Galactosaemia and Immunoreactive Trypsin

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Raised serum or dried bloodspot immunoreactive trypsin (IRT) may occur in:-

Occlusion of pancreatic ductules/ducts

Hyperconcentration of ductular fluid
(for example in Cystic Fibrosis where the Cystic Fibrosis Transmembrane Regulator (CFTR) is defective)

Exocrine pancreatic inflammation/necrosis

- In infancy raised serum or bloodspot IRT is a characteristic finding in Cystic Fibrosis

- Used as the basis of newborn screening programmes for CF

IRT is not ideal!

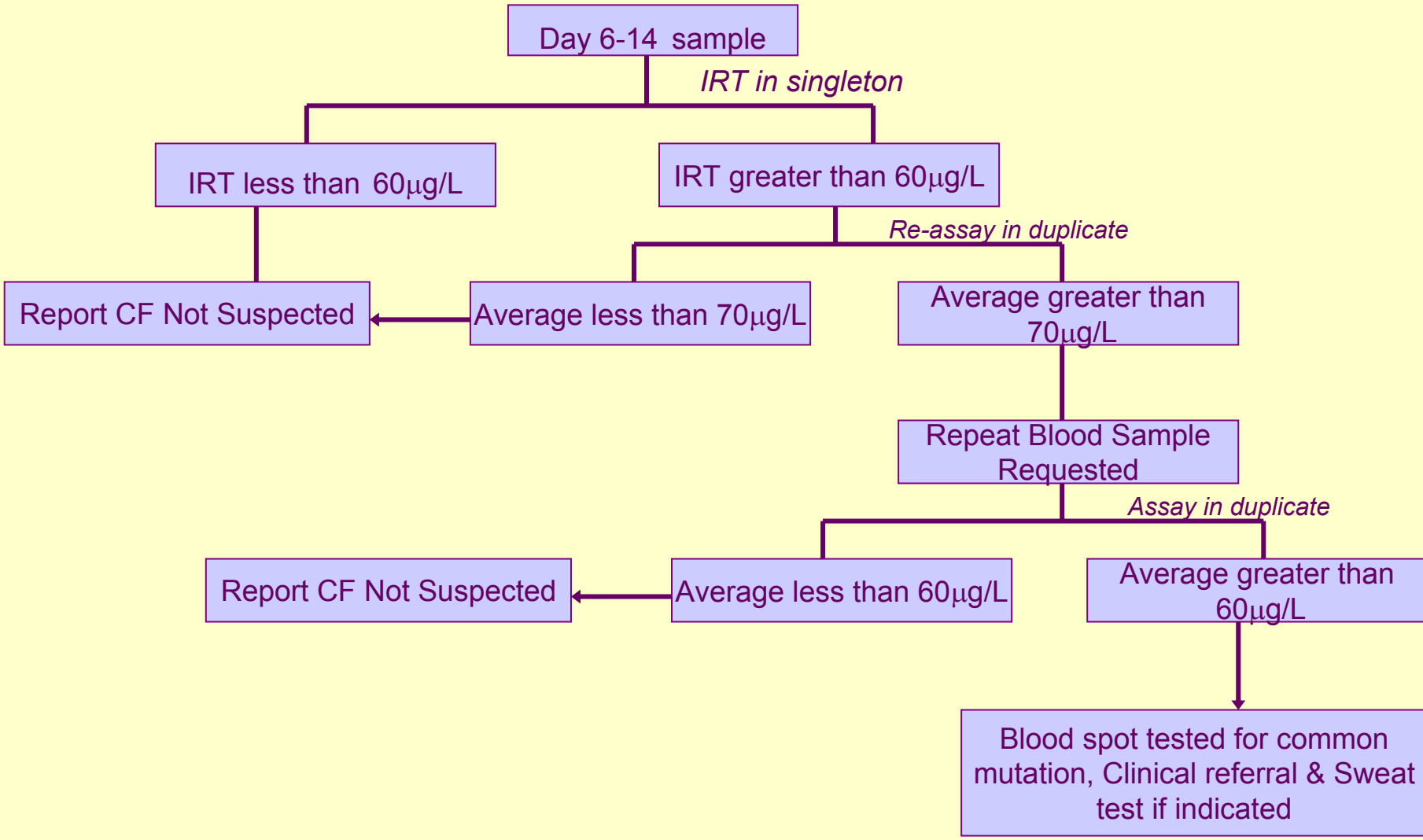
- Newborns have higher values of IRT than adults
- Values decrease to adult levels by about 6 wks of age
- Most babies with CF (~98%) will have raised values of IRT on days 5-8

