Abnormalities of Intermediary Metabolism in Barth Syndrome

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Is Barth Syndrome a Mitochondrial Disease?

1. Muscle biopsies often have normal mitochondrial enzymology and histology

2. Profound muscle fatigue and weakness occur without biochemical signs of mitochondrial dysfunction

3. Severity of growth delay is out of proportion to biochemical signs of mitochondrial dysfunction

4. ATP synthesis is normal in cultured cells and in living tissue

Evidence For a Leucine-Independent Origin of 3-Methylglutaconic Acid

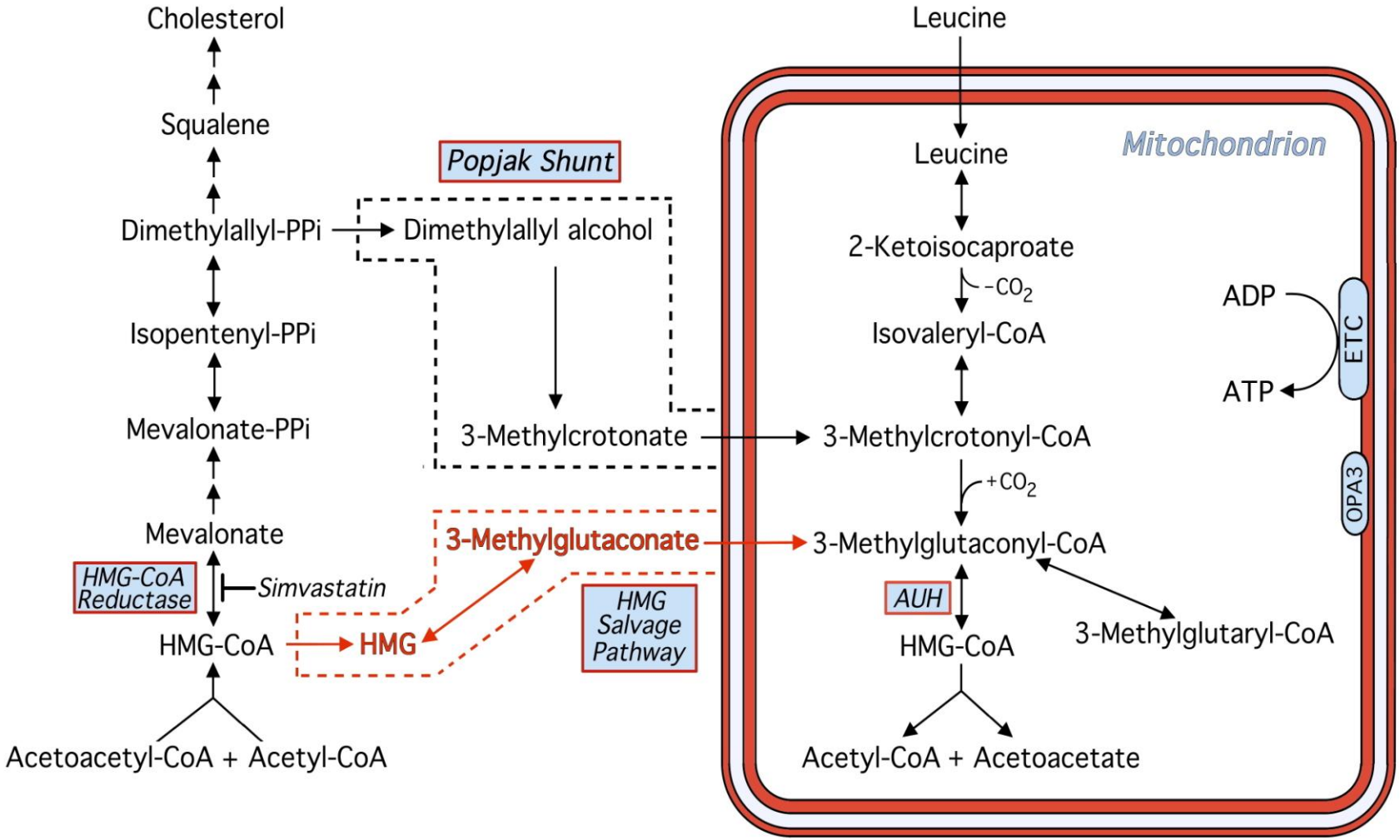
Normal increase after leucine-loading

No increase after prolonged fasting

Normal levels in children with inborn errors of leucine catabolism

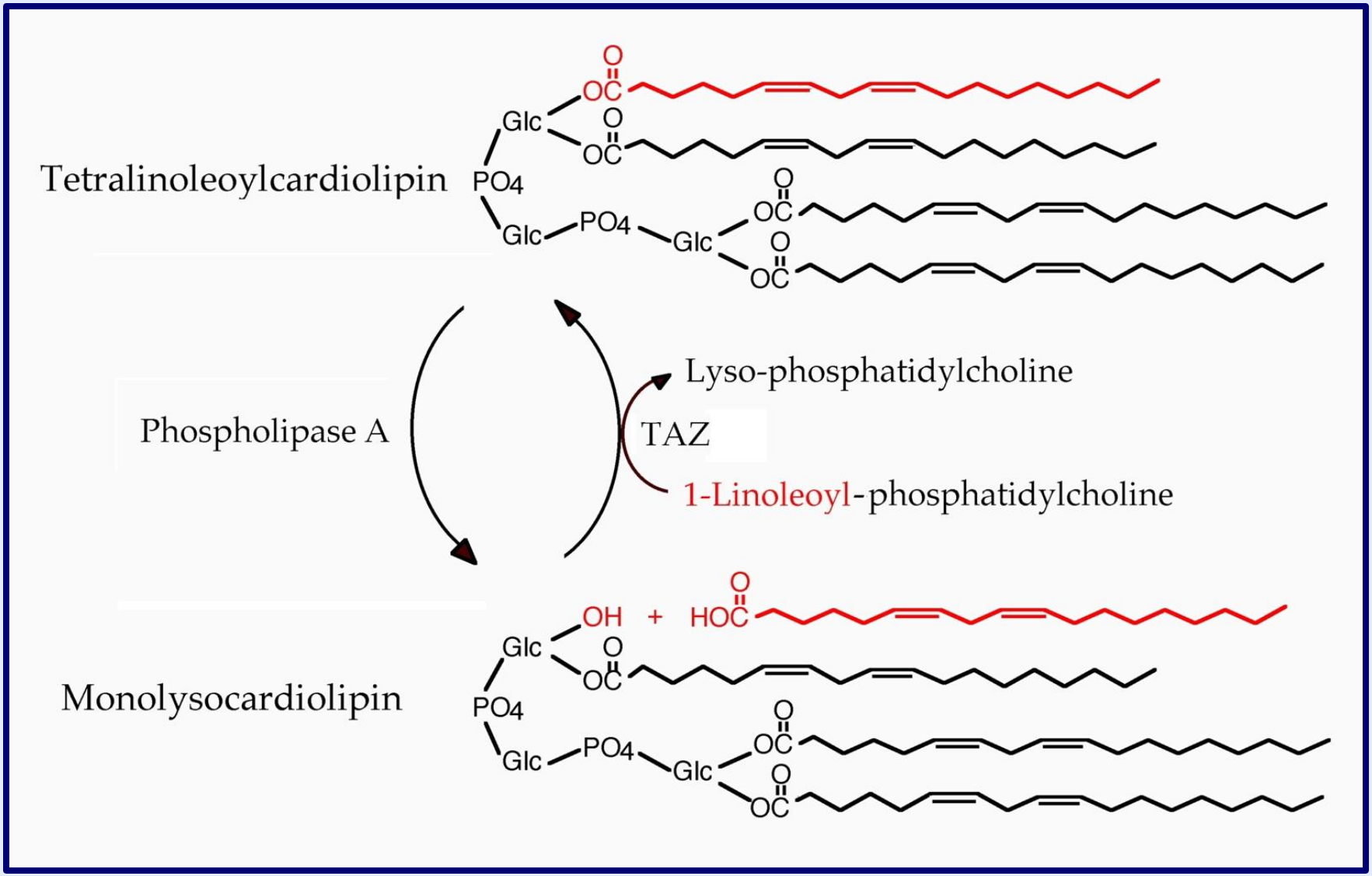
3-MGC labels with ^{13}C-acetate

3-Methylglutaconate Metabolism

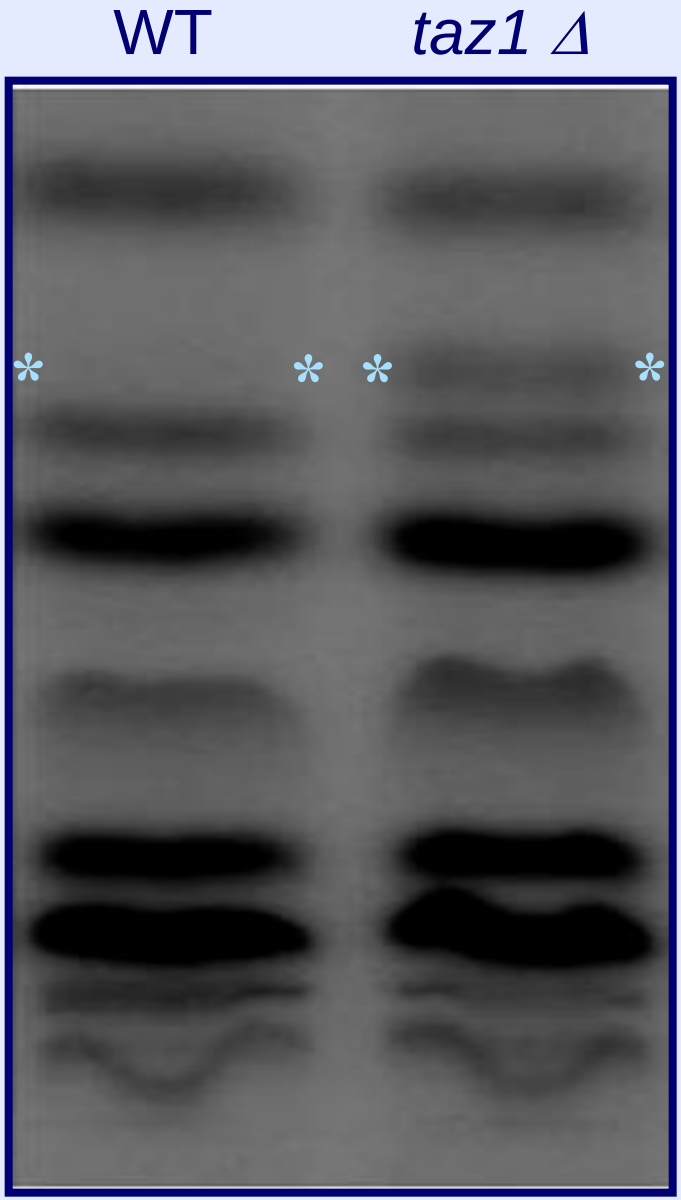


Pei W, et al., Development 137, 2587-2596 (2010)

Cardiolipin Remodeling Cycle



Phospholipids in *taz1* Mutant Yeast



Cardiolipin

Monolyso-cardiolipin

Phosphatidic acid

Phosphatidylethanolamine

Phosphatidylserine

Phosphatidylinositol

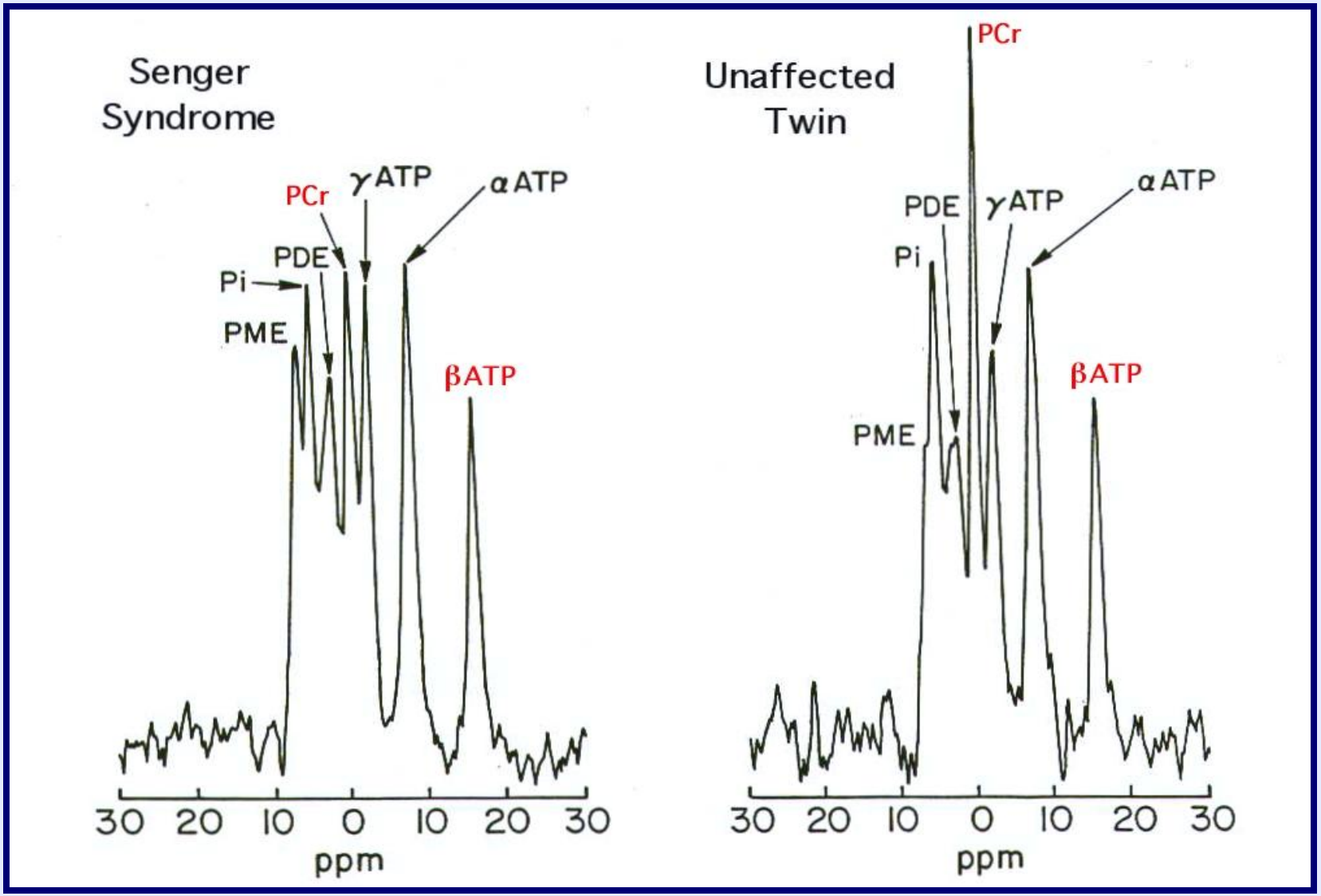
Phosphatidylcholine

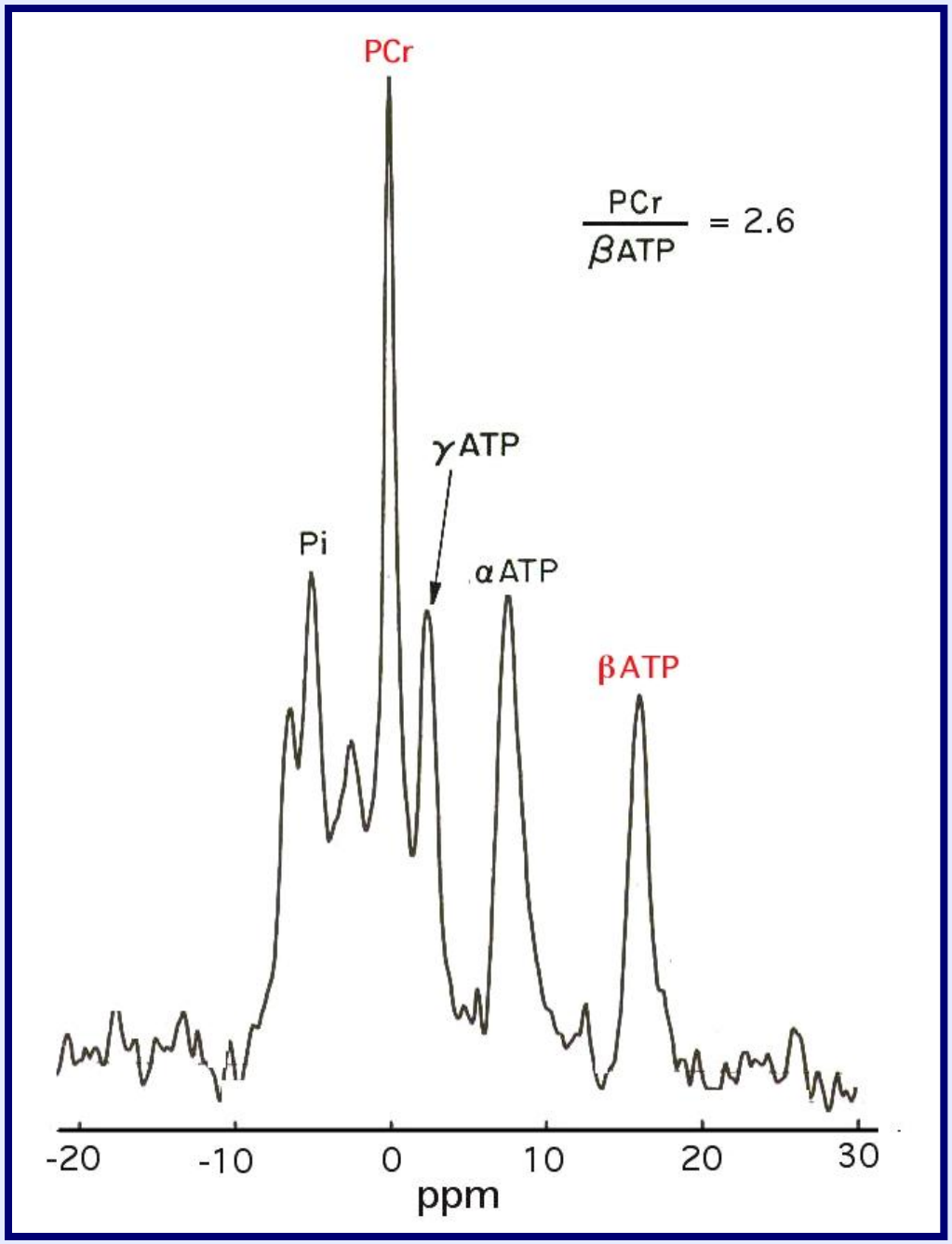
M.Greenberg, 2002
Personal communication

Barth Syndrome – Cardiac Pathology

Prenatal:	+/- Dilated cardiomyopathy Ventricular non-compaction in some
Postnatal:	Stable or progressive cardiomyopathy, variable severity and course Mild left ventricular thickening
Childhood:	Increasing risk of ventricular arrhythmia even when cardiac function is normal
Biopsy:	Endocardial fibroelastosis in some Mitochondria are normal or variably abnormal histologically and enzymatically

