Liver Transplant for Organic Acid Disorders

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Disclosures

- Consultant – Moderna Therapeutics, Horizon Pharma, Natera
- Clinical trials – BioElectron, Stealth Therapeutics
- DSMB – Biomarin, Audentes Therapeutics, Amicus, RegenxBio, Neurovia

Liver Transplant for Organic Acidemias

- Liver the major site of branched-chain amino acid metabolism
- MMA and PA
- Poor outcome for severe cases

Pediatric Liver Transplant for Organic Acidemias

- United Network for Organ Sharing (UNOS) 2002-2012
- 5672 pediatric LT
- 323 (5.4%) liver-only for UCDs/OAs
  - Proportion increased from 4.3% to 7/4%
- 17 LKT (all with MMA)
- 96% deceased donor transplants
- 59% transplanted <2 y

Liver Transpl 20:89-99, 2014

Pediatric Liver Transplant for Organic Acidemias

TABLE 1. Classification of UCDs and OAs

	UCDs (n = 186)	OAs (n = 137)
Mechanism	Defect in 1 of 6 urea cycle enzymes	Defect in an enzyme that metabolizes branched-chain amino acids or lysine or in another step of amino acid metabolism
Types	• Carbamyl phosphate synthetase deficiency • *N*-Acetylglutamate synthetase deficiency • Ornithine transcarbamylase deficiency (X-linked) • Argininosuccinic acid synthetase deficiency (citrullinemia) • Argininosuccinate lyase deficiency • Arginase deficiency	• MSUD • PA • MMA • Homocysteinuria/methylmalonic aciduria • Isovaleric acidemia • Biotin-unresponsive 3-methylcrotonyl coenzyme A carboxylase deficiency • 3-hydroxy-3-methylglutaryl coenzyme A lyase deficiency • Ketothiolase deficiency • Glutaric acidemia type I

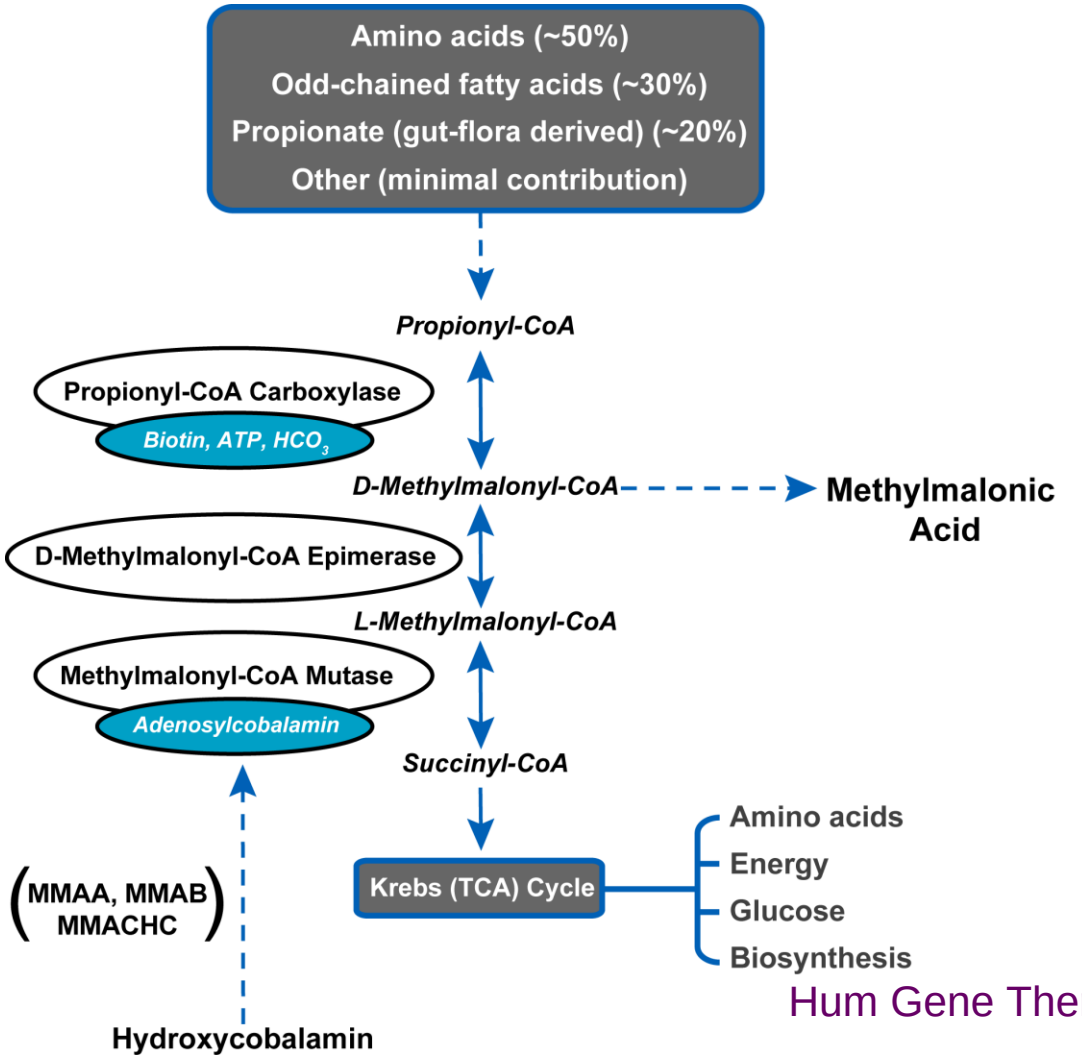
Liver Transpl 20:89-99, 2014

Methylmalonyl-CoA mutase

- Nuclear encoded
- Mitochondrial localized
- Homodimer
- Requires 5’-deoxyadenosylcobalamin (Adocbl)

Propionyl-CoA Carboxylase

- Nuclear encoded
- Mitochondrial localized
- Dodecamer (PCCA and PCCB subunits)
- Biotin-dependent reaction



Hum Gene Ther 19:53-60, 2008

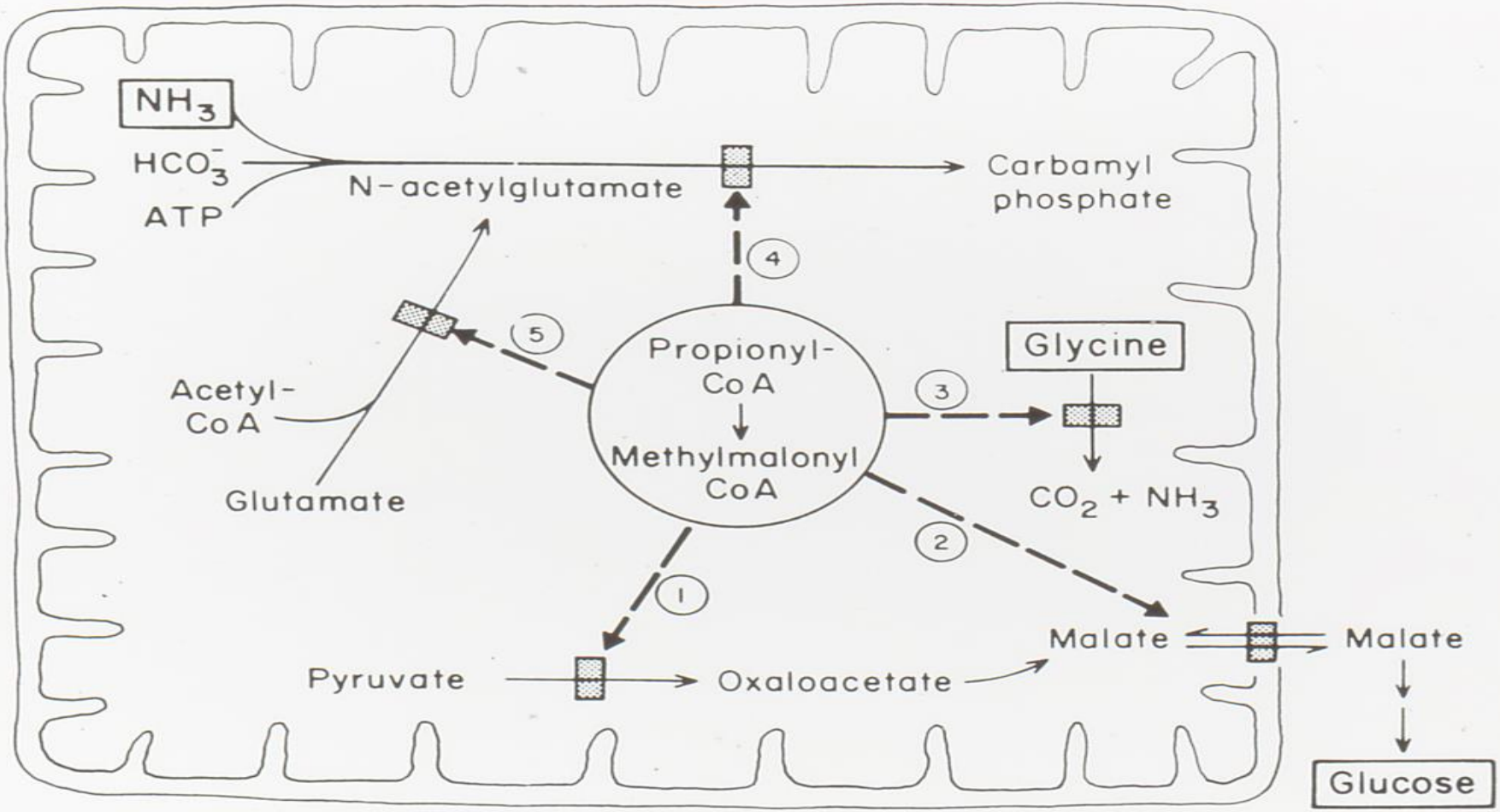

NH3
HCO3-
ATP
N-acetylglutamate
Carbamyl phosphate
4
5
Acetyl-CoA
Glutamate
Propionyl-CoA
Methylmalonyl CoA
Glycine
3
CO2 + NH3
2
1
Pyruvate
Oxaloacetate
Malate
Malate
Glucose

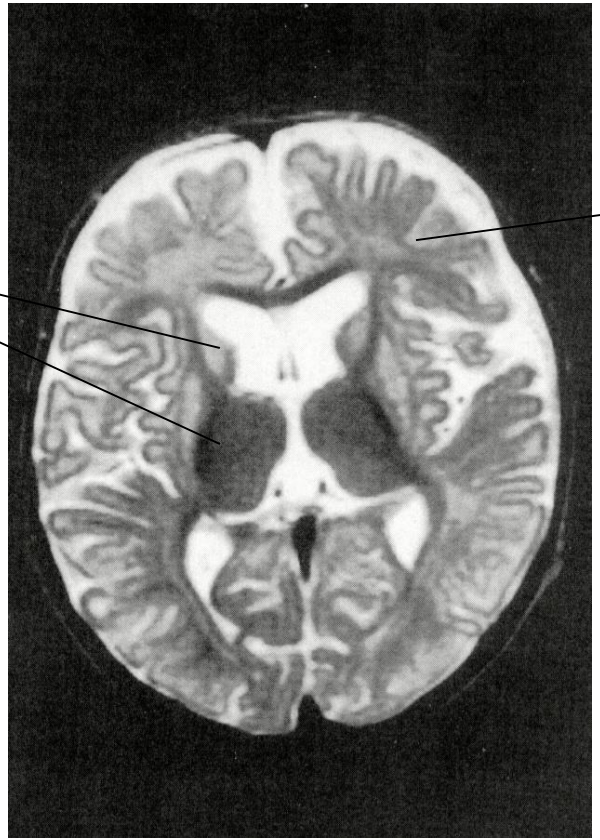
Table 3 Acute and chronic presentations of MMA/PA

Acute presentation	Chronic presentation
Neonatal sepsis-like picture, temperature instability, respiratory distress, hyperventilation	**Often episodic characteristic signs and symptoms**
Nervous system	**Nervous system**
• **Altered level of consciousness (from lethargy and somnolence to coma) mimicking encephalitis or drug intoxication**	• Hypotonia
	• **Developmental delay** (learning disabilities, intellectual disability)
• Acute encephalopathy	• **Movement disorders/dystonia**
• Seizures (in general not isolated but in the context of altered level of consciousness)	• Seizures
• Movement disorders (more frequent in PA)	• Optic atrophy
• Stroke-like episodes (more frequent in MMA)	• *Psychiatric symptoms (hallucinations, psychotic attacks)*
Gastrointestinal system	**Gastrointestinal system**
• **Vomiting and feeding difficulties**	• **Recurrent vomiting with ketoacidosis**
	• Abnormal feeding behavior (anorexia)
	• **Failure to thrive**
	• Constipation
	• Pancreatitis
Hematologic findings	**Hematologic findings**
• Neutropenia, pancytopenia	• Neutropenia, pancytopenia
	• *Secondary hemophagocytosis (rare)*
Heart	**Heart (more frequent in PA)**
• Acute cardiac failure (mostly on basis of cardiomyopathy)	• Cardiomyopathy
• Arrhythmias	• Prolonged QTc interval in ECG
	Kidney (more frequent in MMA)
	• **Chronic renal failure in MMA**
	Other
	• *Dermatitis*
	• *Hearing loss*

Orphanet J Rare Dis 9:130, 2014

BRAIN INJURY IN ORGANIC ACIDEMIAS

caudate and putamen hyperintensity



delayed myelination

Pediatr Res 40:404-9, 1996

MMA Therapy

- Special low-protein diet
- Emergency/sick-day protocols
- Carnitine supplementation
- Vitamin B_{12} in some cases
- Dialysis – ↑NH_3, metabolic acidosis, renal failure
- Liver or combined liver/kidney transplantation
- Kidney transplantation

PA Therapy

- Special low-protein diet
- Emergency/sick-day protocols
- Carnitine supplementation
- Carbamylglutamate
- Nitrogen-scavenging medications
- Liver transplantation
- Heart transplantation

Gene Therapy with a Scalpel

- Liver transplantation
- Kidney transplantation
- Combined liver/kidney transplantation

Combined liver-kidney transplantation in methylmalonic acidemia

W. G. van't Hoff, BSc, MD, MRCP, M. Dixon, BSc, SRD, J. Taylor, BM, MRCP, P. Mistry, PhD, MRCP, K. Rolles, MS, FRCS, L. Rees, MD, FRCP, and J. V. Leonard, PhD, FRCP

A 13-year-old boy with non-B12-responsive methylmalonic acidemia (MMA) had chronic renal failure. Hemodialysis led to symptomatic and biochemical improvement. He subsequently received a combined liver-kidney transplant. After 16 months of follow-up he has a normal lifestyle and a marked reduction in plasma and urine methylmalonate. (J Pediatr 1998;132:1043-4.)