- When tested on days 5-8 there is overlap between unaffected babies and CF babies

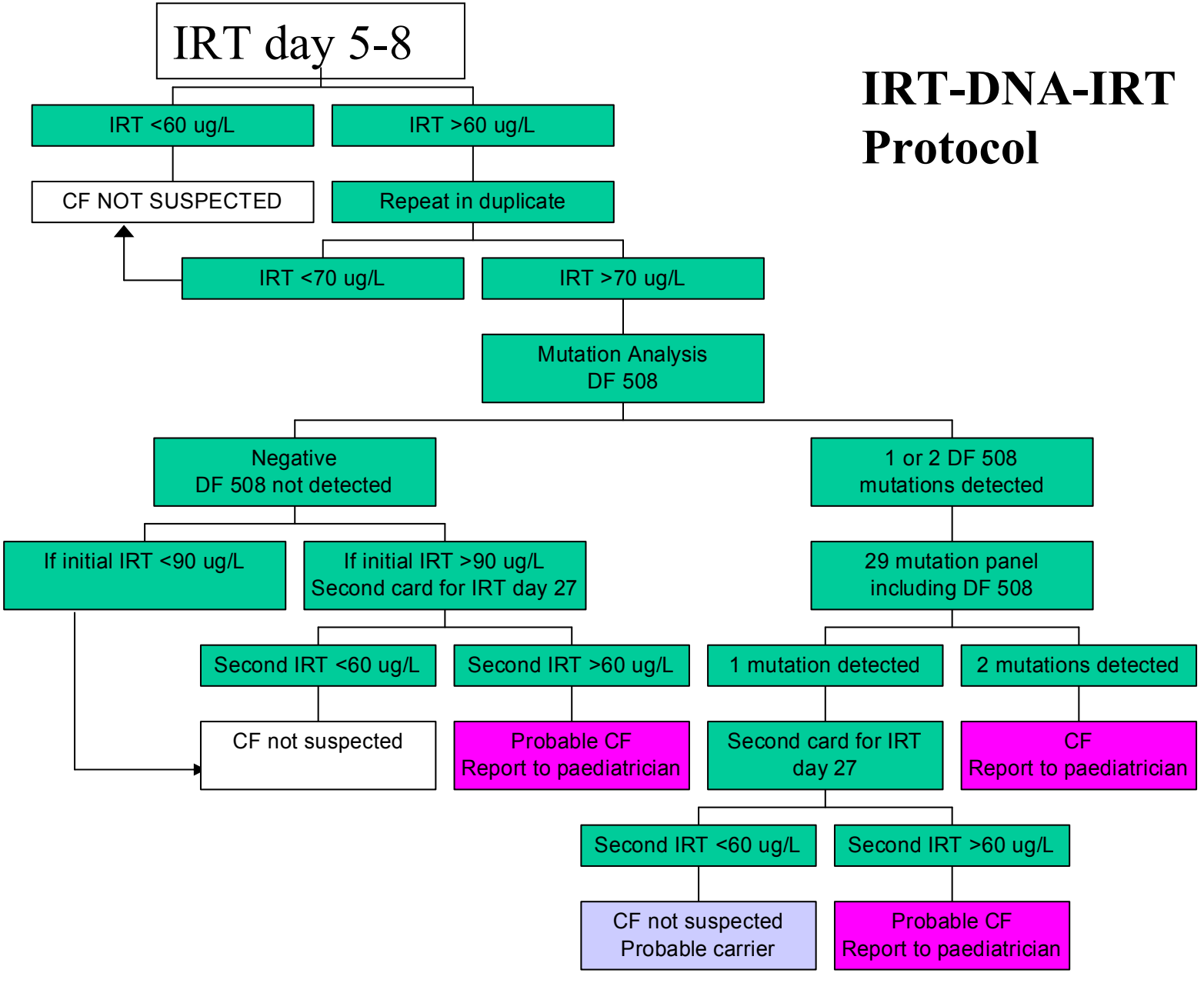
- Therefore a second step is added to the screening process. This can be a second IRT, a DNA test or a combination of both

