Diagnosing Genetic Epilepsies – Impact on Treatment
Cleveland Clinic Foundation Epilepsy Course 2020
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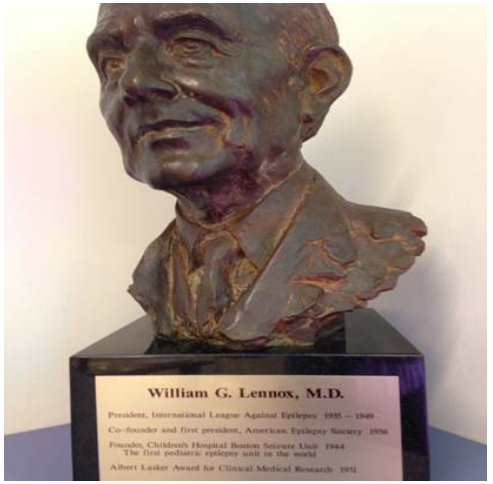

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OBJECTIVES

- 1. Explore the genetic and metabolic basis of epilepsies.
- 2. Decide when to order genetic testing.
- 3. Identify cardinal signs of inherited metabolic epilepsies.
- 4. Intervene with targeted therapy when available in genetic-metabolic epilepsies.

Historical Underpinnings of Epilepsy

- But this disease seems to me to be no more divine than others; but it has its nature such as other diseases have, and a cause whence it originates, and its nature and cause are divine only just as much as all others are, and it is curable no less than the others…Its origin is hereditary, like that of other diseases.

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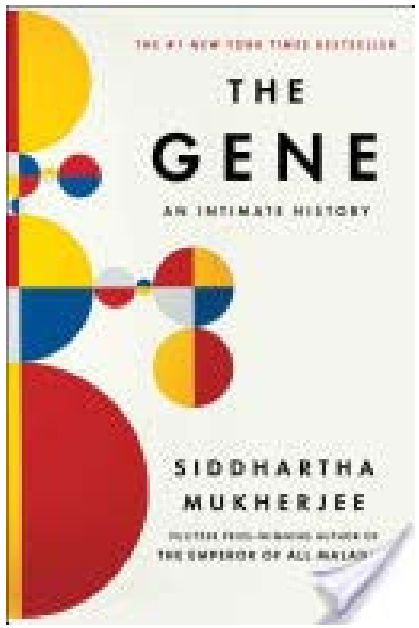
  On the Sacred Disease

  Hippocrates

  400 BCE

Epilepsy Genetics: Milestones



- Familial epilepsy
  - Lennox, twin studies
- "Idiopathic epilepsy"
  - Presumed genetic
- Mapping of the Human Genome
  - 2001
- Personalized, Targeted Therapies

Why Do Genetic Testing?

- 1. Diagnostic Certainty
  - End the odyssey.
  - Aid in prognosis.
- 2. Genetic Counseling
- 3. Impact on Treatment

Impact on Treatment? YES.

- Loss of function – haploinsufficiency
  - SCN1A – Dravet syndrome (avoid Na blockers)
  - SLC2A1 – GLUT1 deficiency (ketogenic diet, carnitine, alpha-lipoic acid)
- Gain of function
  - SCN8A (respond to Na channel blockers)
  - KCNT1 (migratory focal epilepsy of infancy)

Who To Test?

- "Epilepsy Plus"
  - Dysmorphism, cognitive deficit, autism
- CMA
  - Yield of 1.5-5% (Olson et al, Ann Neuro 2014; Lindy et al, Epilepsia 2018)
- Panel or exome sequencing
  - Yield of 15-50% (Helbig et al, Genet Med 2016; Lindy et al, Epilepsia 2018)

What about the “Genetic Generalized Epilepsies”?

- ¼ of all epilepsies
- Generalized spike/wave, normal imaging, normal cognition
- Typical absence, myoclonic, GTCS
- Genetic contribution is suspected
- Monozygotic vs Dizygotic concordance is 76% vs 33% (strongly but not exclusively genetic)
- 8% siblings affected
- Polygenic inheritance presumed

Case Study 1

- 14 yo girl with JME
- FH positive for epilepsy (maternal side)
- Does genetic testing add?
  - If regression occurs, consider other dx'es.
  - Pharmacogenetics may add in Rx choice.
  - Not especially helpful in the more common epilepsies.

Study 2

- A 6 month old boy with infantile spasms, normal MRI.
- A pathogenic variant in an “epilepsy gene” would allay further studies and potentially repeated imaging.
- Possible impact on Rx choice.

Categories of Inherited Metabolic Epilepsies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

When to Suspect a Metabolic Epilepsy

- Onset during neonatorum, infancy, or early childhood
- Typical clinical presentation: newborn with poor feeding, hypotonia, lethargy, respiratory distress, or lactic acidosis
- Myoclonic seizures
- EEG: burst-suppression, hypsarrhythmia
- Family history of metabolic disorder
- Poor response to traditional antiepileptic drug

Treatable Metabolic Epilepsies

Specific Disorders

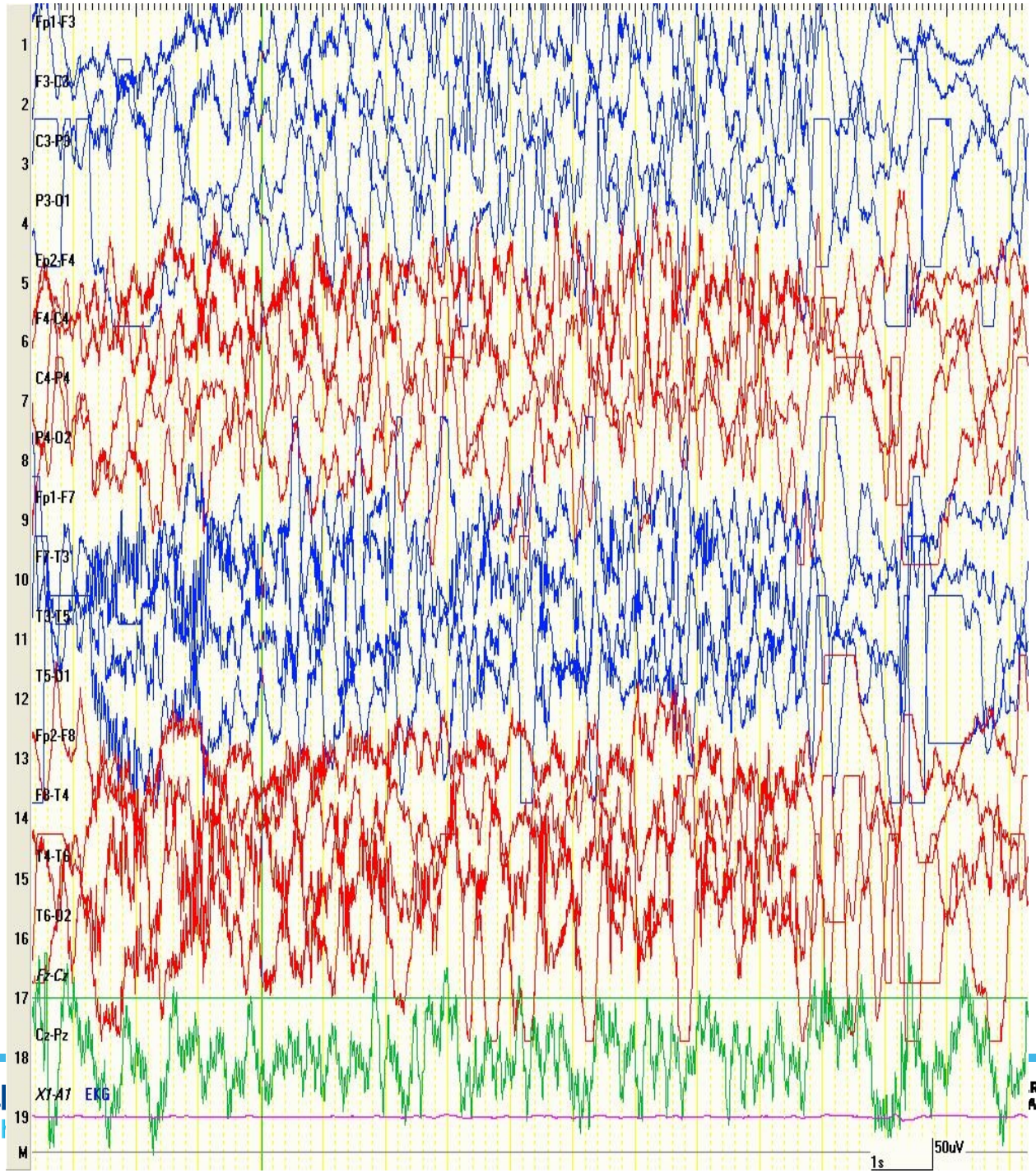
- **Vitamin Responsive Disorders**
  - Pyridoxine, P5P, Folinic acid, Biotin
- **Transportopathies**
  - GLUT1, Cerebral Folate Deficiency, Thiamine Transporter
- **Amino and Organic Acid Disorders**
  - MSUD, propionic, isovaleric, cobalamin C
- **Lysosomal Storage Diseases**
  - LINCL/CLN2
- **Mitochondrial Disorders**
  - PDHC deficiency
- **Purine Synthesis Disorders**
  - MoCo deficiency type A
- **Urea Cycle Disorders**
  - OTC deficiency
- **Neurotransmitter Disorders**
  - BH4, GABA-transaminase
- **Disorders of Glucose Homeostasis**
  - DEND, HI/HA

Neonatal Seizures

- FT NBN 3220 gms
- Abnormal eye movements, grunting 12 hrs
- EEG: episodic suppression, bilateral sharp waves
- Rx phenobarbital, levetiracetam, pyridoxine: seizure-free X 6 wks
- Hospitalized at 3.5 months for stiffening, Rx topiramate
- Mycolonic & tonic-clonic seizures; steroids ineffective

- Pyridoxal-5-phosphate stopped seizures with first dose. Breakthrough events as dose becomes due.
- CSF levels for P5P = 23 (23-64), ↑ thr; extra peak (suspected pyridoxine phosphate) .
- PNPO sequencing: homozygous mutation, conserved area, gly>arg.

- Pearl, Hyland, Chiles et al: *Partial pyridoxine responsiveness in PNPO deficiency*. J Inherit Metab Dis 2013.


Fp1-F3
F3-C3
C3-P3
P3-O1
Fp2-F4
F4-C4
C4-P4
P4-O2
Fp1-F7
F7-T3
T3-T5
T5-O1
Fp2-F8
F8-T4
T4-T6
T6-O2
Fz-Cz
Cz-Pz
X1-A1
EKG
1s
50uV

PNPO Deficiency

- Clinical Triad
  - Rotatory eye movements, hyperexcitability, hypersalivation (I Tein 2015)
- CSF Profile
  - Elevated glycine, threonine
  - Depressed [P5P]

PYRIDOXINE DEPENDENCY: REPORT OF A CASE OF INTRACTABLE CONVULSIONS IN AN INFANT CONTROLLED BY PYRIDOXINE

By ANDREW D. HUNT, JR., M.D.,* JOSEPH STOKES, JR., M.D., WALLACE W. McCRORY, M.D., AND H. H. STROUD, M.D.
Philadelphia

THE IMPORTANCE of pyridoxine in animal and human nutrition has been a subject of wide interest since its original description as a B factor by György[1] in 1934. Unlike the majority of vitamins, however, no pathologic condition in humans has been described which occurred spontaneously and was corrected solely by the administration of pyridoxine. The authors recently observed an infant with a severe convulsive disorder who responded in an extraordinary manner to regular administration of pyridoxine. This phenomenon was thought to be unique and to warrant the following case report.

CASE REPORT

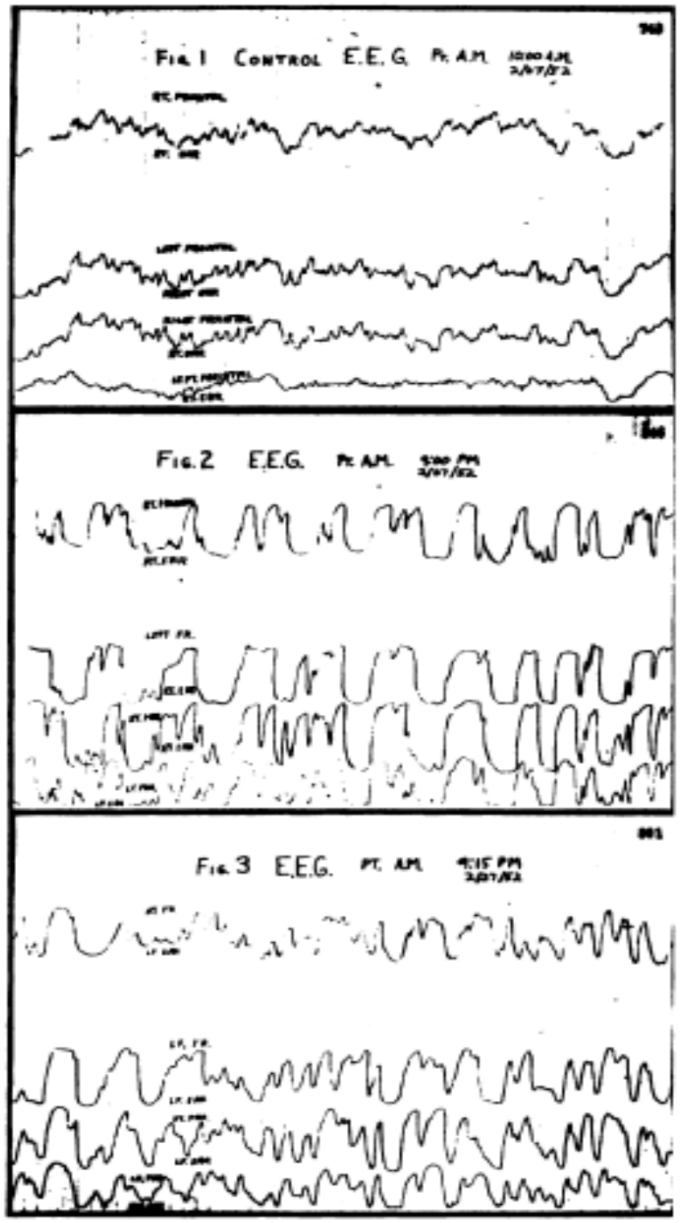
A. M., a female infant, was admitted to The Children's Hospital of Philadelphia at the age of 13 days because of constant and intractable

Mrs. M.'s first pregnancy had been normal, devoid of illness or significant nausea and vomiting. The second pregnancy, however, was accompanied by severe nausea and vomiting which was treated with injections of pyridoxine and thiamine during the 1st 4 mo. of gestation.