Combined Liver-Kidney Transplantation in MMA

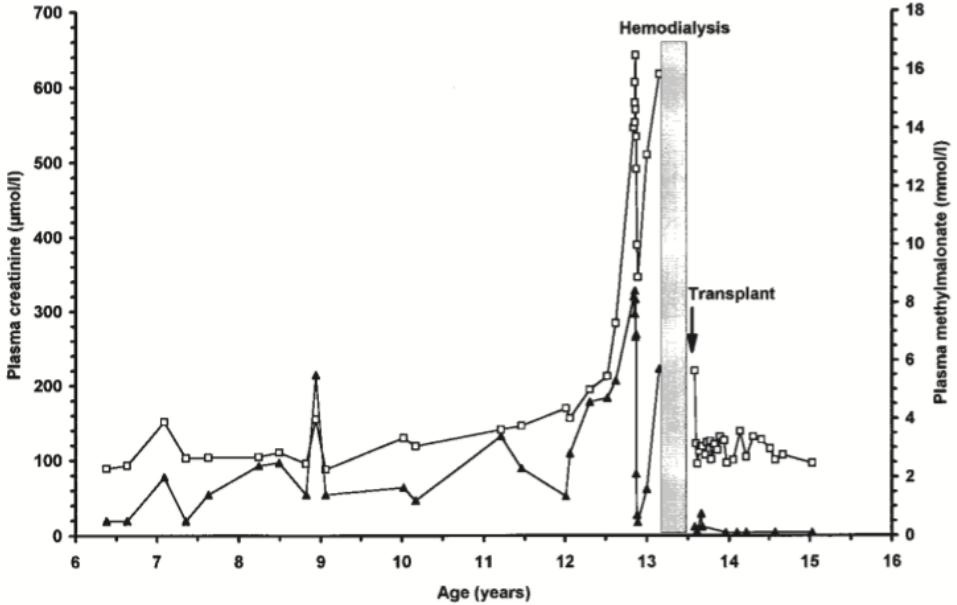
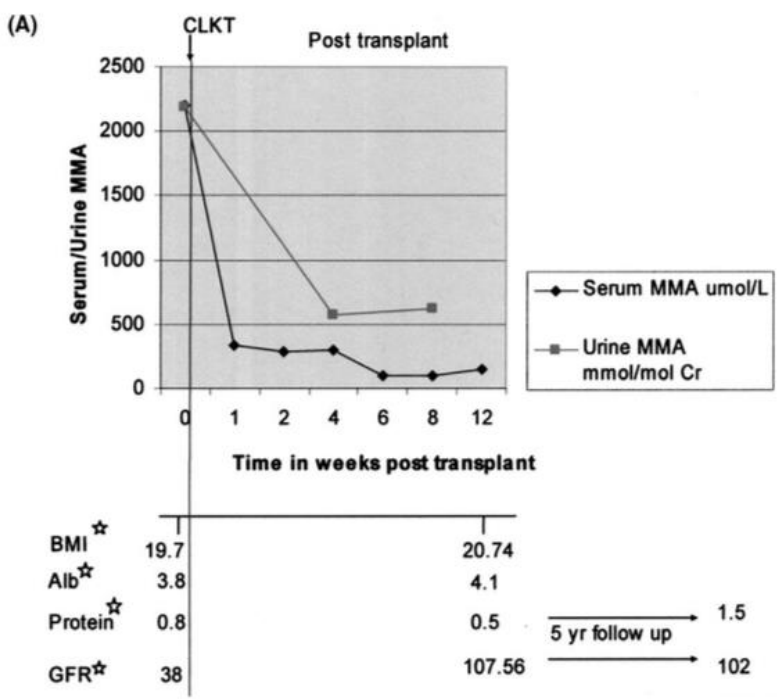
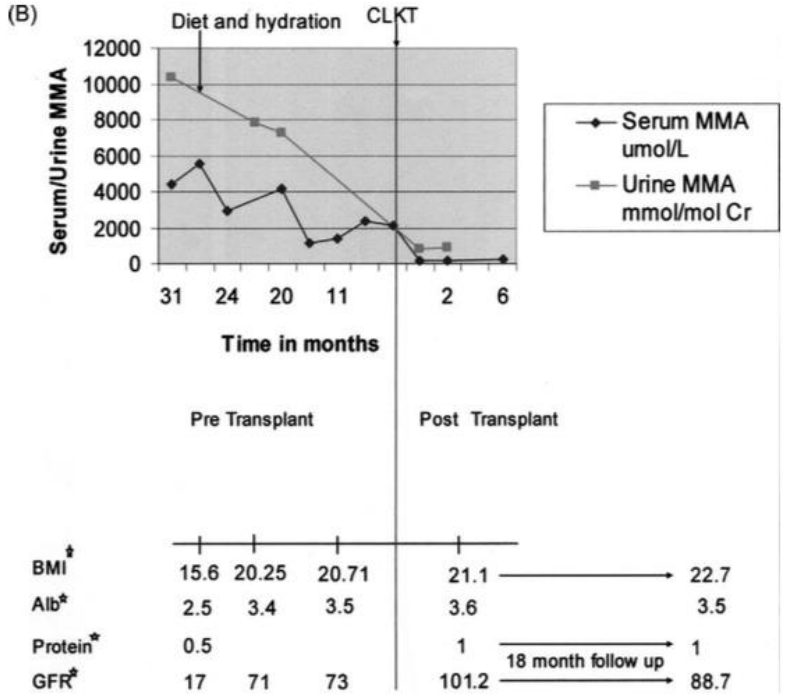


Fig. 1. Change in plasma creatinine (*empty squares*) and methylmalonate (*MMA*) concentrations (*closed triangles*) before and after transplantation.

J Pediatr 132:1043-4, 1998

Combined Liver-Kidney Transplantation in MMA





JIMD 28:517-524, 2005

J Inherit Metab Dis (2014) 37:899–907
DOI 10.1007/s10545-014-9730-7

ORIGINAL ARTICLE

A detailed analysis of methylmalonic acid kinetics during hemodialysis and after combined liver/kidney transplantation in a patient with *mut*^{0} methylmalonic acidemia

Hilary J. Vernon · C. John Sperati · Joshua D. King · Andrea Poretti · Neil R. Miller · Jennifer L. Sloan · Andrew M. Cameron · Donna Myers · Charles P. Venditti · David Valle

Methylmalonic Acid Kinetics

- 28 y *mut*^{0} MMA
- Hemodialysis
  - 54% reduction in plasma MMA
  - Rapid reaccumulation of MMA over 24 h
- Following combined LKT
  - 97% reduction in plasma MMA
- Post-operative
  - Worsening vision (pre-existing optic neuropathy)
  - Seizures
  - Transient, focal leukoencephalopathy

JIMD 37:899-907, 2014

Methylmalonic Acid Kinetics

Table 1 Plasma metabolite measurements during first week of dialysis

	Day 1		Day 2		Day 3			Day 4	Day 5
	Pre-dialysis	2 h post dialysis	Pre-dialysis	2 h post dialysis	Pre-dialysis	2 h post dialysis	4 h post dialysis	22 h post diaysis	52 h post diaysis
Plasma MMA (umol/L)	6,127	3,914	5,818	2,986	4,156	*	37.73	*	61.87
Plasma C3 (umol/L)	57.62	*	51	39.51	51.41	*	37.73	*	61.87
Plasma C4DC (umol/L)	8.66	*	7.25	3.03	4.76	*	3.38	*	9.23

* Indicates sample not available

JIMD 37:899-907, 2014

Methylmalonic Acid Kinetics

Table 2 Plasma and CSF metabolites measured at post operative days (POD) 28, 48, 53, 55, and 57

	Methylmalonate (umol/L)	C3 (umol/L)	C4DC (umol/L)	Glycine (umol/L)	Alanine (umol/L)	Glutamine (umol/L)	Serine (umol/L)
Normal range in CSF	0.14–0.73	*	*	10.0–32.0	24.0–42.0	320–837	28–61
Normal range in plasma	0.11–0.43	<0.92	<0.18	87.0–323.0	136.0–440.0	337.0–673.0	67–171
Average CSF/plasma	1.61	*	*	0.039	0.098	0.828	0.227
CSF (POD 28)	1439	5.97	0.49	5	49	399	16
Plasma (POD 28)	338	26.52	1.08	203	416	567	60
CSF/plasma	4.26	0.23	0.45	0.02	0.12	0.70	0.27
CSF (POD 48)	1274	2.22	0.42	5	51	376	16
Plasma (POD 48)	153	13.95	0.46	191	400	477	71
CSF/plasma	8.33	0.16	0.91	0.03	0.13	0.79	0.23
CSF (POD 55)	565	0.5	0.1	6	36	396	27
Plasma (POD 53)	135	**	**	165	334	541	63
Plasma (POD 57)	104	12.52	0.81	224	301	540	72
CSF/plasma (D55+D57/2)	5.43	0.04	0.12	0.03	0.12	0.73	0.40

JIMD 37:899-907, 2014

THE JOURNAL OF PEDIATRICS • www.jpeds.com

ORIGINAL ARTICLES

Treatment of Methylmalonic Acidemia by Liver or Combined Liver-Kidney Transplantation

Anna-Kaisa Niemi, MD, PhD[1], Irene K. Kim, MD[2], Casey E. Krueger, PhD[3], Tina M. Cowan, PhD[4], Nancy Baugh, MS, RD[5], Rachel Farrell, MS[1,6], Clark A. Bonham, MD[2], Waldo Concepcion, MD[2], Carlos O. Esquivel, MD, PhD[2], and Gregory M. Enns, MB, ChB[1]

J Pediatr 166:1455-61, 2015

Table I. Demographic, diagnostic, and surgical data of patients (n = 14) who received LKT or LT at Lucile Packard Children's Hospital at Stanford between December 1997 and May 2012

Patient	Sex	Time of diagnosis	Identified by NBS	Diagnosis*	Age at Tx	Procedure	Graft	Long-term immunosuppression	Complications
1	M	Neonatal	no	Non-B12-responsive clinically	10 y 9 mo	LKT	Whole	Prednisone, tacrolimus	
2	M	Neonatal	no	fibroblast assay, mut^0	20 y 8 mo	LKT, bilateral nephrectomy	Whole	Prednisone, tacrolimus, sirolimus	1. Re-exploration, bleeding 2. Post-transplant diabetes mellitus and hypertension attributed to immunosuppressive regimen
3	M	Neonatal	no	Fibroblast assay, mut^0	5 y 11 mo	LKT, bilateral nephrectomy, splenectomy	Whole	Prednisone, tacrolimus, azathioprine	1. Spontaneous splenic rupture → splenectomy 2. Re-exploration, bleeding 3. Seizure POD12 (high tacrolimus level)
4	M	Neonatal	no	†	11 y 2 mo	LKT, right nephrectomy, splenectomy	Whole	Sirolimus	
5	F	Neonatal	yes	c.682C>T (p.R278X), c.1106 G>A (p.R369H)	3 y 3 mo	LT	Whole	Tacrolimus, mycophenolate	Mild acute rejection 4 weeks post-transplantation, received steroids
6‡	F	3 mo	no	c.322C>T (p.R108C)	15 y 4 mo	LKT	Whole	Prednisone, tacrolimus, mycophenolate	
7	F	Neonatal	yes	c.682C>T (p.R228X), c.581C>T (p.P194L)	11 mo	LT	Whole	Tacrolimus	
8	F	9 mo	no	c.572C>A (p.A191E)	17 y 6 mo	LKT, splenectomy	Whole	Tacrolimus, mycophenolate	Re-exploration, drainage of subphrenic abscess
9	M	Neonatal	no	c.349G>T (p.E117X), c.1038_1040 delTCT	8 y 10 mo	LKT	Whole	Tacrolimus, mycophenolate	Acute rejection 3 weeks post-transplant
10	M	2 y	no	Fibroblast assay, mut^0	16 y 1 mo	LKT	Whole	Prednisone, tacrolimus, mycophenolate	
11	F	Neonatal	yes	c.682C>T (p.R228X)	10 mo	LT	1. Whole 2. Whole	Tacrolimus	1st transplantation: Hepatic artery thrombosis POD5 → re-transplantation. 2nd transplantation: No complications
12§	F	Neonatal	yes	c.1399C>T (p.R467X)	1 y 1 mo	LT	Seg 2-4	Tacrolimus	Mild acute rejection POD10, received dose of steroids
13§	F	Neonatal	yes	c.1399C>T (p.R467X)	1 y 2 mo	LT	Seg 2-4	Tacrolimus	
14	F	Neonatal	yes	c.682C>T (c.R228X) p.A732WFSX3	1 y 8 mo	LT	Whole	Tacrolimus	

J Pediatr 166:1455-61, 2015

LT or LKT for MMA

- Mean age for transplantation 8.75 ± 7 years (0.8-20.7 y)
- LKT 13.3 ± 4.9 years (5.9-20.7 y)
  - 88% underwent pre-operative hemodialysis
- LT 1.5 ± 0.9 years (0.8-3.3 y)

J Pediatr 166:1455-61, 2015

Postoperative period

- Mean follow-up 3.3y

Patient survival	**100%**
Liver allograft survival	**93%** (hepatic artery thrombosis, n=1)
Kidney allograft survival	**100%**

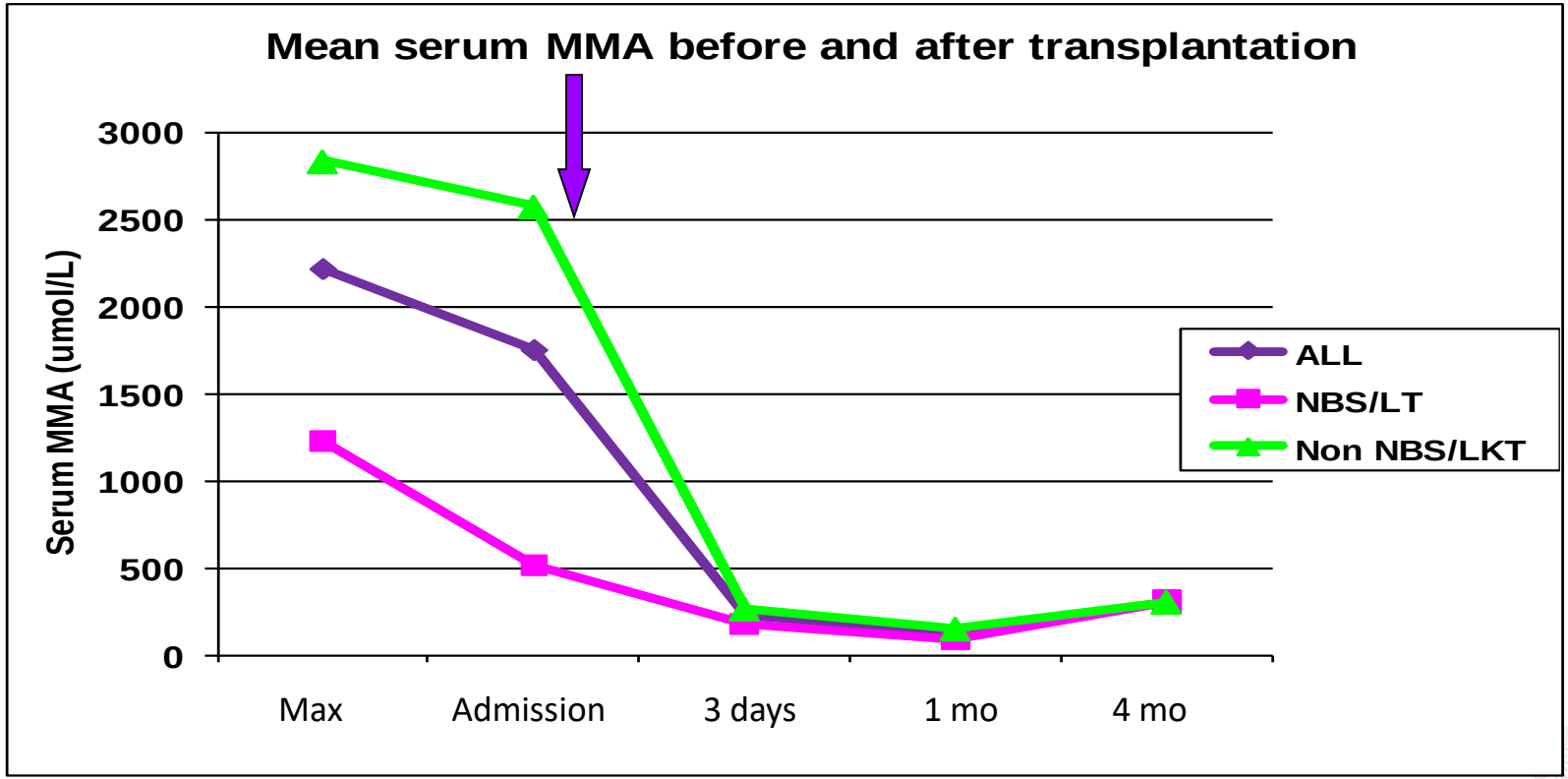
J Pediatr 166:1455-61, 2015

Table II. Mean maximum serum MMA (μmol/L ± 1 SD, normal <0.3) and range, and mean serum MMA at the time of admission for transplantation, and 3 days, 1 month, and 4 months after transplantation in all patients, in patients with LKT, and in those who received LT

Patient group	Mean maximum serum MMA in μmol/L (range)	Serum MMA in μmol/L on admission for transplantation	Serum MMA in μmol/L three days post-transplantation	Serum MMA in μmol/L 1 mo post-transplantation	Serum MMA in μmol/L 4 mo post-transplantation
All patients (n = 14)	2107 ± 1427 (210-5452)	1647 ± 1492 (99-4420)	210 ± 154 (73-622) *(87% decrease)*	125 ± 94 (27-343) *(93% decrease)*	305 ± 108 (143-600) *(83% decrease)*
LKT (n = 8)	2840 ± 1365 (1450-5452)	2580 ± 1451 (1030-4420)	264 ± 209 (99-622) *(90% decrease)*	149 ± 119 (38-343) *(94% decrease)*	303 ± 196 (143-600) *(88% decrease)*
LT (n = 6)	1129 ± 821 (507-2330)	529 ± 259 (99-732)	164 ± 83 (73-261) *(69% decrease)*	97 ± 48 (27-153) *(82% decrease)*	307 ± 61 (236-383) *(51% decrease)*

Decrease in MMA level (%) from the level on admission to transplantation in italics.