^{31}P-NMR Spectroscopy of Heart Muscle

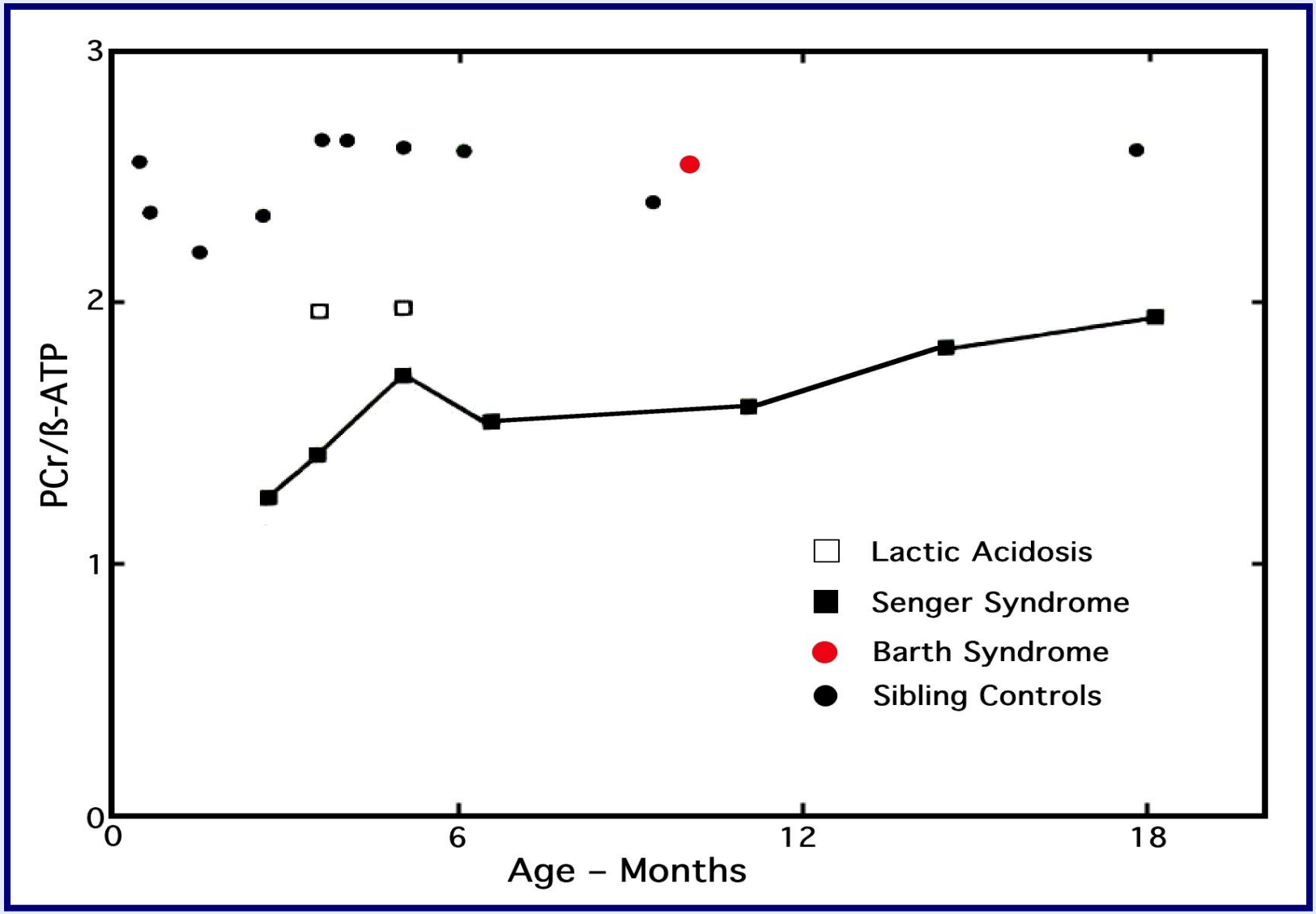




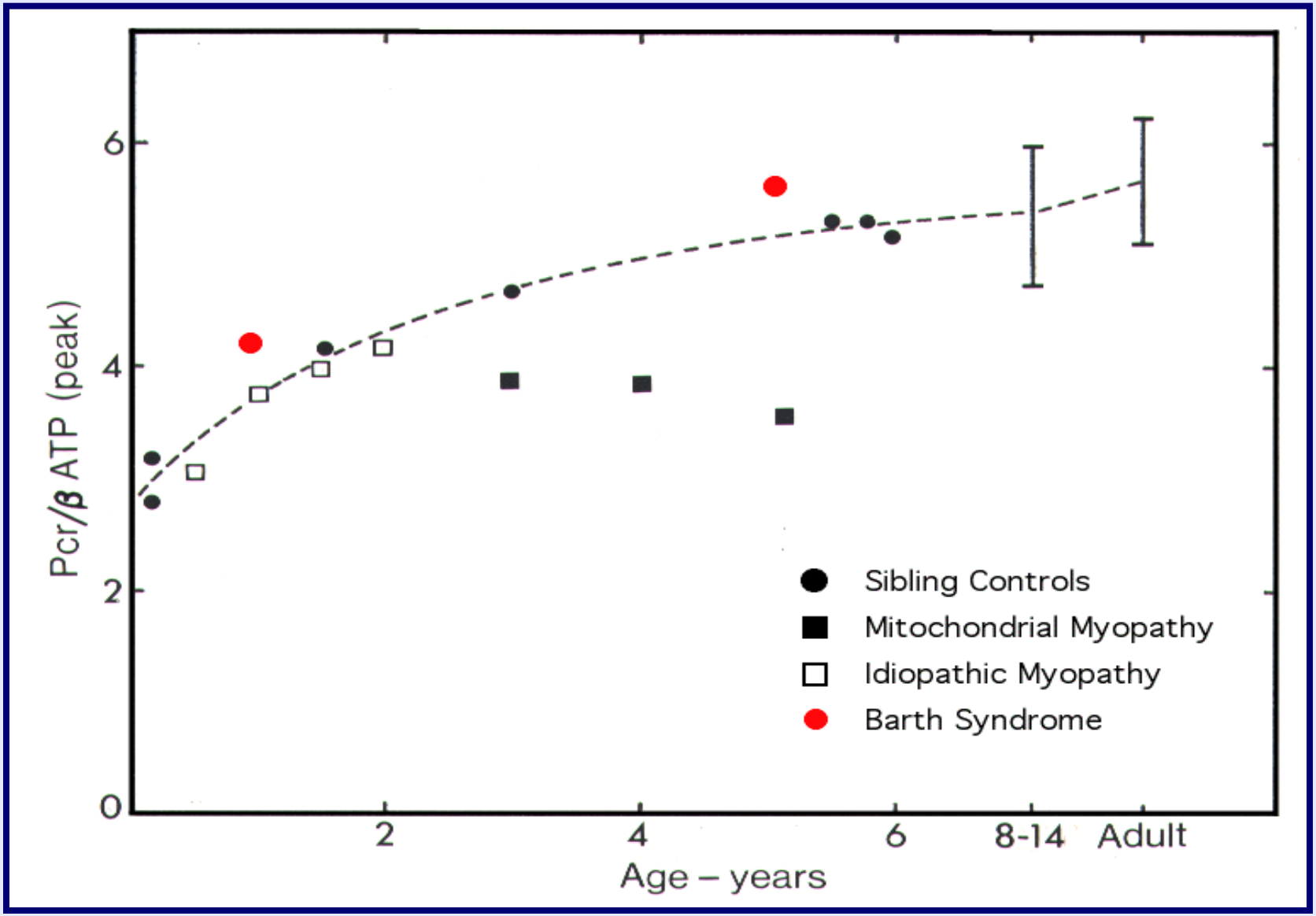
Barth Syndrome: ^{31}P-NMR Spectroscopy of Cardiac Muscle

Age: 8 months
Dx: dilated cardiomyopathy

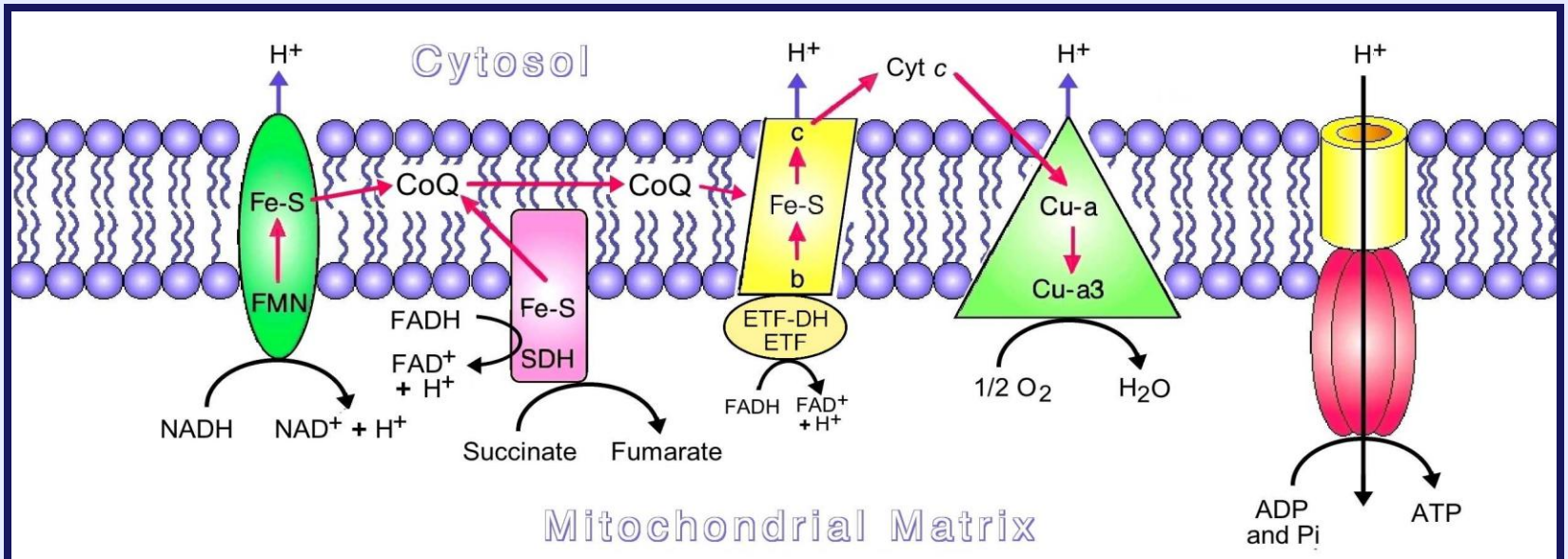
^{31}P-NMR Spectroscopy of Cardiac Muscle



^{31}P-NMR Spectroscopy of Gastrocnemius



Mitochondrial Electron Transport Chain



Complex	I	II	III	IV	V
Mitochondrial Genes	7	0	1	3	2
	ND1–6, ND4L		*Cytb*	*COXI – III*	*ATPase6, 8*
Nuclear Genes	36	4	10	10	11
	NDUFS1, 2, 4, 7, 8 NDUFV1	*SDHA, B, C, D*	*BCS1L*	*SURF1, SCO1 SCO2, COX10*	
Clinical Syndrome	Leigh syndrome Leukodystrophy Cardioencephalo-myopathy	Leigh syndrome Paraganglioma Pheochromocytoma	Hepatopathy Encephalopathy	Leigh syndrome Cardioencephalo-pathy Hepatopathy	Cardioencephalo-pathy Hepatopathy

Plasma Amino Acids – Normal 4 to 6-Hour Fasting

Amino Acid	μmol/L	Min/Max	Mean
Aspartic Acid	11	1 - 17	9
Threonine	109	24 - 160	92
Serine	107	67 - 171	119
Asparagine	39	28 - 96	62
Glutamic Acid	38	1 - 85	43
Glutamine	510	337 - 673	505
Proline	128	51 - 271	161
Glycine	228	87 - 323	205
Alanine	281	136 - 440	288
Citrulline	30	10 - 34	22
Valine	195	78 - 326	202
Cystine	54	44 - 96	70
Methionine	24	7 - 43	27
Isoleucine	55	21 - 89	55
Leucine	95	40 - 172	106
Tyrosine	42	20 - 108	64
Phenylalanine	41	25 - 81	53
Ornithine	40	22 - 94	58
Lysine	110	69 - 205	137
Histidine	79	37 - 125	81
Arginine	48	15 - 115	65

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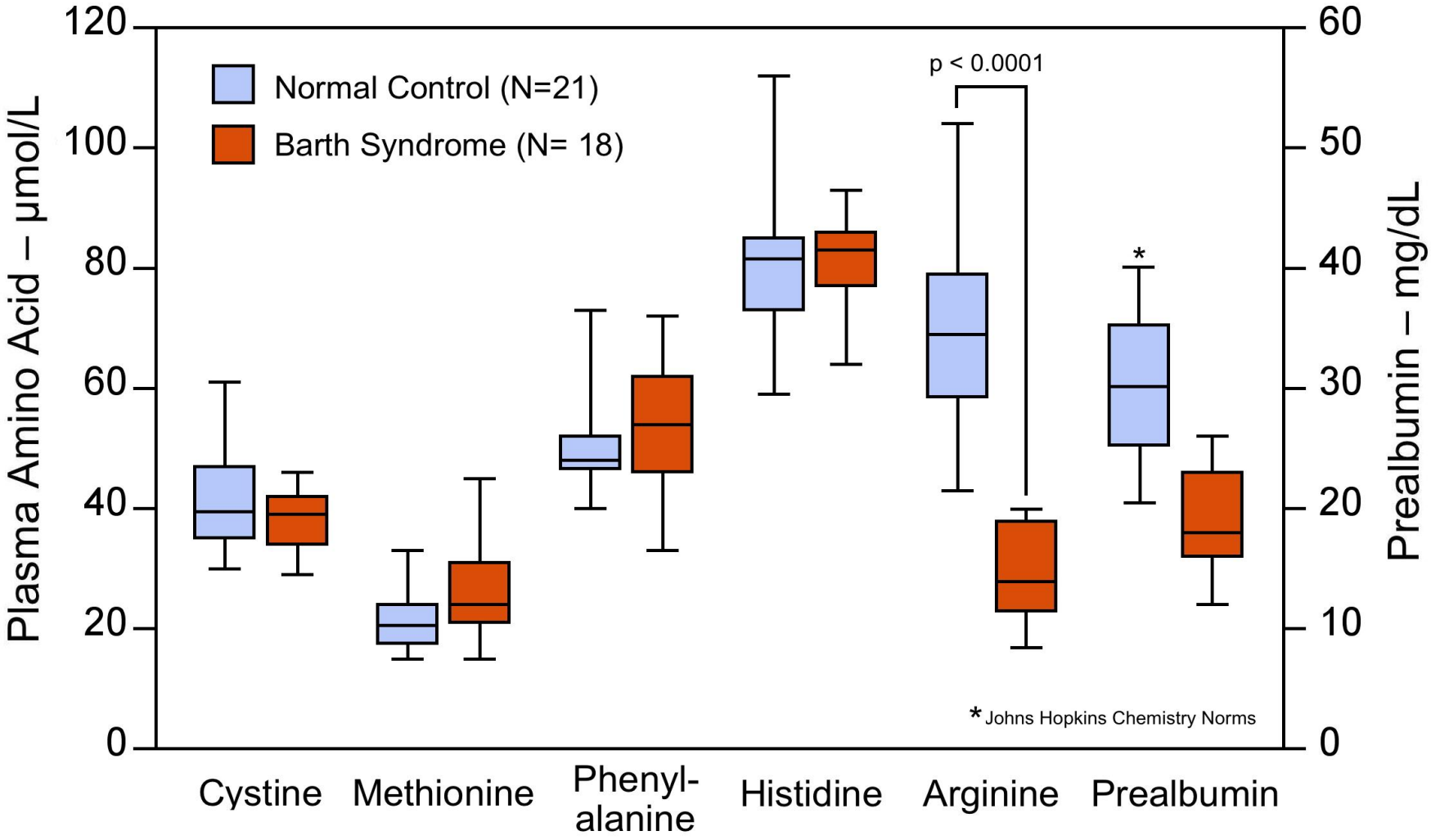
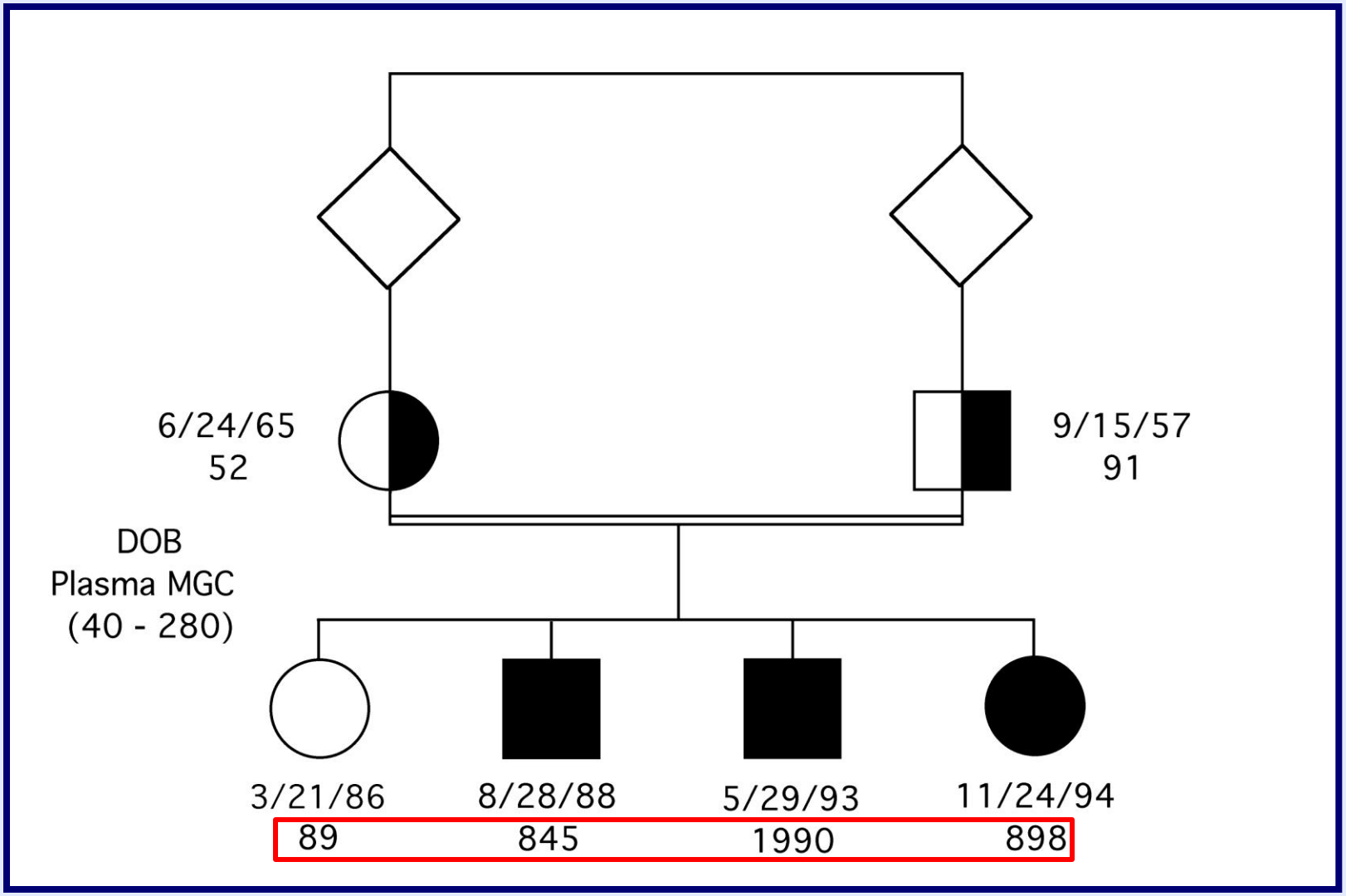
Barth Syndrome – Plasma Amino Acids



