Amino acid disorders (PKU, MSUD, HT,HCU)

Biochemical monitoring of amino acids is integral to:

- manage & adjust dietary treatment
- optimise treatment and outcome
- maintain amino acid(s) in target treatment range
- prevent deficiency - low amino acid concentrations
- prevent toxicity - high amino acids concentrations
- manage intercurrent illnesses

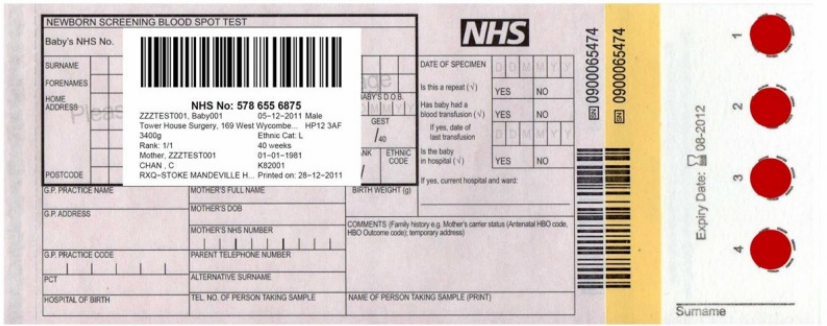
Amino acid disorders (PKU, MSUD, HT,HCU)

- accumulation of precursor amino acid(s) in plasma
- measure the accumulating amino acid(s)
- normal reference ranges
- target treatment reference ranges for amino acid(s)
- target plasma reference range >normal reference range
- *measurement of uncertainty (MU)*

- adjust diet based on result and other factors
- families informed of result by phone, letter, e-mail, text, graph of results at clinic

Home blood sampling and collection

- blood spot on blood cards (PKU, Tyr, MSUD, HCU)
  - heel/finger prick
  - importance of good sample to obtain accurate results !
  - convenient, simple, achievable
  - enables regular, accurate monitoring of diet
- blood in Sarstedt microvette tube



Frequency of blood sampling: a guide

PKU, MSUD, HT(1,11,111)

- weekly in infancy and early childhood
- 2 weekly in toddlers, young children
- monthly in older children

HCU:

- less easy to monitor from home
- hospital phlebotomy for prompt blood separation
- dried blood spot for tHc (LC–MS/MS), methionine

  (Adam Gerrard, Mary Anne Preece, BCH)
- weekly in infants until stable, then 2 weekly

Sampling time of day – standardise ?

- diurnal variation of amino acids
- ideally sample at least 3½ hours after end of last meal, avoid high aa's due to postprandial absorption
  Bachmann C, J Inher Metab Dis 2008
- PKU - phenylalanine is highest after overnight fast
  Macdonald A et al, Arch Dis Child 1997
- aim is to at least collect at same time of day
  eg: PKU bath time on a Sunday – warm, good blood flow
- try to document time delay between last meal & sampling

Tandem mass spectrometry (MS/MS) - blood spots for

- for PKU, Tyrosinaemia
- HCU (and paired plasma sample 1/mth)

Ultra High performance/pressure liquid chromatography

- BCAA blood spot

Results

- daily - phe, tyr, BCAA
- weekly - tHc, methionine (BCH)

Interpretation of amino acid results

- look at trends
- adjust diet
- repeat blood test to follow up dietary change
- timing of sample in relation to food
- consider clinical status
  - is child well ?
  - is child on ER ?
- growth
- age (puberty)
- compliance with diet
- no of days since last increased exchanges/protein

Reasons for high concentrations of amino acid(s) on treatment

- acute catabolism: infection, stress, surgery
- chronic catabolism due to inadequate intake of:
  - precursor free aa's
  - energy
- too much natural protein
- wrong protein substitute product or low protein foods
- non-adherence
- medicine (Betaine in HCU)