- In CF babies the IRT remains elevated when retested at approximately 4 wks of age

Two Stage IRT Screening



IRT-DNA-IRT Protocol



Non-CF causes of raised IRT

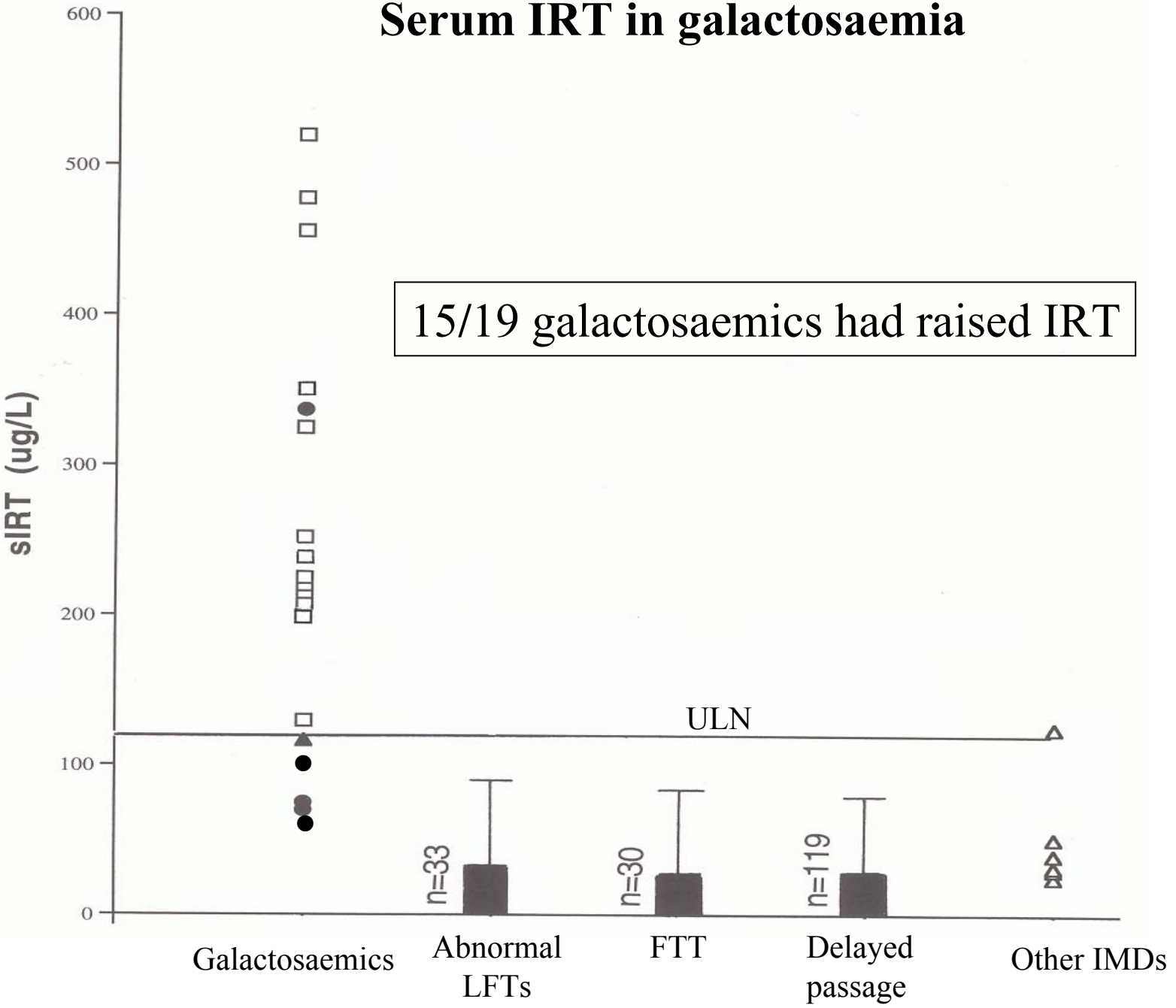
Faecal contamination of the bloodspot card