Pregnancy with the patient also resulted in severe nausea and vomiting, sufficiently so to require hospitalization on 1 or 2 occasions for intravenous fluids. During the second, third, fourth and fifth months of this pregnancy she was given, 3 to 4 times weekly, an intramuscular injection consisting of pyridoxine HCl 50 mg., and thiamine HCl, 50 mg. No adverse reactions were noted during this therapy.

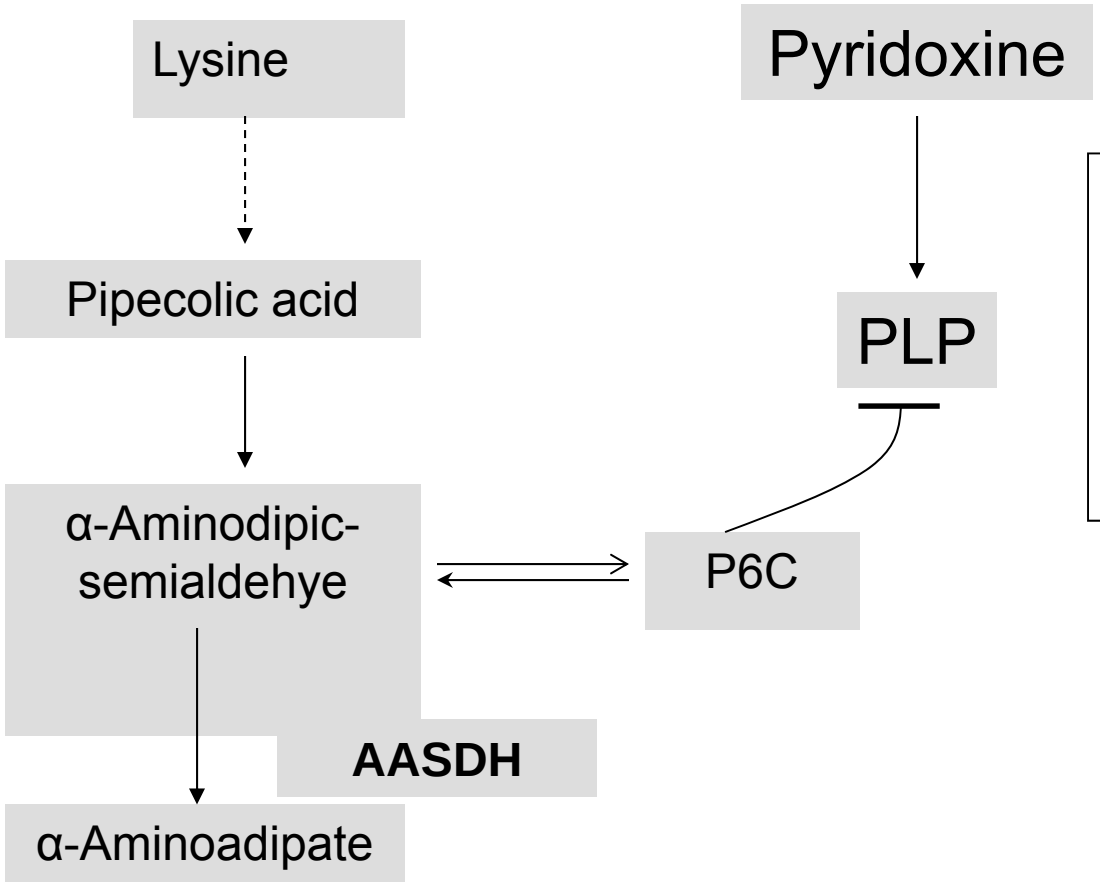
Labor had a spontaneous onset, occurred at term, and was of 4 hr.'s duration. No difficulties were encountered during delivery, birth weight was 3.2 kg., respirations began spontaneously, and the baby's color was considered good. However, 3 hr. after birth, generalized twitching accompanied by shrill cries made its

Hunt et al. Pyridoxine Dependency: Report of a Case of Intractable Convulsions in an Infant Controlled by Pyridoxine. Pediatrics 1954



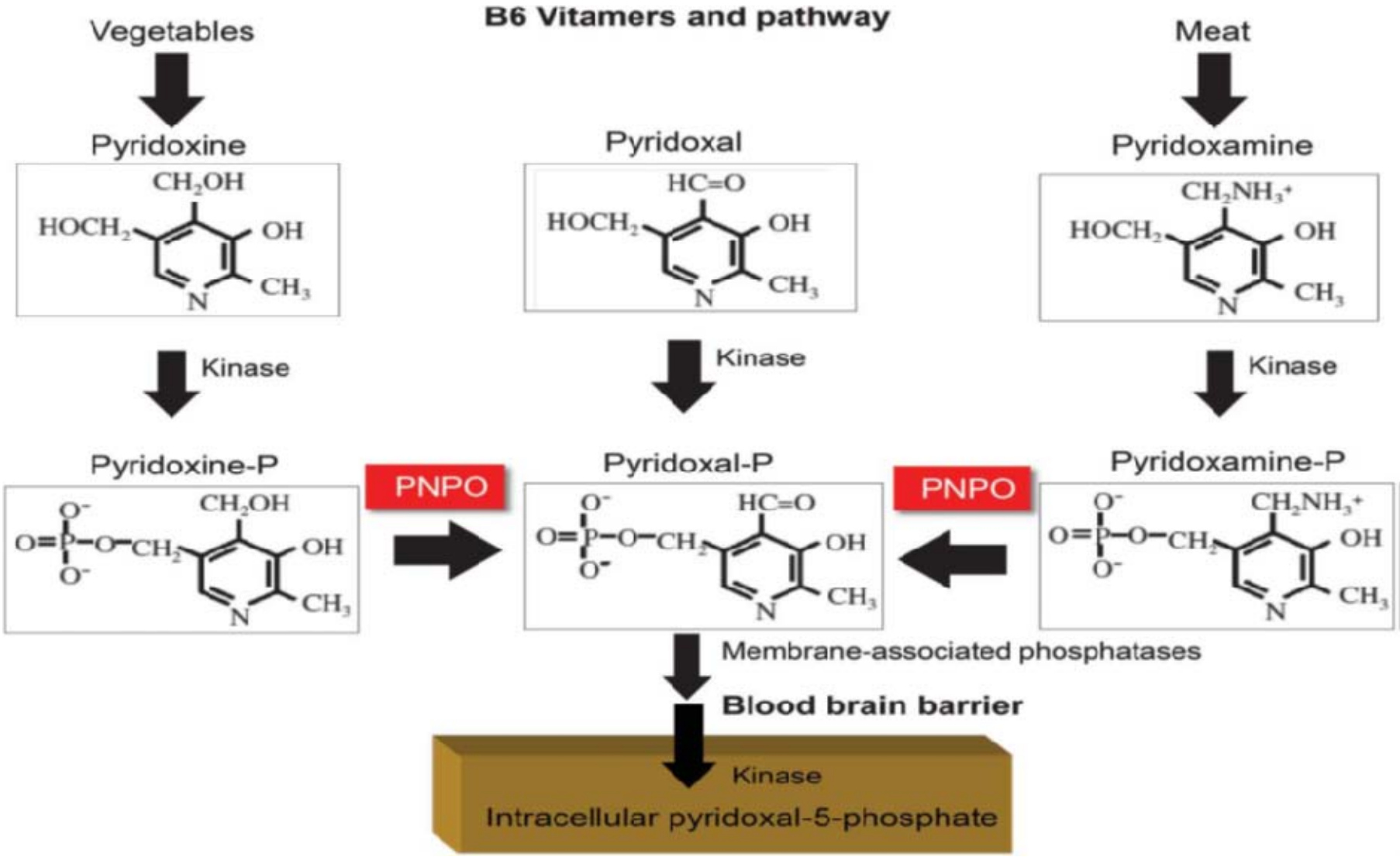
Hunt et al. Pyridoxine Dependency: Report of a Case of Intractable Convulsions in an Infant Controlled by Pyridoxine. Pediatrics 1954

Antiquitin (AASDH) deficiency in Pyridoxine dependent epilepsy



When AASDH (antiquitin) activity is deficient, pipecolic acid and P6C both accumulate. P6C sequesters PLP, the biologically active form of pyridoxine.

AASDH = alpha-Aminodipic-semialdehyde Dehydrogenase *(antiquitin);* **P6C =** delta-Piperideine-6-carboxylate; **PNPO** = Pyridox(am)ine oxidase; **PLP** = Pyridoxal-5-phosphate


B6 Vitamers and pathway
Vegetables
Pyridoxine
Kinase
Pyridoxine-P
PNPO
Pyridoxal
Kinase
Pyridoxal-P
PNPO
Meat
Pyridoxamine
Kinase
Pyridoxamine-P
Membrane-associated phosphatases
Blood brain barrier
Kinase
Intracellular pyridoxal-5-phosphate

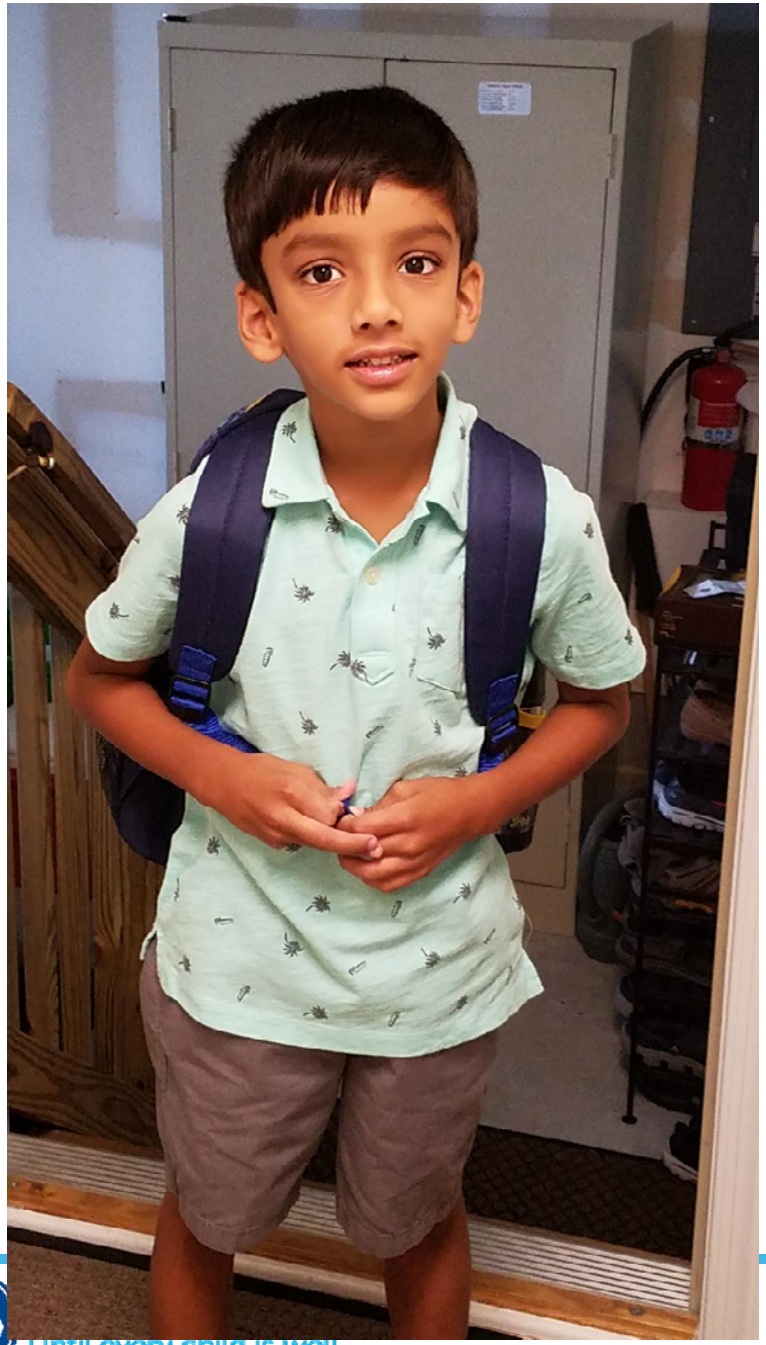
Approach to Intractable Neonatal Seizures Suspect for Pyridoxine Related Dependency

Diagnostics:

- Blood, urine for AASA, pipecolic acid
- DNA for ALDH7A1 or PNPO molecular analysis