J Pediatr 166:1455-61, 2015


Mean serum MMA before and after transplantation
Serum MMA (umol/L)
3000
2500
2000
1500
1000
500
0
Max
Admission
3 days
1 mo
4 mo
ALL
NBS/LT
Non NBS/LKT

Other clinical outcomes

- No hyperammonemia or metabolic acidosis

- Renal function normal on those with LT only
  - mean follow-up 1.1 years

Neurological outcomes

14

LT 6

Pretransplant:

1/6 global developmental delay

3/6 mild developmental delay

2/6 mild motor delay, otherwise age appropriate

Post:

3/5 gained motor skills

No neurological deterioration

LKT 8

Pretransplant:

4/8 global developmental delay

2/8 mild developmental delay

Post:

Maintained previous level

No neurological deterioration

Liver or combined LKT for MMA

Table IV. Average daily protein intake of total protein, natural protein, and medical food protein (and range) at the time of admission for transplantation and at the time of last available nutritional assessment after transplantation among all patients, those who received LKT, and those who received LT

	On admission for transplantation			At the time of last nutritional assessment		
Patient group	Total protein	Natural/whole protein	Protein from medical foods	Total protein	Natural/whole protein	Protein from medical foods
All patients (n = 14)	1.6 g/kg/d (0.9-2.8)	0.9 g/kg/d (0.3-1.9) (n = 13)*	0.8 g/kg/d (0.4-1.7) (n = 13)*	1.6 g/kg/d (0.6-2.6)	0.9 g/kg/d (0.6-1.8) (n = 13)*	0.7 g/kg/d (0-1.4) (n = 13)*
LKT (n = 8)	1.2 g/kg/d (0.9-1.7)	0.6 g/kg/d (0.3-1.0) (n = 7)*	0.7 g/kg/d (0.4-1.2) (n = 7)*	1.3 g/kg/d (0.6-1.9)	0.7 g/kg/d (0.6-1.0) (n = 7)*	0.6 g/kg/d (0-0.9) (n = 7)*
LT (n = 6)	2.1 g/kg/d (1.3-2.8)	1.1 g/kg/d (0.8-1.9)	1.0 g/kg/d (0.5-1.7)	2.0 g/kg/d (1.5-2.6)	1.2 g/kg/d (0.9-1.8)	0.8 g/kg/d (0.6-1.4)

*Documentation of exact natural vs medical food intake is not available on 1 patient who received LKT (patient 1).

J Pediatr 166:1455-61, 2015

Table V. Neurodevelopmental and neurocognitive functioning*

Case	Transplant age	Pre-transplant developmental functioning (age)	Post-transplant developmental functioning (age)
1	10 y 9 mo	PIQ = 55, 1st %ile; VIQ = 87, 19th %ile (9 y 5 mo)	PIQ = 82, 12th %ile; VIQ = 89, 23rd %ile (12 y 2 mo)
2	20 y 8 mo	Assessment not available for review; mild delays, B and C grades in high school, completed 1 y college	No decline, day program, lives with family
3	5 y 11 mo	Assessment not available for review; GDD dx in records	Increased fine motor skills, attention, and mood by parent report
4	11 y 2 mo	Assessment not available for review; GDD dx in records	Increased motor skills and energy level by parent report
5	3 y 3 mo	Cognitive = 55, <1st %ile; Language = 50, <1st %ile; Motor = 55, <1st %ile (3 y 2 mo)	No changes noted
6	15 y 4 mo	PIQ = 92, 30th %ile; VIQ = 92, 30th %ile (14 y 11 mo)	Attended college, no concerns or changes noted
7	11 mo	Assessment not available for review; mild delays reported in dev. milestones, sits and crawls (11 mo)	Walks and jumps; receives ST, PT, OT (2 y 9 mo)
8	17 y 6 mo	PIQ = 53, <1st %ile; VIQ = 61, <1st %ile (17 y 6 mo)	No change, Adult school program, lives at home (19 y)
9	8 y 10 mo	GDD, cognitive function estimated at 12-24 mo (8 y 1 mo)	GDD, no changes
10	16 y 1 mo	All scores within the average range (15 y 7 mo)	No concerns or changes
11	10 mo	Cognitive = 9 mo; Motor = 5 mo (7 mo)	Gross motor skills increased by clinician report; energy and social skills increased by parent report
12	1 y 1 mo	Cognitive = 62, 1st %ile; Language = 68, 2nd %ile (7 mo)	Cognitive = 67, 1st %ile; Language = 80, 9th %ile (17 mo)
13	1 y 2 mo	Cognitive = 62, 1st %ile; Language = 62, 1st %ile (7 mo)	Cognitive = 65, 1st %ile; Language = 82, 12th %ile (17 mo)
14	1 y 8 mo	Cognitive = 85, 16th %ile; Language = 109, 73rd %ile; Motor = 67, 1st %ile (16 mo)	Not assessed

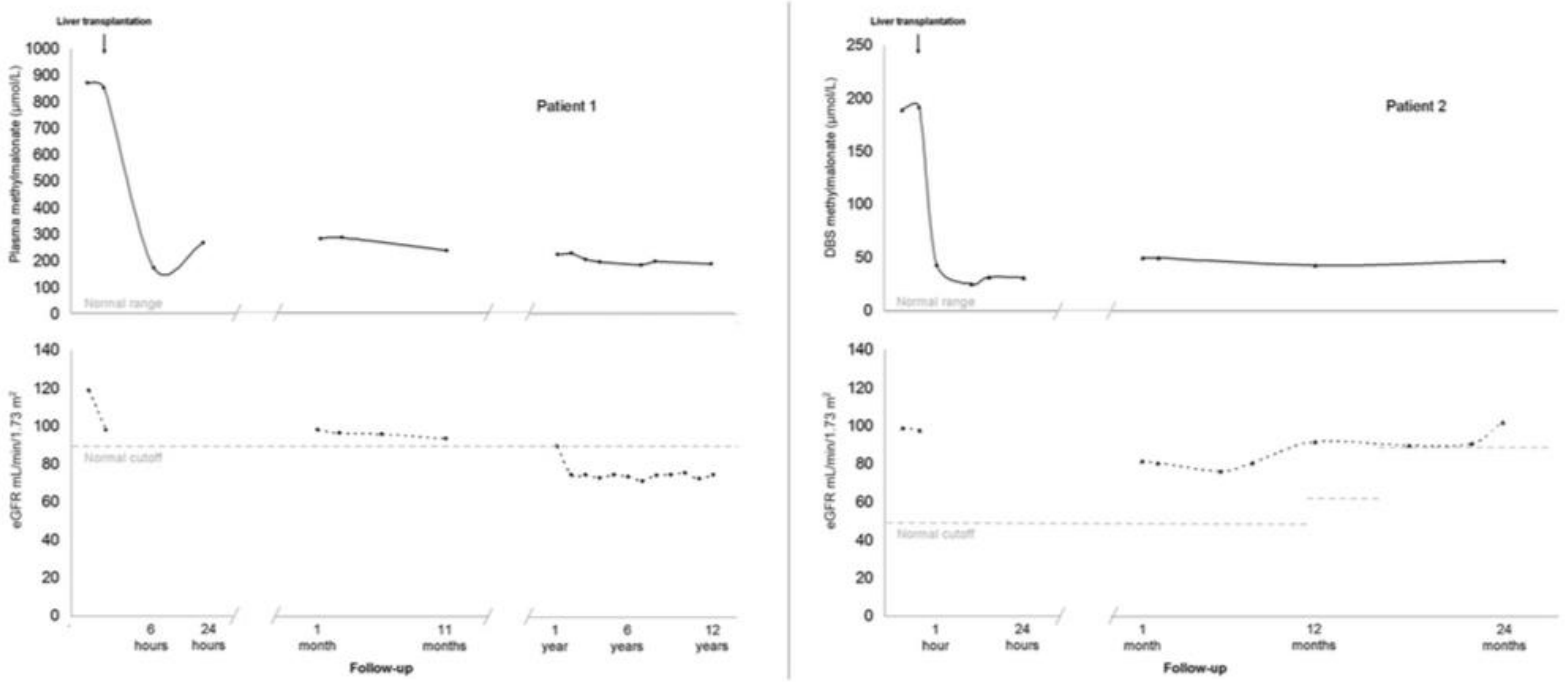
J Pediatr 166:1455-61, 2015

Early Liver Transplantation for MMA

- 2 patients with severe neonatal disease
- LT at 3 y and 9 m
  - No preoperative dialysis
  - Before significant neurological or renal morbidity
- Follow-up 12 y and 2 y
  - Mild renal impairment
- Normal tolerance to fasting catabolism
- Increased protein tolerance
  - 0.8 g/kg/d to 1.5-1.8 g/kg/d

Pediatrics 136:e252-6, 2015

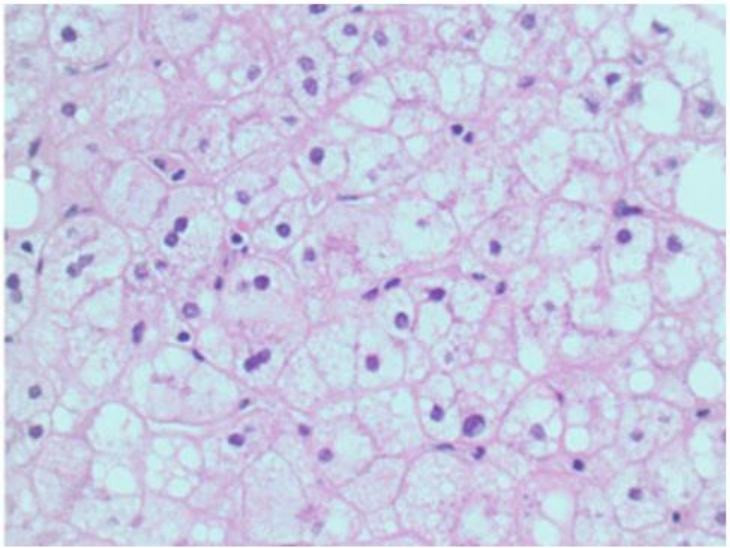
Early Liver Transplantation for MMA



Pediatrics 136:e252-6, 2015

Domino Liver Transplantation in MMA

- 28 y *mut*^{0} MMA
  - Frequent episodes of metabolic crises after age 21 y
  - Increasing neurological disability
- Domino recipient
  - 61 y
  - primary sclerosing cholangitis, biliary cirrhosis



MMA explant: 25-30% macrosteatosis

JIMD Rep 2015

Pre-operative Dialysis for MMA

TABLE 3. *Peri- and postoperative plasma methylmalonic academia levels (normal range, 0.35 ± 0.22 nmol/mL)*

Patient number	Admission to hospital	Just before dialysis	Just after dialysis	Just before surgery	Anhepatic time	After reperfusion	Just after operation	POD 1	POD 7	POD 30
1	268.0	92.2	58.5	70.8	62.0	64.0	47.8	37.5	37.7	99.4
2	47.0	40.0	33.6	50.7	44.7	48.2	48.8	60.9	65.1	59.2
3	143.0	31.7	32.9	25.9	32.9	38.1	39.6			36.4
4	39.0	14.4	13.4	12.3	19.0	26.8	38.4	37.0	28.1	29.3
5	375.0	137.3	87.1	86.4	114.6	106.4	100.2	190.5	109.6	87.8
6	1970.0	357.0	140.5	251.0	199.0	181.5	146.4	329.0	176.9	232.0
7	166.0	107.3	38.7	38.7	46.3	35.2	31.5	44.4	22.7	13.8
8	278.0	NA	NA	342.0	298.0	337.0	251.0	175.8	77.7	59.6
9	702.0	NA	NA	302.0	303.0	230.0	191.0	117.5	147.9	124.4
10	255.0	NA	NA	160.0	147.0	119.0	88.0	129.0	33.8	8.5

NA, not applicable; POD, post operative day.

Therap Apher Dial 15:488-92, 2011

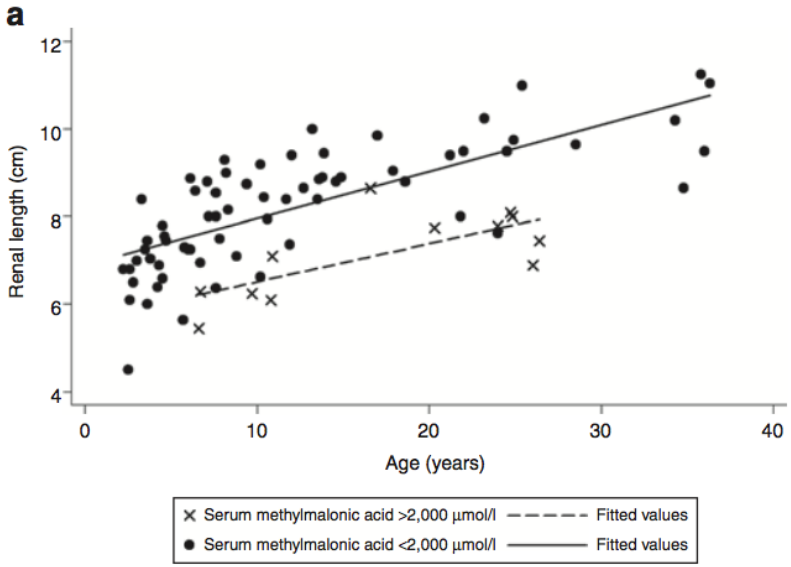
Cystatin C

- 13 Kda endogenous cystein proteinase inhibitor
- Major role in intracellular catabolism of peptides
- Produced by all nucleated cells, synthesized at a relatively constant rate, released into plasma
- 99% filtered by glomeruli
- Elevated urine cystatin C may indicate tubular epithelial damage