Autosomal Recessive Barth Syndrome – Cardiomyopathy-Parkinsonism Syndrome

- Postnatal growth retardation
- Mild chronic neutropenia
- Dilated cardiomyopathy – mild to severe
- Sudden death from presumed arrhythmia
- Ataxia - Parkinsonian tremor
- Moderate developmental delay, +/- seizures
- Apparent autosomal recessive inheritance (Xq28 excluded)

Autosomal Recessive Barth Syndrome



Autosomal Recessive Barth Syndrome: Laboratory abnormalities

Chronic neutropenia - mild to moderate

Hypocholesterolemia

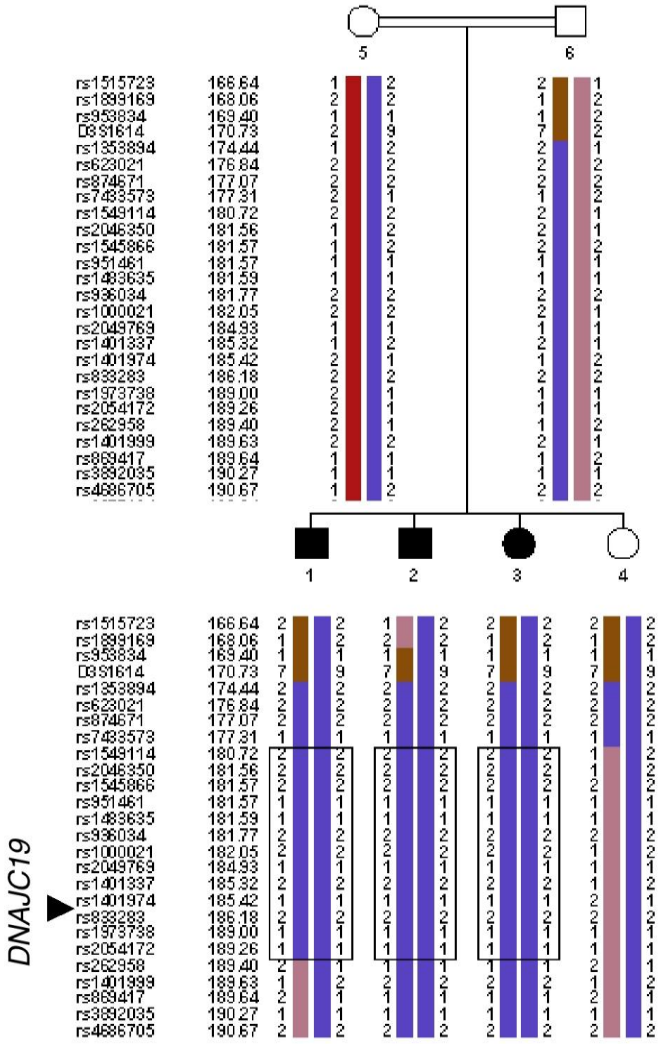
3-Methylglutaconic aciduria

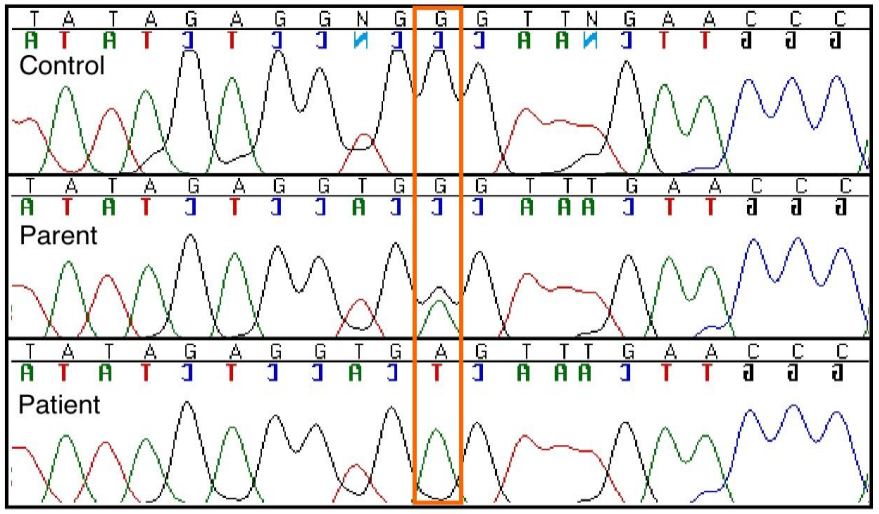
Citric acid cycle organic aciduria

Low plasma arginine levels

Multifocal white matter lesions on MRI

Autosomal Recessive Barth Syndrome: *DNAJC19*



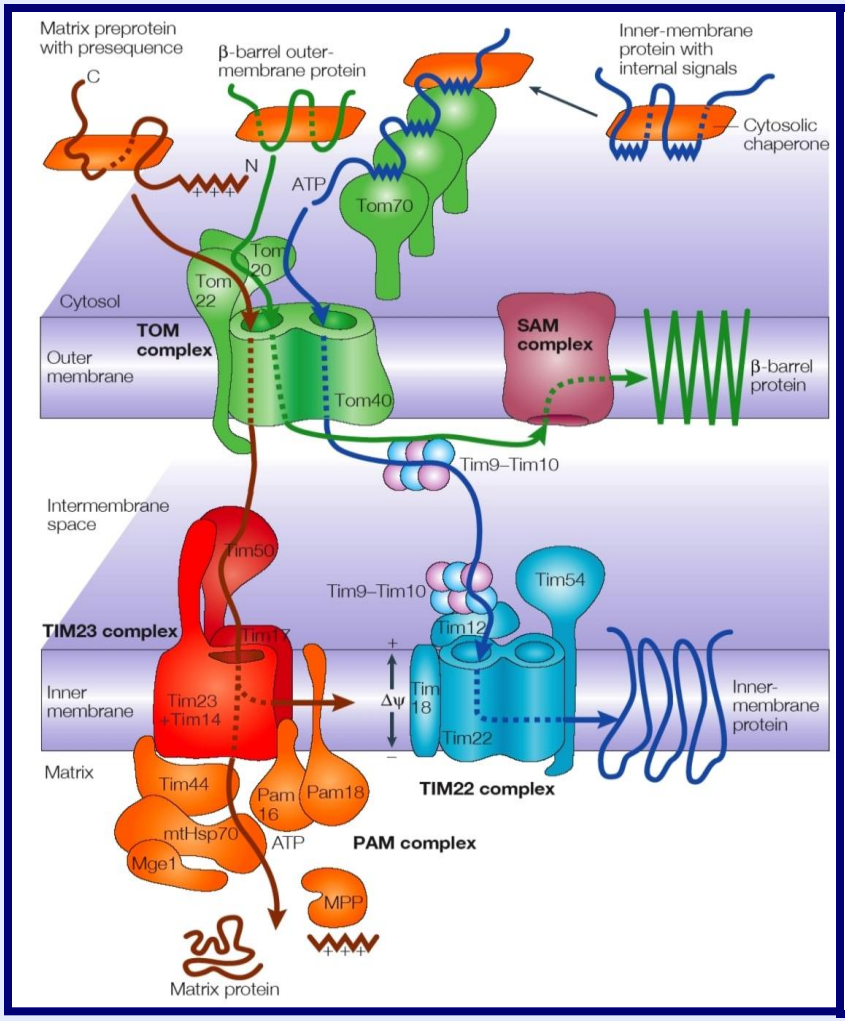


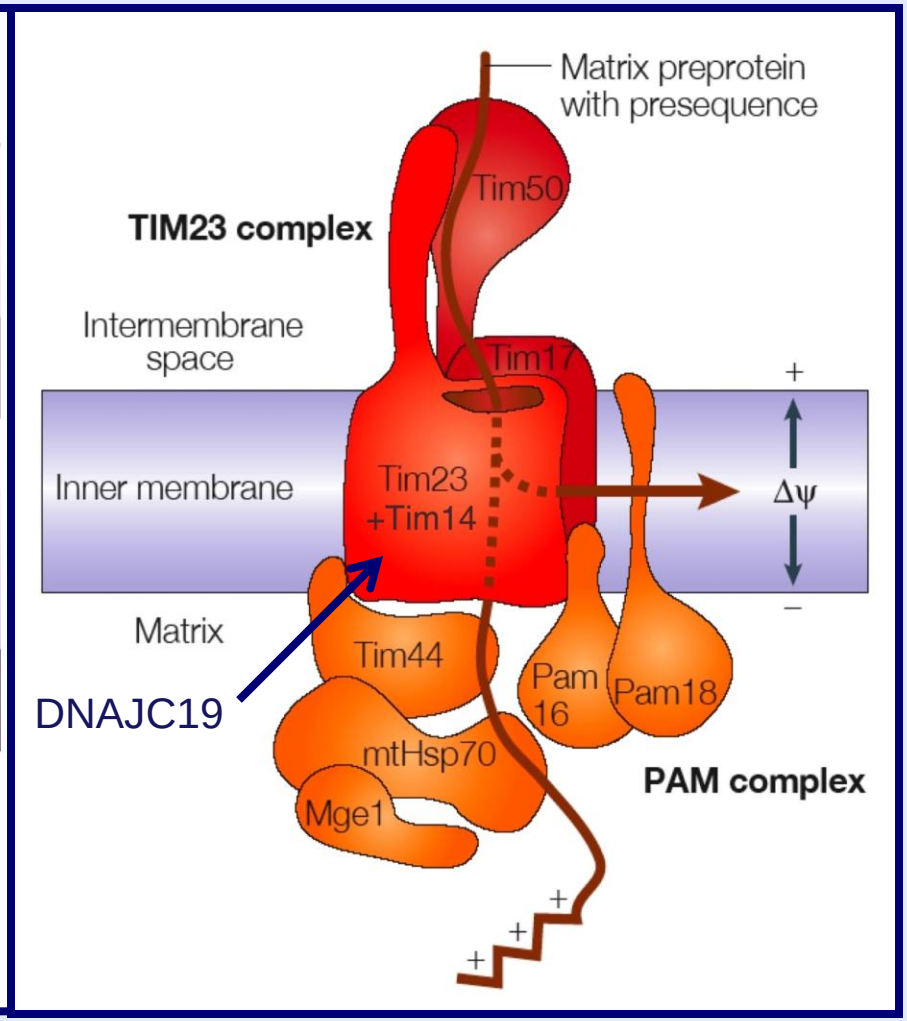
c.158G>A G53E

DNAJC19

Homo	49	YYRGGFEPKMTKREAALILGVS-PTANKGKIRDAHRRIML
Pan	49	YYRGGFEPKMTKREAALILGVS-PTANKGKIRDAHRRIML
Canis	57	YYRGGFEPKMTKREAALILGVS-PTANKGKIRDAHRRIML
Mus	49	YYRGGFEPKMTKREAALILGVS-PTANKGKIRDAHRRIML
Gallus	48	YYRGGFEPKMTKREAALILGVS-PTANRSKIREAHRRIML
Drosophila	52	YYKGGFDPKMNKREASLILGVS-PSASKIKIKDAHKKIML
Anopheles	31	YYRGGFDAKMNKREASLILGVS-PSASKAKVKDAHKKIML
Schiz	41	---GGFESKMSRAEAIQILSLNNRTLTRQKIKEAHRRLML