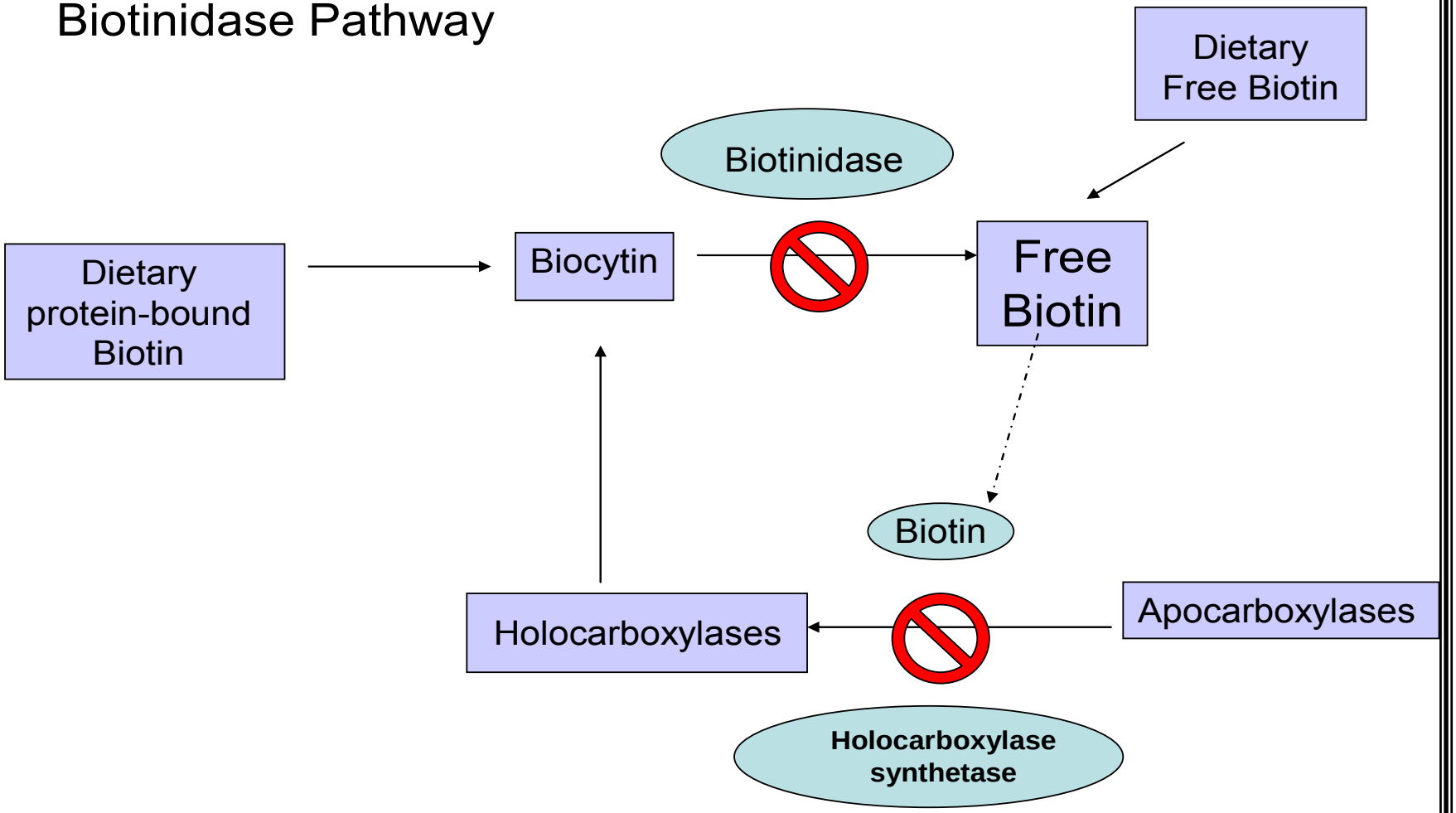
Treatment:

- B6 100 mg IV bolus (5-10 mins) with EEG + cardiorespiratory monitoring.
- If no response, repeat 100-500 mg IV B6 bolus.
- In responders, observe as inpatient a minimum of 48 hours.
- While biomarkers are pending, consider oral/enteral B6 15-30 mg/kg/day divided BID.
- Begin P5P 30-50 mg/kg/day divided 4-6 X/day when available X 3-5 days.
- Folinic acid 3-5 mg/kg/D divided BID X 3-5 days.




CAREER OPPORTUNITIES

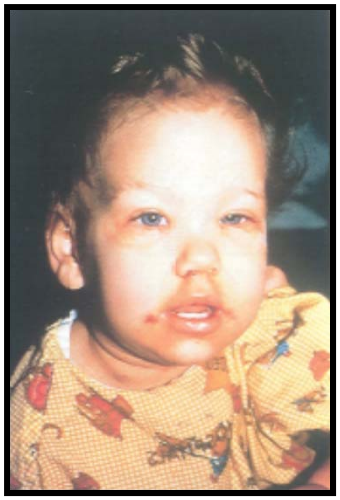
Biotinidase Pathway

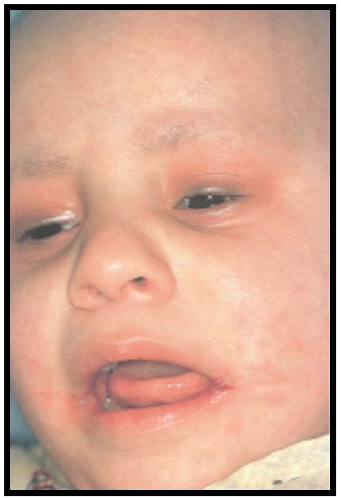


Biotinidase Deficiency: Phenotypic Components

- Developmental delay
- Hypotonia
- Seizures
- Ataxia
- Alopecia, perioral rash
- Episodic metabolic acidosis
- Hearing loss
- Vision loss, optic atrophy
- Lactic and propionic acidemia

Patients with Biotinidase Deficiency





Nyhan WL, Barshop BA, Ozand PT:
Atlas of Metabolic Diseases (2005).

Secondary biotinidase deficiency (beyond multiple carboxylases)

- 1. Dietary deficiency (vegan diets)
- 2. Malabsorption
- 3. Hemodialysis
- 4. Parenteral nutrition
- 5. Drugs, e.g. valproic acid

Treatment of Biotinidase Deficiency

- Gratifying response to biotin 10 mg/day.
- Visual and sensorineural hearing loss, once established, persist.

Clinical Landmines: Biotinidase Deficiency

1. Misdiagnosed as “atypical” or “childhood” multiple sclerosis. Patients may present in *adolescence with spastic paraparesis*. Dermatologic manifestations misdiagnosed as acrodermatitis enteropathica or anhidrotic ectodermal dysplasia.

2. Seizures (generalized, myoclonic, or infantile spasms) occur in the majority of patients and may be the *only obvious symptom.* Testing for biotinidase deficiency is warranted in any patient with unexplained seizures.

Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
  - Glucose
  - Folate, thiamine, riboflavin, manganese – infantile onset and later
- Amino and Organic Acid Disorders
- Lysosomal Storage Diseases
- Mitochondrial Disorders
- Purine Metabolism Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

BRIEF REPORTS

DEFECTIVE GLUCOSE TRANSPORT ACROSS THE BLOOD–BRAIN BARRIER AS A CAUSE OF PERSISTENT HYPOGLYCORRHACHIA, SEIZURES, AND DEVELOPMENTAL DELAY

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RONALD I. JACOBSON, M.D.,
GABRIEL M. RONEN, M.D.,
RAMIN A. BEHMAND, B.S.,
AND SAMI I. HARIK, M.D.

GLUCOSE (D-glucose) is an essential fuel for the brain and many other tissues. Five glucose-transporter proteins facilitate the diffusion of glucose across lipophilic plasma membranes.[1-3] This process is saturable and stereospecific, but it is not concentrative, energy dependent, or influenced by sodium. In humans, the erythrocyte glucose transporter (type 1 glucose transporter) has been studied most extensively. It accounts for 2 to 5 percent of the erythrocyte-membrane protein and seems to be identical in molecular weight and antigenic properties to the glucose transporters in the endothelial cells of brain capillaries.[4-7] Brain capillaries contain large amounts of messenger RNA for the type 1 glucose transporter,[8,9] and the density of these transporters in brain capillaries is approximately 10 times greater than in tissues other than erythrocytes.[4,5,10,11] The presence of the type 1 glucose transporter in erythrocytes and brain capillaries provides an opportunity to study genetic conditions that may affect the transport of glucose across the blood–brain barrier. The movement of water-soluble molecules across this barrier is limited by the occluding junctions between endothelial cells and by the scarcity of pinocytosis.[12,13] Dick et al.[4] estimated that capillary endothelial cells in the brain transport about 10 times their weight of glucose per minute to support the glucose requirements of the brain.[4,14] A defect of the glucose-transporter protein of brain capillaries should interfere with cerebral energy metabolism and brain function. We have studied two children with persistent hypoglycorrhachia (low concentrations of glucose in cerebrospinal fluid), seizures, and delayed development who seemed to have a genetic defect involving the type 1 glucose transporter. Both responded dramatically to treatment with a ketogenic diet. We believe that these two children have a primary defect of glucose transport into the brain.

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CASE REPORTS

Patient 1

The gestation of a male infant was complicated by mild oligohydramnios and a viral illness in the mother during the fifth month of pregnancy. At birth the infant weighed 3235 g, and his Apgar scores were 6 and 7 at one and five minutes, respectively. He had transient neonatal respiratory distress, suckled poorly, and had mild hypotonia. He was treated with oxygen for one day and was apparently well when he went home at the age of five days. His first seizure occurred at the age of 2.5 months. The seizures were described as myoclonic jerks of one limb lasting up to eight minutes and associated with staring. On examination, the infant was clinically well, his head circumference was in the 50th percentile, and results of electroencephalography and cranial magnetic resonance imaging were normal. The cerebrospinal fluid concentration of glucose was low (Table 1), and the ratio of cerebrospinal fluid glucose to blood glucose was 0.29 (normal, 0.65). The concentration of protein in cerebrospinal fluid was normal in this and later examinations. Treatment with phenobarbital was started. Clonazepam therapy was added at three months because of recurrent seizures. A second lumbar puncture at the age of 4.5 months again demonstrated hypoglycorrhachia (Table 1), and serial blood glucose measurements ruled out occult hypoglycemia. The concentration of lactate in cerebrospinal fluid was low (0.41 mmol per liter). When the patient was six months old, a lumbar puncture performed five hours after a morning feeding again revealed hypoglycorrhachia (Table 1). Brief, subtle myoclonic limb jerking and staring alternating with eye-rolling continued despite treatment with phenobarbital and clonazepam. The infant achieved head control at the age of 3 months, turned over at 4 months, and sat unsupported at 10.5 months. At the age of 7.5 months, he had a head circumference of 43.4 cm (less than the third percentile), but he was alert and friendly. He was in the 50th percentile for height and the 90th percentile for weight. The sagittal suture and anterior fontanelle were closed. Limb tone was mildly increased, and tendon reflexes were brisk. Cerebrospinal fluid concentrations of glucose and lactate again were low (Table 1). Electroencephalography demonstrated a right frontal focus admixed with a vertex transient wave. Cranial magnetic resonance imaging suggested a mild delay in myelination.

A defect in the transport of glucose across the blood–brain barrier was suspected because of the persistently low glucose and lactate values in cerebrospinal fluid. In an effort to provide an alternative fuel source for the brain, a ketogenic diet was started. The patient stopped having seizures within four days after beginning the diet, and the seizures did not recur after clonazepam was discontinued at the age of 11.5 months or after phenobarbital was discontinued at the age of 14 months. Neurologic development was mildly delayed. He began sitting at the age of 10.5 months, pulling to stand and cruising at 11 months, and walking at 16 months. At 27 months, his vocabulary was limited to 10 to 15 words, and occasionally he put two words together in a simple phrase.

The parents were of Italian descent, and there was no known consanguinity. The mother and two older sisters of the proband had microcephaly, with head circumferences below the third percentile, but they were neurologically normal.

Table 1. Blood and Cerebrospinal Fluid Values for Glucose and Lactate in Two Children with Defective Glucose Transport across the Blood–Brain Barrier.*

AGE	BLOOD GLUCOSE	BLOOD LACTATE	CSF GLUCOSE	CSF LACTATE†	CSF GLUCOSE:BLOOD GLUCOSE‡
mo		*millimoles per liter*			
Patient 1					
2.5	5.5	—	1.6	—	0.29
4.5	4.7	—	0.88	0.41	0.19
6.0	4.8	—	1.06	—	0.22
7.5	5.8	1.3	1.89	0.31	0.33
Patient 2					
5.5	—	—	1.4	1.0	—
5.8	6.7	—	1.6	1.3	0.24
6.0	4.6	1.0	1.5	0.9	0.33
9.0	9.4§	2.4	1.8	1.5	0.19
17.5	5.2	1.3	1.8	1.2	0.35
27.8	5.5	1.4	1.7	1.3	0.31

*CSF denotes cerebrospinal fluid.
†Normal values are 1.0 to 2.8 mmol per liter.
‡Normal value is 0.65.
§Measured two hours after an infusion of 15 percent glucose and an injection of glucagon.

De Vivo DC, Trifiletti RR, Jacobson RI, Ronen GM, Behmand RA, Harik, SI: NEJM 1991

nature neuroscience

GLUT1 reductions exacerbate Alzheimer's disease vasculo-neuronal dysfunction and degeneration

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The glucose transporter GLUT1 at the blood-brain barrier (BBB) mediates glucose transport into the brain. Alzheimer's disease is characterized by early reductions in glucose transport associated with diminished GLUT1 expression at the BBB. Whether GLUT1 reduction influences disease pathogenesis remains, however, elusive. Here we show that GLUT1 deficiency in mice overexpressing amyloid β-peptide (Aβ) precursor protein leads to early cerebral microvascular degeneration, blood flow reductions and dysregulation and BBB breakdown, and to accelerated amyloid β-peptide (Aβ) pathology, reduced Aβ clearance, diminished neuronal activity, behavioral deficits, and progressive neuronal loss and neurodegeneration that develop after initial cerebrovascular degenerative changes. We also show that GLUT1 deficiency in endothelium, but not in astrocytes, initiates the vascular phenotype as shown by BBB breakdown. Thus, reduced BBB GLUT1 expression worsens Alzheimer's disease cerebrovascular degeneration, neuropathology and cognitive function, suggesting that GLUT1 may represent a therapeutic target for Alzheimer's disease vasculo-neuronal dysfunction and degeneration.

The glucose transporter GLUT1, encoded by *SLC2A1*, mediates glucose transport into the brain[1,2]. GLUT1 is expressed at the BBB but not in neurons[1,2]. GLUT1 exists in two isoforms: a 55-kDa isoform in brain endothelial cells and a 45-kDa isoform in adjacent astrocyte endfoot processes[3]. The crystal structure of human GLUT1 has been recently reported[4]. Brain glucose uptake correlates with GLUT1 levels in cerebral microvessels[5–7]. GLUT1 deficiency is found in patients with epilepsy, movement disorders and cognitive impairment[8]. In mice, GLUT1 haploinsufficiency (*Slc2a1*$^{+/-}$) diminishes cerebrospinal fluid (CSF) glucose levels and leads to microcephaly[9,10]. In the zebrafish, GLUT1 is required for the formation of the BBB[11], raising the possibility of a dual role similar to that of the major facilitator family domain–containing-2a (MFSD2A) transporter, which transports essential fatty acids across the BBB into the brain and regulates BBB integrity[12–14].