Reasons for low concentrations of amino acids (s) on treatment

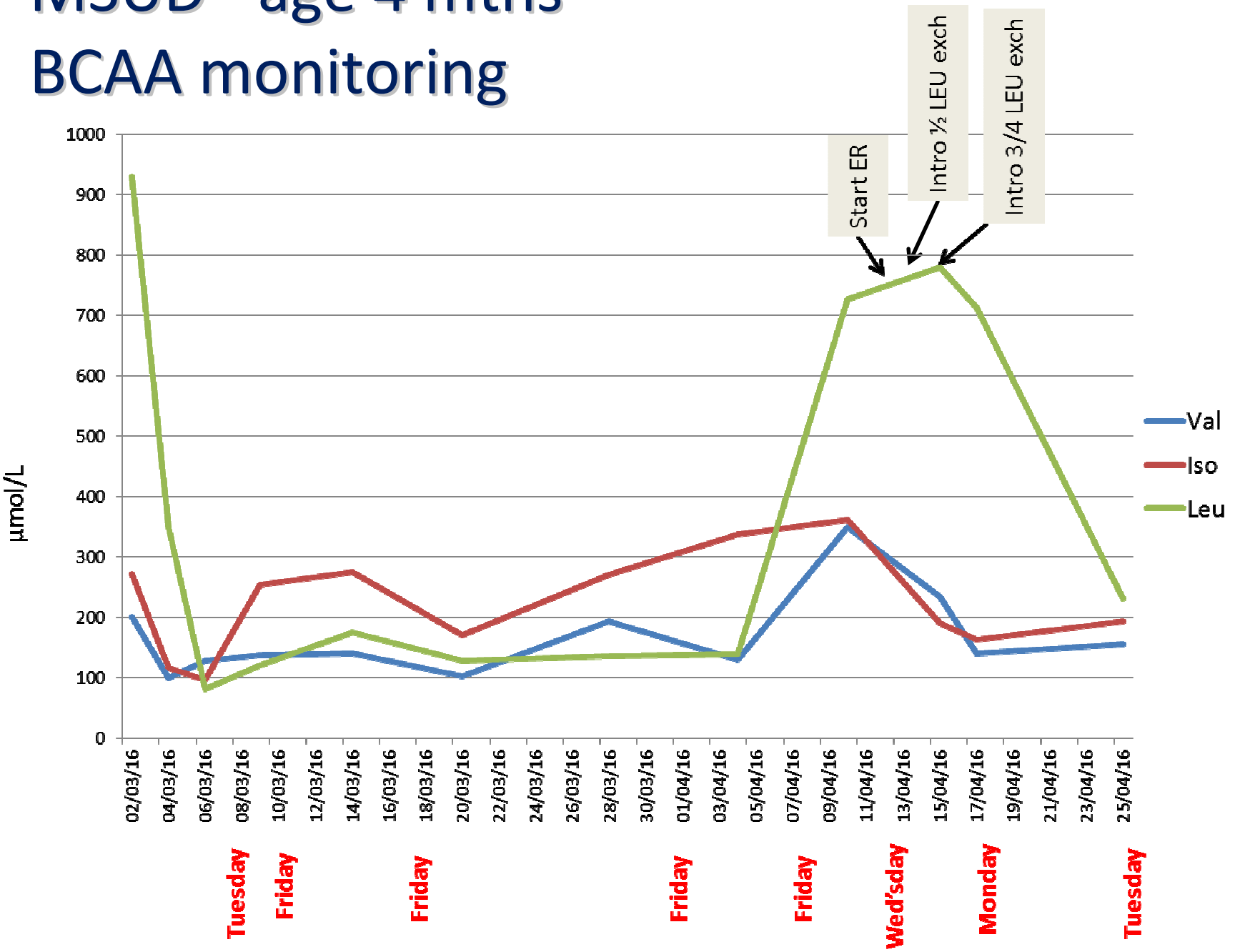
- inadequate natural protein intake
- inadequate protein substitute or single amino acids
- increased requirement post illness
- growth spurt
- inadequate synthesis and supplementation
  - cysteine - HCU
  - tyrosine - PKU

PKU Age(years)	Treatment aim Phe range µmol/l MRC Arch Dis Child 1993	Treatment aim Phe range µmol/l European PKU Guidelines 2016 (unpublished)
0-4	120-360 (35-100 normal)	120-360
5-10	120-480	120-360
>11	120-700	120-600

MSUD	Plasma reference ranges µmol/l	
	normal	target treatment range
Leucine	65-220	200-400 75-200 < 5y 75-300 > 5y
Isoleucine	26-100	200-400
Valine	90-300	200-400

Frazier DM et al, 2014

MSUD - age 4 mths
BCAA monitoring



Tyrosinaemia type 1	Plasma reference ranges µmol/l normal	blood spot treatment aims
Tyrosine	30 -120	200 - 400
Phenylalanine	35 -100	35 -100

HT1 – teenager age 13 years

Diet:13g natural protein, Tyr Cooler x 3 (45g aa's)

Date		Tyrosine aim 200-400µmol/l	Phenylalanine 35-100µmol/l
14.11.09	am	557	34
	pm	398	75
05.12.09	am	747	64
	pm	589	53
05.01.10	am	986	100
	pm	925	70
16.01.10	am	458	51
	pm	361	36
23.01.10	am	388	33

HCU	Treatment aims			
	plasma		homocysteine	
	methionine µmol/l	cysteine µmol/l	free µmol/l	total µmol/l
diet alone	normal range	normal range	< 10	< 80 -100
betaine	high up to 1000	normal range	< 10	< 80 -100