Curr Op Crit Care 16:533-9, 2010

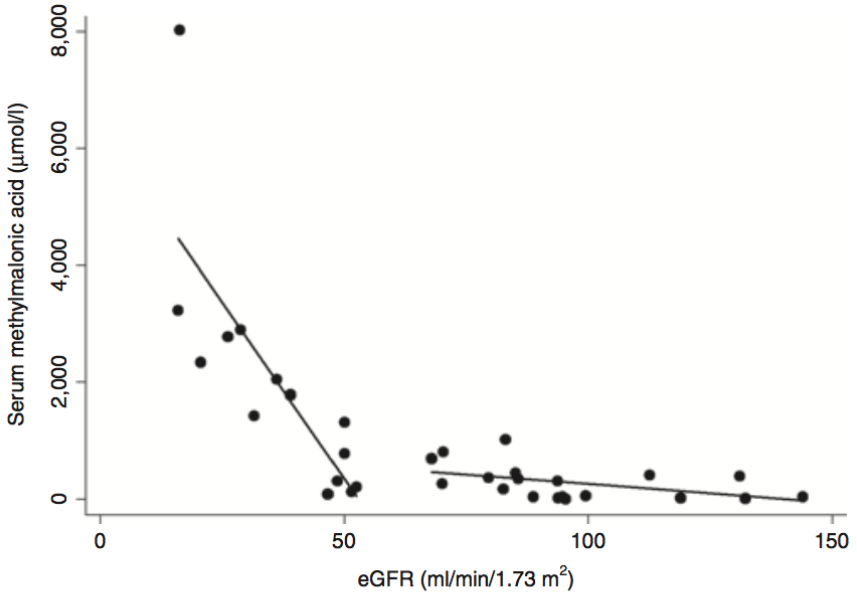
Renal Growth in MMA

- MMA (n=50)
- 2004-2011
- Renal length decreased over time v. controls
- Cystatin C and [MMA] highly correlated with decreased renal function and smaller kidneys



Genet Med 15:990-6, 2013

Renal Disease in MMA



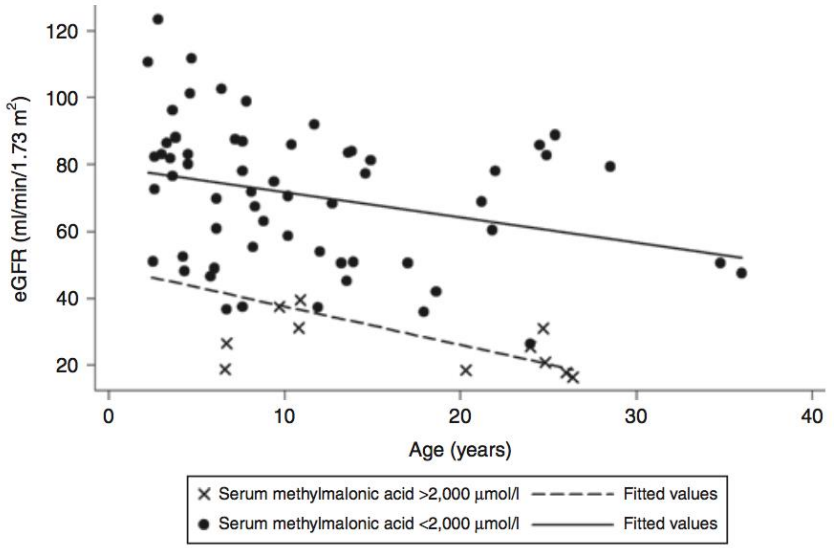


Figure 3 Serum methylmalonic acid (MMA_S) for all enzymatic subtypes versus estimated glomerular filtration rate (eGFR) creatinine–cystatin C.

Genet Med 15:990-6, 2013

Pediatr Nephrol (2001) 16:848–851

BRIEF REPORT

R. Lubrano · P. Scoppi · P. Barsotti · E. Travasso
S. Scateni · S. Cristaldi · M.A. Castello

Kidney transplantation in a girl with methylmalonic acidemia and end stage renal failure

	PreTx	PostTx							
Months		2	6	12	18	24	30	36	42	48
UMM acid	8.16	0	0	3.2	3.5	5.1	7.2	9.4	4.1	4.6
UMM acid/creatinine ratio	14.84	0	0	0.25	0.39	0.47	0.85	1.36	0.48	0.5
UMM acid/BUN ratio	0.028	0	0	0.0014	0.0022	0.0017	0.0031	0.0093	0.0030	0.0045
Serum creatinine (mg/dl)		1	0.9	0.8	1	0.9	0.7	1.1	1	1
BUN (mg/dl)	57	10	11	10	8	9	9	11	13	14
$GFR_{(creatinine)}$ (ml/min/1.73 m^2)		50	65	75	85	80	75	58	59	67
$GFR_{(^{99}TcDTPA)}$ (ml/min/1.73 m^2)				65		64		63		62
Proteinuria (mg/24 h)		90	85	111	76	90	108	85	50	37
RTP (%)		92.84	92.5	92.42	91.25	90.99	82.28	69.35	88.76	97
FENa (%)		0.55	1.05	1.10	0.61	1.61	1.32	1.07	1.12	0.27

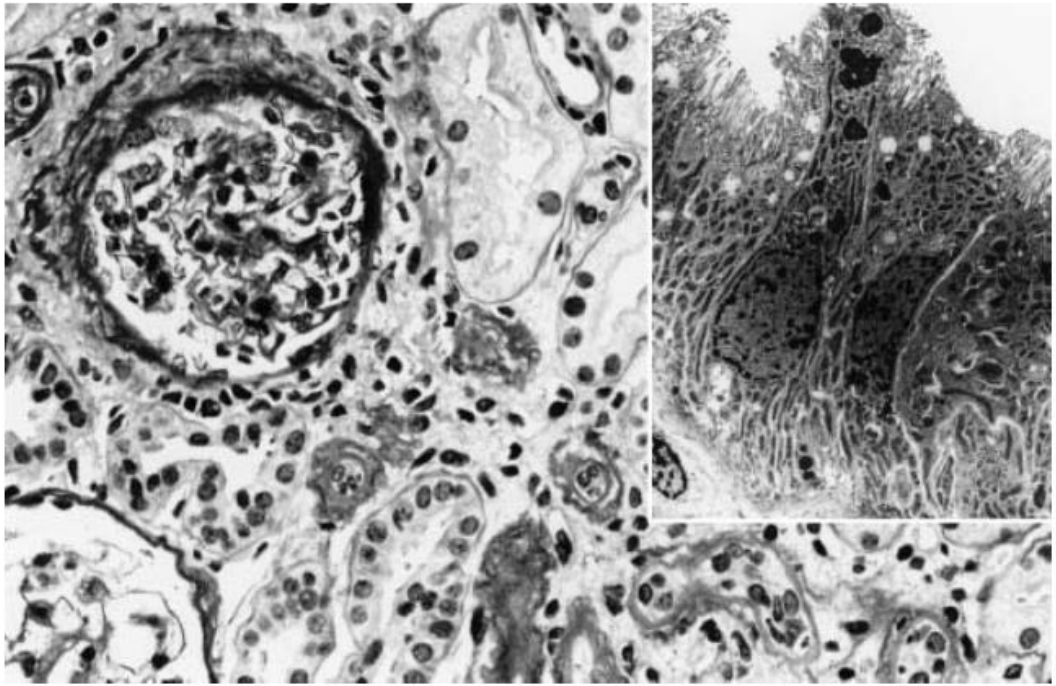
Kidney Transplantation in MMA

- 17 y female – vitamin B_{12}–unresponsive MMA
- Growth retardation at 3 m
- Frequent episodes of vomiting at 4 m
- Hyperammonemic coma at 9 m
- Hemodialysis at 16.5 y
  - Concentric hypertrophic cardiomyopathy

Pediatr Nephrol 16:848-51, 2001

Kidney Transplantation in MMA

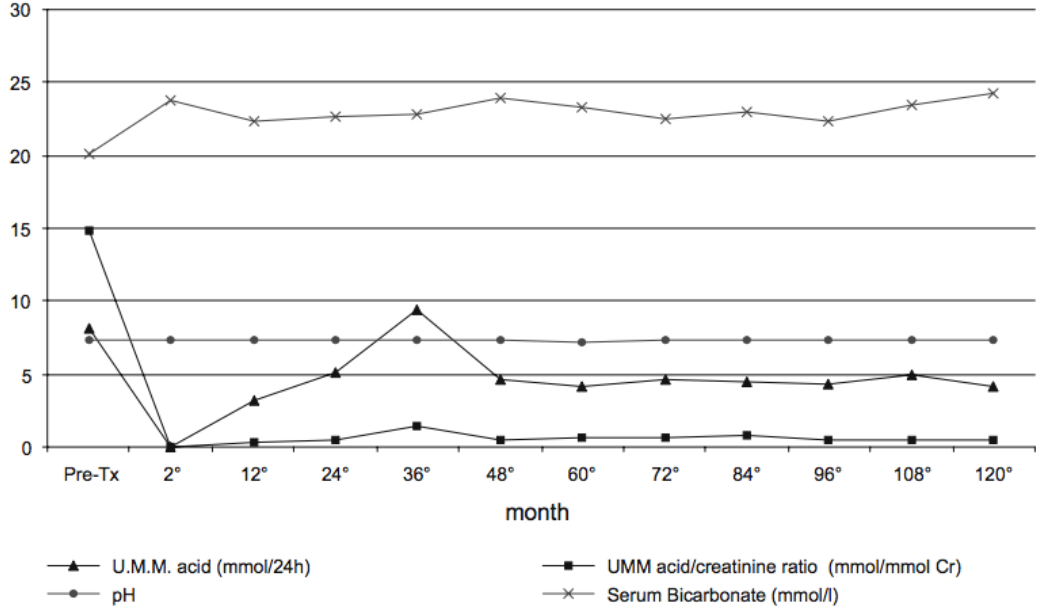
Fig. 1 Renal biopsy showed focal chronic parenchymal damage with interstitial and periglomerular fibrosis, focal tubular atrophy, and infiltrating mononuclear cells; epithelial cells of histologically normal tubuli do not reveal relevant substructural alterations. *Inset* An EM picture from the proximal tubular epithelium. PAS, ×210 (*inset* UrPb, ×1640)



Pediatr Nephrol 16:848-51, 2001

Kidney Transplantation in MMA

- 10 y follow-up



Pediatr Nephrol 22:1209-14, 2007

Kidney Transplantation in MMA

- 16 y 5 m follow up
  - Normal renal function
  - Uneventful pregnancy
- DNA analysis
  - Homozygous c.586C>T (p.Arg196Term) in exon 4
  - MMA cblA type
  - Fibroblasts responsive to vitamin B_{12}
  - Milder clinical course than *mut*^{0} type

Table 1 Results of in vivo and in vitro exposure of patient's fibroblasts to vitamin B_{12}

Exposure to vitamin B_{12}	Analysis	−OHCbl	+OHCbl
In vitro	[1-^{14}C] Propionate incorporation rate in fibroblasts (nmol/10 h/mg protein)	0.08	0.33
In vivo	Urinary MMA (mmol/mol of creatinine)	213.0	76.8
	Serum MMA (μmol/L)	43.3	13.5

MMA, Methylmalonic acidemia

Pediatr Nephrol 28:2067-8, 2013

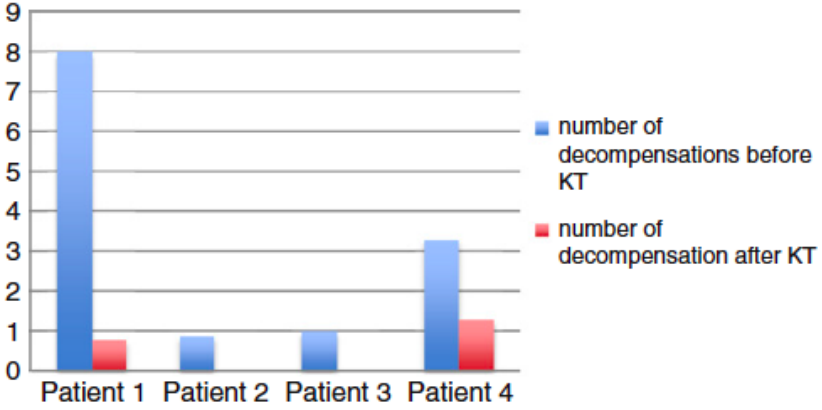
Kidney Transplantation in MMA

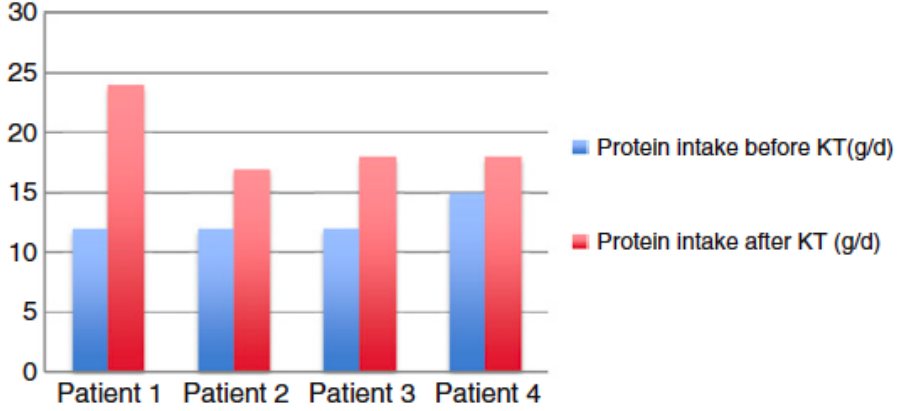
- 4 patients with *mut*^{0} MMA (5 to 10 y)
- End-stage renal disease (n=2); stage III (n=1); normal (n=1)
- No further metabolic decompensations
- Protein intake increased from 0.6 g/kg/d to 0.8 g/kg/d
- 1 death secondary to hepatoblastoma
- 2 neurologically stable; 1 transient improvement of choreoathetosis

Mol Genet Metab 110:106-110, 2013

Kidney Transplantation in MMA

a

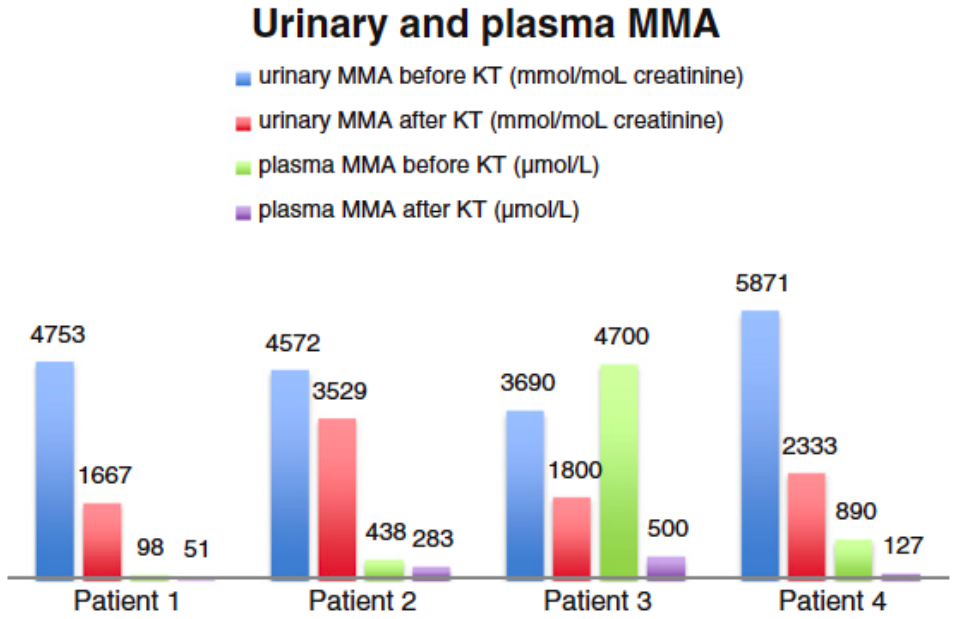




Mol Genet Metab 110:106-110, 2013

Kidney Transplantation in MMA

b



KT: Kidney tansplantation

Mol Genet Metab 110:106-110, 2013

Kidney Transplantation in MMA

- 2/4 with chronic Tacrolimus nephrotoxicity
- 3/4 had stage II chronic kidney disease at follow up (22-55 months)
- 1/4 had normal renal function at 30 months post-transplantation
- 1/4 developed neurological regression and an extrapyramidal syndrome at 18 months; died at 20 months after developing hepatoblastoma