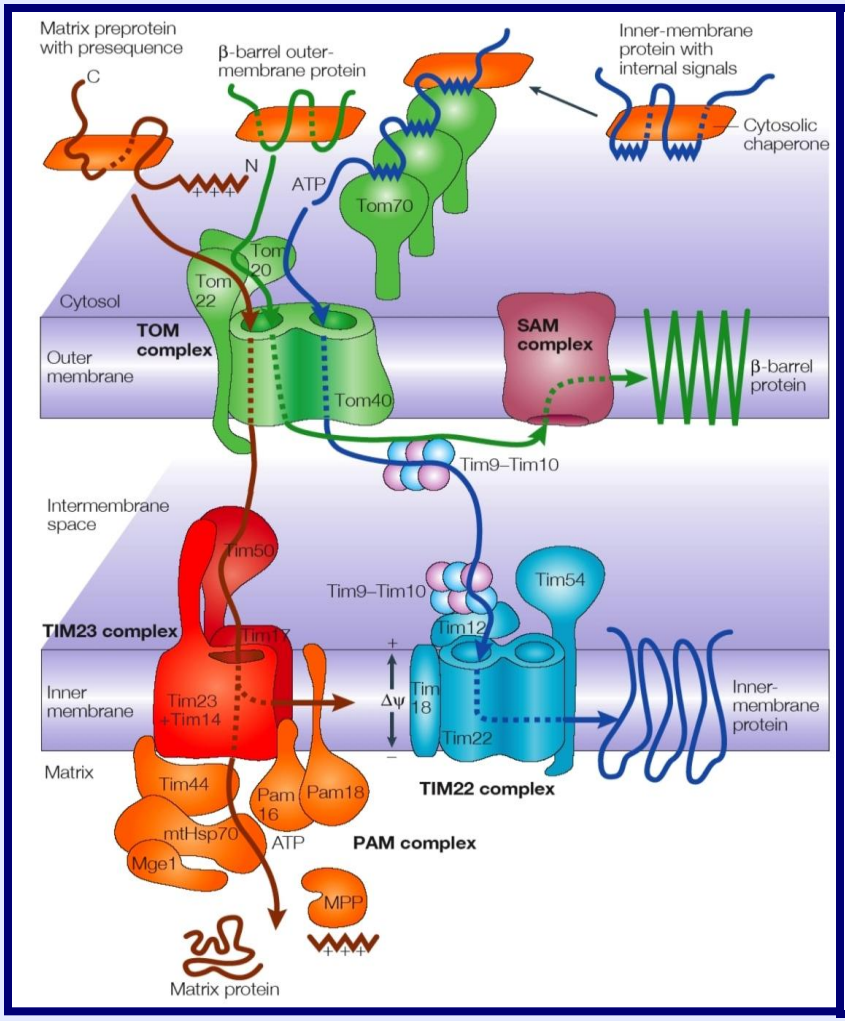
Mitochondrial Protein Import Motors

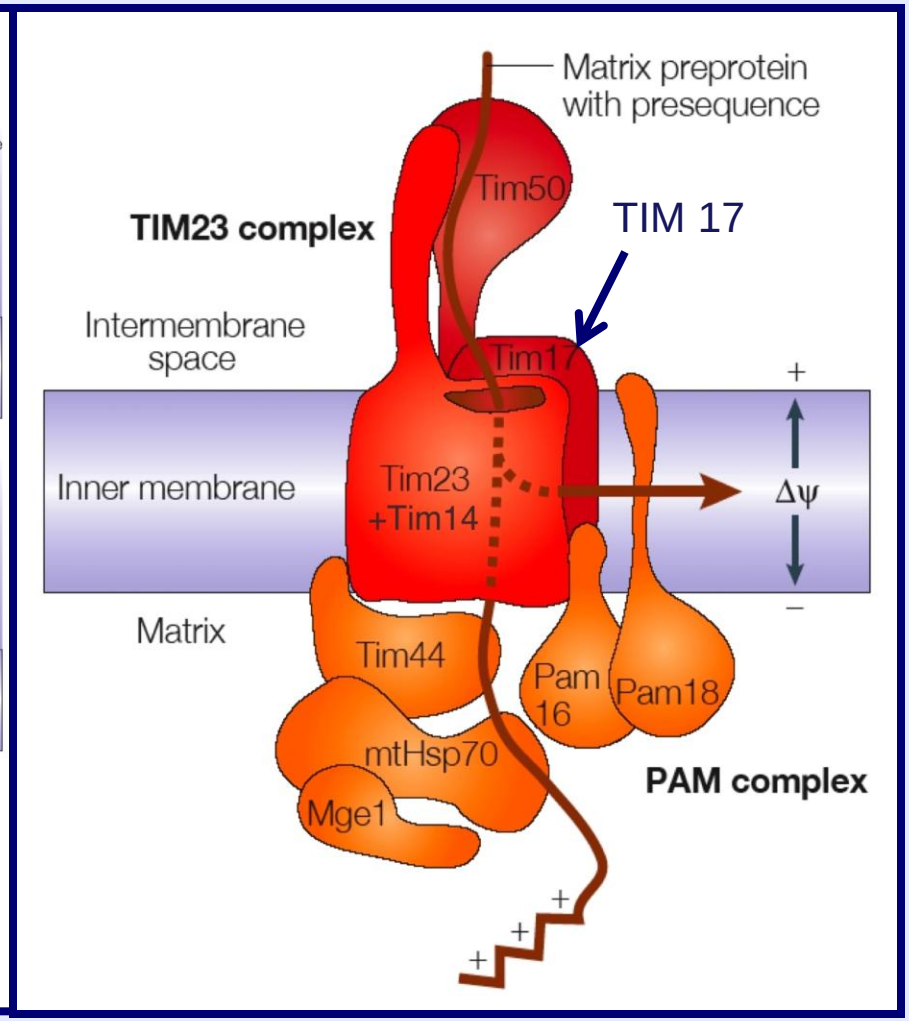




P. Rehling, Nat Reviews, 2004, 5:519-530.

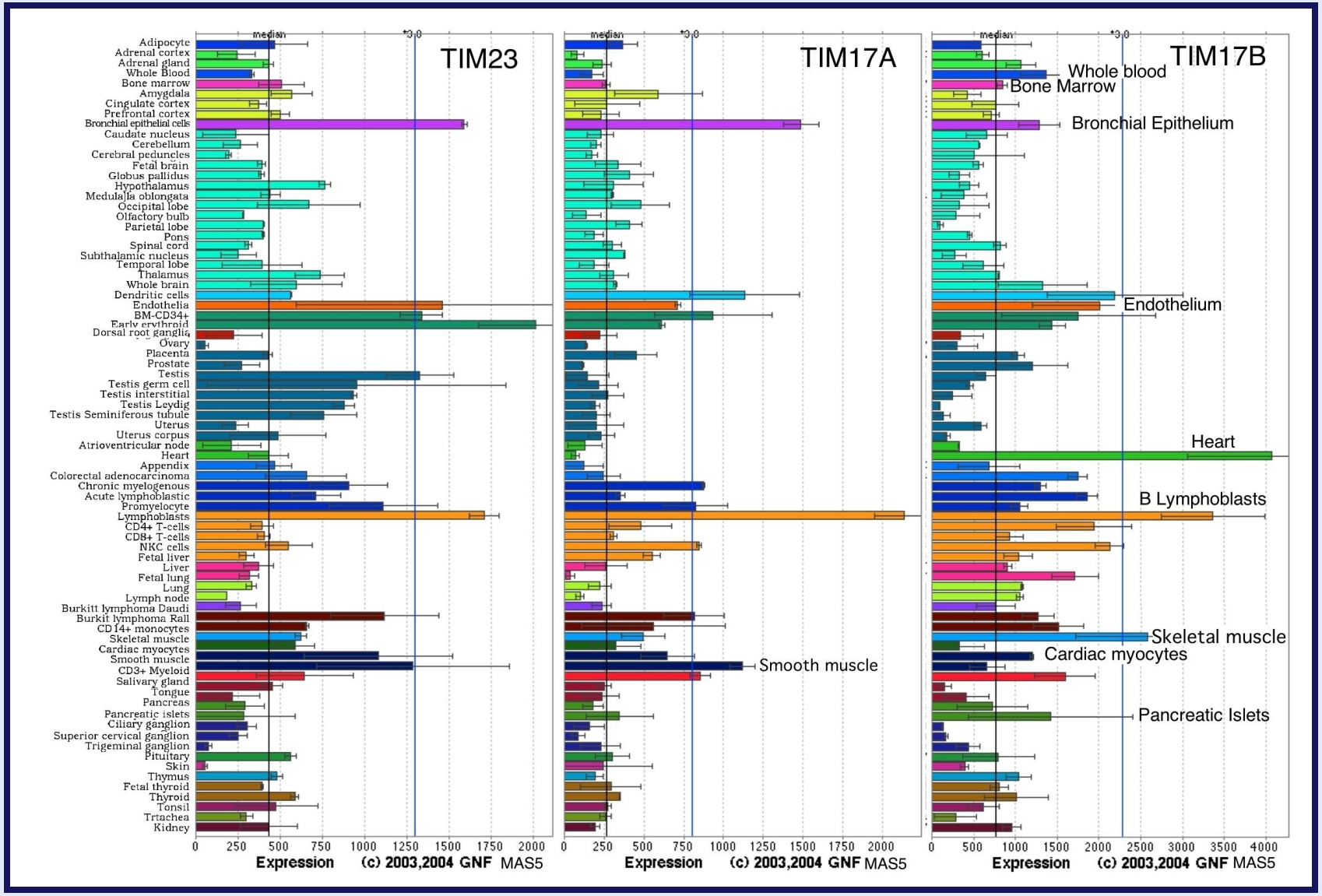
Mitochondrial Protein Import Motors



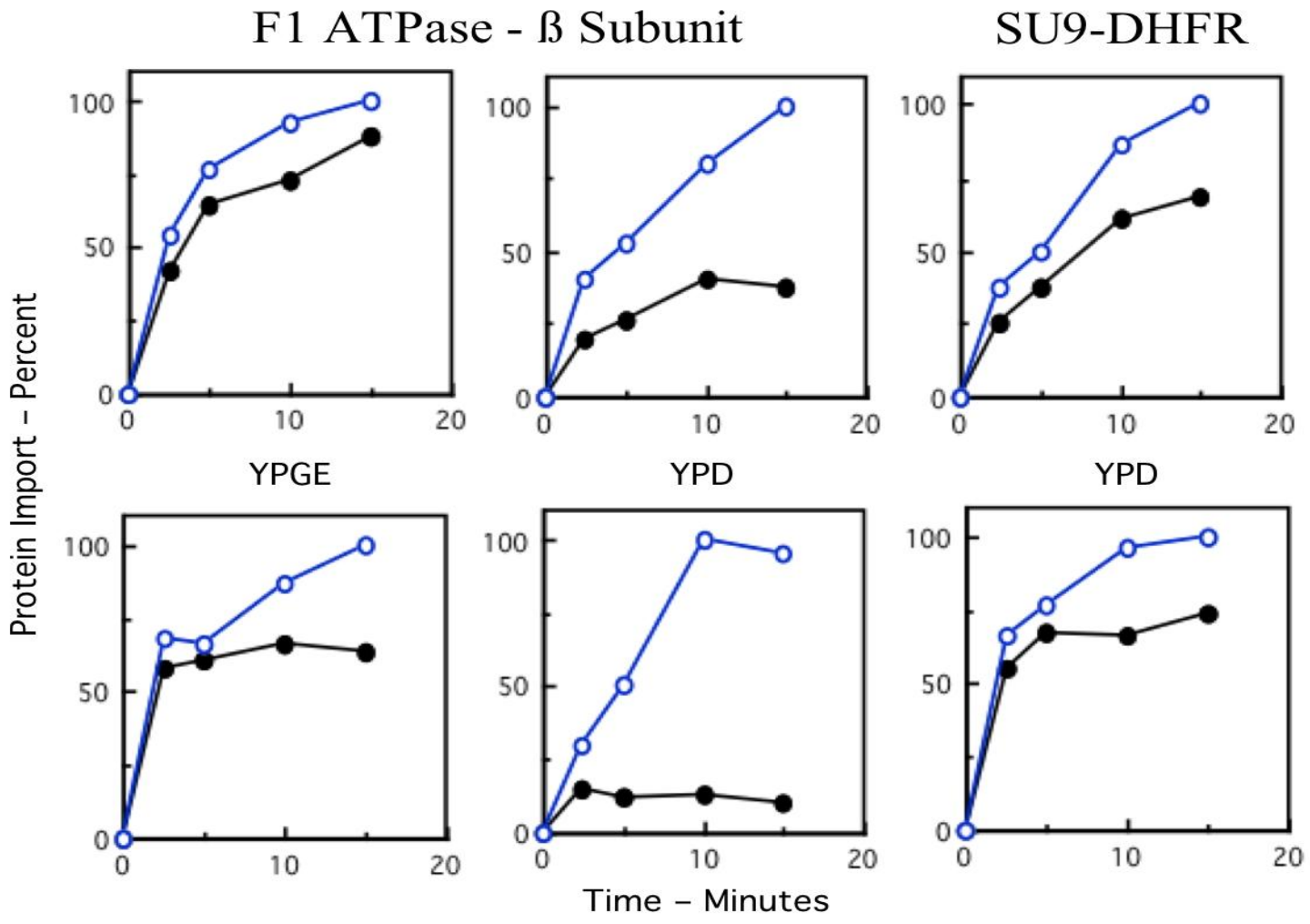


P. Rehling, Nat Reviews, 2004, 5:519-530.

TIM 17 mRNA Expression Profile

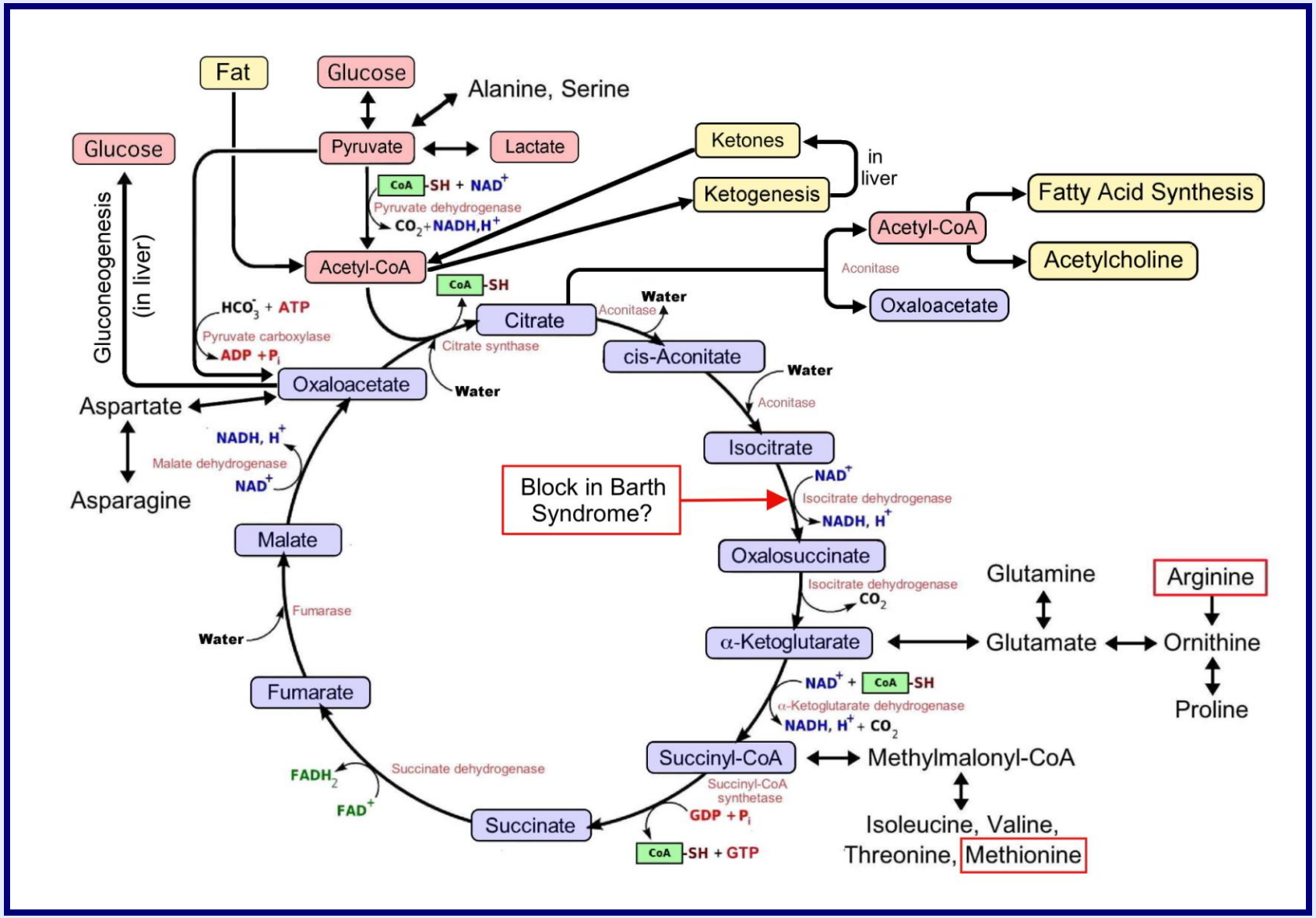


Mitochondrial Protein Import in Yeast Crd1Δ Cardiolipin Mutants

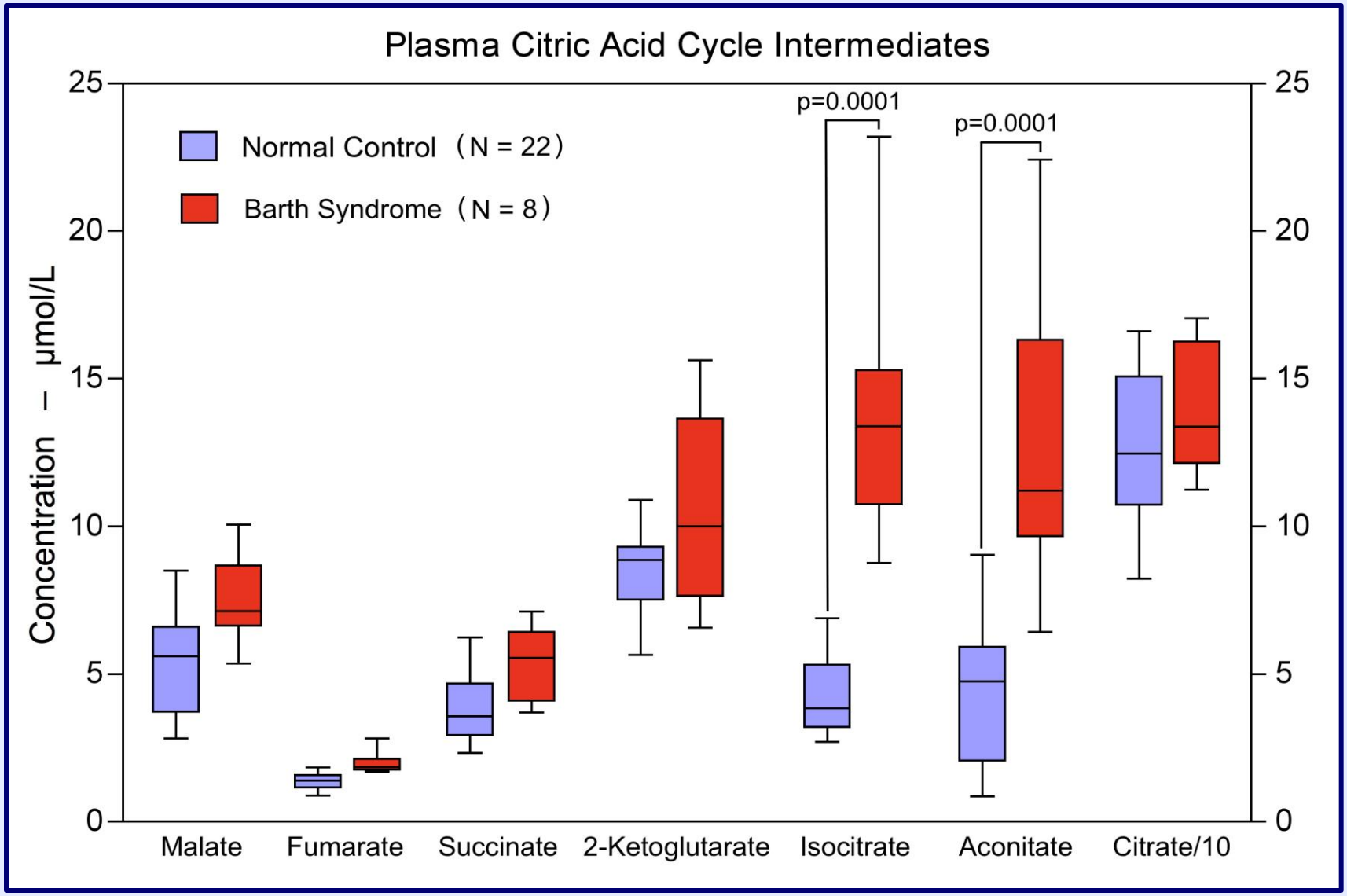


F. Jiang et al, J Biol Chem 275:22387-22394, 2000

Citric Acid Cycle



Plasma Citric Acid Cycle Intermediate Levels



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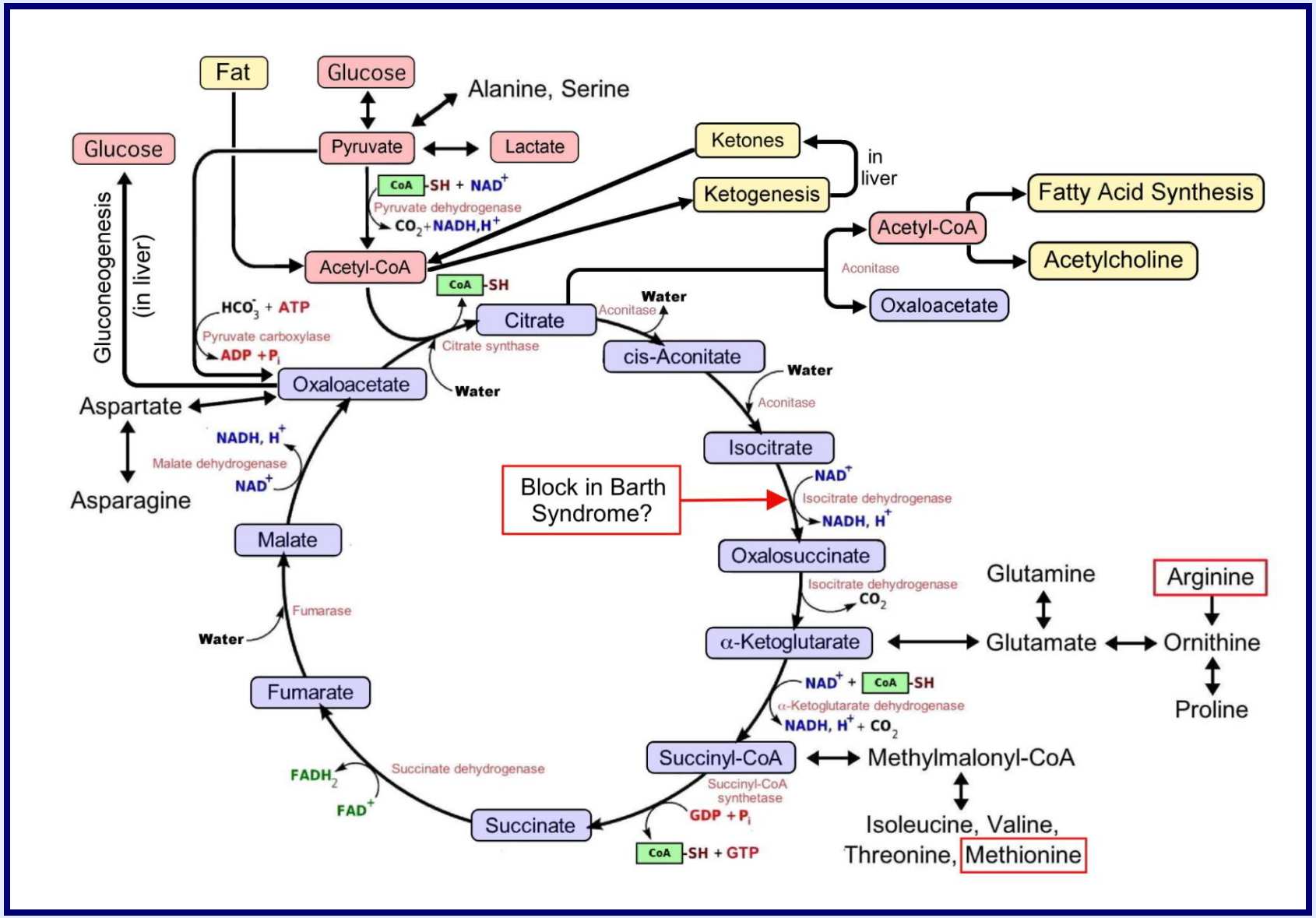