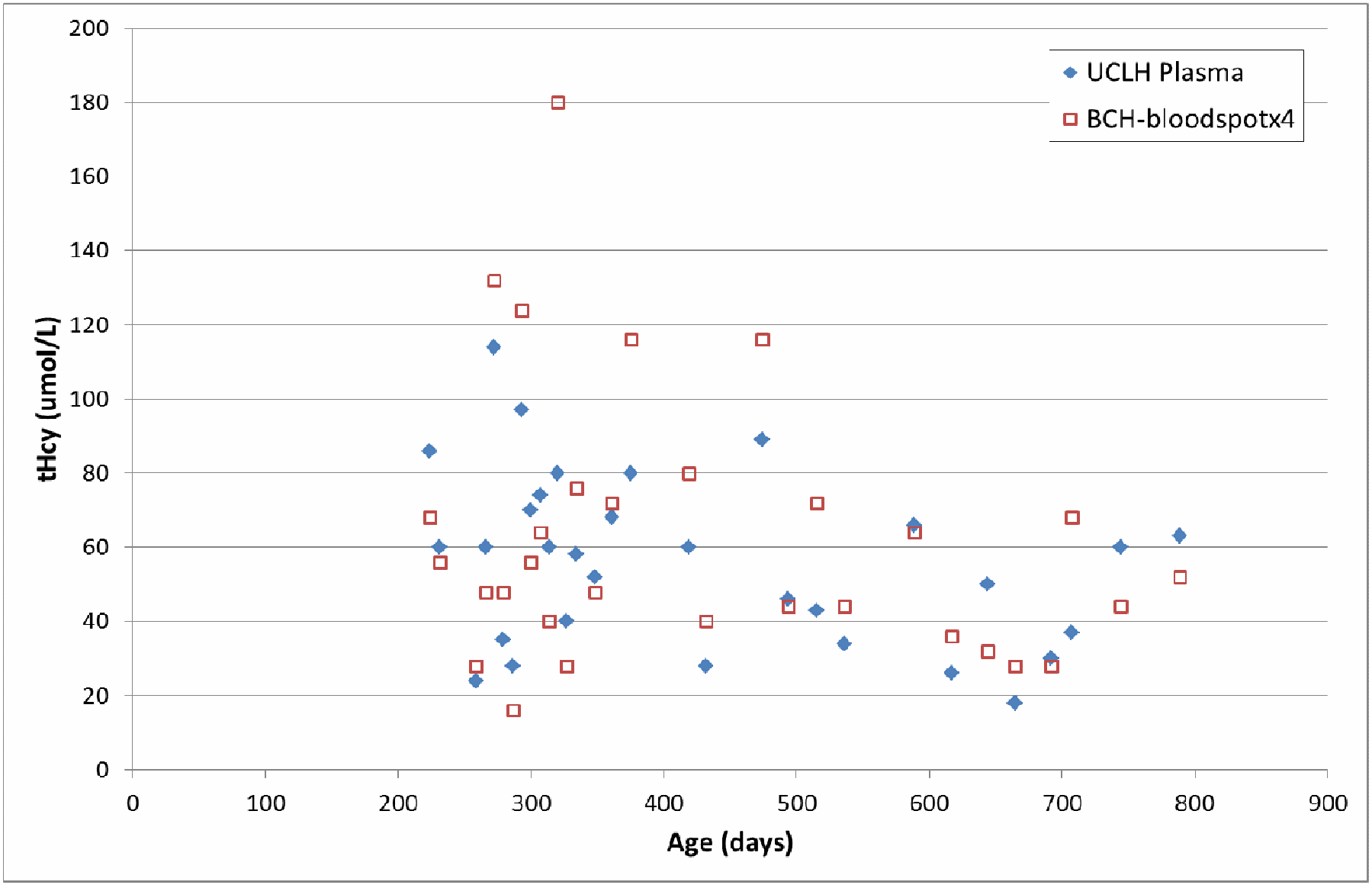
- Lifetime - free Hcy <10 µmol/l associated with good outcome
- tHcy > 60µmol/l before observe free Hcy
- Dried blood spot - apply a factor of x 4

Summary

- biochemical monitoring is integral to dietetic management
- monitoring both single and trends of results is important
- repeated/regular monitoring to review interventions
- cannot interpret in isolation need to consider other factors - clinical picture, growth, dietary intake, compliance

Measurement of uncertainty

- should MU be considered when interpreting results for monitoring?
- would it be helpful to have MU reported with results ?
- is this more of a problem for higher results ?
  - apply same MU then greater range
  - Leuc 400µmol/L (apply MU of 15) = 340 to 460
  - Leuc 800µmol/L (apply MU of 15) = 680 to 920


UCLH Plasma
BCH-bloodspotx4
tHcy (umol/L)
Age (days)
0
20
40
60
80
100
120
140
160
180
200
0
100
200
300
400
500
600
700
800
900