, PBG, Total porphyrins (Uro, Copro)

d) Feces: Total porphyrins (Uro, Copro, Proto, Iso-copro)

Liver and skin: Biopsy !!!!



# **Specific Biochemical Condition in Acute Porphyric Attacks**

- **Markedly increased activity of ALA synthase  
In the liver**
- **Increased production, accumulation and  
excretion of ALA and PBG**
- **specific porphyrin excretion profile  
depending on the location of the enzymatic  
defect**

# Molecular analysis:

- a) Sequence of candidate genes
- b) RT-PCR → cDNA-PCR
- c) Gel Electrophoresis (PAGE)
- d) Electromobility Shift Assay (
- e) Extra-long PCR
- f) MLPA
- g) New potential genes**

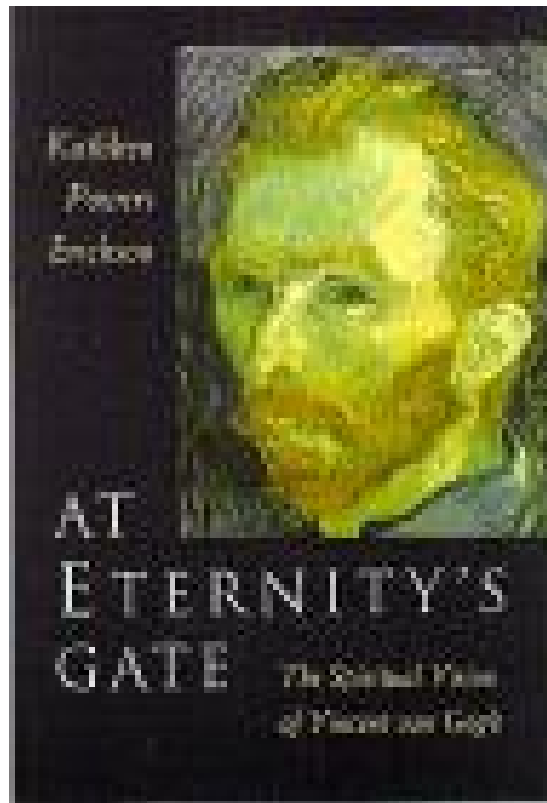
# Management of an acute porphyric attack

- Admission to hospital
- Withdrawal of all common precipitants (drugs, alcohol, fasting, infection...)
- Opiates and chlorpromazine
- Carbohydrates (200-300 g/day)
- Early Normosang® (human hemin) infusion (5 mg/kg/24 hours x 4 days)

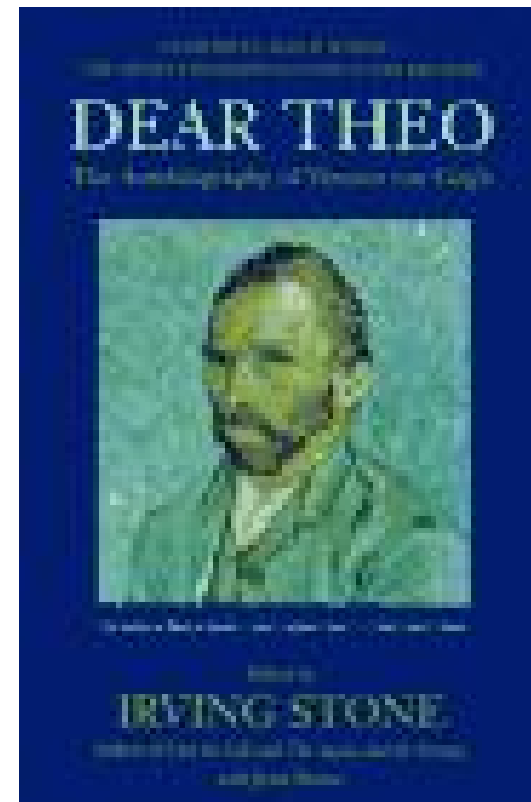
# Normosang<sup>®</sup> in Acute Attacks

## *Rapid clinical improvement*

- Mean abdominal pain duration 2-3 d
- Decreased urinary ALA/PBG 2-3 d
- Hospital duration < 5 d
- No further neurological complications
- No major side effects
- No known risk during pregnancy

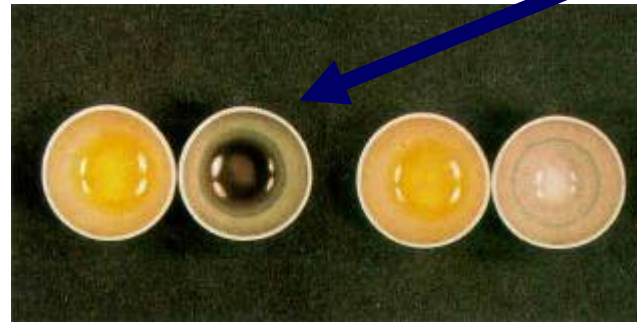


van Gogh ?





# King George III



**Royal family Physicien  
Sir George Backer,  
18 Ottobre 1788**



# *Porphyrias at the European level*