Dementia due to Alzheimer's disease is characterized by progressive metabolic disturbances[15], neurovascular dysfunction[1,2] and BBB breakdown[16]. Diminished glucose uptake in the hippocampus, parietotemporal cortex and/or posterior cingulate cortex has been shown by 2-[^{18}F]fluoro-2-deoxy-D-glucose positron emission tomography (FDG-PET) in individuals who are at genetic risk for Alzheimer's disease[17,18], have a positive family history[19], and/or have mild or no cognitive impairment but go on to develop Alzheimer's disease[20,21]. FDG-PET changes precede brain atrophy and neuronal dysfunction in humans[15,22] and transgenic Alzheimer's disease models[23], which may reflect reductions in BBB glucose transport[24,25]. Reduced GLUT1 levels in cerebral microvessels were also found in Alzheimer's disease[26–28]. Whether GLUT1 reductions can lead to cerebrovascular damage contributing to and/or accelerating Alzheimer's disease–like neurodegeneration remains, however, elusive.

To address this question, we crossed transgenic mice overexpressing human amyloid β-peptide (Aβ) precursor protein (*APP*$^{Swe/Ind}$)[29] with GLUT1-deficient *Slc2a1*$^{+/m}$ mice[9]. We also used conditional *Slc2a1*$^{flox/flox}$ mice[30] to determine the effects of cell-specific GLUT1 deletions from endothelium and astrocytes on the BBB phenotype. We found that GLUT1 was necessary for the maintenance of proper brain angioarchitecture, cerebral blood flow (CBF) and BBB integrity, and that GLUT1 reductions in *APP*$^{Swe/Ind}$ mice accelerated Aβ accumulation and led to progressive neuronal dysfunction, behavioral deficits, neuronal loss and neurodegeneration that developed after initial cerebrovascular changes. We also show that GLUT1 deficiency in endothelium, but not in astrocytes, initiates the BBB breakdown. Our data suggest that GLUT1 reductions at the BBB have an early pathogenic action in neuronal demise in an Alzheimer's disease–like neurodegenerative process.

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Boston Children's Hospital
Until every child is well™

GLUT1 Deficiency

- 3 “phenotypes” appear to be an ontogenic spectrum
  - 1. “Classic”: neonatal seizures, microcephaly
  - 2. Infancy: Delay, dysarthria, dystonia
  - 3. Later: Choreoathetosis, dystonia, paroxysmal exertional dyskinesias
- CSF glc < 40-60; CSF/serum < 0.4 X 3
- MRI: T2 hyperintensities, subcort U fibers
- SLC2A1 mutations in 10% early onset absence, and in MAE of Doose

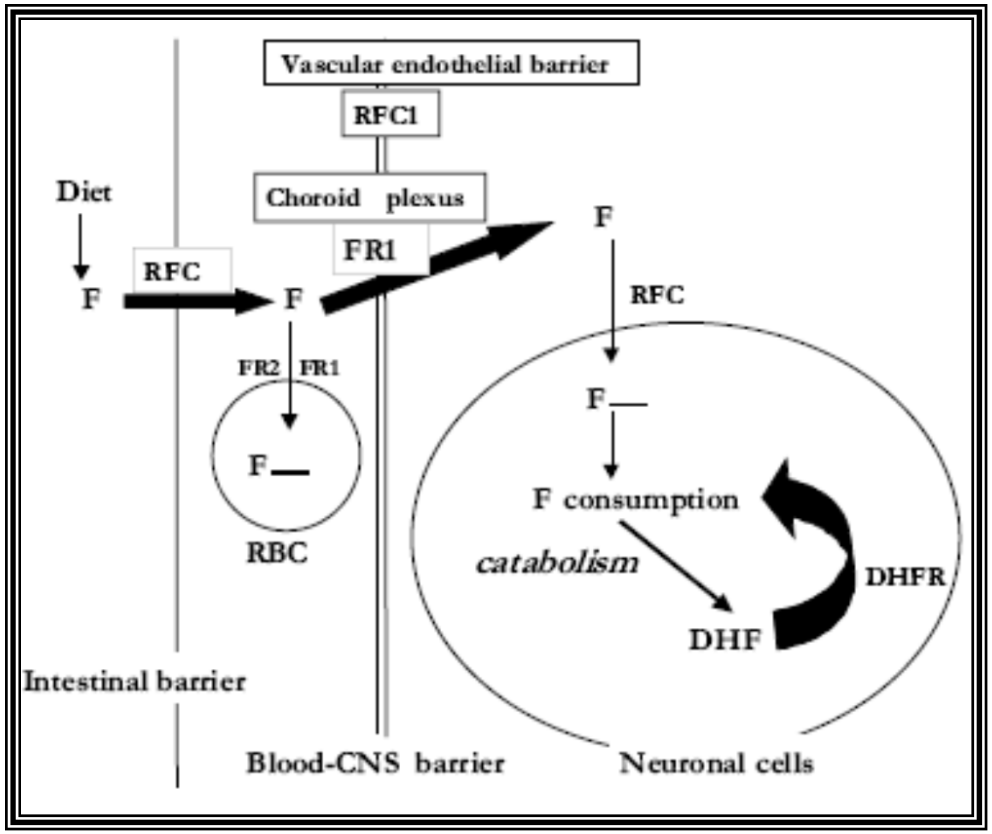
GLUT1 DS: Treatment

- Ketogenic diet, carnitine (to augment diet)
- Alpha lipoic acid
  - augments GLUT1 function
- Avoid PBS, DZP, Cl hydrate, VPA
  - inhibit GLUT1 function

Secondary causes of Hypoglycorrhachia

- Meningitis (esp bacterial, TB)
- Status epilepticus
- Mitochondrial disorders
- Systemic hypoglycemia
- Subarachnoid hemorrhage
- Meningeal carcinomatosis

FRI Mediated Endocytosis Across the BBB



Manifestations: Cerebral Folate Deficiency

- Infantile onset (4-6 months)
  - Irritability
  - Decelerating head growth
  - Seizures
  - Psychomotor retardation
  - Cerebellar ataxia
  - Pyramidal tract signs
  - Ballismus, choreoathetosis
- After three years of age:
  - Optic atrophy
  - Cortical blindness

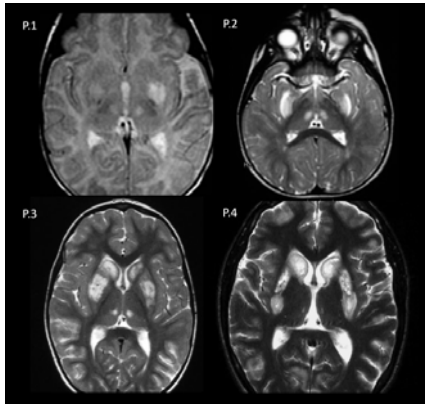
Differential Diagnosis of CSF 5MTHF

- 1. FOLR1 mutations, blocking/binding Abs
- 2. 5,10-MTHFR deficiency
- 3. 3-phosphoglycerate dehydrogenase def.
- 4. DHFR/DHPR def. (BH4 synthesis/recycling)
- 5. Hereditary folate malabsorption
- 6. Rett, Aicardi-Goutieres, mitochondrial (KSS)
- 7. Drugs, e.g. valproate
- 8. Deficiency dietary intake
- 9. Proton-coupled folate transporter 1 (PCFT1) deficiency
- 10. KCNH1 mutations

Thiamine Transporter-2 Deficiency

- Formerly biotin responsive basal ganglia disease
- SLC19A3 mutations
- Acute encephalopathy, dystonia, seizures
  - Ozand et al: Biotin-responsive basal ganglia disease: a novel entity. Brain 1998.
  - Zeng et al: Biotin-responsive basal ganglia disease maps to 2q36.3 and is due to mutations in SLC19A3. Am J Hum Gen 2005.
  - Alfadhel et al: Biotin-responsive basal ganglia disease should be renamed biotin-thiamine-responsive basal ganglia disease. Orphanet J Rare Dis 2013.
  - Distelmaier et al: Biotin-responsive Basal Ganglia Disease: a treatable differential diagnosis of Leigh Syndrome. JIMD Rep 2014.

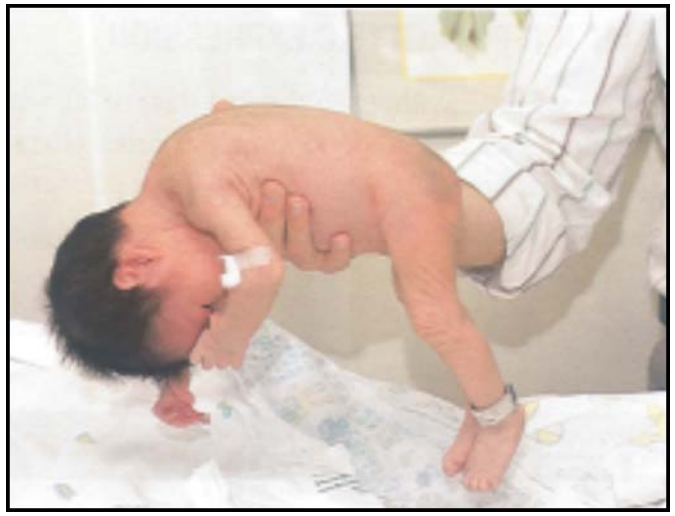
Figure: Ortigoza-Escobar et al: Thiamine transporter-2 deficiency: outcome and treatment monitoring. Orphanet J Rare Dis 2014.

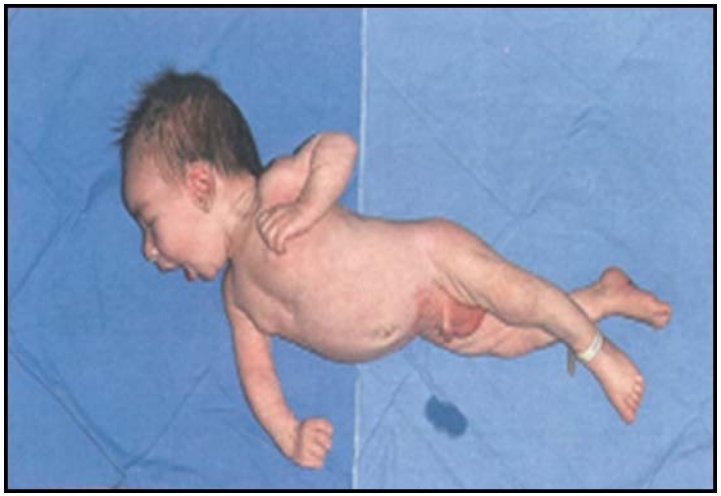


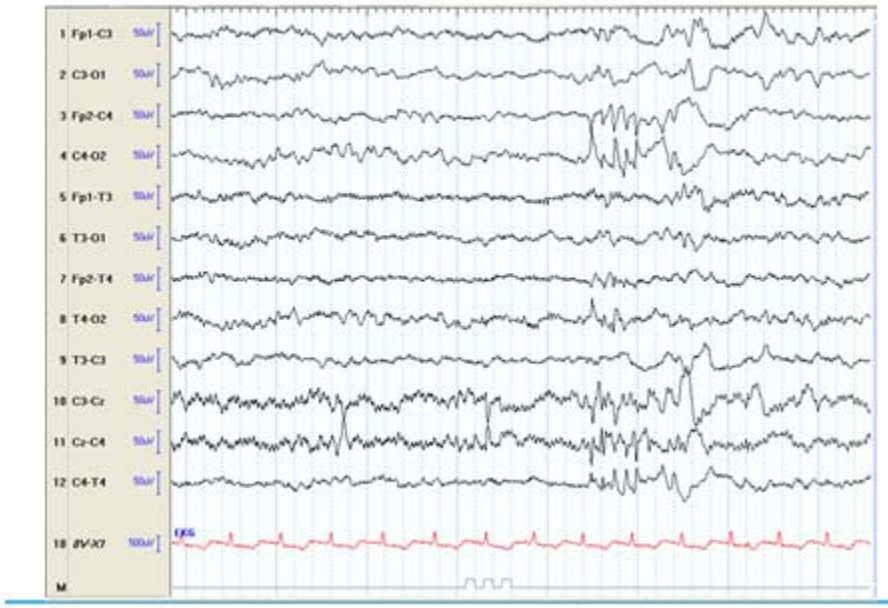
Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
  - Propionic, methylmalonic, isovaleric acidemias
  - Serine synthesis deficiencies
  - Creatine synthesis deficiencies
  - Many require rapid recognition to reduce ammonia, ICP, dietary restrictions. Assess toxic neonate with negative sepsis evaluation for lactic acid & NH3.
- Lysosomal Enzyme Disorders
- Mitochondrial Disorders
- Purine Metabolism Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