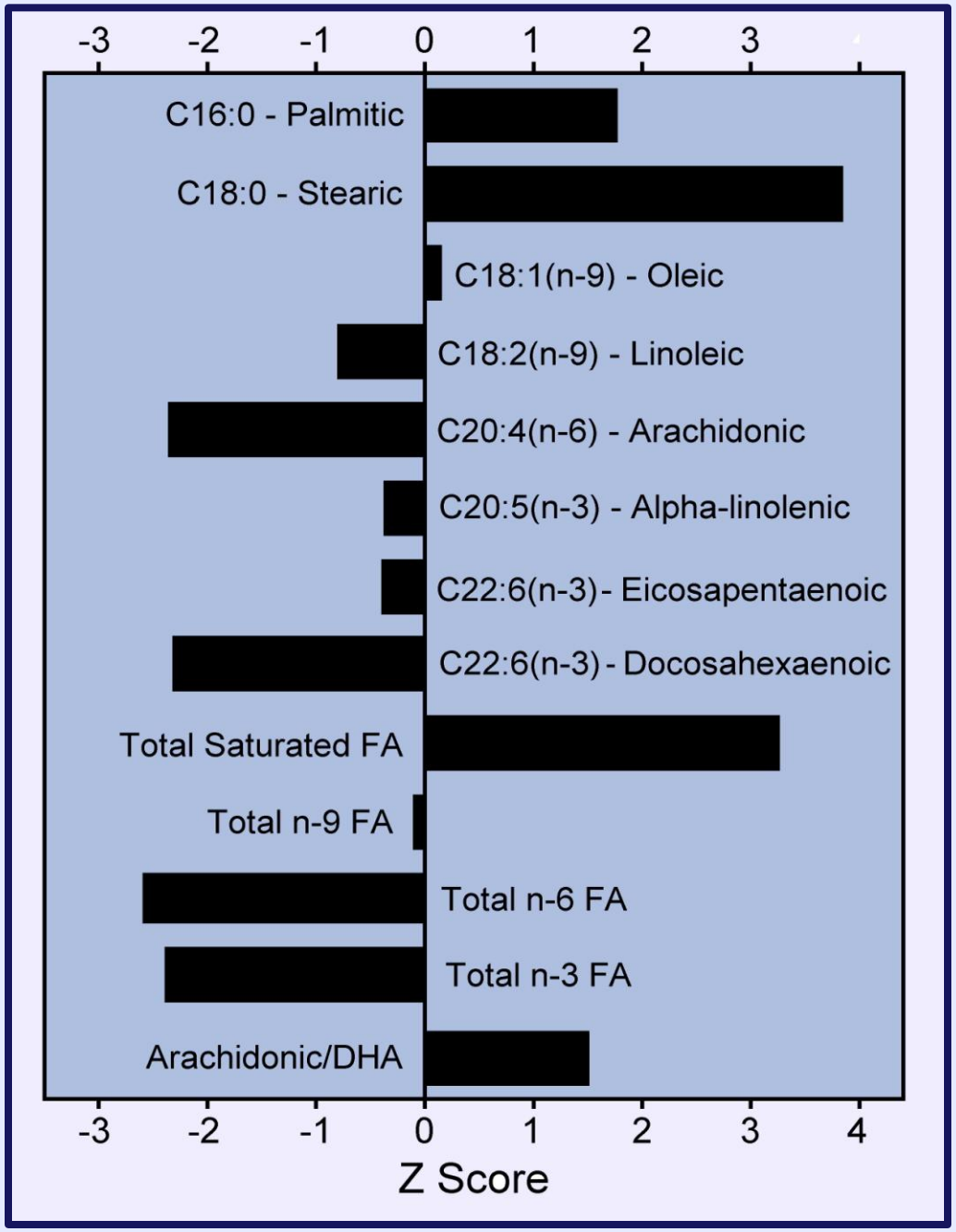

Barth Syndrome

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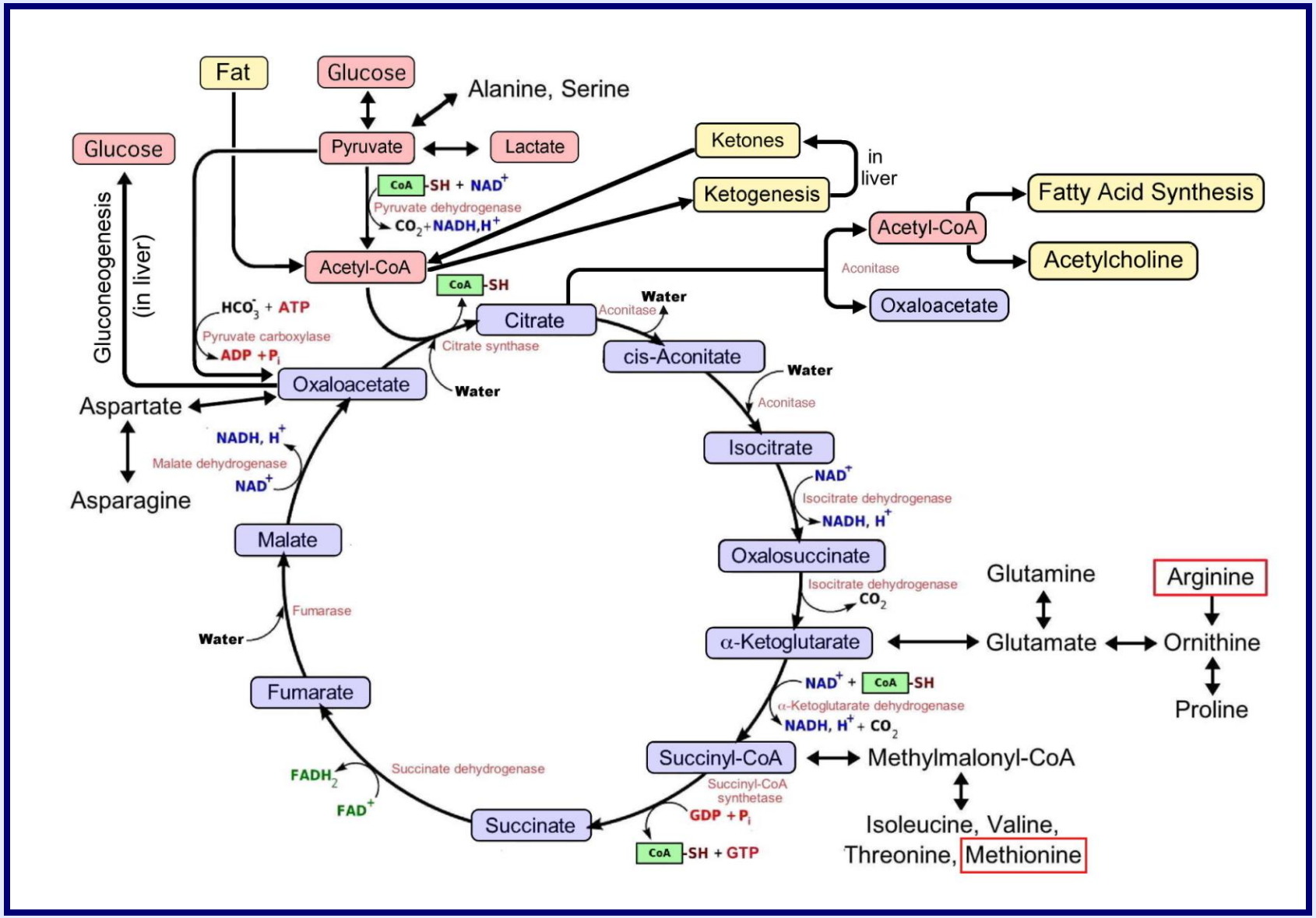
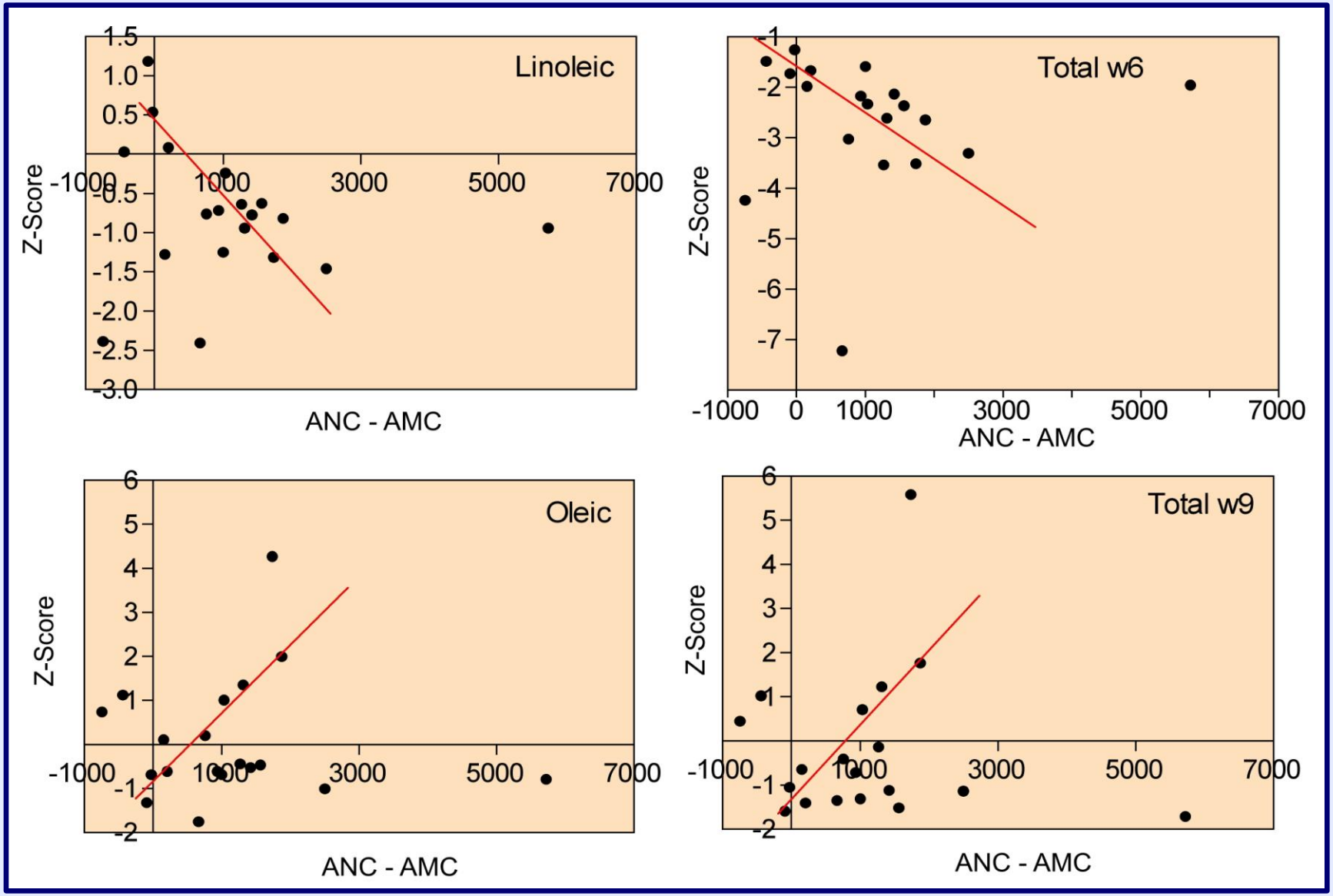
Erythrocyte Total Lipid Fatty Acid Levels

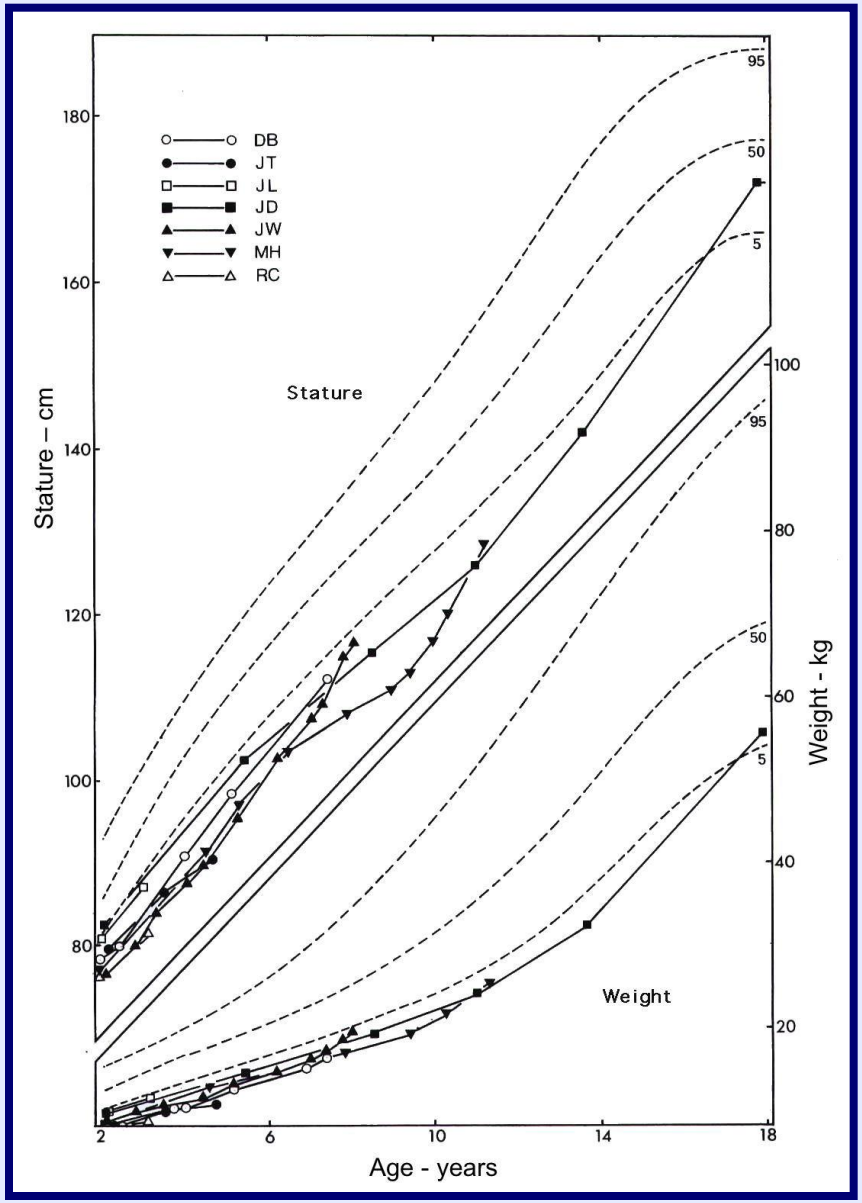
Citric Acid Cycle

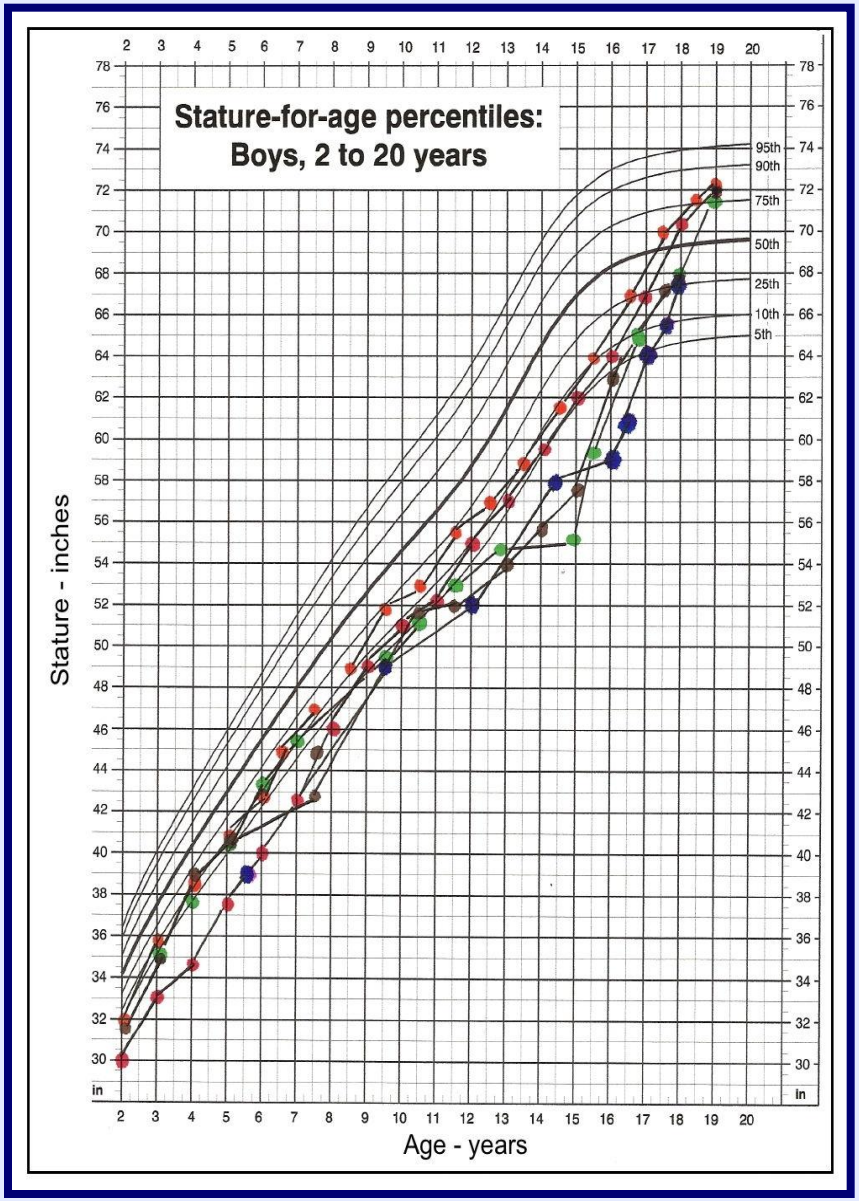


RBC Lipid Fatty Acid Levels vs. Neutrophil Index



Growth in Barth Syndrome





Barth Syndrome: Conclusions & Speculations

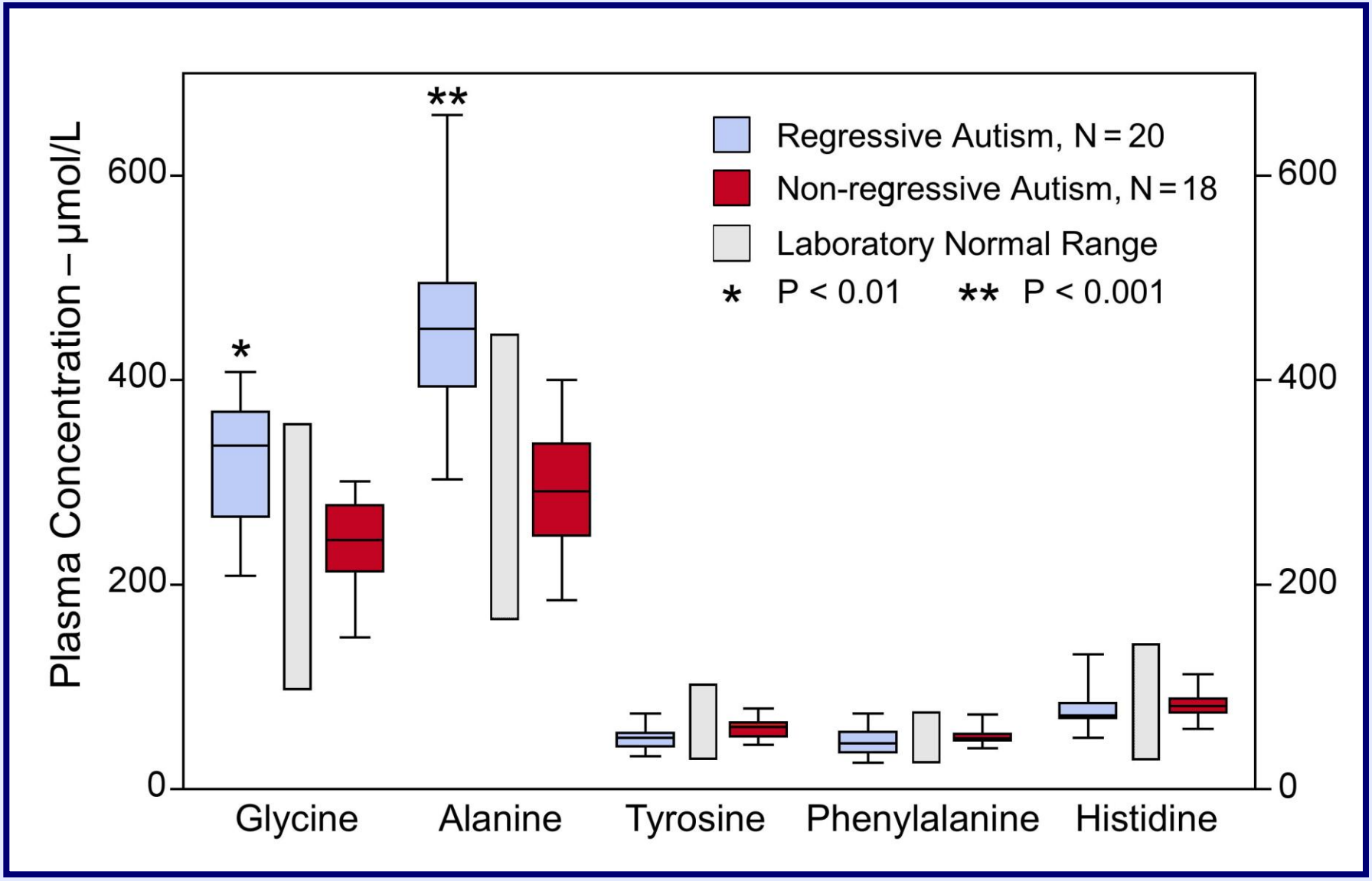
Cardiomyopathy is largely nutritional, due to endogenous amino acid depletion