Mol Genet Metab 110:106-110, 2013

Kidney Transplantation in MMA

- 12 y with *MMAB*

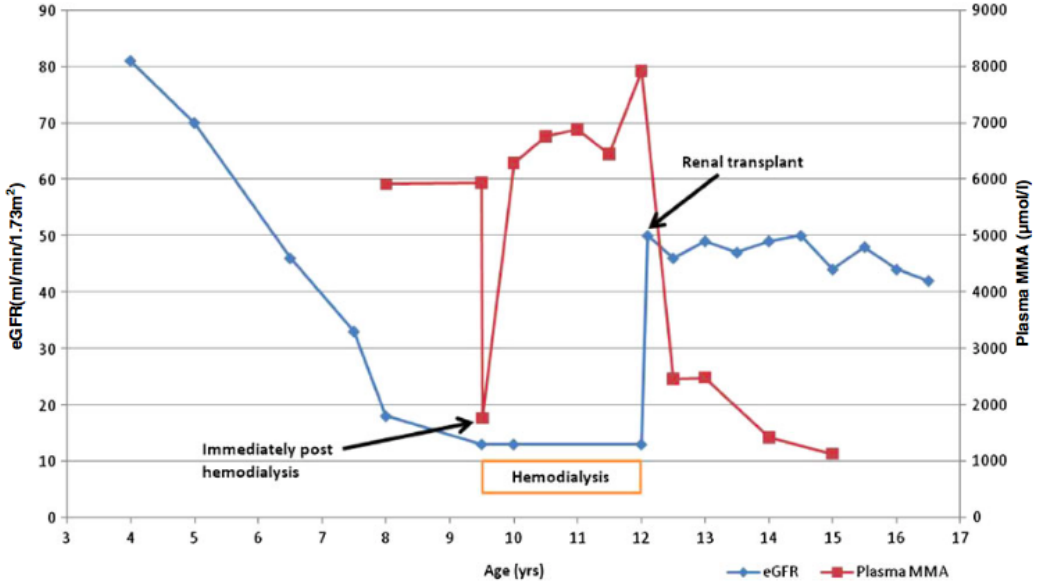
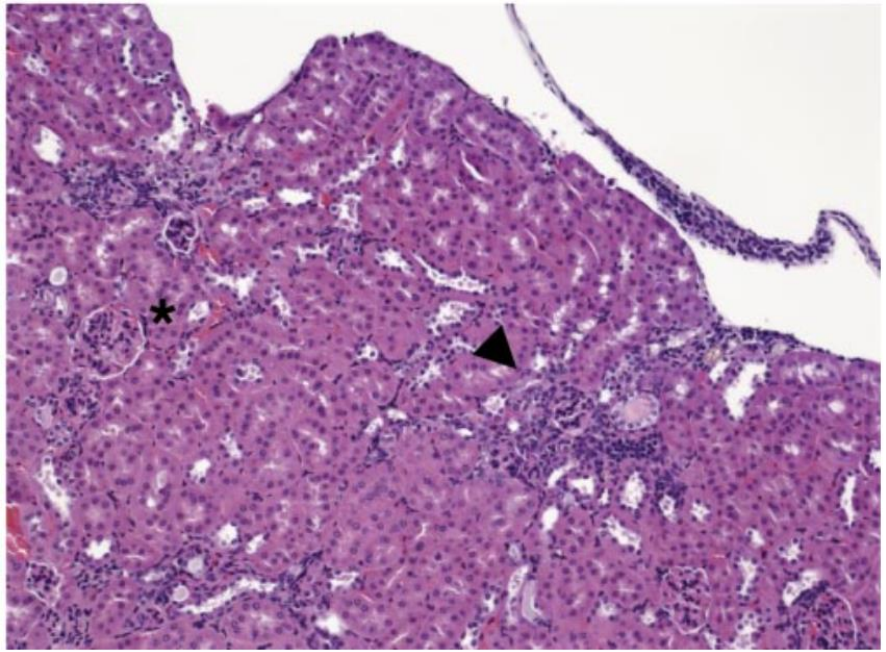
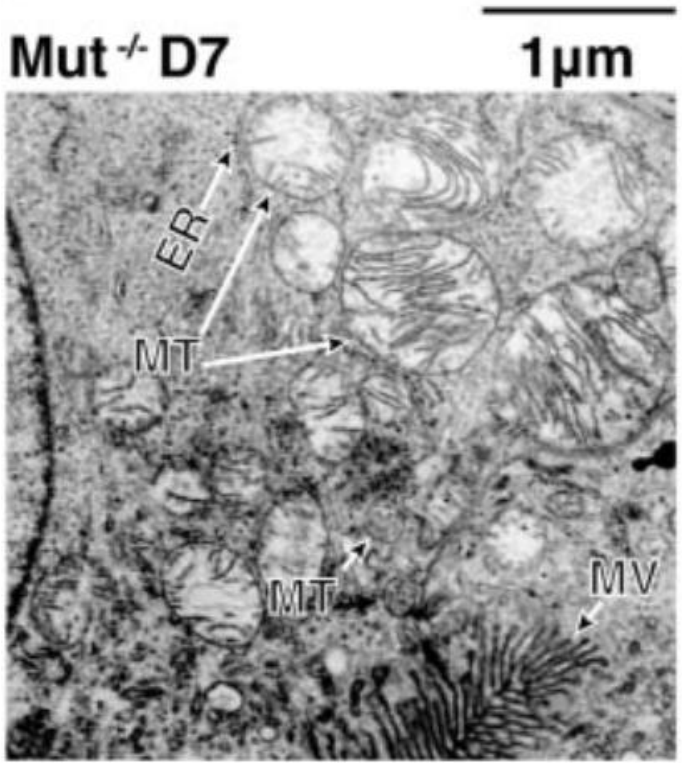


Fig. 2 Serum methylmalonate concentrations and eGFR pre-dialysis, during hemodialysis and after renal transplantation

JIMD 34:695-700, 2011

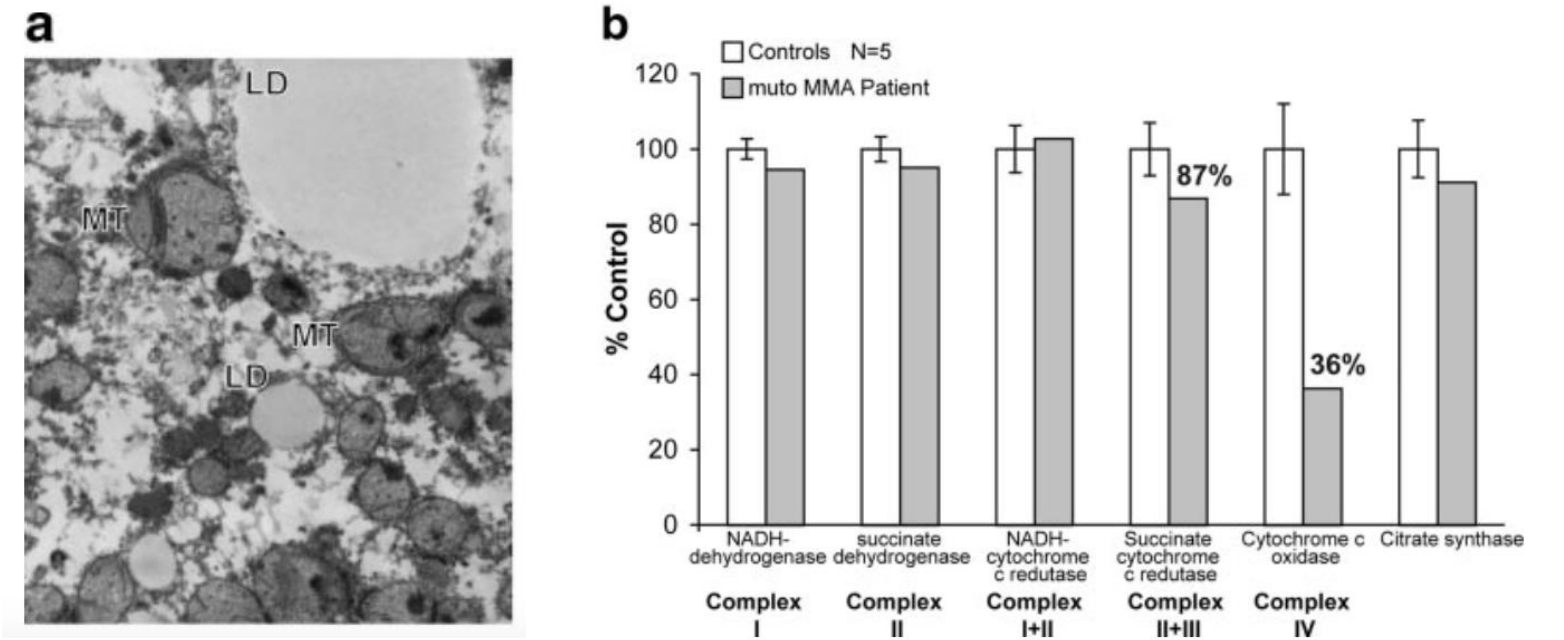
Abnormal Kidney Mitochondria in MMA





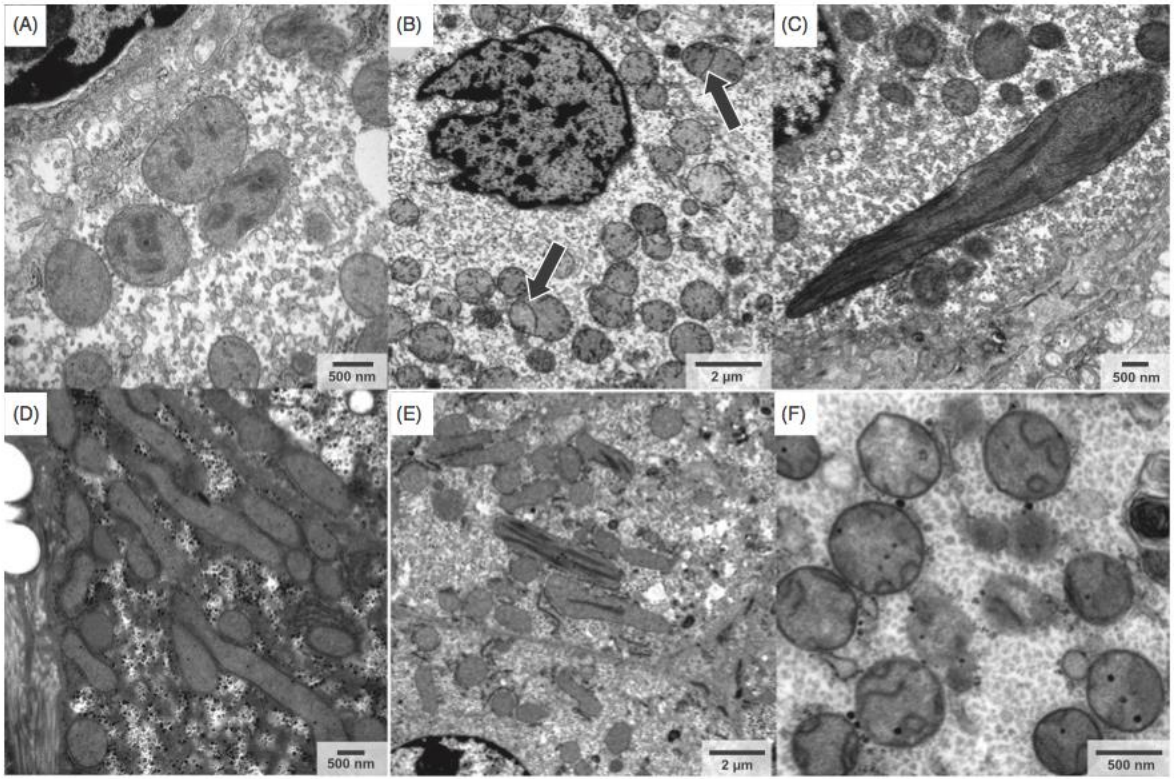
FASEB J 23:1252-61, 2009

Abnormal Liver Mitochondria in MMA



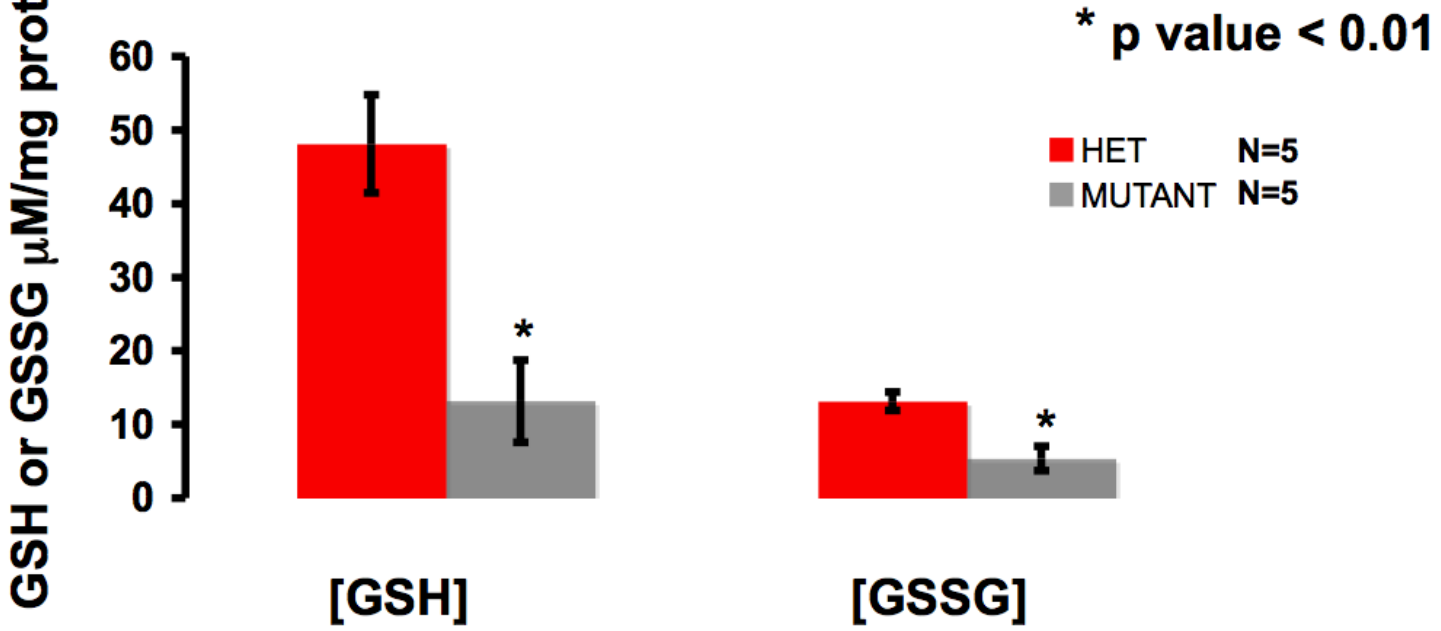
FASEB J 23:1252-61, 2009

Abnormal Liver Mitochondria in MMA



Ultrastruct Pathol 38:309-14, 2014

GLUTATHIONE (GSH, GSSG) IN MURINE LIVER EXTRACTS



Courtesy Tina Cowan

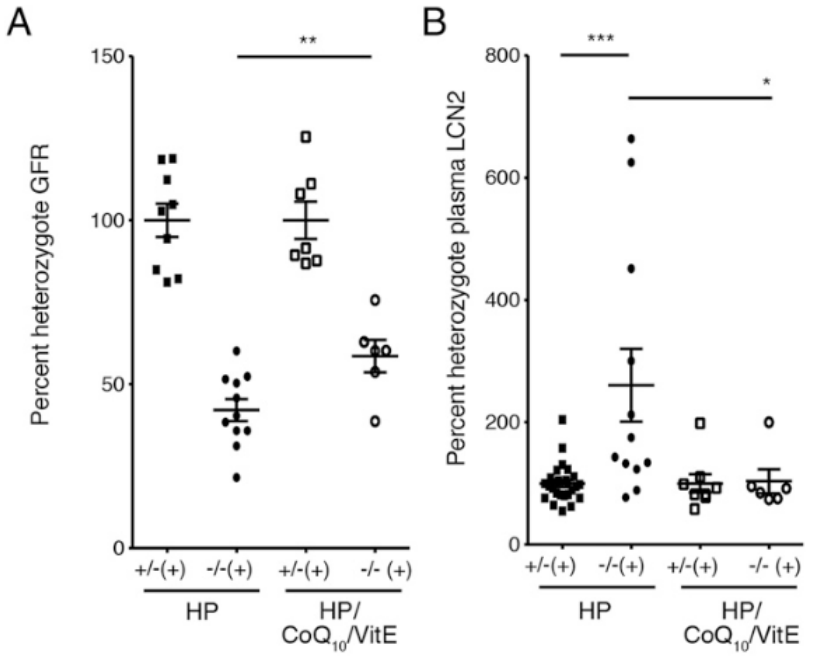
Glutathione in Organic Acidemias

- Glutathione levels can be low in organic acidemias, especially in times of crisis
- Possible role for antioxidants
- Careful attention to protein status
- Glutathione levels post-transplantation?