4 day old: ↓ feeding, crying, alternating flaccidity/opisthotonus

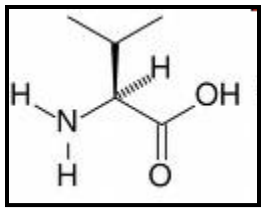




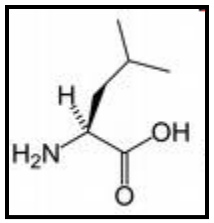


Maple Syrup Urine Disease

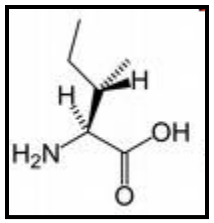
- Branched-chain ketoaciduria
  - Deficiency in branched-chain α-keto acid dehydrogenase (BCKD) complex
  - Accumulation of branched amino acids: leu, ile, val



valine

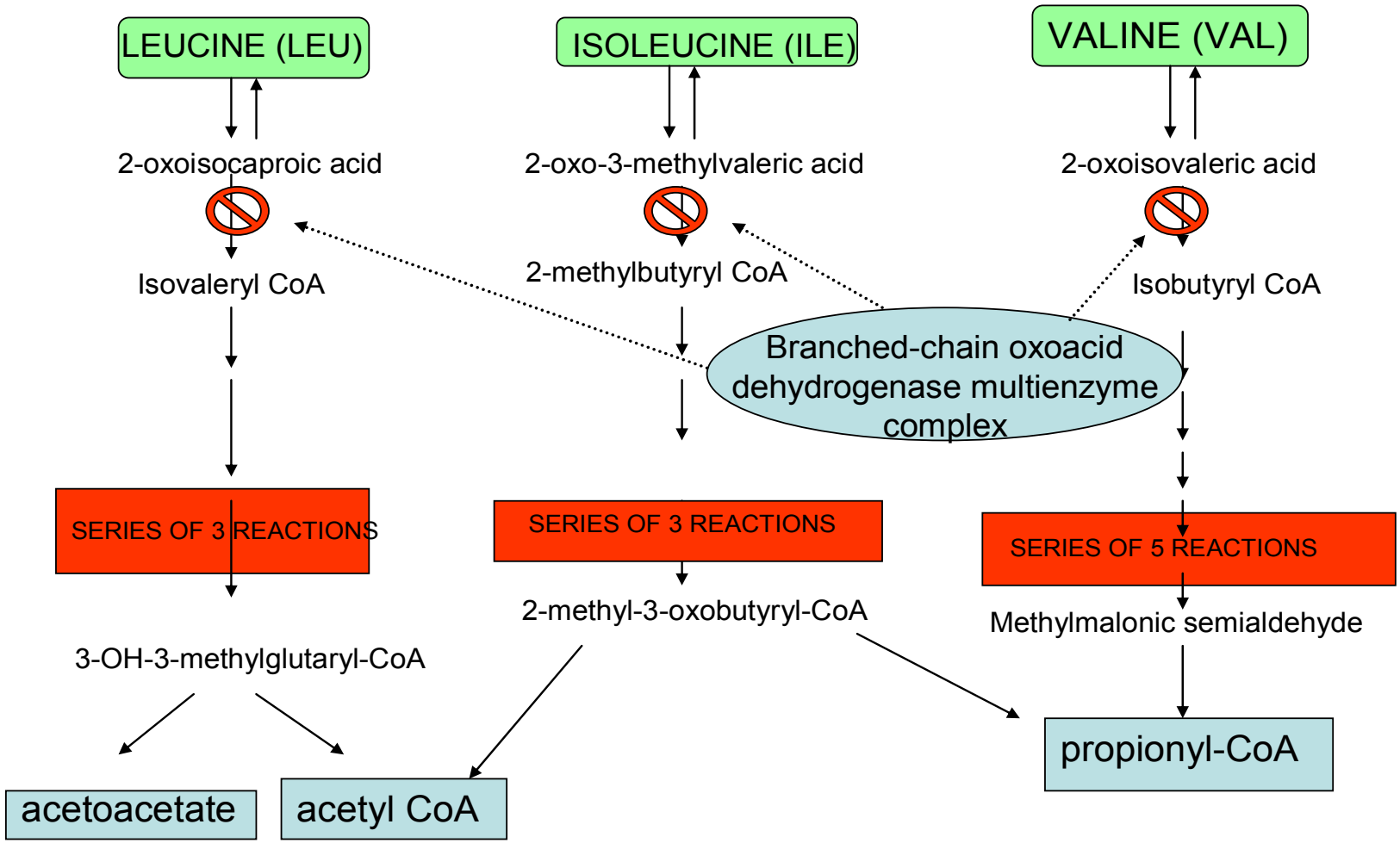


leucine



isoleucine

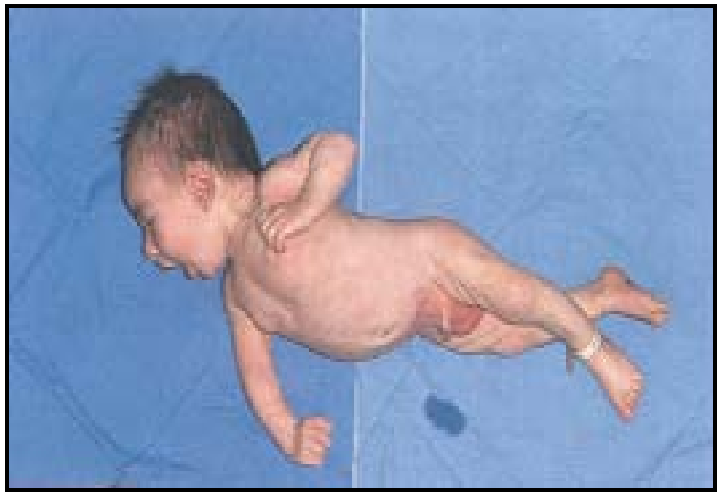
Maple Syrup Urine Disease: Branched Chain Amino Acid (BCAA) Pathways:

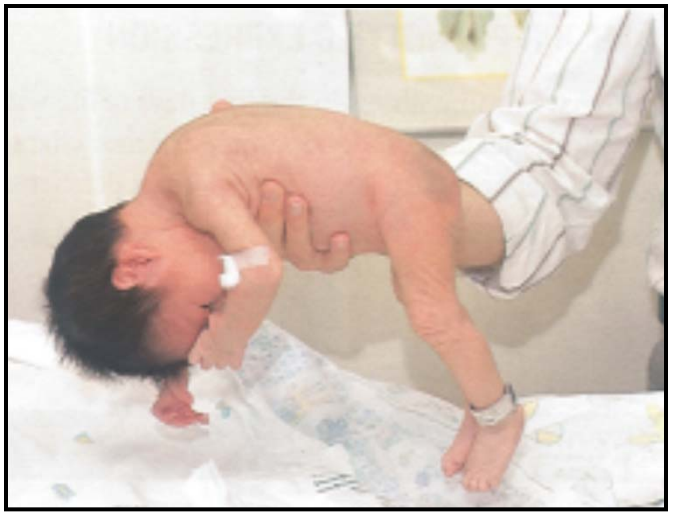


Major Phenotype

- Overwhelming illness in first days of life: lethargy to coma
- Opisthotonus
- Convulsions
- Recurrent episodes

Two patients with MSUD





From: Atlas of Metabolic Diseases 2005

Teenage girl with MSUD

From: Atlas of Metabolic Diseases 2005

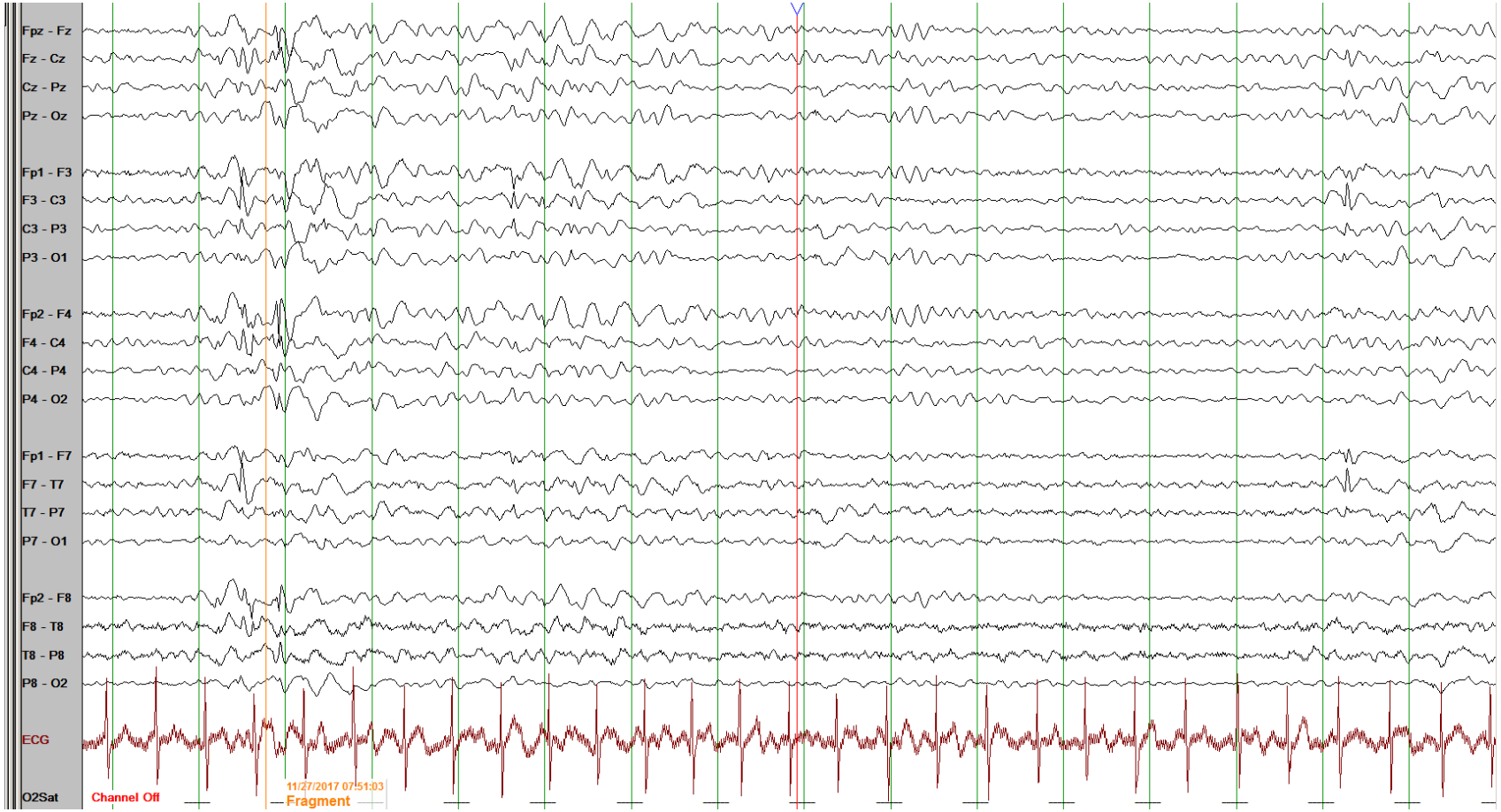
Categories of Inherited Metabolic Epilepsies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Lysosomal Storage Diseases
- Mitochondrial Disorders
- Purine Metabolism Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

Neuronal Ceroid Lipofuscinoses

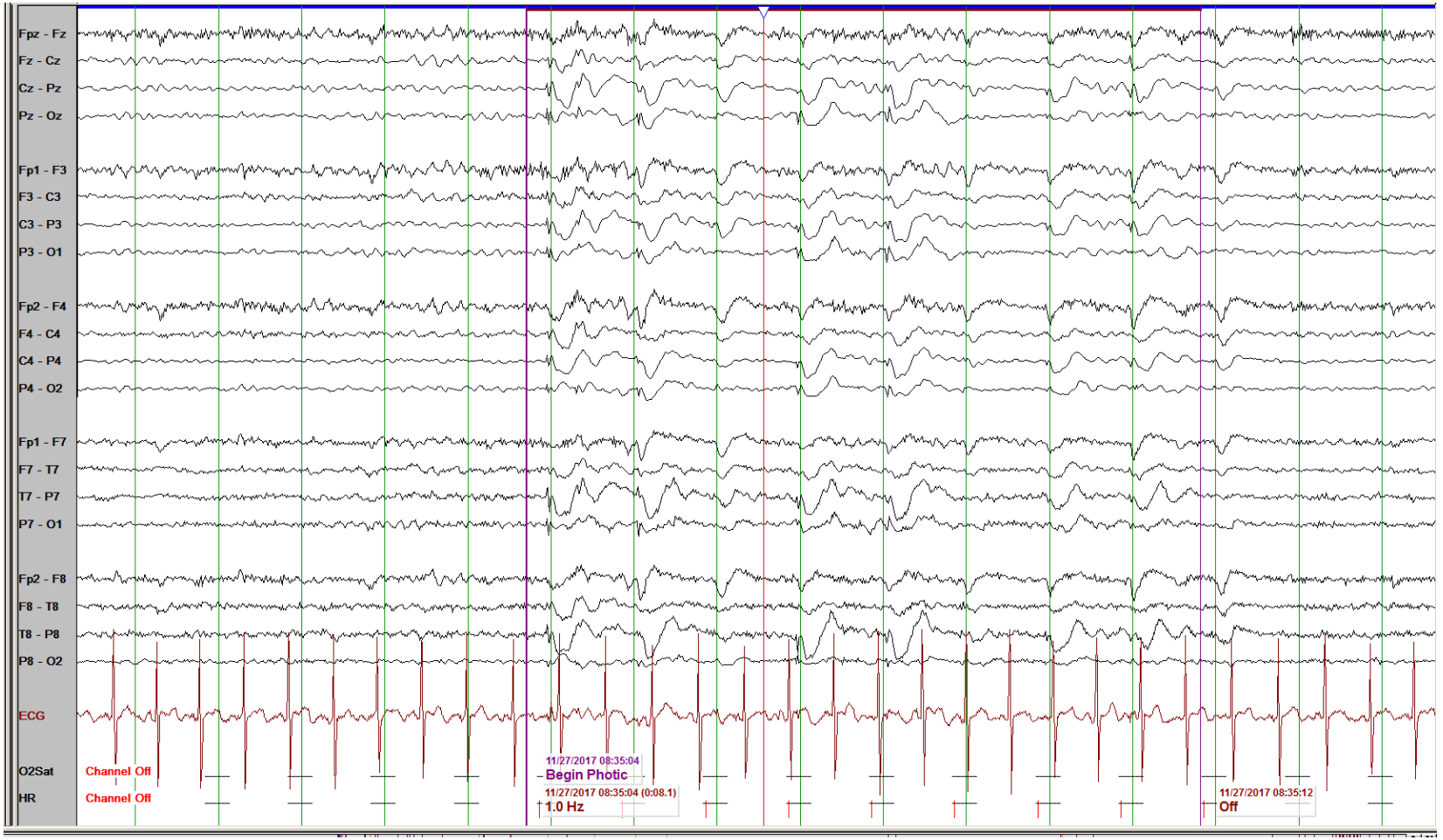
- 13 reported genes, 160+ mutations
- Late infantile NCL2: CLN2
- TPP1 gene
- Deficiency of tripeptidyl peptidase 1
- Onset 2-4 years
- Dementia, seizures, visual impairment
- Seizures: GTCS, atonic, astatic, myoclonic, absence, focal onset
- Action myoclonus