A major effect of cardiolipin deficiency is impaired import of citric acid cycle enzymes

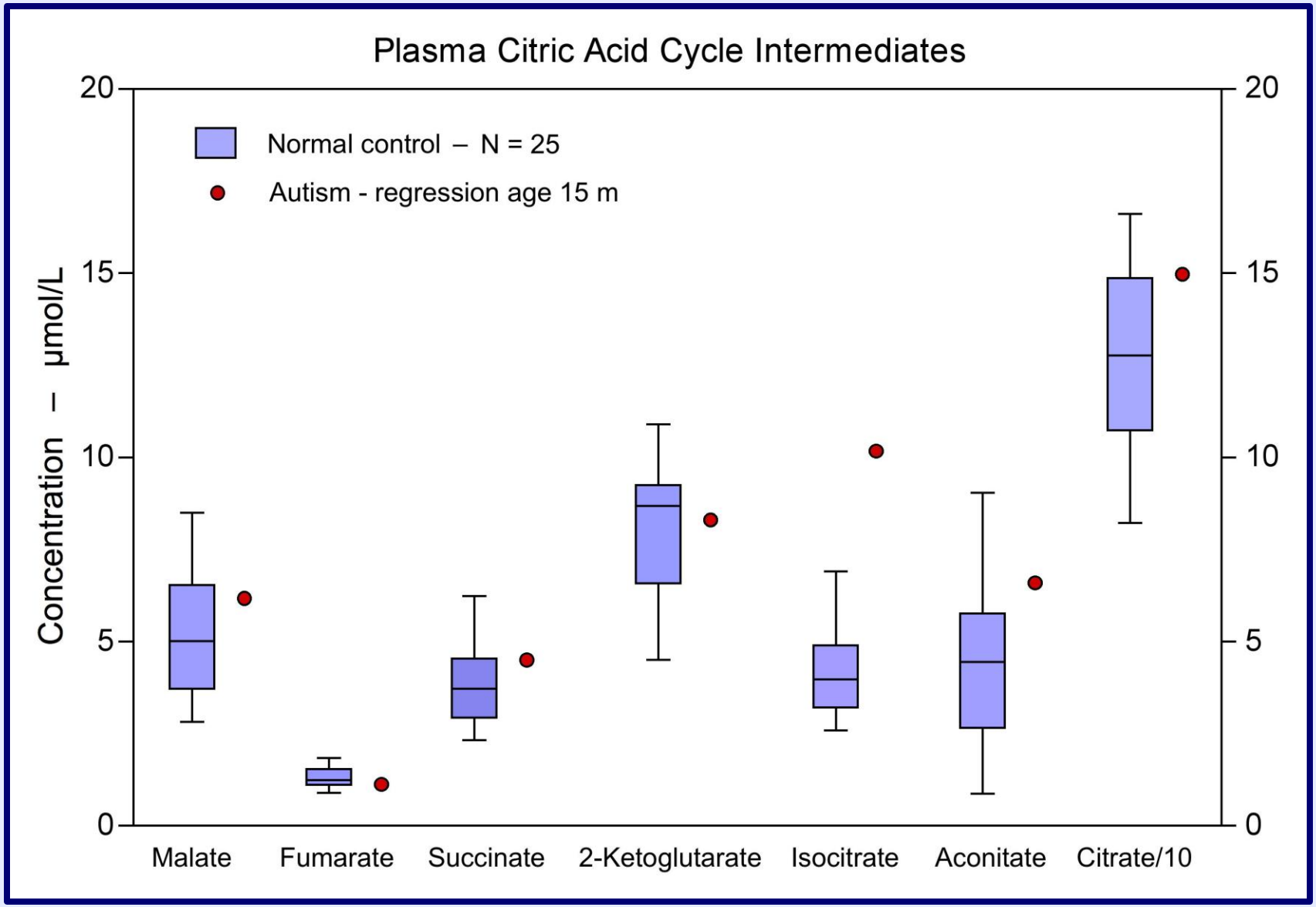
TAZ deficiency affects non-mitochondrial lipids

Neutropenia & growth abnormalities could be caused by abnormal receptor response

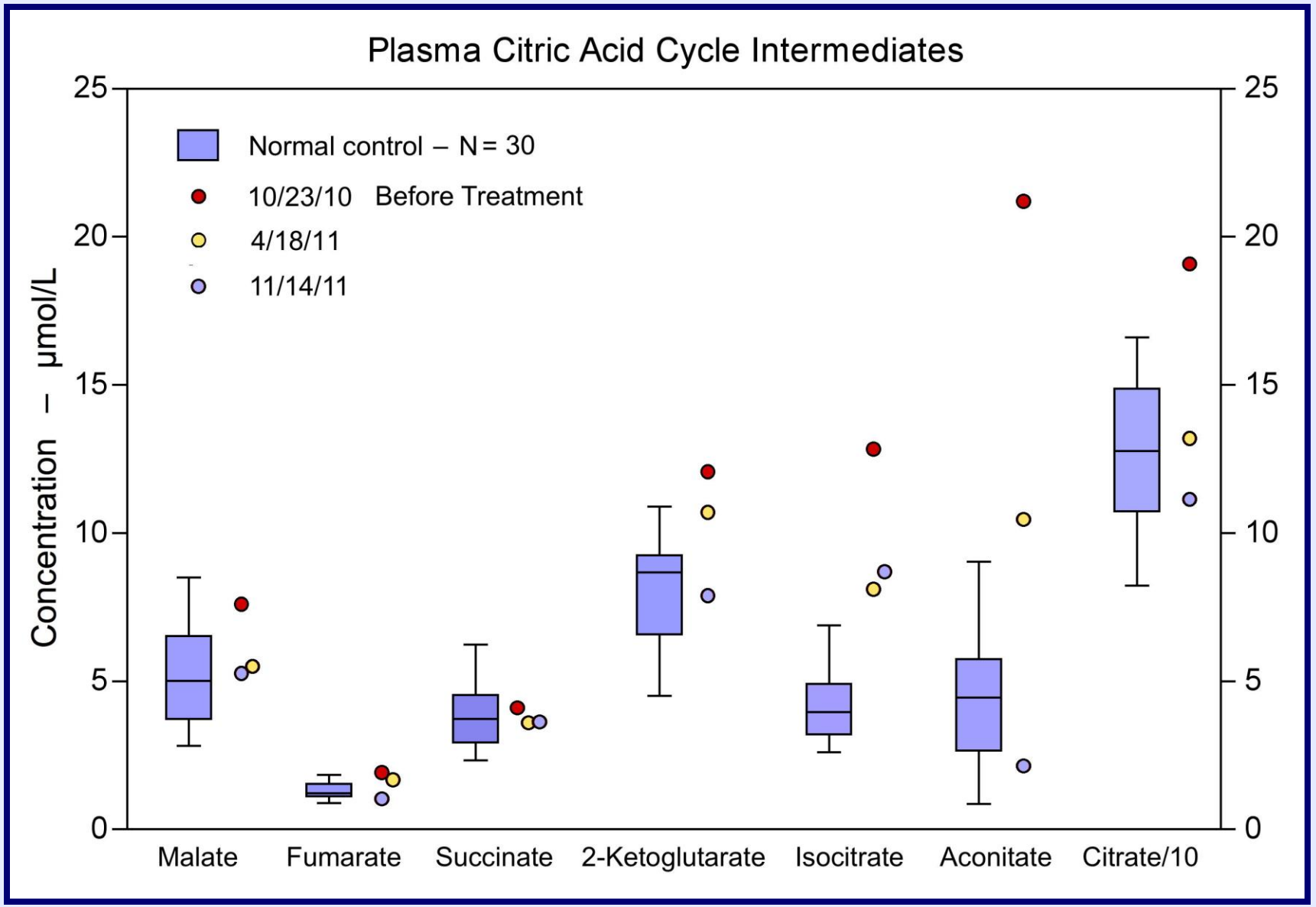
Plasma Amino Acids In Pediatric Autism



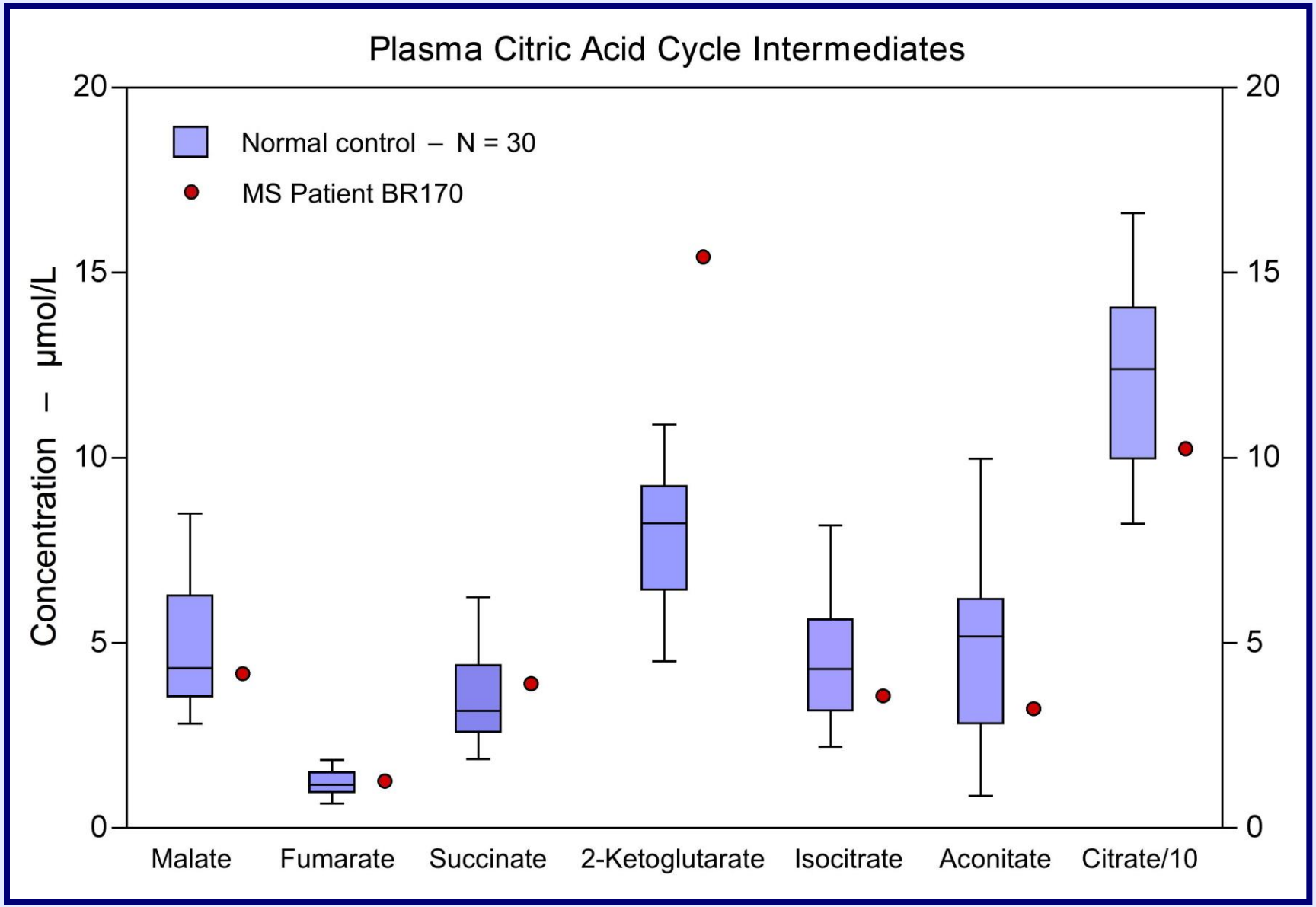
TCA Intermediates in Pediatric Autism



Mitochondrial Treatment of Regressive Autism



TCA Intermediates in Multiple Sclerosis



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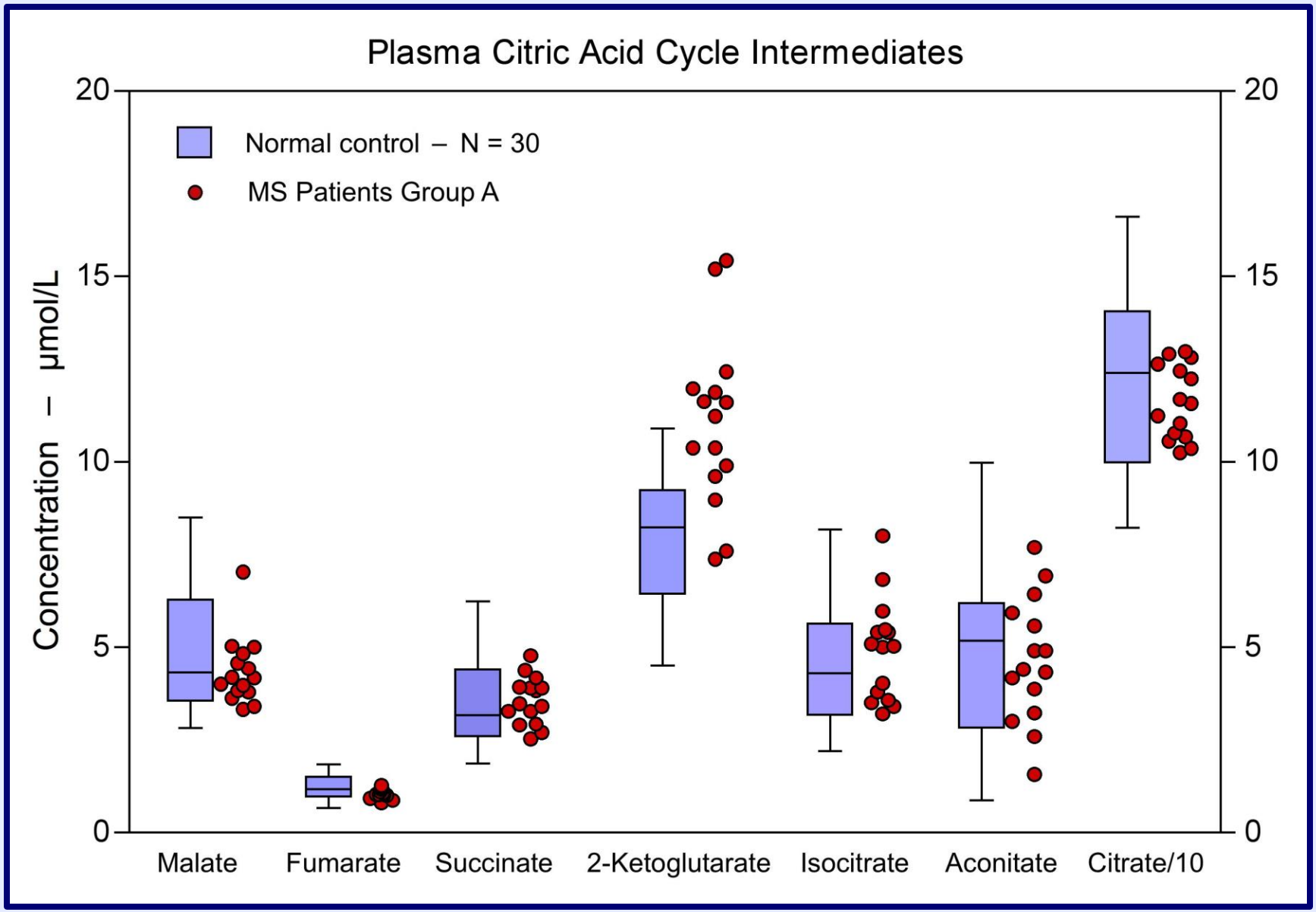
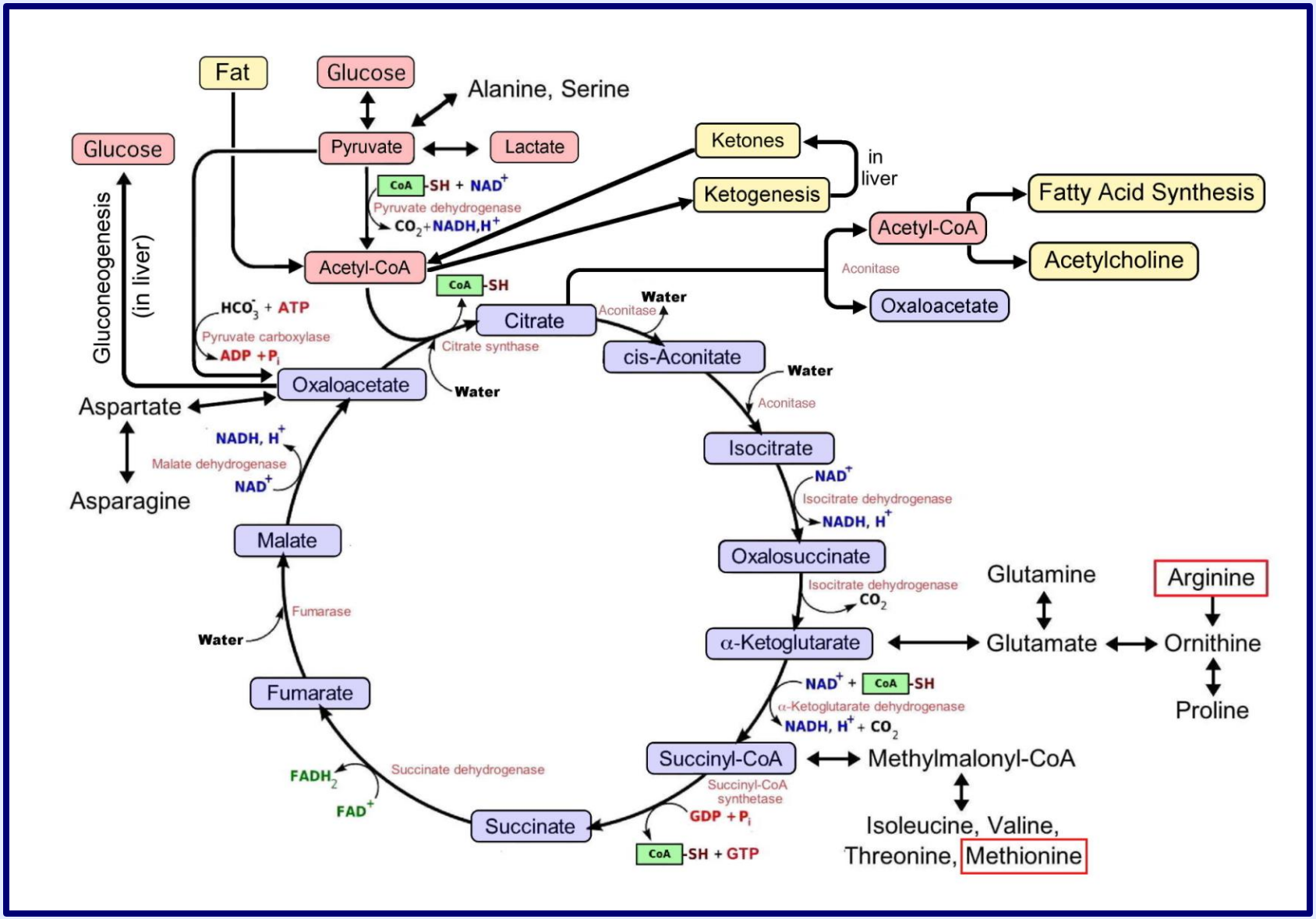
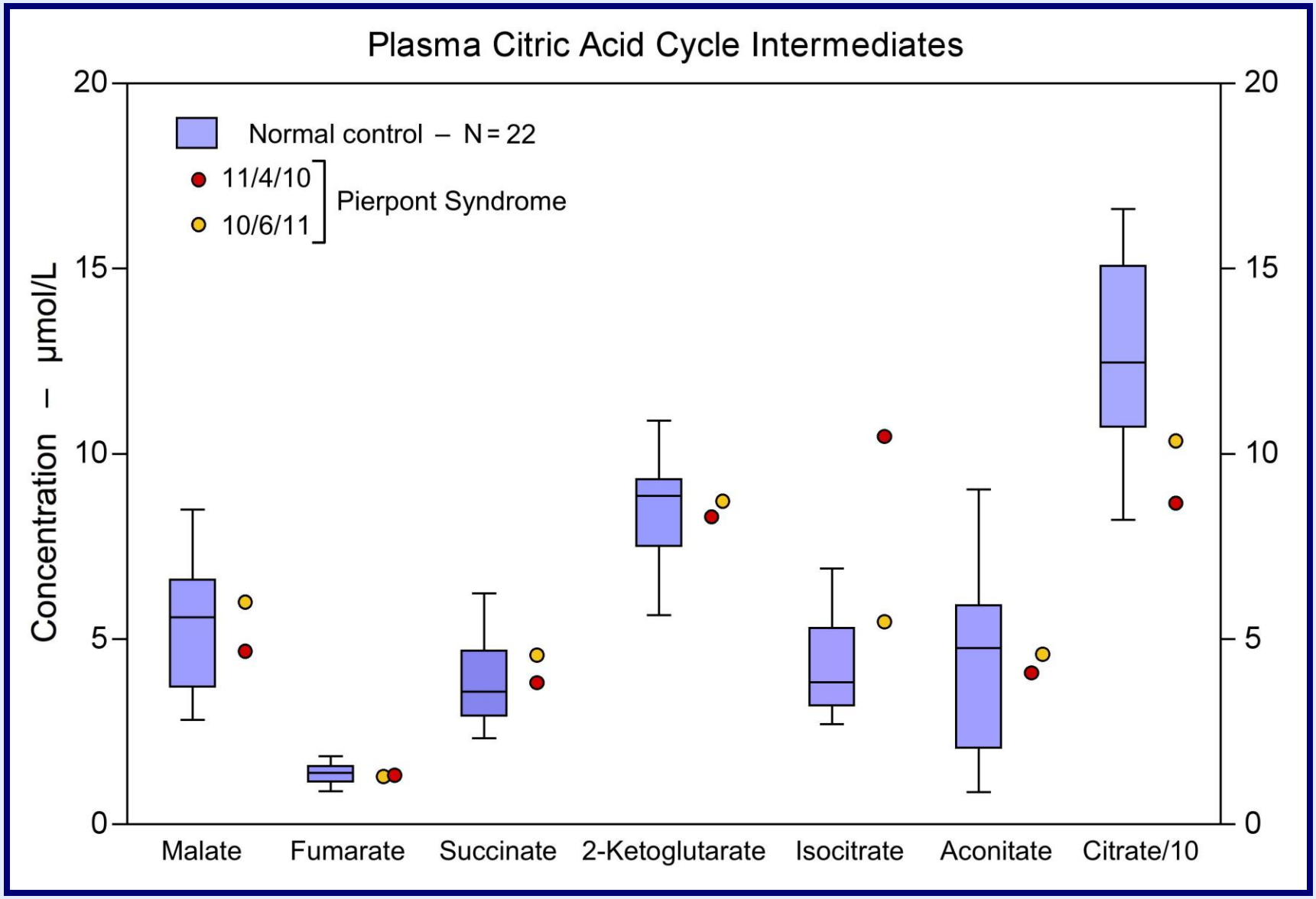