ΔF508 heterozygotes
Hypoxic insult to pancreas
Renal insufficiency
Congenital heart disease
Nephrogenic DI

Spina bifida
Gastroschisis
Viral infections
Trisomies 13, 18

Mechanisms not always clear!

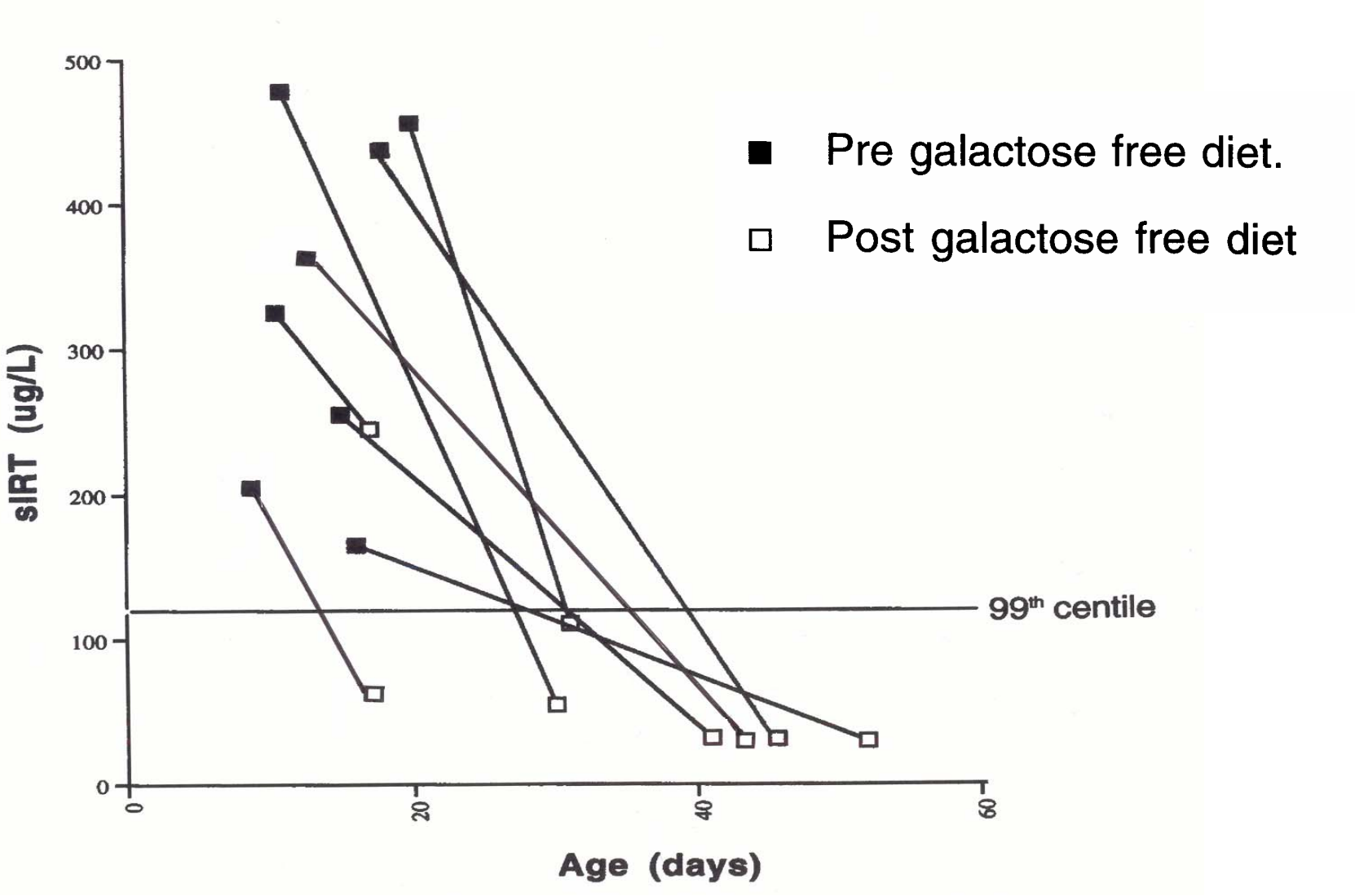
Serum IRT in galactosaemia