Interictal – CLN2



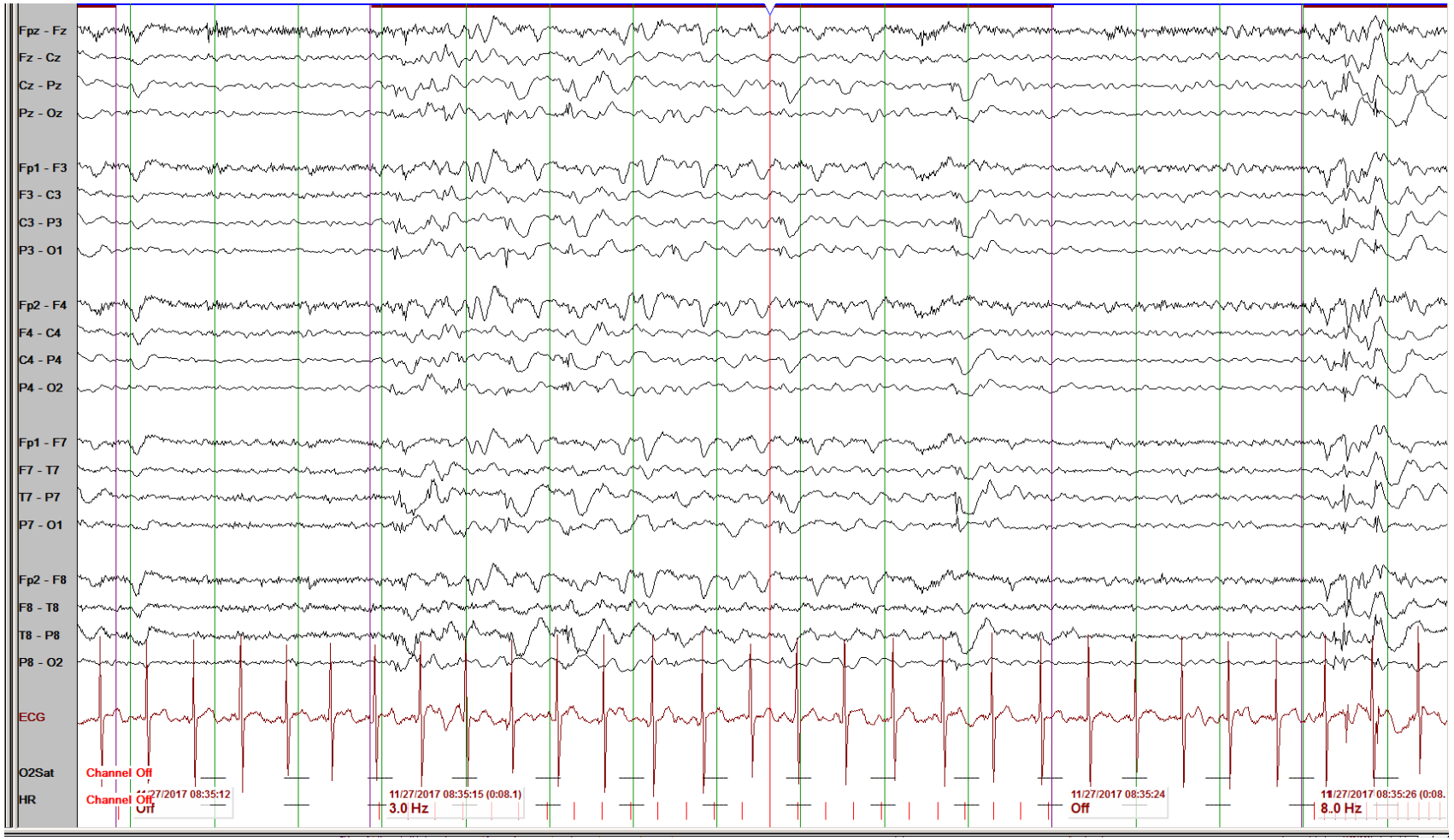
EEG courtesy of Elaine Wirrell MD, Mayo Clinic

1 Hz PPR



EEG courtesy of Elaine Wirrell MD, Mayo Clinic

3 Hz PPR



EEG courtesy of Elaine Wirrell MD, Mayo Clinic

Newly approved Treatment for CLN2/LINCL

Recombinant human TPP1

Cerliponase alfa

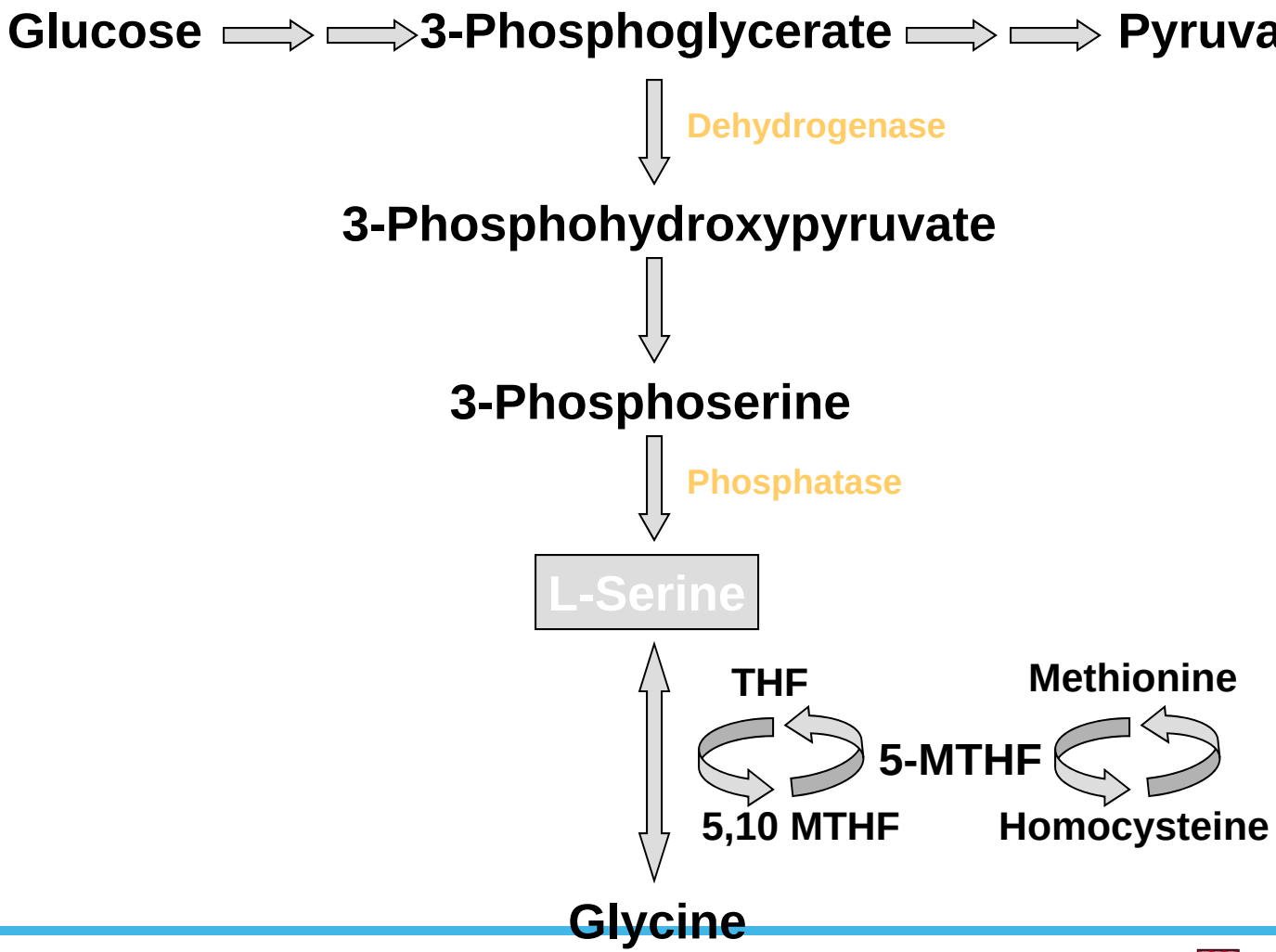
Rx Brineura, FDA approved 2017

Intraventricular delivery

Efficacy studied in 22 patients 3-8 years old.

Fewer declined in walking ability compared to natural history study.

Congenital microcephaly, neonatal seizures, infantile spasms

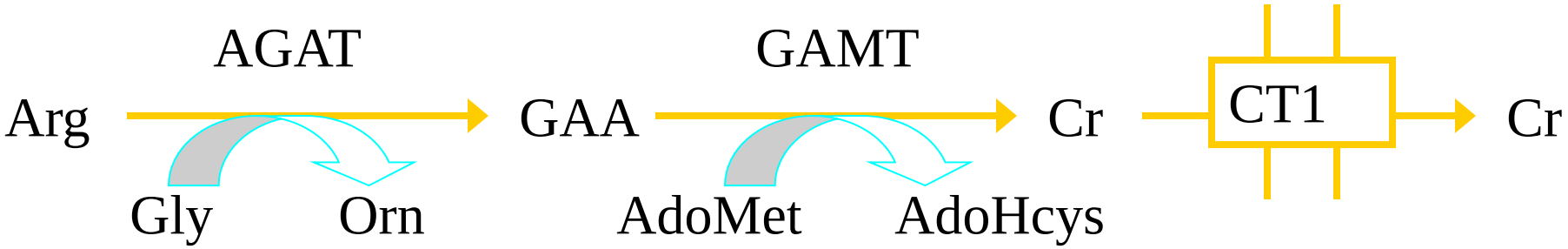


Serine Biosynthesis Disorder

- Low CSF and (fasting) plasma serine
- Treatable with serine supplementation (400-600 mg/kg/day) and glycine (200-300 mg/kg/day).
- Normal outcome with pre- and post-natal Rx
  - De Koning, T.J. et al (2004).

Creatine Synthesis/Transport

Check plasma/urine creatine and GAA.



AGAT: *Arginine:Glycine Aminidotransferase*

GAMT: *Guanidinoacetate* N-*Methyltransferase*

Metabolic Disorders of Creatine

- First described in 1994: GAMT deficiency.
- GAA level: ↑ GAMT; ↓ AGAT; nl transporter defect
- Rx: creatine (GAMT, AGAT deficiencies); arginine restriction, ornithine supplementation (GAMT)
- Normalization of outcome in presymptomatic neonatal intervention (Schulze, Hoffmann, Bachert et al. Neurology 2006)

Clinical Symptoms in Disorders of Creatine Metabolism

	GAMT	AGAT	Creatine Transporter 1
Reduced somatic growth		X	
Early developmental delay	X	X	
Neurologic regression	X		
Intellectual deficiency	X	X	X
Autistic behavior	X		
Hypotonia	X		X
Epilepsy	X	X	X
Movement disorder	X		
MRI: abnormal pallidal signal	X		

Categories of Inherited Metabolic Epilepsies

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- Amino and Organic Acid Disorders
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- Mitochondrial Disorders
- Purine Metabolism Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

New hope for Mb cofactor deficiency

- Mb dependent enzymes
  - Sulfite oxidase, xanthine oxidase, nitrate reductase, nitrogenases
  - Type A: lack cyclic pyranopterin monophosphate (cPMP)
- Early presentation: EIEE
- Later presentation: GDD
- Laboratory: decreased uric acid, + urine sulfites, elevated U S-sulfocysteine, xanthine, hypoxanthine

Efficacy and safety of cyclic pyranopterin monophosphate substitution in severe molybdenum cofactor deficiency type A: a prospective cohort study

Bernd C Schwahn*, Francjan J Van Spronsen*, Abdel A Belaidi, Stephen Bowhay, John Christodoulou, Terry G Derks, Julia B Hennermann, Elisabeth Jameson, Kai König, Tracy L McGregor, Esperanza Font-Montgomery, José A Santamaria-Araujo, Saikat Santra, Mamta Vaidya, Anne Vierzig, Evangeline Wassmer, Ilona Weis, Flora Y Wong, Alex Veldman*, Günter Schwarz*

Summary

Background Molybdenum cofactor deficiency (MoCD) is characterised by early, rapidly progressive postnatal encephalopathy and intractable seizures, leading to severe disability and early death. Previous treatment attempts have been unsuccessful. After a pioneering single treatment we now report the outcome of the complete first cohort of patients receiving substitution treatment with cyclic pyranopterin monophosphate (cPMP), a biosynthetic precursor of the cofactor.

Methods In this observational prospective cohort study, newborn babies with clinical and biochemical evidence of MoCD were admitted to a compassionate-use programme at the request of their treating physicians. Intravenous cPMP (80–320 μg/kg per day) was started in neonates diagnosed with MoCD (type A and type B) following a standardised protocol. We prospectively monitored safety and efficacy in all patients exposed to cPMP.

Findings Between June 6, 2008, and Jan 9, 2013, intravenous cPMP was started in 16 neonates diagnosed with MoCD (11 type A and five type B) and continued in eight type A patients for up to 5 years. We observed no drug-related serious adverse events after more than 6000 doses. The disease biomarkers urinary S-sulphocysteine, xanthine, and urate returned to almost normal concentrations in all type A patients within 2 days, and remained normal for up to 5 years on continued cPMP substitution. Eight patients with type A disease rapidly improved under treatment and convulsions were either completely suppressed or substantially reduced. Three patients treated early remain seizure free and show near-normal long-term development. We detected no biochemical or clinical response in patients with type B disease.

Interpretation cPMP substitution is the first effective therapy for patients with MoCD type A and has a favourable safety profile. Restoration of molybdenum cofactor-dependent enzyme activities results in a greatly improved neurodevelopmental outcome when started sufficiently early. The possibility of MoCD type A needs to be urgently explored in every encephalopathic neonate to avoid any delay in appropriate cPMP substitution, and to maximise treatment benefit.

Funding German Ministry of Education and Research; Orphatec/Colbourne Pharmaceuticals.

Introduction

Human molybdenum cofactor deficiency (MoCD) usually manifests during the first few postnatal days with exaggerated startle reactions, alterations in muscle tone, lethargy, intractable seizures, and autonomic dysfunction.[1] At the onset of clinical symptoms, brain imaging reveals global white matter and deep grey matter involvement, followed by rapidly evolving widespread subcortical necrosis. Multicystic lesions appear within days, with subsequent brain atrophy and secondary microcephaly.[2] Some patients display developmental anomalies, such as brain malformations or mildly dysmorphic facial features at birth. Surviving patients can develop marfanoid features and lens dislocation.[3,4] Milder phenotypes with late onset of symptoms have been described.[5] Symptoms are predominantly caused by sulphite toxicity due to a functional loss of sulphite oxidase, one of four molybdenum cofactor-dependent enzymes in human beings. Treatment of severely affected children is supportive. Encephalopathy and intractable epilepsy often cause severe distress to patients and carers.