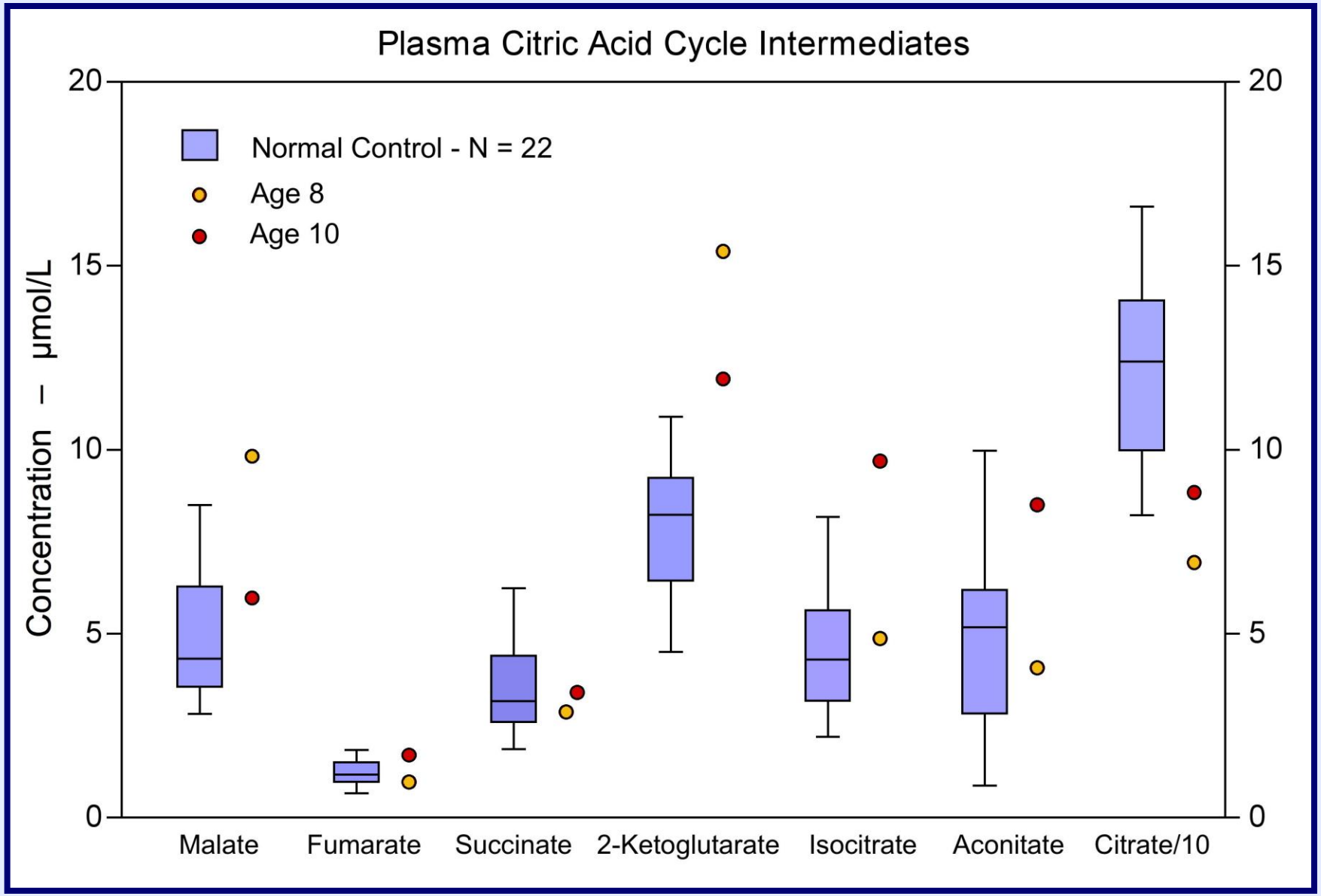
Citric Acid Cycle



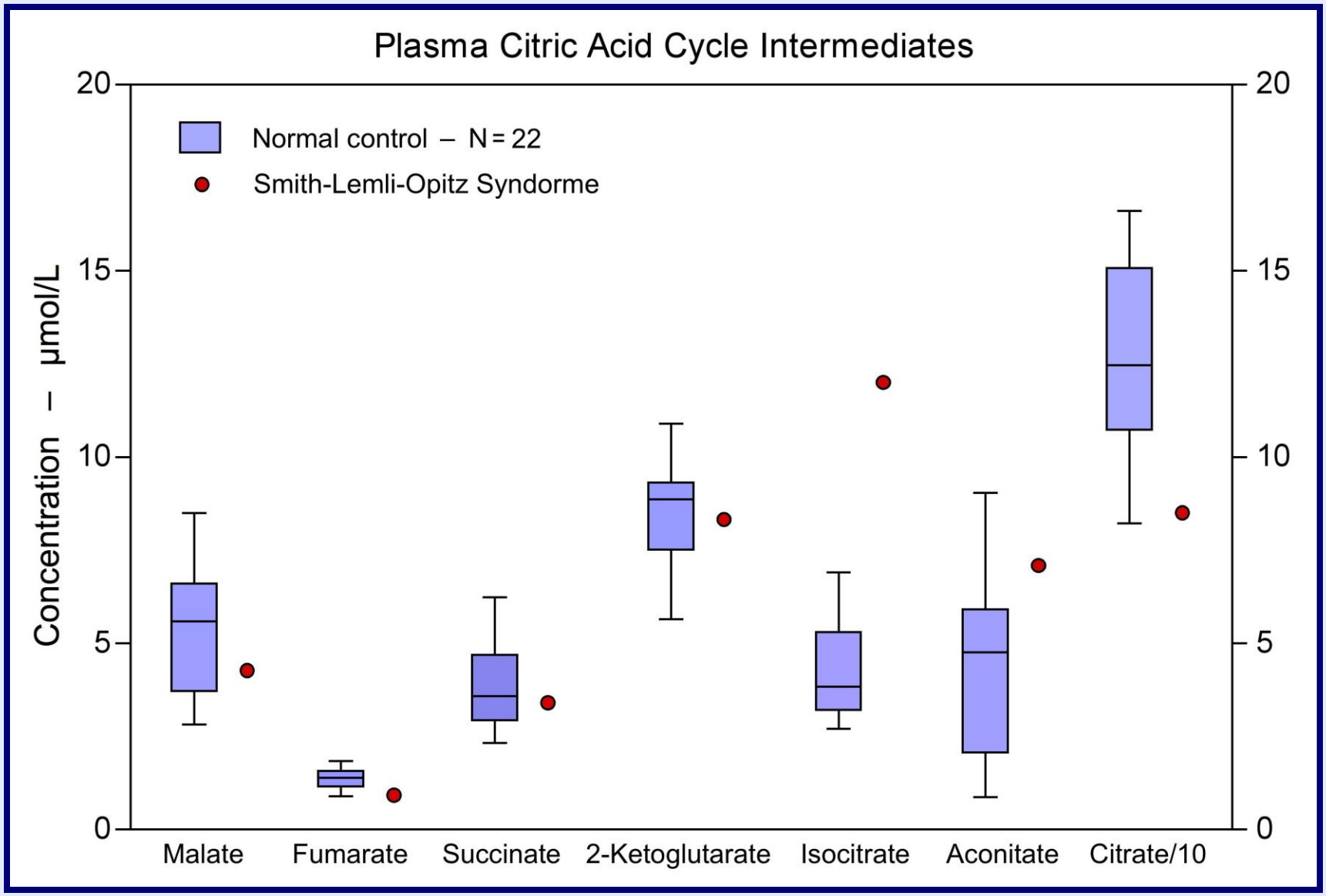
TCA Intermediates in Pierpont Syndrome



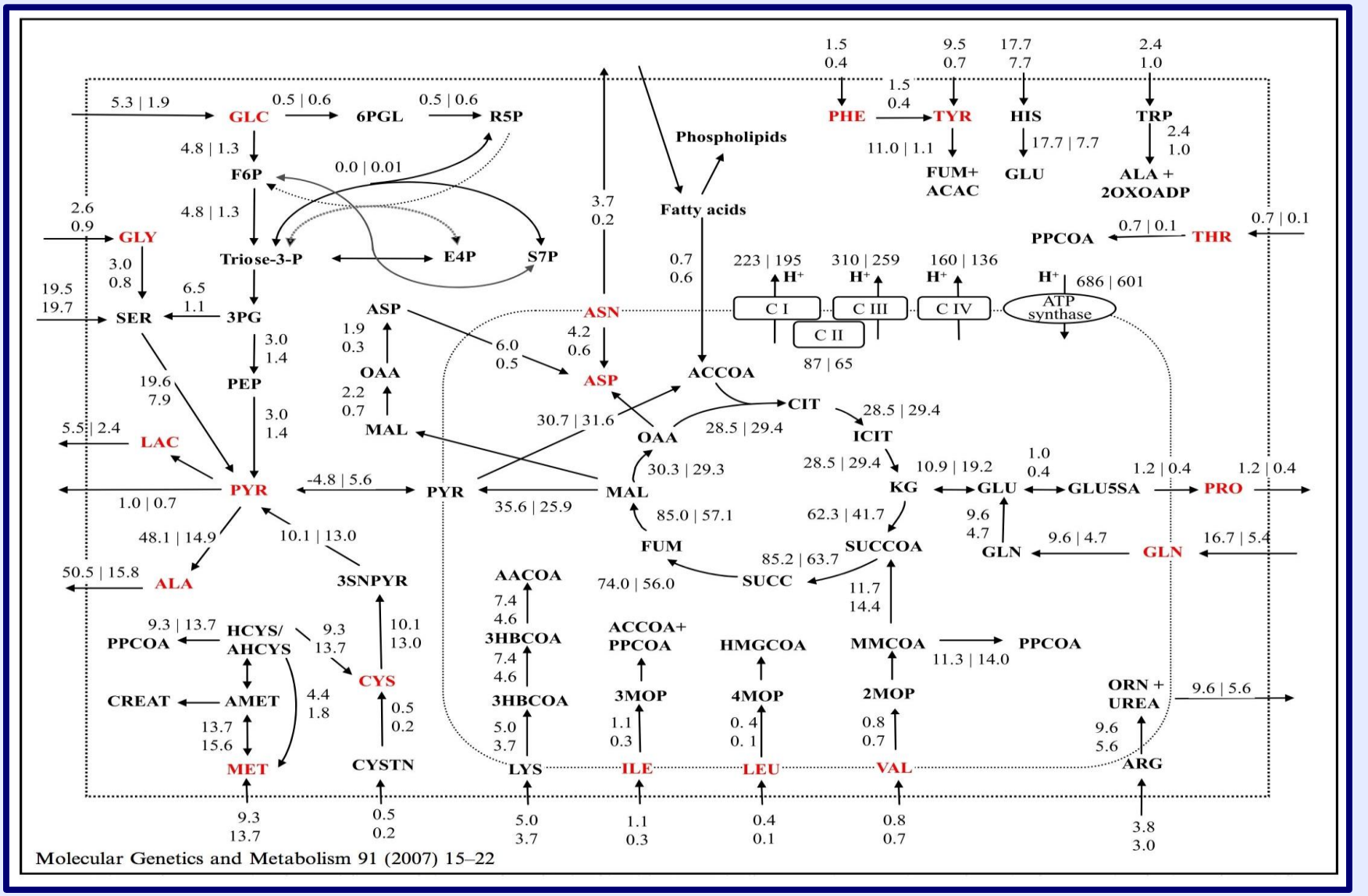
TCA Intermediates in Adrenoleukodystrophy