J Pediatr 129:445-8, 1996
Acta Paediatr 101:e505-8, 2012

Mitochondrial Dysfunction in MMA

- *Mut*$^{-/-}$;Tg$^{\text{INS-Alb-}Mut}$ mice
- Rescued from neonatal lethality
- Develop chronic tubulointerstitial nephritis
- Lipocalin-2 a biomarker of kidney disease
- Antioxidant therapy ameliorated the renal disease of MMA



PNAS 110:13552-7, 2013

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Children undergoing liver transplantation for treatment of inherited metabolic diseases are prone to higher oxidative stress, complement activity and transforming growth factor-β1

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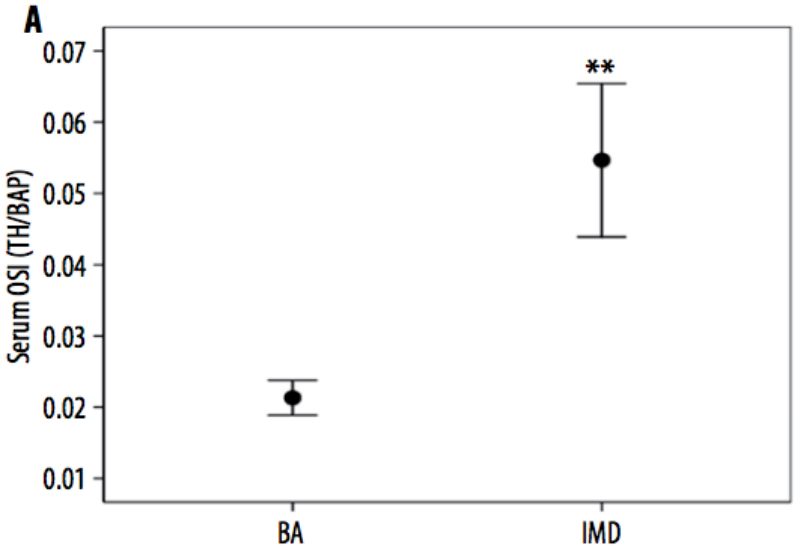
Authors' Contribution:
A Study Design
B Data Collection
C Statistical Analysis
D Data Interpretation
E Manuscript Preparation
F Literature Search
G Funds Collection

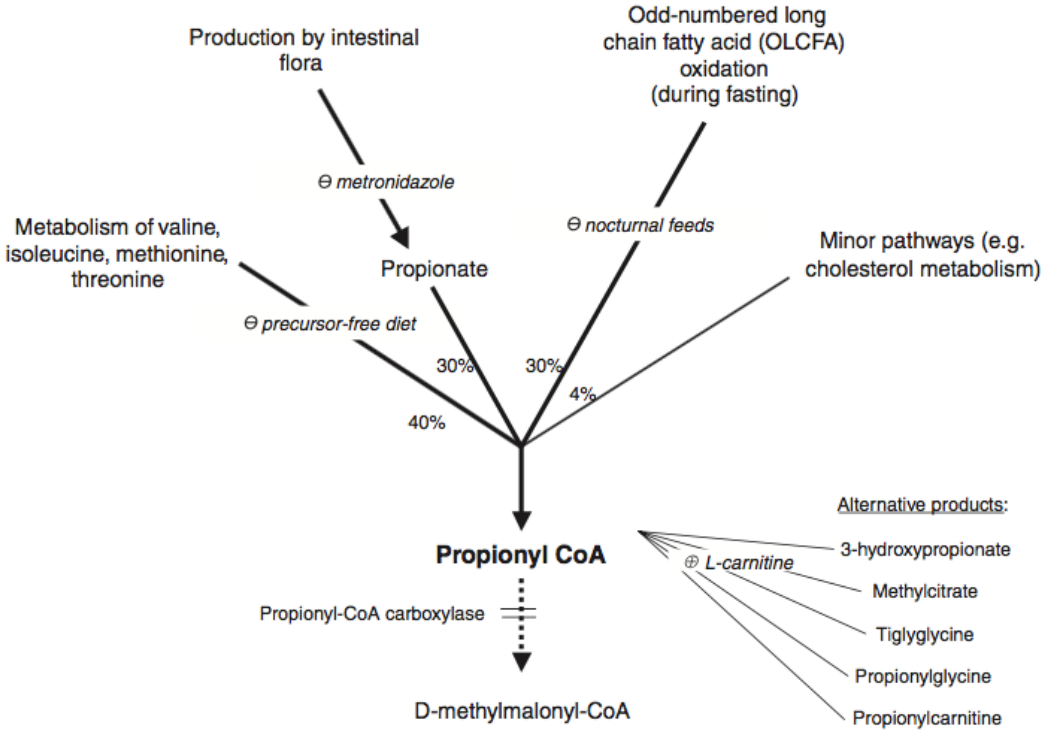
Oxidative Stress in LT for IEM

- Total hydroperoxide:biologic antioxidant potential ratio (TH/BAP)
- Biliary atresia (n=10)
- “Inherited metabolic disease” (n=6)
  - MMA, PA, arginase deficiency, GSD1a, tyrosinemia (n=2)



Ann Transplant 18:63-8, 2013

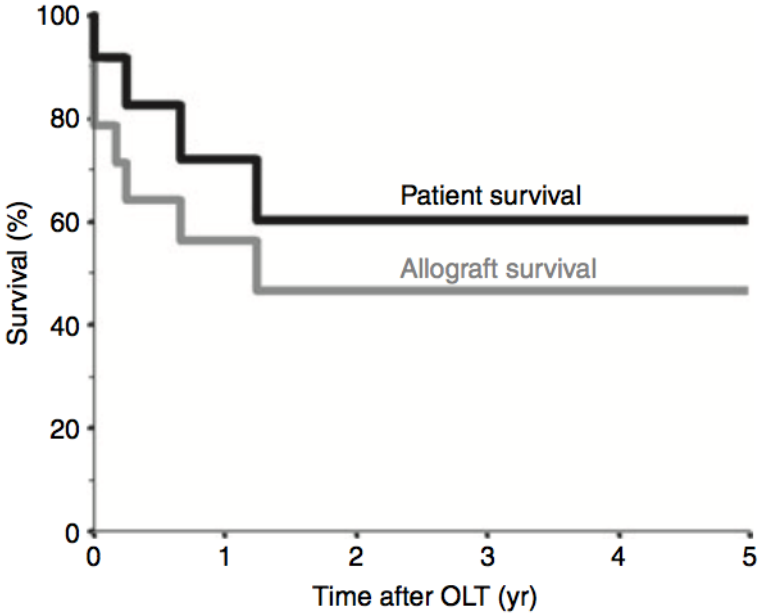
Liver Transplantation in PA



Pediatr Transplant 10:773-81, 2006

Liver Transplantation in PA

- 12 patients, 14 OTL
- 72% 1-year survival, 56% 5-year survival
- 56% 1- year allograft survival, 47% 5-year allograft survival
- Stabilization and improvement of neurological function



Pediatr Transplant 10:773-81, 2006

Liver Transplantation in PA

- 12 patients, 17 OTL
- 3/12 dilated cardiomyopathy
- 6/12 had renal dysfunction pre-transplant
- Graft survival 60% at 5 years
- All patients had renal dysfunction post transplant
- Improved quality of life
- Stabilization of neurological status
- No metabolic decompensations
- Major reduction of propionate metabolites

Am J Transplant 15:786-91, 2015

Liver Transplantation in PA

- Mortality 58%
- 3/12 primary graft non-function
- 3/12 died from heart failure
- All with cardiomyopathy had normalized heart function
- 4/12 ARDS
- 6/12 hepatic artery thrombosis
- 1/12 acute encephalopathy – recovered after stopping Tacrolimus

Am J Transplant 15:786-91, 2015

TABLE 1. Liver transplantation for Propionic Acidemia (N = 20)

	References	Age of onset (age of LT, y)	PCC activity	Type of graft	Main indication	Protein restriction after LT, $g \cdot kg^{-1} \cdot day^{-1}$	Mean follow-up, y	Overall outcome
1	Murphy et al (4)	(26 m)	—	—	—	—	—	Retransplanted; died of heart failure 3 days later.
2	Saudubray et al (9)	16 d (7)	—	LDLT	PMC	None	–	Boy: chronic hyperammonaemia and neurologic sequelae; died 15 mo after LT
3		3d (9)		LDLT		None	5	Girl: no specific complications
4	Rela et al (10)	21 d (1.8)	—	ALT	PMC	None	15	
5		0 d (1,1)		LDLT (LR)	FH		11.4	No further metabolic decompensation
6	Vara et al (3)	3 d (0.8)		LDLT	FH		7.3	Development status unchanged after LT
7		5 d (7)		LDLT	PMC		4.9	
8		3 d (1.1)		LDLT	Elective		2.2	
9	Yorifuji et al (11)	Neo (2)	2%	LDLT (LR)	PMC	2	3.9	
10		Neo (5)	—	LDLT (LR)	PMC	1.8	1.4	Reduced metabolic decompensations

J Pediatr Gastroenterol Nutr 64:e73-6, 2017

TABLE 1. Liver transplantation for Propionic Acidemia (N = 20)

	References	Age of onset (age of LT, y)	PCC activity	Type of graft	Main indication	Protein restriction after LT, $g \cdot kg^{-1} \cdot day^{-1}$	Mean follow-up, y	Overall outcome
11		Neo (1 y)	—	LDLT (LR)	PMC	1.5	0.7	
12	Kayler et al (12)	Neo (3)		cadaveric	—	—	—	Died 3 mo after LT
13	Barshes et al (2)	2 d (1.25)	<5%	Cadaveric	PMC	—	3.7	Development improvement
14		Neo (2)		cadaveric	PMC		0.5	Persistent development delays
15	Romano et al (7)	10 m (9 y)	0.001*	Cadaveric	PMC + CMP	None	13	No further metabolic decompensation Regression of CMP within 1 year after LT
16		3 d (6.5 y)	0.015*	Cadaveric	PMC + CMP	None	0.5	
17	Kasahara et al (8)	3 d (7 m)	<1%	LDLT (LR)	PMC	2	1.7	Neurologic improvement
18	Nagao et al (13)	7d (2y)	0.0*	LDLT (LR)	PMC		—	No further metabolic decompensation
19		44 d (2 y 2 m)	0.0*	LDLT (LR)	PMC		—	Normal cardiac function and development
20	Ryu et al (5)	7d (22 m)	—	LDLT	PMC	—	—	Death on D4: hepatic failure and severe metabolic acidosis

J Pediatr Gastroenterol Nutr 64:e73-6, 2017

Liver Transplantation in PA

- Stop some medications
  - Sodium benzoate
  - Metronidazole
- Continue L-carnitine
- Liberalize protein
  - Natural protein 1 mg/kg/d
- Decreased metabolic crises; no hyperammonemia
- Lower C3-acylcarnitine and C3/C2 ratio
- Improved quality of life

J Pediatr Gastroenterol Nutr 64:e73-6, 2017

Liver Transplantation in PA

- Dilated cardiomyopathy is a frequent complication of PA
  - Toxic metabolites – inhibition of energy pathways
    - Methylcitrate (Krebs cycle enzymes)
    - Propionyl-CoA (pyruvate dehydrogenase, ETC activity, succinyl-CoA synthetase)
    - Methylmalonyl-CoA (succinate-supported respiration)
  - Anapleurotic defect
    - Decreased succinyl-CoA (TCA cycle function)
- Cardiomyopathy develops independent of any specific metabolic profile
- OTL reverses cardiomyopathy

J Pediatr 156:128-34, 2010
Transpl Int 28:1447-50, 2015

MMA and PA Pre-operative Management

- Dietician evaluation
- Nephrology evaluation (MMA)
  - Cystatin C
- CHDF (MMA)
  - If kidney failure
- Cardiology evaluation (PA)

Perioperative Management

- Prevent catabolism
- Avoid transplantation during metabolic decompensation
- Infusion of 10% dextrose (+ sodium bicarbonate) at 1.5 times normal maintenance rate
- Attention to acid-base status, glucose, lactate levels
- Pre-operative dialysis
  - Renal failure
  - High levels of abnormal metabolites

Pediatr Anesth 26:694-702, 2016
J Pediatr Gastroenterol Nutr 64:e73-6, 2017

MMA Peri-operative Management

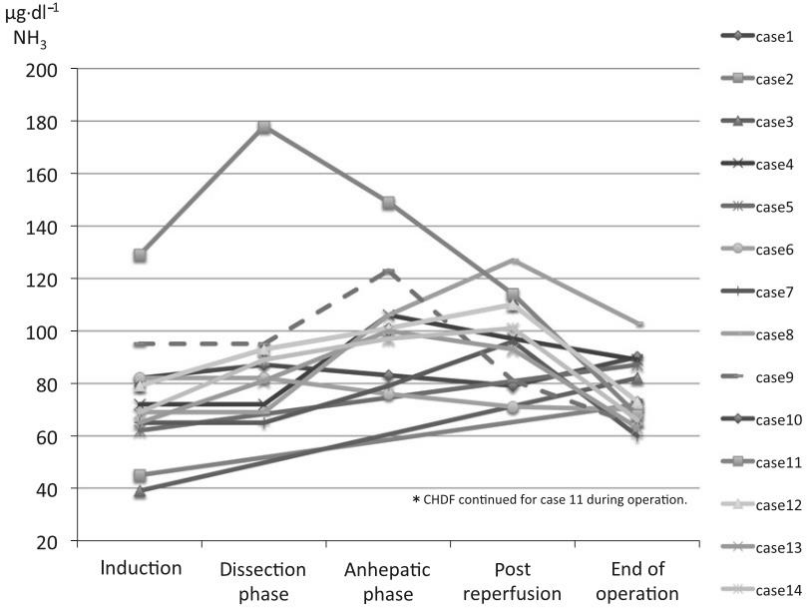


Figure 3 Intraoperative NH_3 change in all cases.