Molybdenum cofactor is synthesised by a complex biosynthetic pathway involving three steps (figure 1) that we have described in detail elsewhere.[6,7] The true incidence of MoCD is not known, but is estimated at between one in 100 000 and one in 200 000 newborn babies worldwide.[1] Around two-thirds of patients with MoCD have mutations in *MOCS1*, resulting in the inability to synthesise the first intermediate in the biosynthetic pathway, cyclic pyranopterin monophosphate (cPMP), classified as MoCD type A.[1] Almost all remaining patients have a defect in *MOCS2*, resulting in the accumulation of cPMP that cannot be further used, and are classified as MoCD type B.[8–10] MoCD type A and B and the very rare type C (associated with mutations in *GPHN*) are autosomal recessively transmitted, and are clinically indistinguishable.

After successful preclinical studies in an animal model for MoCD,[11] we reported the initial treatment

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See Comment page 1924

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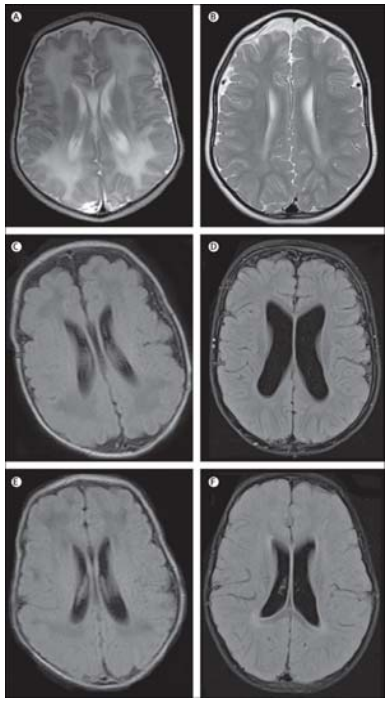
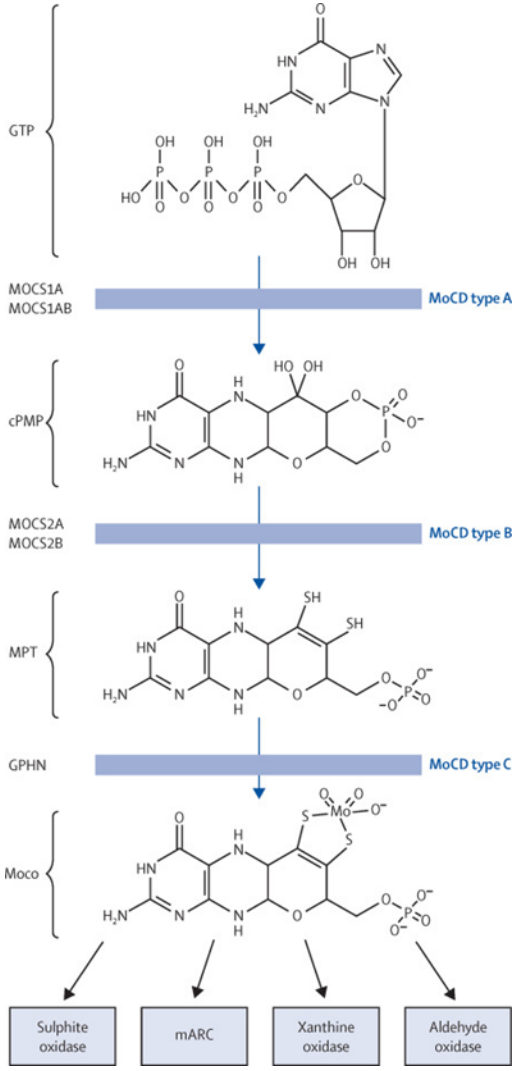


Top: 3 days; 37 mos
Middle: 5 h; 47 mos
Bottom: 4 h; 24 mos

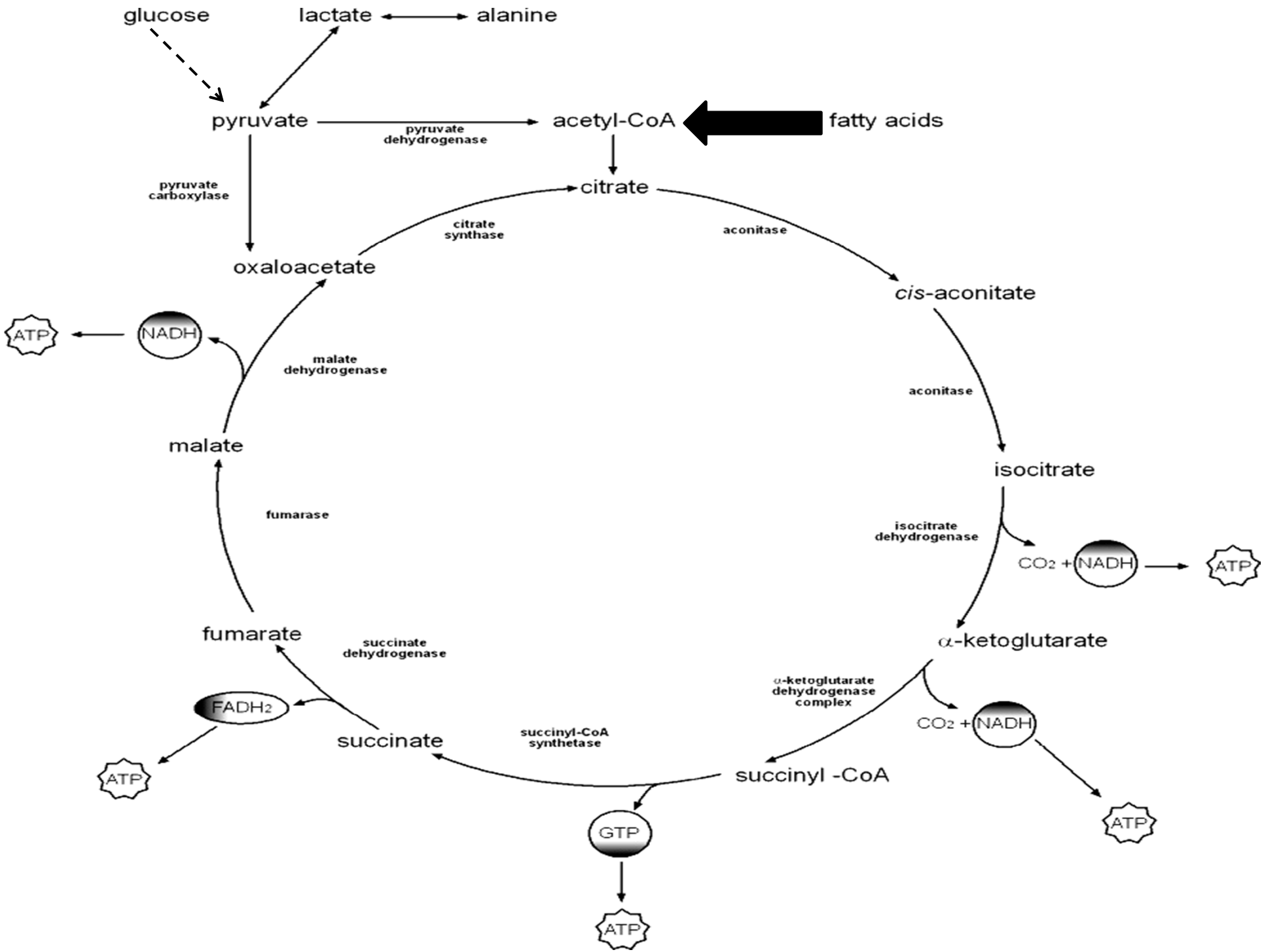


Boston Children's Hospital
Until every child is well

HARVARD MEDICAL SCHOOL TEACHING HOSPITAL

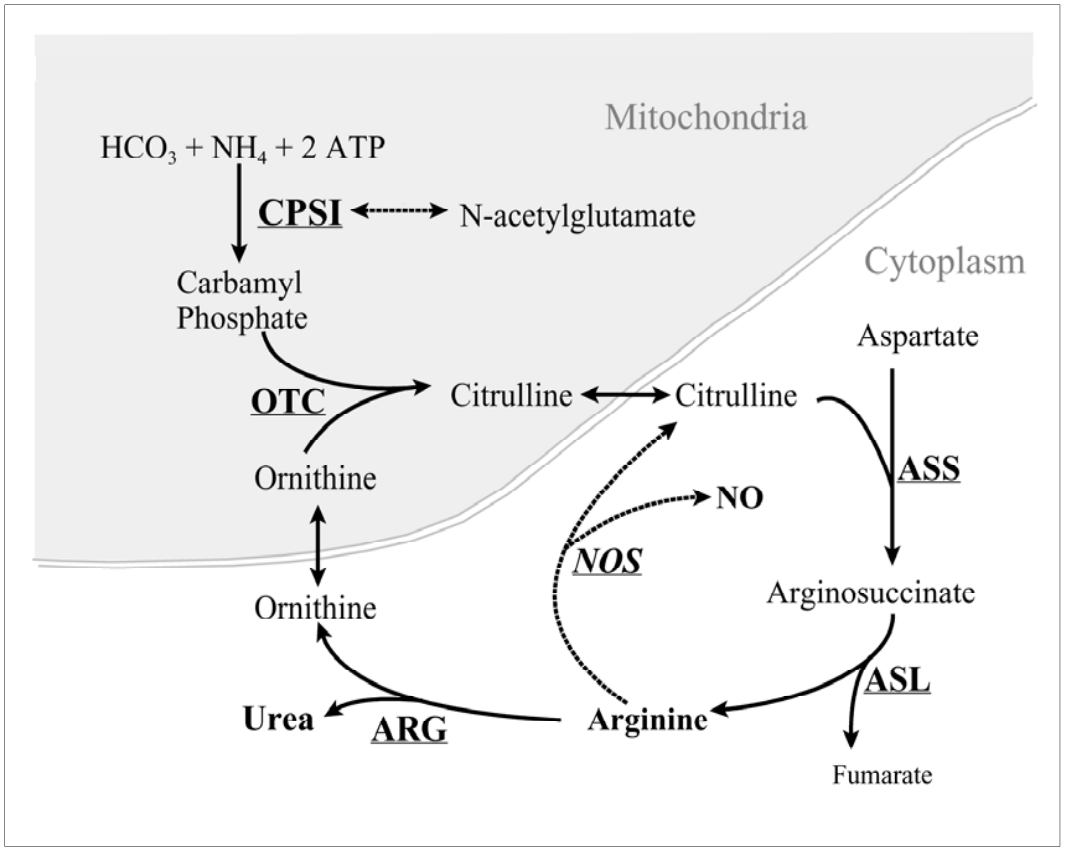
Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
  - Pyruvate dehydrogenase deficiency (Rx – ketogenic diet)
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis


glucose
lactate
alanine
pyruvate
pyruvate dehydrogenase
acetyl-CoA
fatty acids
pyruvate carboxylase
citrate
citrate synthase
aconitase
oxaloacetate
cis-aconitate
ATP
NADH
malate dehydrogenase
aconitase
malate
isocitrate
fumarase
isocitrate dehydrogenase
CO2 + NADH
ATP
fumarate
α-ketoglutarate
succinate dehydrogenase
α-ketoglutarate dehydrogenase complex
FADH2
CO2 + NADH
succinate
succinyl-CoA synthetase
ATP
succinyl -CoA
ATP
GTP
ATP

Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
- Urea Cycle Disorders
  - Na benzoate/Na phenylacetate, dialysis
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis


Mitochondria
$HCO_3 + NH_4 + 2$ ATP
CPSI
N-acetylglutamate
Carbamyl Phosphate
Cytoplasm
Aspartate
OTC
Citrulline
Citrulline
ASS
Ornithine
NO
NOS
Ornithine
Arginosuccinate
ASL
Urea
ARG
Arginine
Fumarate

Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis

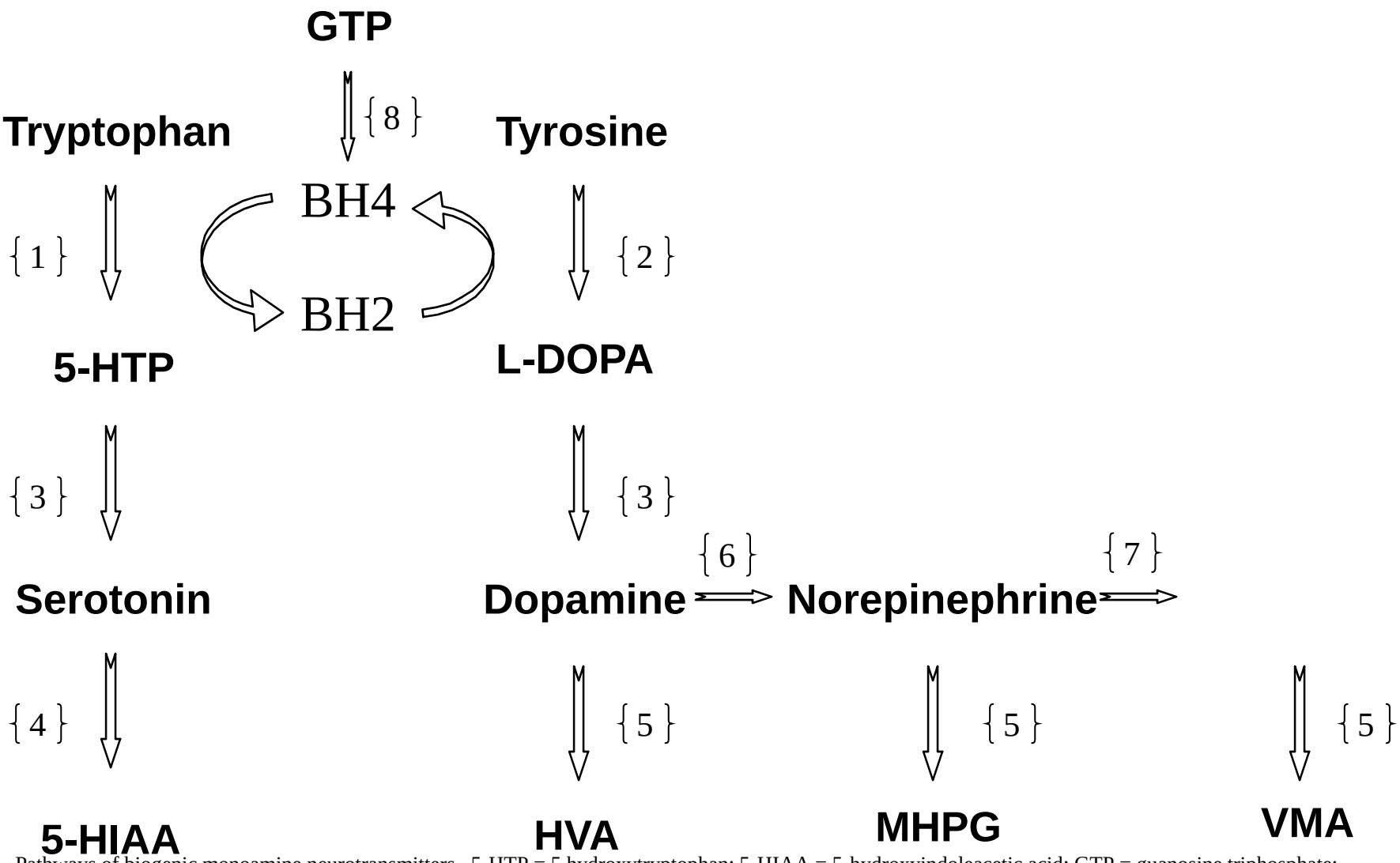
Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
  - BH4, monoamine precursors, MAO-inhibitors
- Disorders of Glucose Homeostasis