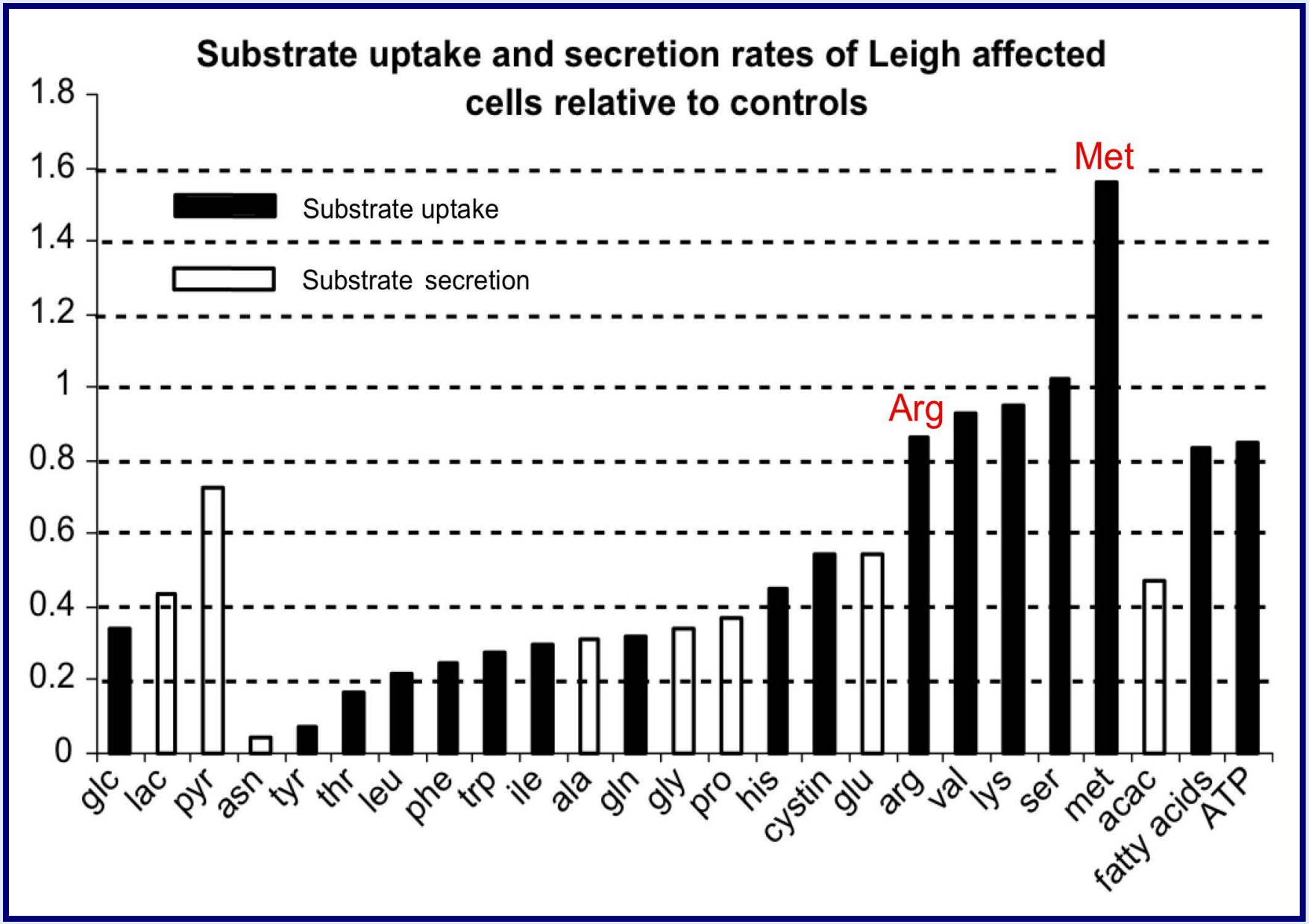


TCA Intermediates in Smith-Leml-Opitz Syndrome

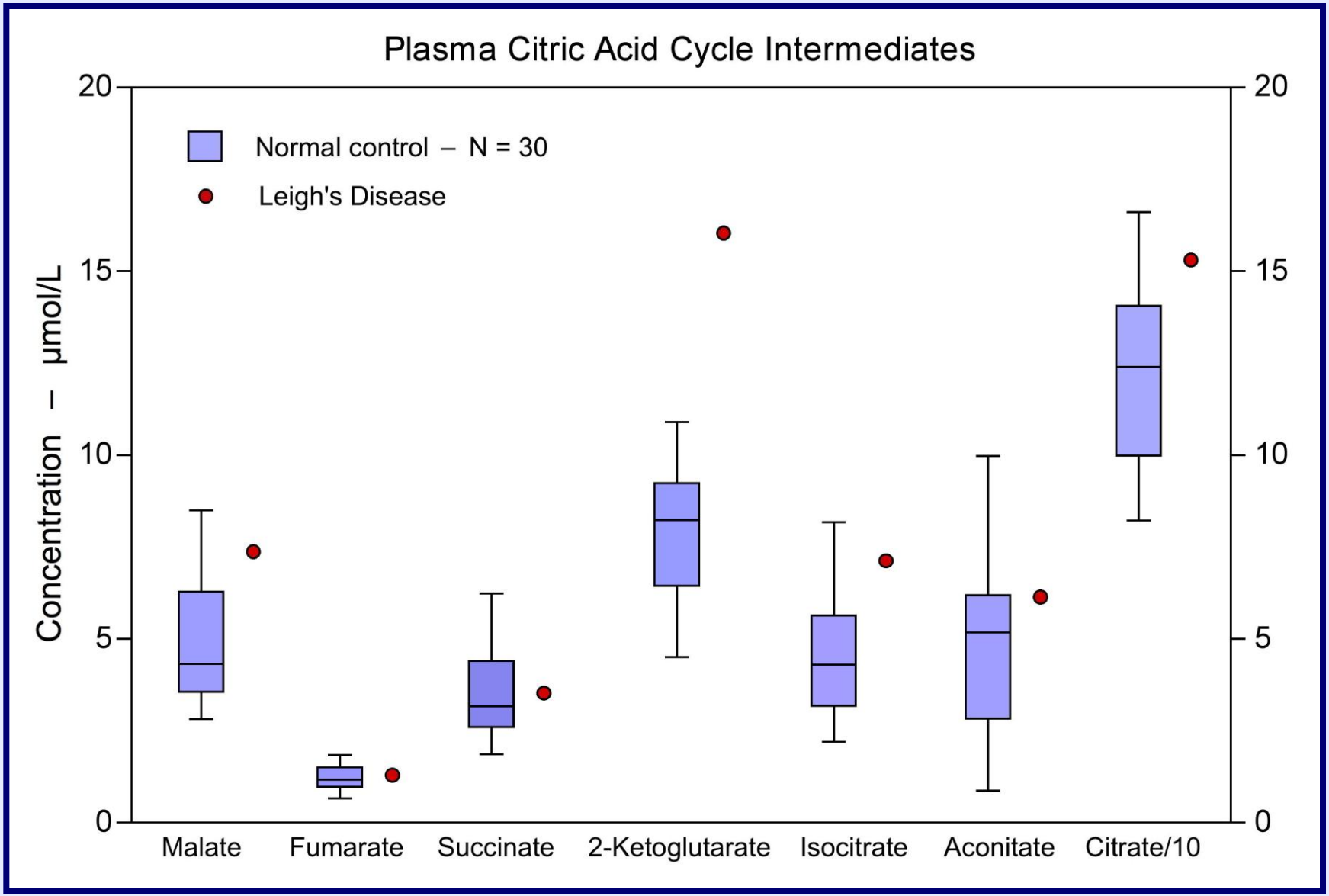


Substrate Fluxes in Leigh's Disease Fibroblasts

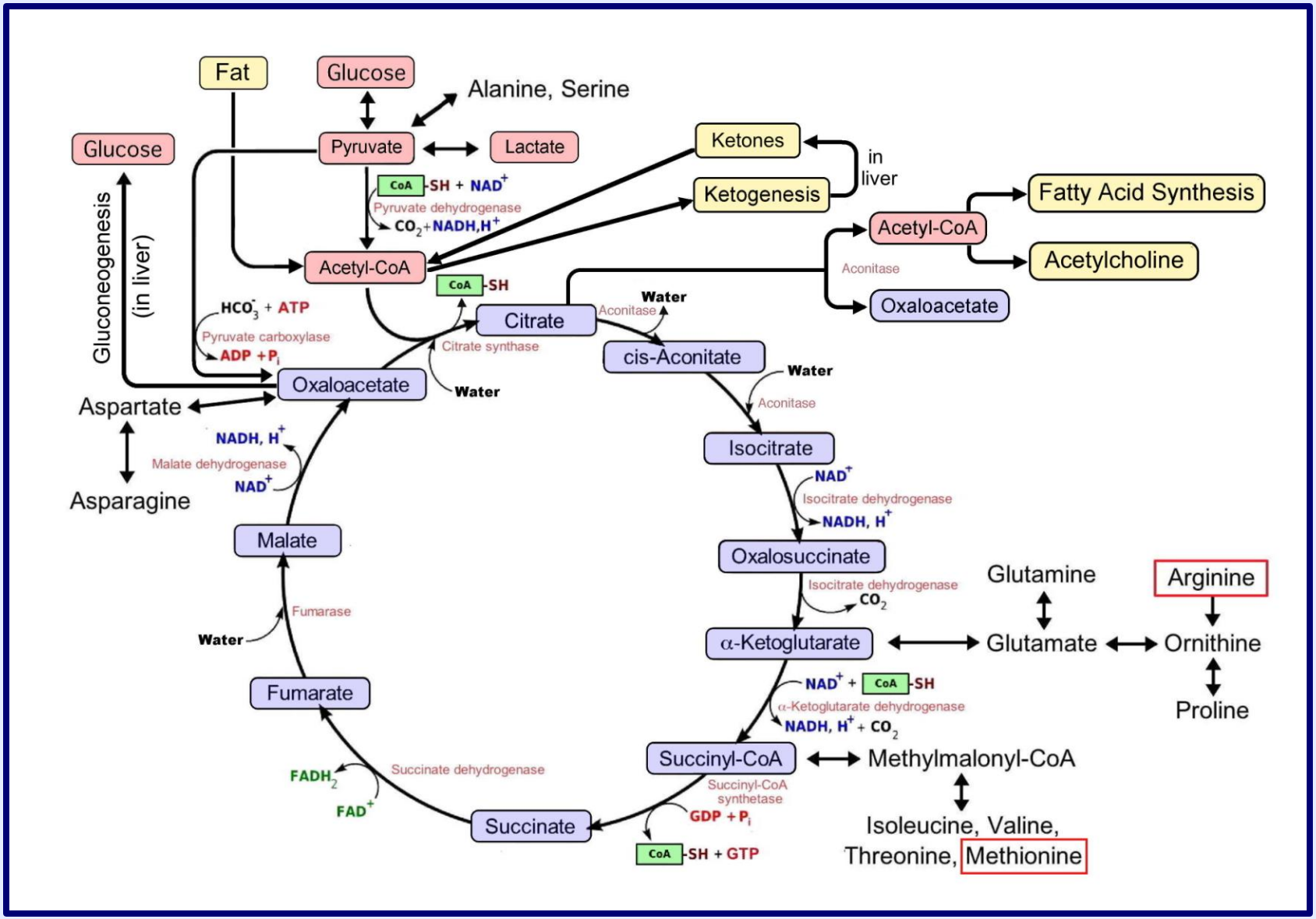



Substrate uptake and secretion rates of Leigh affected cells relative to controls
1.8
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
Substrate uptake
Substrate secretion
Met
Arg
glc
lac
pyr
asn
tyr
thr
leu
phe
trp
ile
ala
gln
gly
pro
his
cystin
glu
arg
val
lys
ser
met
acac
fatty acids
ATP

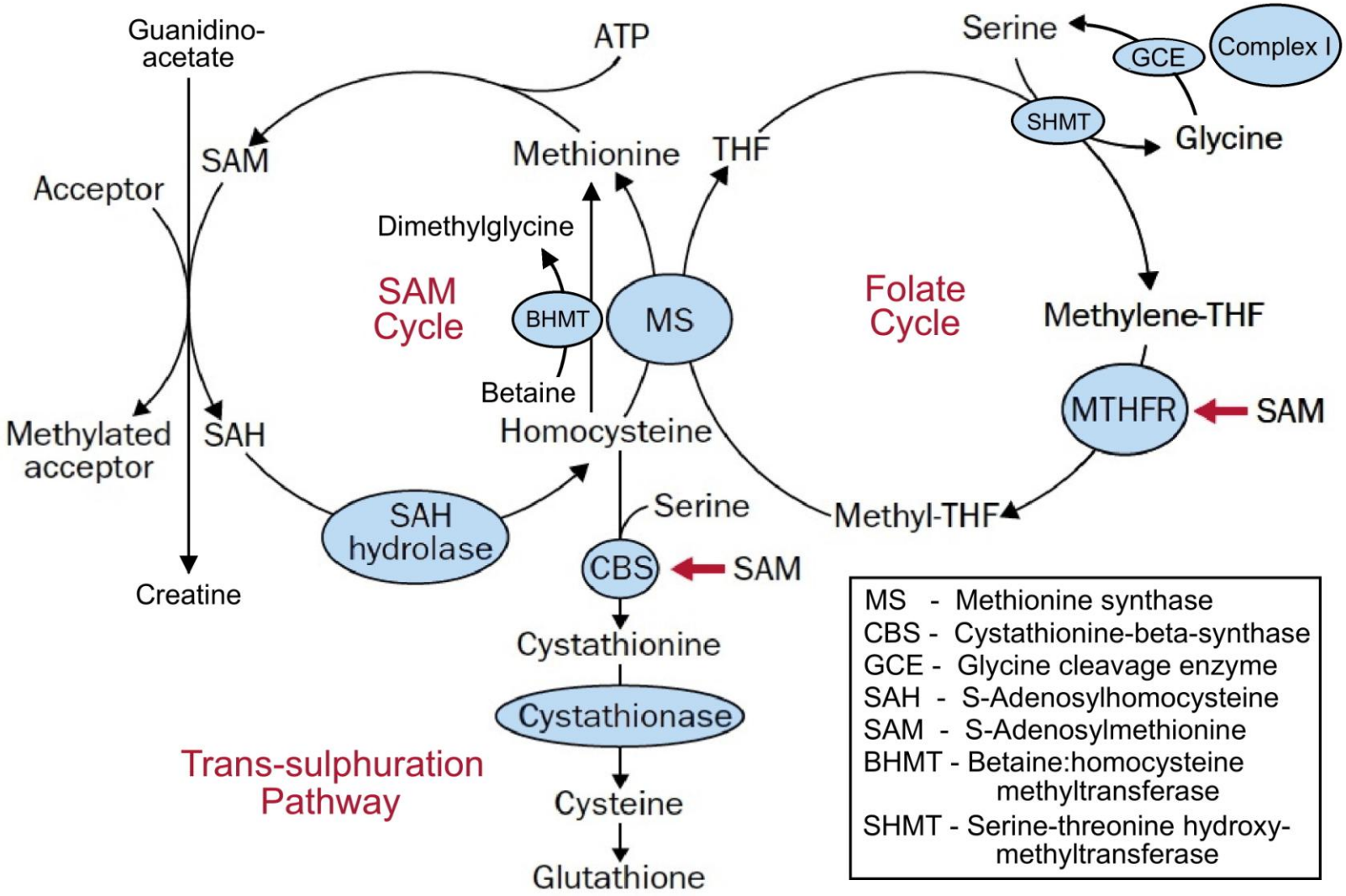
TCA Intermediates in Leigh's Disease



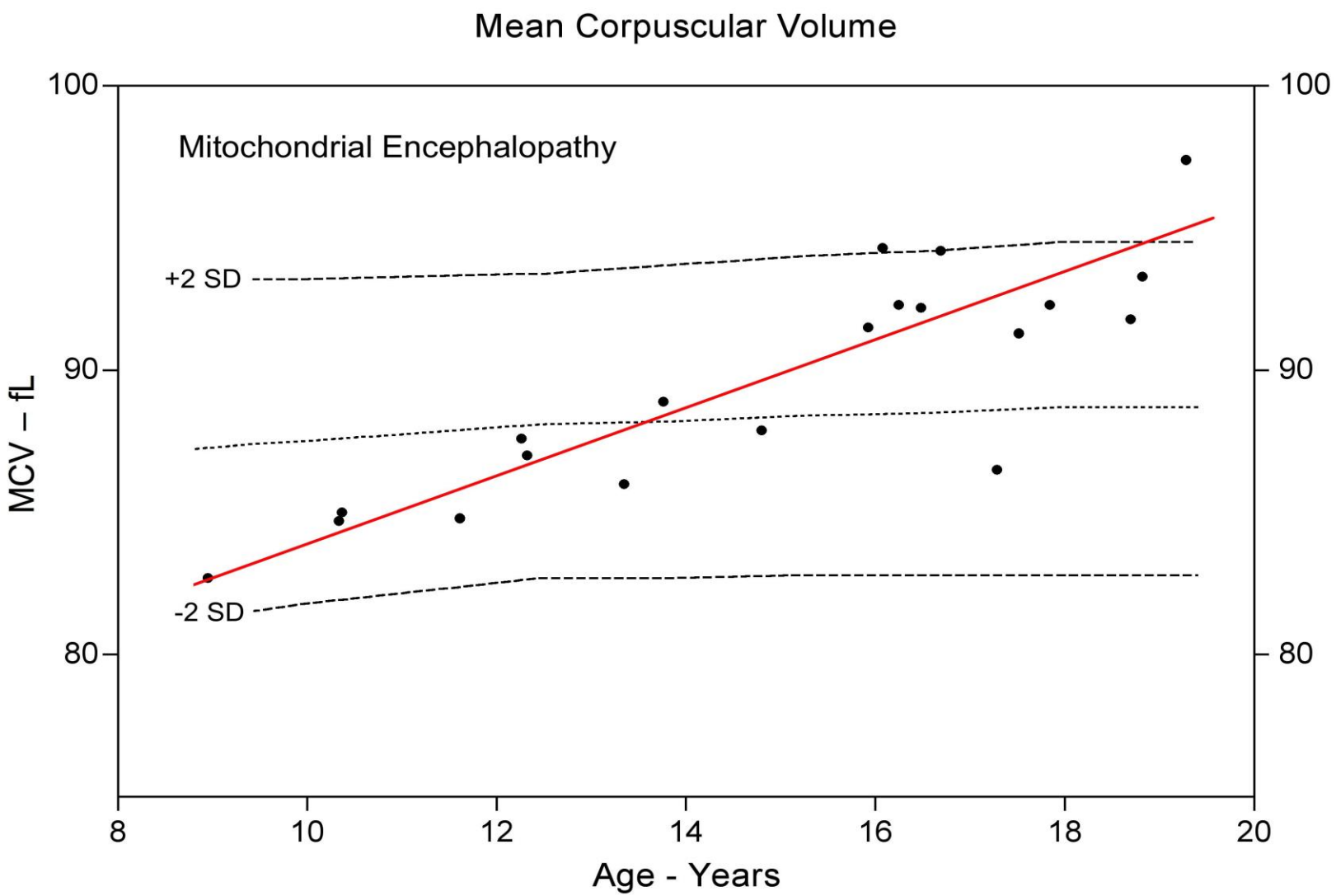
Citric Acid Cycle



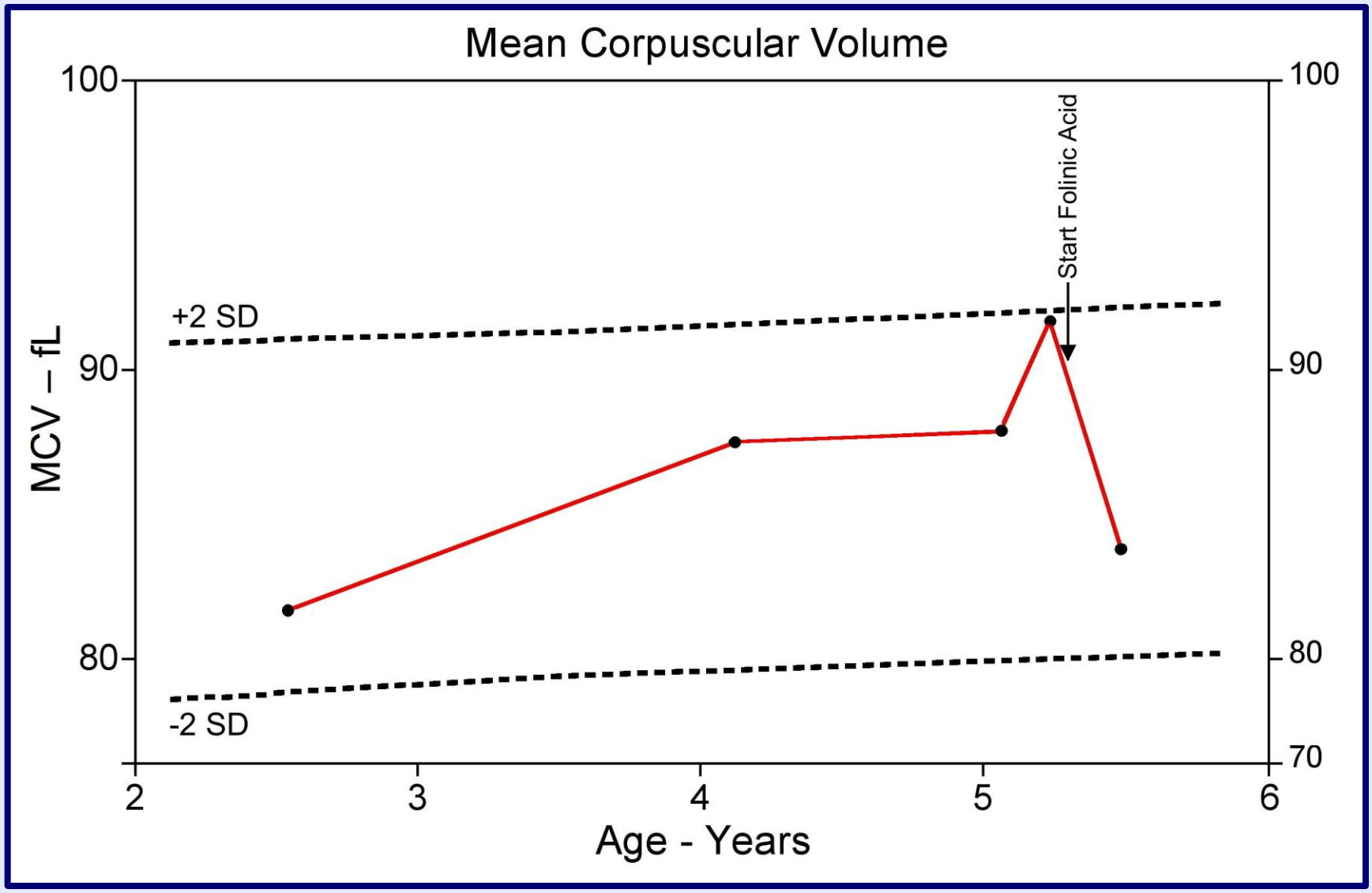
Folate – Methionine Cycle



Red Cell Volumes in Leigh's Disease



Red Cell Volumes in Leigh's Disease



Disorders in which Mitochondrial Dysfunction Contributes to the Disease Process

1. ATP deficiency may not be the primary of cellular damage in mitochondrial diseases.
2. In Barth syndrome and many classical mitochondrial diseases, extramitochondrial substrate depletion is a major cause of cellular and organ pathology.
3. Careful analysis of plasma amino acid and TCA cycle intermediates can identify both the essential pathology of a mitochondrial disease and its treatment.

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