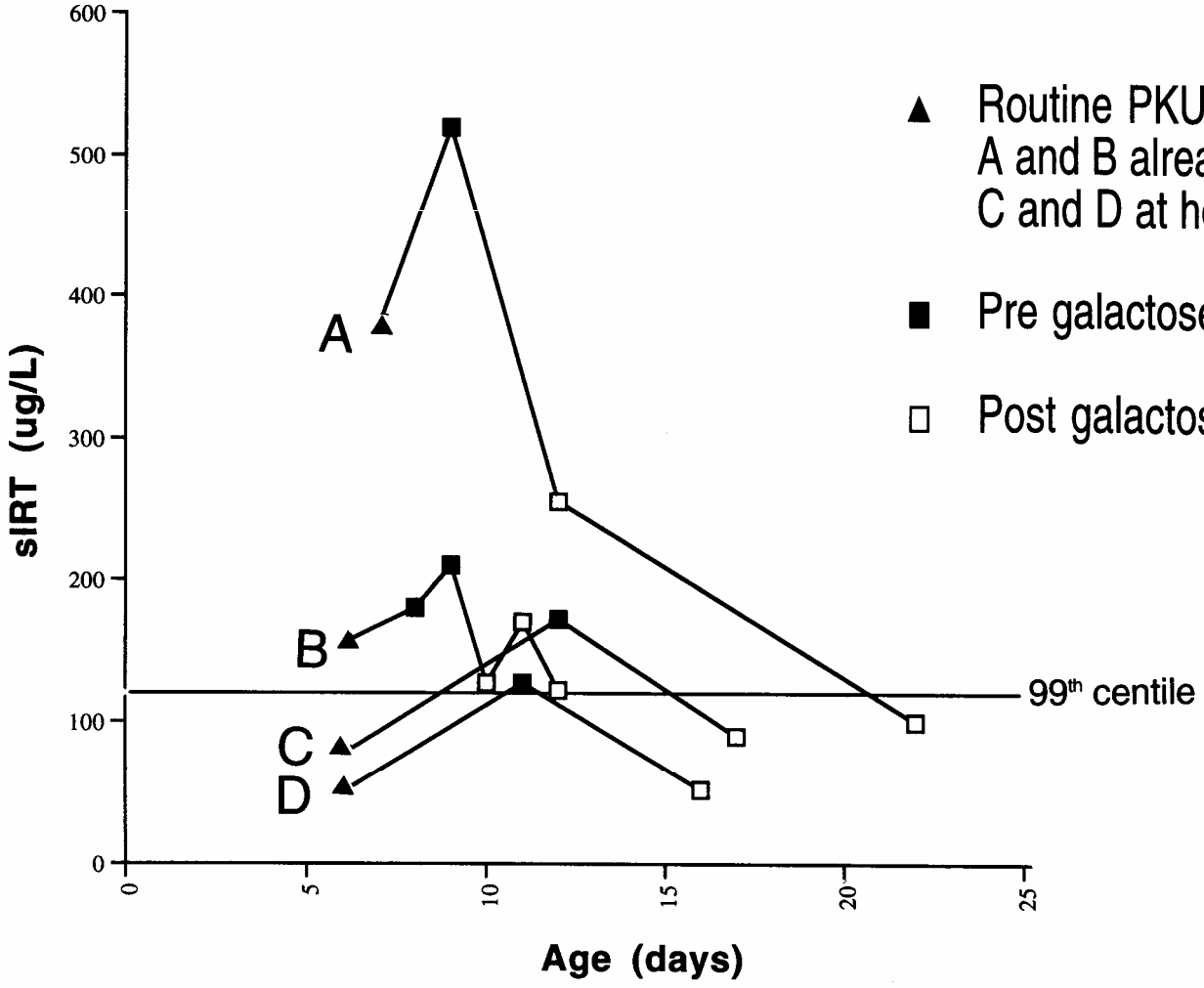
- Raised IRT was not seen in other inherited metabolic disorders with renal Fanconi syndrome, including those with liver damage (Wilson's, tyrosinaemia type 1) – intracellular trapping of phosphate is not a feature of these disorders