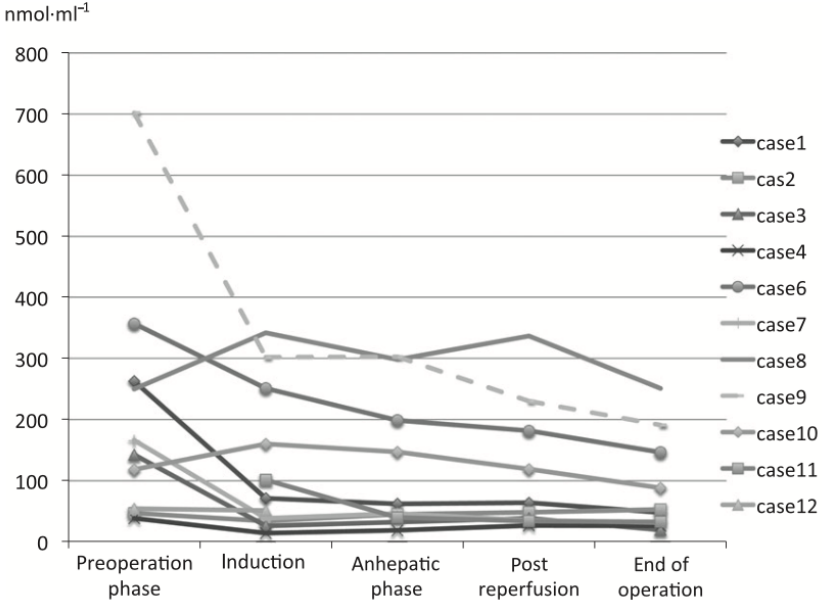


Figure 4 Methylmalonic acid change through LT.

Pediatr Anesth 26:694-702, 2016

Anesthesia Considerations

- Avoid drugs metabolized to propionic acid, odd-chain organic acids, alcohols, or fatty acids
- Avoid propofol
  - Small amount of soybean oil fats metabolized to propionate
- Avoid muscle relaxants metabolized by ester hydrolysis (succinylcholine, cisatracurium, atracurium)
  - Metabolites include odd-chain organic molecules

Anesth Analg 91:309-11, 2000
J Pediatr Gastroenterol Nutr 64:e73-6, 2017

MMA and PA Post-operative Management

- TPN + IL
  - POD 1 start 0.5 g/kg/d amino acids
  - POD 2 start 0.8 g/kg/d amino acids
  - POD 3 start 1.0 g/kg/d amino acids
- Carnitine supplementation
- Close monitoring blood glucose
- Transition from TPN to enteral/oral feeds
- Maintain same dietary plan post-operatively
  - Slowly liberalize protein as tolerated (months)
- Wean off metabolic formula as possible as DRI reached

MMA and PA Post-operative Management

- Start enteral or oral feeds as soon as possible when stable
- Wean TPN either by decreasing volume or altering amino acids in TPN depending on other needs for fluids and calories
- Wean lipids once calories from enteral feeds approach goal

MMA and PA Post-operative Management

- Wean off metabolic formula as possible as DRI reached
- This is typically a slow, stepwise process
  - Clinic visit interval
  - Monitoring labs (e.g., plasma amino acids, nutrition labs)
    - Complete blood count, prealbumin, C-reactive protein, zinc, selenium, essential fatty acids, 25-hydroxy vitamin D, vitamin B_{12}

Liver Transplantation Complications

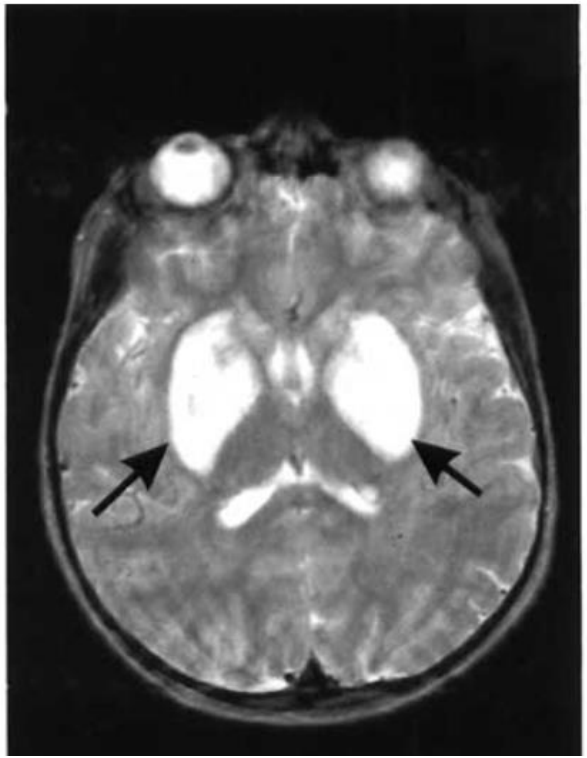
- Mortality
- Lactic acidemia
- Metabolic decompensation
- ‘Metabolic stroke’
- Graft rejection
- Post-op pancreatitis

- Immunosuppression complications
  - Diabetes
  - Hypertension
  - Seizures
  - Infections
  - Nephrotoxicity
- Surgical complications
  - Hepatic artery thrombosis
  - Subphrenic abscess
  - Splenic rupture

J Pediatr 140:261-3, 2002
Ther Apher Dial 15:488-92, 2011
J Pediatr 166:1455-61, 2015
Pediatr Anesth 26:694-702, 2016

Metabolic Stroke in MMA

- LT at 9 months
- Age 5 ½ y developed pneumonia
- Acute neurological decompensation 1 w later while stable on IV antibiotics



J Pediatr 140:261-3, 2002

Post-transplant Complications in MMA

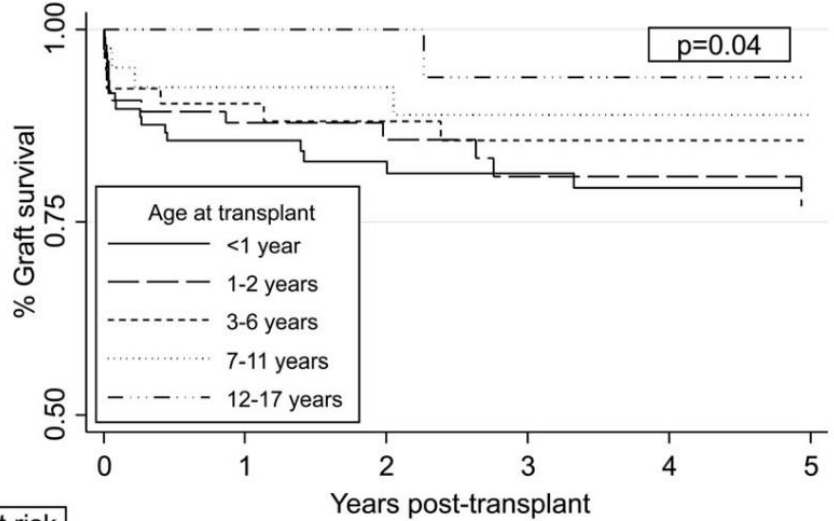
- LT at 22 y
- 3 m after LT developed kidney failure
- Normal brain CT and MRI
- Progressive neurological findings
  - Limp
  - Lost voluntary control of legs and, later, arms
  - Spasmodic leg contractions

Eur J Pediatr 161:377-9, 2002

Persisting Morbidity

- Biochemical abnormalities persist
- Renal insufficiency after LT
- Risk of metabolic strokes remains

Pediatric Liver Transplant for Organic Acidemias



Number at risk						
<1 year at transplant	97	71	53	48	39	29
1-2 years	77	54	39	31	25	20
3-6 years	52	41	38	30	24	21
7-11 years	41	34	30	24	19	15
12-17 years	26	18	16	12	10	8

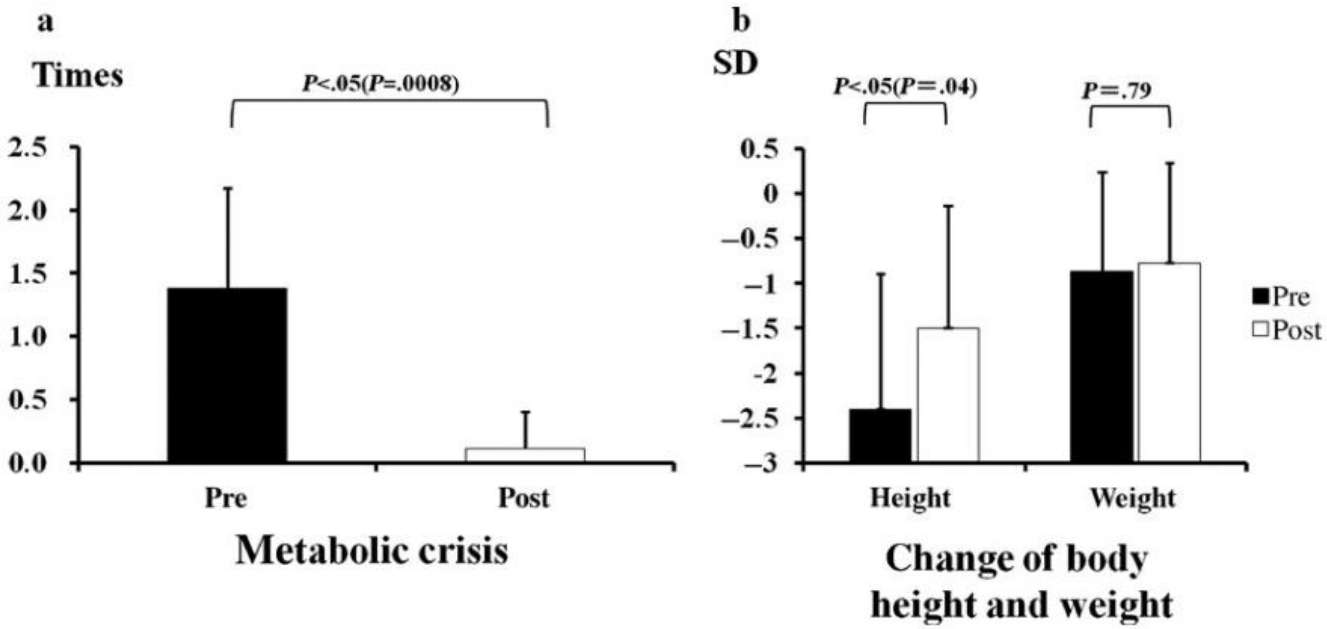
Liver Transpl 20:89-99, 2014

Pediatric Liver Transplant for Organic Acidemias (UNOS)

- 5-y post-transplant survival 78% (<2 y) and 88% (≥ 2 y)
- Vascular thrombosis caused 44% of graft losses
- 65% of graft losses occurred in children <2 y
- Children with UCDs/OAs more likely to have cognitive and motor delays compared to those who underwent transplantation for other indications

Liver Transpl 20:89-99, 2014

MMA Post-transplant Outcomes



Pediatr Transpl 20:1081-6, 2016

MMA Post-transplant Outcomes

- 13 MMA patients
- 9 *mut*^{0}, 2 *mut*$^{-}$, 2 unknown
- Living donor transpant
- 4 to 16 y follow up
- Protein-restricted diet
- Mean DQ before and after LDLT did not differ significantly
- ↓ mean plasma MMA and C3
- One patient developed renal failure

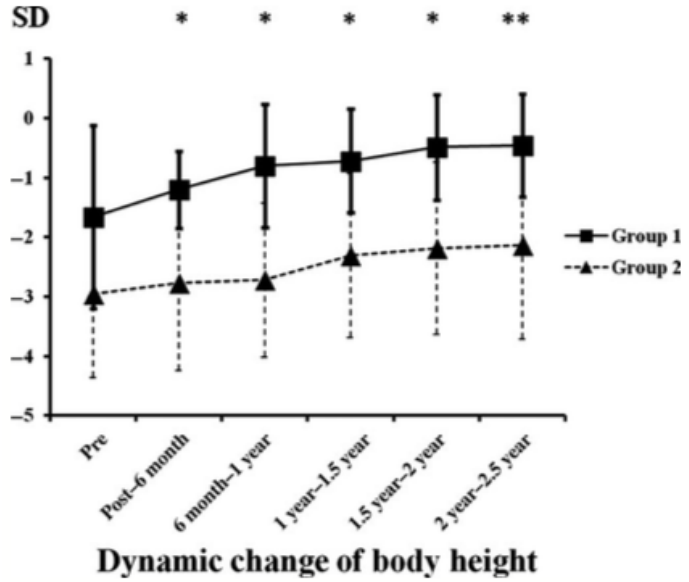


FIGURE 2 Change in height after LDLT; ■ data of patients who underwent LDLT before the age of 1 y (n=7); ▲ data of patients who underwent LDLT after the age of 1 y (n=6) *P<.05, ** P=.053

Pediatr Transpl 20:1081-6, 2016

Liver Transplantation Cost-Effectiveness

- 1.5 more life years lived
- 7.9 more QALYs
- Savings of $582,369 for lifetime societal cost per individual (compared to nutritional support)
- LT more effective and less costly in all 1-way sensitivity analyses
- LT a dominant treatment strategy in newborns with classical MMA or PA

Liver Transpl 21:1208-18, 2015

Organ Prioritization for MMA Transplantation

- Autonomy
  - Right to request transplantation
  - Request might not be honored (scarcity of organs)
- Beneficence
  - Dietary management
  - Consideration of benefits of transplantation
- Nonmaleficence
  - Risks of procedure and immunosuppression
  - Long-term neurological outcomes
- Justice
  - Consider interests of communities v. individual
  - Consider utility

AMA J Ethics 18:153-5, 2016

MMA Therapy in Development

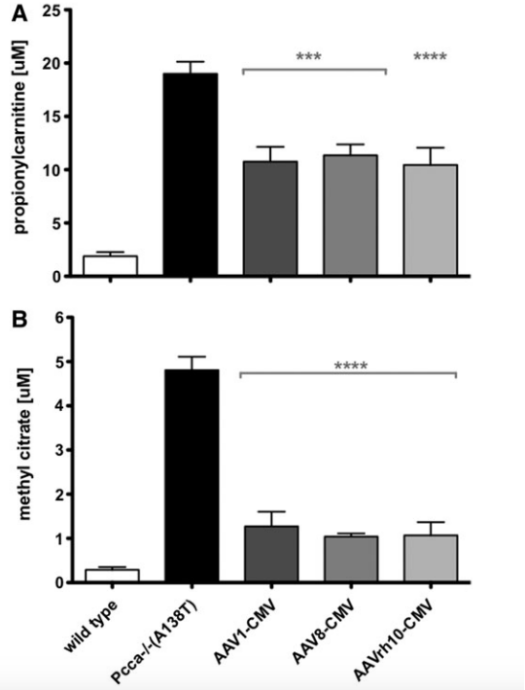
- Gene therapy
- mRNA therapy
- Hepatocyte transplantation

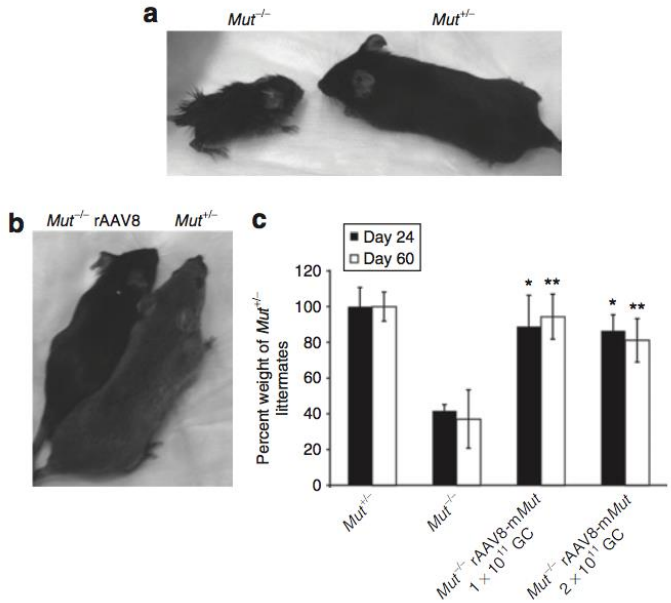
MMA Therapy in Development

- Gene therapy
  - Transgenic model
  - Hepatic targeting > phenotypic correction
- AAV
  - Hepatic genotoxicity
  - Immune responses
    - Neutralizing antibodies