Disorders of Recycling or Synthesis of BH_4

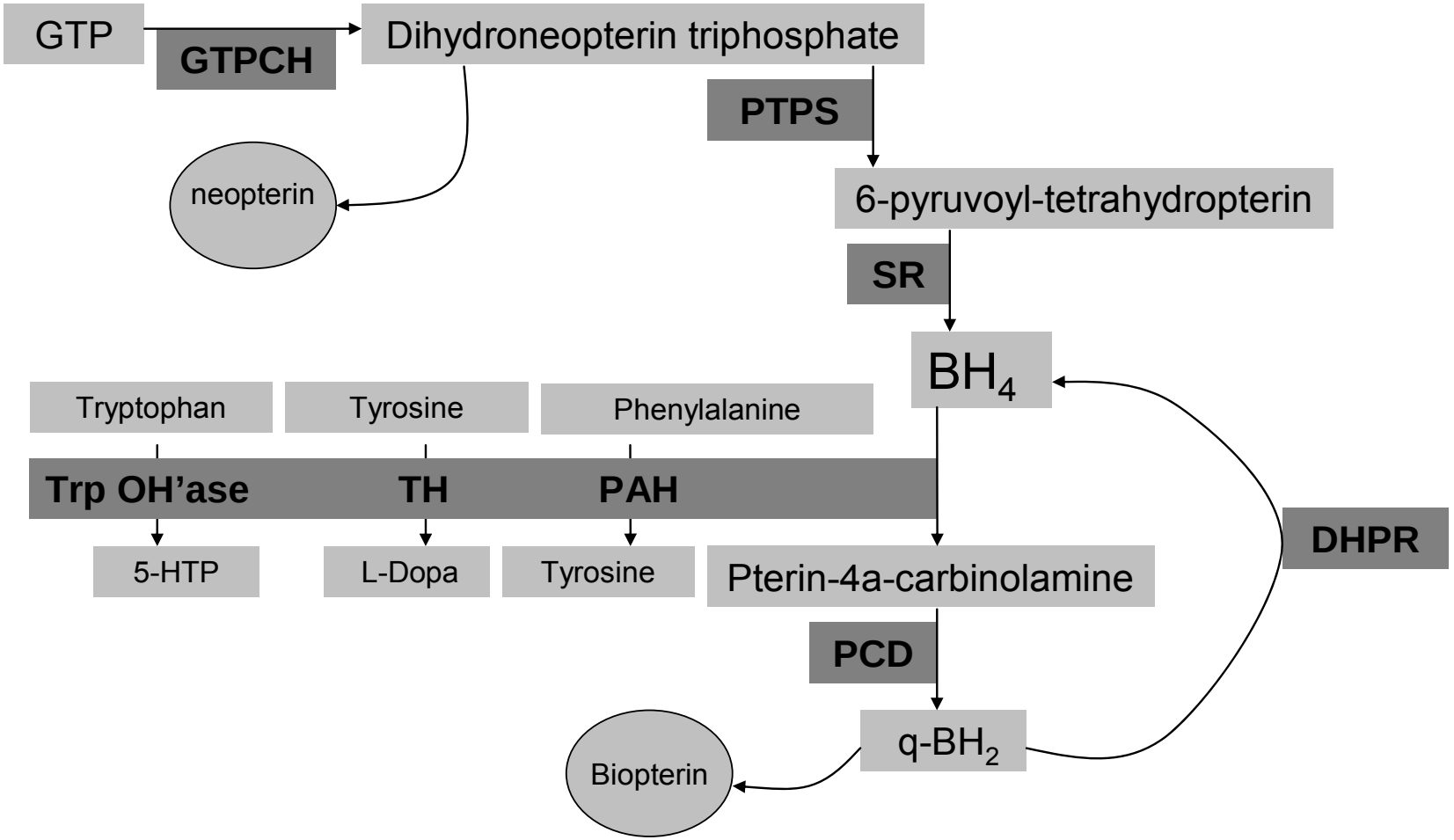
- Usually diagnosed due to hyper-phenylalanine on the newborn screen.

- Some of these conditions are associated with normal blood [phe].
  - Evaluation for a disorder in the BH4 pathway should be done in infants with unexplained neurologic disease.



Pathways of biogenic monoamine neurotransmitters. 5-HTP = 5 hydroxytryptophan; 5-HIAA = 5-hydroxyindoleacetic acid; GTP = guanosine triphosphate; BH4 = tetrahydrobiopterin; BH2 = quinonoid dihydrobiopterin; L-DOPA = levodopa; HVA = homovanillic acid; MHPG = 3-methoxy-4-hydroxyphenylglycol; VMA = vanillylmandelic acid.{1}tryptophan hydroxylase;{2}tyrosine hydroxylase; {3}aromatic-L-amino acid decarboxylase; {4}monoamine oxidase; {5}monoamine oxidase, aldehyde dehydrogenase, catechol-O-methyltransferase; {6}dopamine β-hydroxylase; {7}phenylethanolamineN-methyltransferase; {8}GTP-cyclohydroxylase I

Tetrahydrobiopterin (BH_4) Metabolism

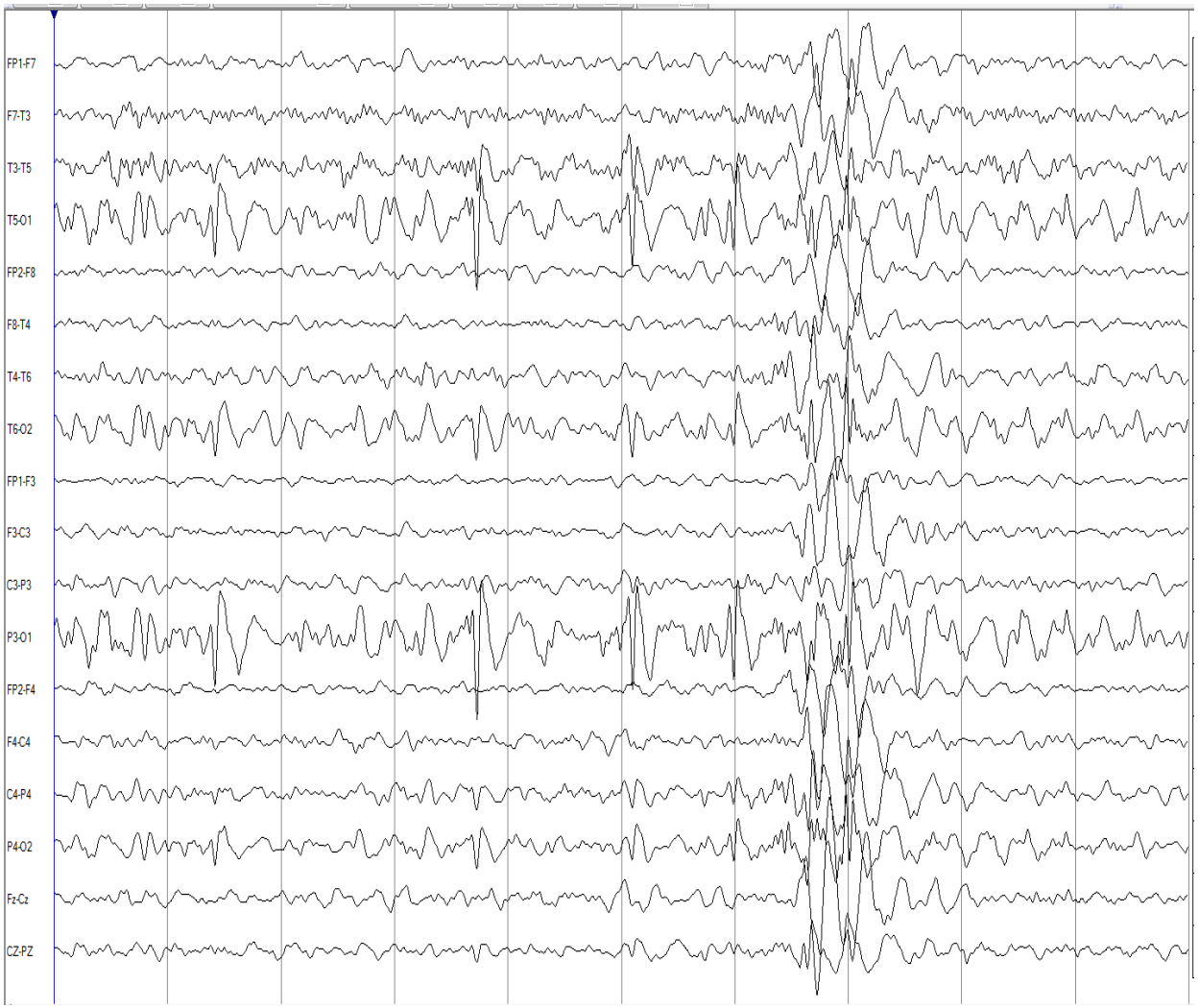


GTP = Guanine triphosphate; **GTPCH** = GTP Cyclohydrolase I; **PTPS** = 6-pyruvoyl-tetrahydropterin synthase; **SR** = Sepiapterin Reductase; **BH_4** = Tetrahydrobiopterin; **DHPR** = Dihydropterin Reductase; **Trp OH'ase** = Tryptophan Hydroxylase; **TH** = Tyrosine Hdyroxylase; **PAH** = Phenylalanine Hydroxylase; **5-HTP** = 5-Hydroxytryptophan; **PCD** = Pterin-carbinolamine Reductase; **q-BH_2** = q-Dihydrobiopterin.

Major Phenotype of BH_4 Disorders

- Intellectual disability
- (Myoclonic) seizures
- Muscular rigidity
- Dystonia
- Drooling
- Microcephaly
- Neuroimaging:
  - cerebral atrophy, lucency of the white matter, basal ganglia calcifications

7 yo girl w/DHPR deficiency, Rx delayed to 3 years of age



Acute but Reversible Severe Epileptic Encephalopathies

- Vitamin Responsive Disorders
- Transportopathies
- Amino and Organic Acid Disorders
- Mitochondrial Disorders
- Urea Cycle Disorders
- Neurotransmitter Disorders
- Disorders of Glucose Homeostasis
  - Neonatal diabetes – treat with sulfonylureas, not insulin
  - Congenital hyperinsulinism – HI/HA – treat with antiseizure medicines, diazoxide

Treatable Metabolic Epilepsies

- **Vitamin Responsive Disorders**
  - Pyridoxine, P5P, Folinic acid, Biotin
- **Transportopathies**
  - GLUT1, Cerebral Folate Deficiency, Thiamine Transporter
- **Amino and Organic Acid Disorders**
  - MSUD, propionic, isovaleric, cobalamin C
- **Lysosomal Storage Diseases**
  - LINCL/CLN2
- **Mitochondrial Disorders**
  - PDHC deficiency
- **Purine Synthesis Disorders**
  - MoCo deficiency type A
- **Urea Cycle Disorders**
  - OTC deficiency
- **Neurotransmitter Disorders**
  - BH4, GABA-transaminase
- **Disorders of Glucose Homeostasis**
  - DEND, HI/HA

Audience Response #3

- In a neonate with intractable seizures responsive to vitamin B6 with normal pipecolic acid and ALDH7A1 sequencing, which of the following is the most likely diagnosis?
- A. PNPO deficiency
- B. Biotinidase deficiency
- C. Carnitine deficiency
- D. Folate deficiency

ARS #3 - ANSWER

- In a neonate with intractable seizures responsive to vitamin B6 with normal pipecolic acid and ALDH7A1 sequencing, which is the most likely diagnosis?

* A. *PNPO deficiency*

B. Biotinidase deficiency

C. Carnitine deficiency

D. Folate deficiency

REF: Pearl, Hyland, Chiles et al: *Partial pyridoxine responsiveness in PNPO deficiency*. J Inherit Metab Dis 2013.

ARS #4

- Which of the following is true regarding late infantile neuronal ceroid lipofuscinosis (NCL, also known as CLN2)?
  - A. Presents as myoclonic seizures in a 2-4 year old child
  - B. EEG shows photoparoxysmal response at slow strobe rates (1-3 Hz)
  - C. Recombinant enzyme replacement therapy approved only as intraventricular administration
  - D. All of the above

ARS #4 - Answer

- Which of the following is true regarding CLN2?
  - A. Presents as myoclonic seizures in a 2-4 year old child
  - B. EEG shows photoparoxysmal response at slow strobe rates (1-3 Hz)
  - C. Recombinant enzyme replacement therapy approved only as intraventricular administration
  - *D. All of the above*

REF: de los Reyes et al, Pediatr Neurol 2020.

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Diagnosing Genetic Epilepsies – Impact on Treatment
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