- However number of cases very small

Serum IRT pre and post treatment in galactosaemia



sIRT Pre and post treatment



▲ Routine PKU/CHT/CF screen (E. Anglia)
A and B already acutely ill in hospital.
C and D at home.

■ Pre galactose free diet.

□ Post galactose free diet

Biochemical results

Patient A	At diagnosis	Post treatment
Bilirubin (µmol/L)	325	139
ALP (IU/L)	2648	1378
ALT (IU/L)	467	139
Phe (µmol/L)	357	normal
Tyr (µmol/L)	923	523
Met (µmol/L)	90	normal
Amino aciduria	gross	normal
IRT (µg/L)	181	normal

Biochemical results

	At diagnosis	Post treatment
Patient B		
Amino aciduria	Gross	Mild
IRT (µg/L)	175	Normal
Patient D		
Amino aciduria	Gross	Normal
IRT (µg/L)	130	Normal
Patient E		
Amino aciduria	Gross	Normal
IRT (µg/L)	200	Normal

CFTR

Large number of the more common mutations are located in the nucleotide binding folds of the protein

Normal function is dependent on the adequate supply of ATP for phosphorylation of the regulator domain and allosteric binding of ATP to the nucleotide domains. Also Na^+/K^+ ATPase required to maintain the electrochemical gradient for Cl^- to exit the apical membrane

According to “Scriver”

“Modest inhibition of mitochondrial ATP synthesis, even with small changes in tissue ATP, will disrupt transport processes.”

Do conditions which depress the ATP pool affect the action of CFTR and mimic the pathophysiology in CF??

Babies in intensive care

Day 1 Guthrie's undertaken
(paper chromatography of amino acids)

Observed that babies with raised alanine (usually related to hypoxia) had raised IRTs.

Unexplained lactic acidosis

Female infant aged 4m
Failure to thrive, steatorrhoea, renal tubular acidosis

Raised IRT, normal sweat test, negative for common CF mutations

Plasma lactate = 5.5 mmol/ l
CSF lactate = 4.2 mmol/L

Age 9m – viral gastroenteritis triggered seizures and encephalopathy

Raised IRT persisted

CT scan: Mild diffuse atrophy affecting brain stem, cerebellum and cerebrum

Respiratory chain enzyme complexes normal (muscle biopsy)

Fibroblast pyruvate dehydrogenase assay repeatedly just below the reference range (?PDH heterozygosity)

Summary

Serum IRT concentrations in untreated galactosaemia are comparable to those seen in CF

Values may be normal prior to development of acute symptoms and signs

IRT rapidly declines with dietary treatment

The decline mirrors improving amino aciduria rather than acute markers of liver disease

Is energy-deficit the common link between the observed renal absorption defect and the raised sIRT in galactosaemia?