Gene Therapy (Pre-Clinical)

- Adenovirus, AAV, Lentivirus





Hum Gene Ther 19:53-60, 2008
Mol Ther 18:11-16, 2010
Gene Ther 29:385-91, 2012
Hum Gene Ther 25:529-38, 2014
Hum Gene Ther 25:837-43, 2014

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Systemic Messenger RNA Therapy as a Treatment for Methylmalonic Acidemia

Ding An,[1,3] Jessica L. Schneller,[2,3] Andrea Frassetto,[1] Shi Liang,[1] Xuling Zhu,[1] Ji-Sun Park,[1] Matt Theisen,[1] Sue-Jean Hong,[1] Jenny Zhou,[1] Raj Rajendran,[1] Becca Levy,[1] Rebecca Howell,[1] Gilles Besin,[1] Vladimir Presnyak,[1] Staci Sabnis,[1] Kerry E. Murphy-Benenato,[1] E. Sathyajith Kumarasinghe,[1] Timothy Salerno,[1] Cosmin Mihai,[1] Christine M. Lukacs,[1] Randy J. Chandler,[2] Lin T. Guey,[1] Charles P. Venditti,[2,4,*] and Paolo G.V. Martini[1,4,5,*]

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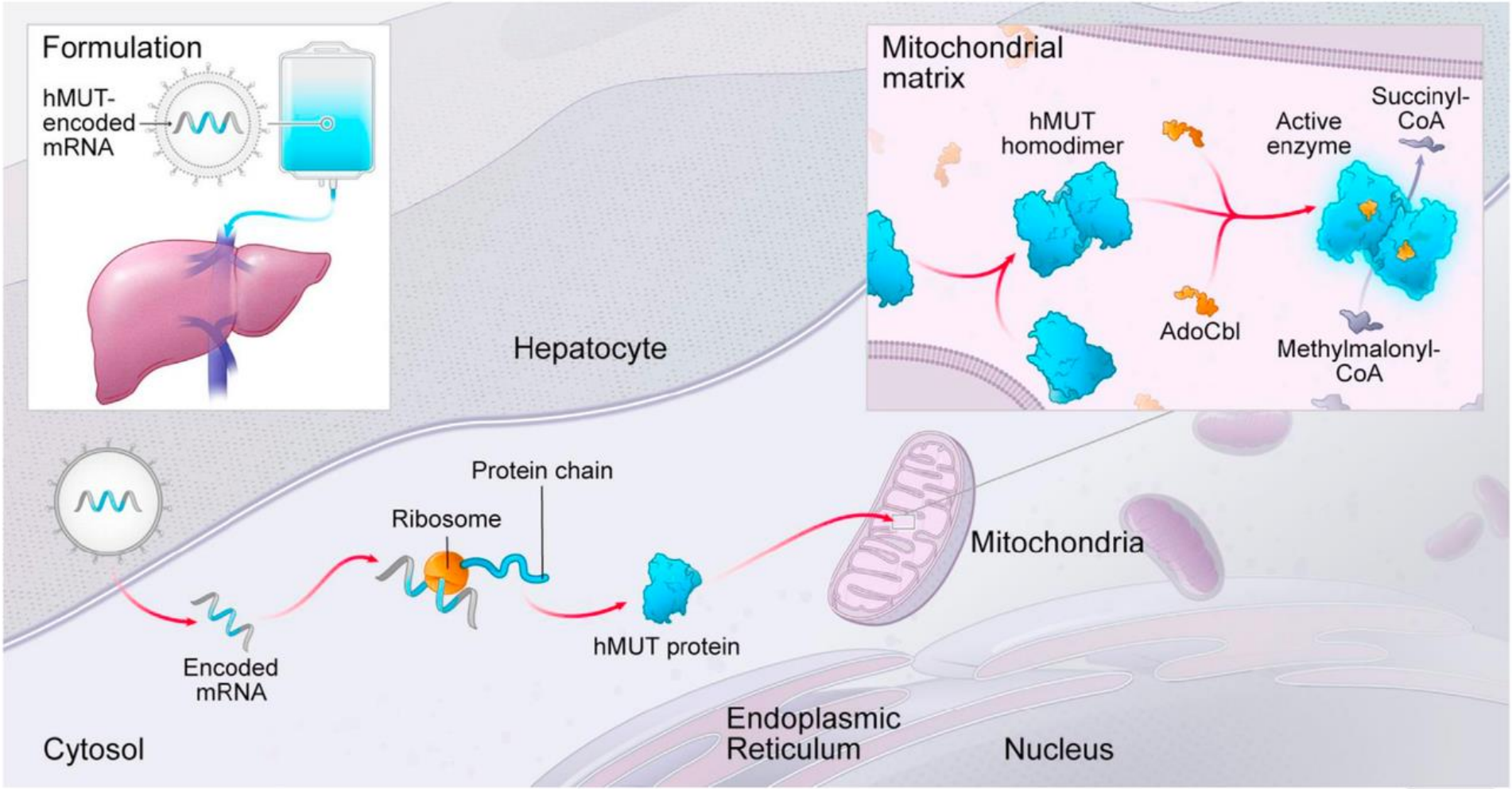
mRNA Therapy

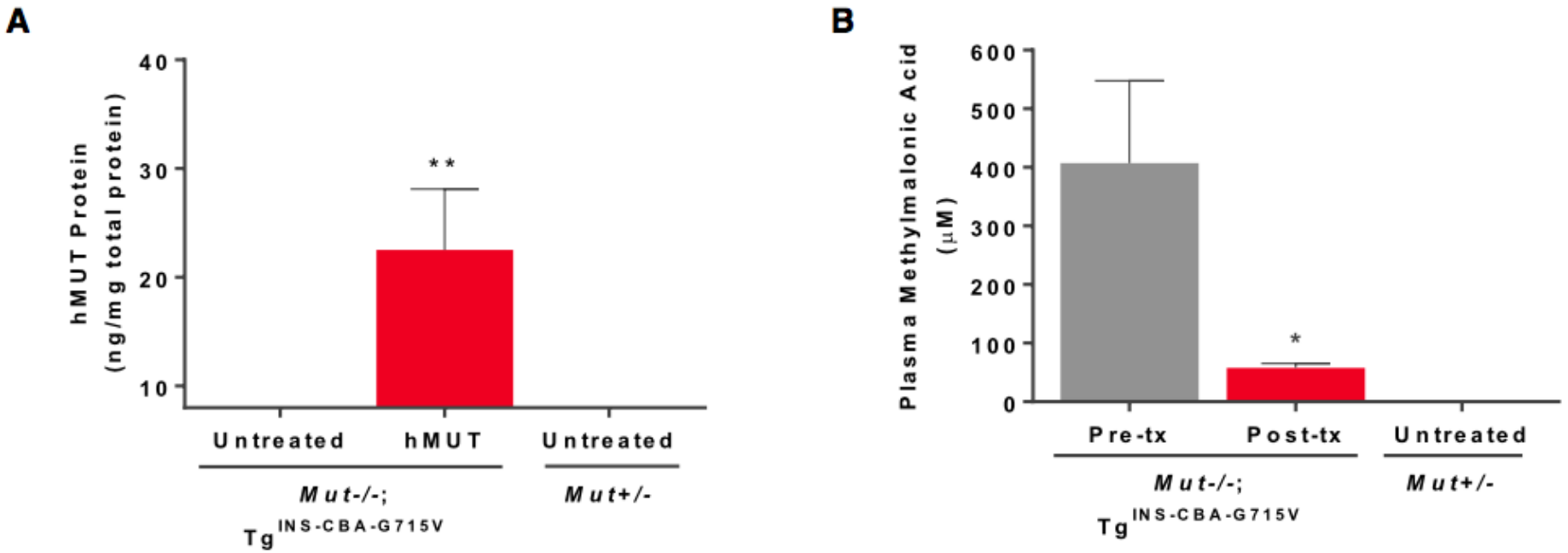
- No insertional mutagenesis
- Avoids constitutive gene activation
- Alternative to conventional ERT
- Being developed for:
  - Hemophilia B
  - Sensory nerve disorders
  - Lung disease
  - Cancer

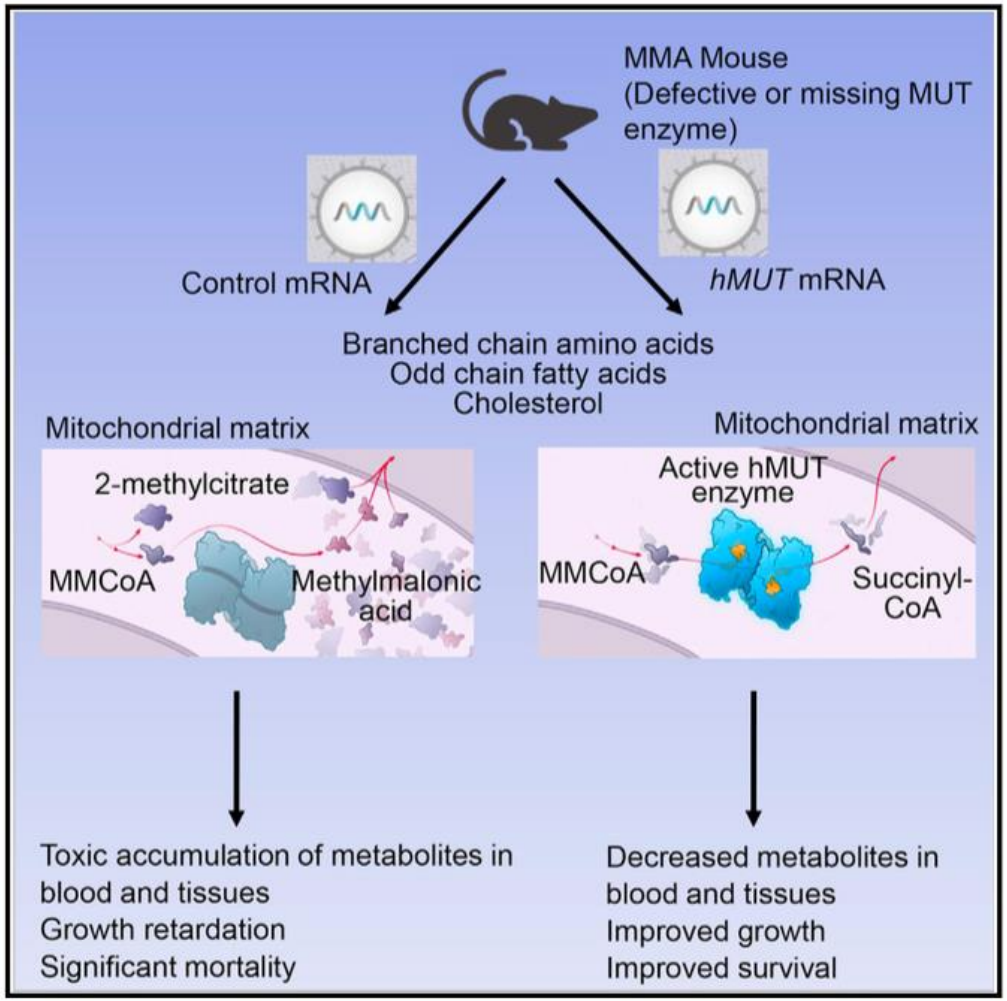
Lipid Nanoparticles

- Bio-degradable
- Liver targeting
- Encapsulate biomolecules
- Deliver systemically


Formulation
hMUT-encoded mRNA
Mitochondrial matrix
hMUT homodimer
Active enzyme
Succinyl-CoA
AdoCbl
Methylmalonyl-CoA
Hepatocyte
Protein chain
Ribosome
Mitochondria
Encoded mRNA
hMUT protein
Cytosol
Endoplasmic Reticulum
Nucleus

Improved Metabolism in *mut*$^{-}$ MMA Mice after i.v. *hMUT* mRNA




MMA Mouse
(Defective or missing MUT enzyme)
Control mRNA
hMUT mRNA
Branched chain amino acids
Odd chain fatty acids
Cholesterol
Mitochondrial matrix
2-methylcitrate
MMCoA
Methylmalonic acid
Mitochondrial matrix
Active hMUT enzyme
MMCoA
Succinyl-CoA
Toxic accumulation of metabolites in blood and tissues
Growth retardation
Significant mortality
Decreased metabolites in blood and tissues
Improved growth
Improved survival

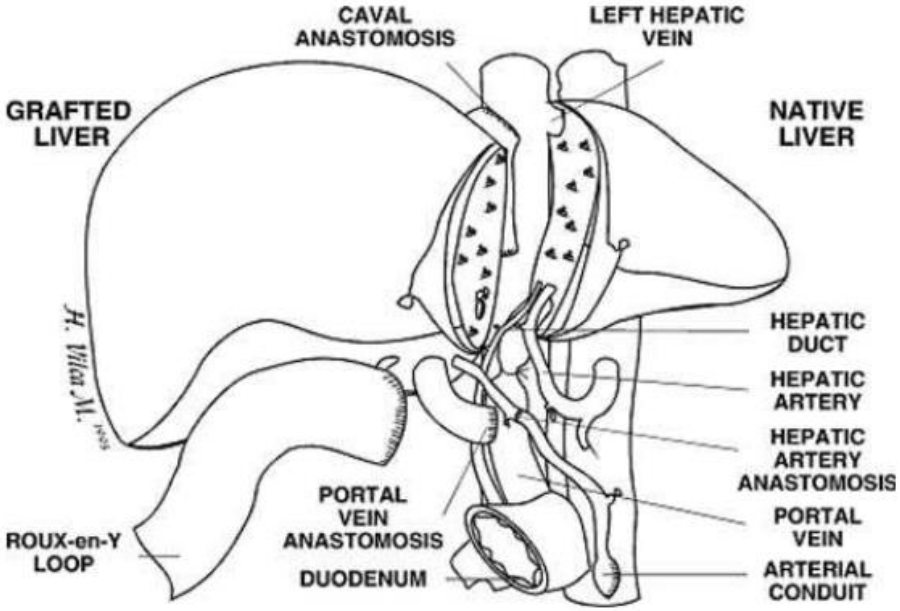
MMA Hepatocyte Therapy

- *Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$ mouse model
  - Muscle-specific promoter
  - Significant hepatic and renal pathology
  - Growth retardation
- 8 week old mice (n=6)
- Direct splenic injection of fresh hepatocytes
- 3 survivors showed improved weight gain and metabolic parameters up to 2 months after transplantation

Mol Ther 20, Suppl1:20, S59, 2012

Auxillary Liver Transplantation in PA

- 2 y underwent ALT
- Normal diet
- Normal growth; acceptable neurological and psychomotor development
- Alternative approach
- Preserves native liver
- Back-up in case of graft failure
- Future gene, stem cell, or mRNA therapy



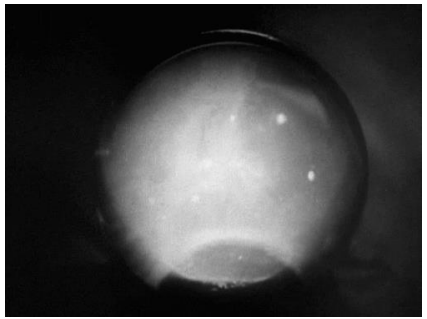
Am J Transplant 7:2200-3, 2007

Summary - Ongoing questions

- Does early LT preserve or decrease decline in renal function in MMA?
- Does transplantation improve neurological outcome?
- Can redox modulation therapy help maintain kidney function or improve neurological outcome?

Summary

- LT, LKT (MMA) or KT (MMA, milder forms?) appear to be viable therapeutic approaches; no reason to use extended criteria donors
- Decreased frequency of metabolic crises and hospitalizations
- Stabilization of neurological function
- Liberalization of diet
- Weight gain
- Improved quality of life
- Consider auxillary LT (gene, stem cell, mRNA therapies)?



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