Child Neurology Open
Volume 8: 1-7

DOI: 10.1177/2329048X20985179
journals.sagepub.com/home/cno
SAGE

The Application of Neurodiagnostic Studies to Inform the Acute Management of a Newborn Presenting With Carbamoyl Phosphate Synthetase 1 Deficiency

Meaghan McGowan, BA[1], Carlos Ferreira, MD[2], Matthew Whitehead, MD[3], Sudeepta K. Basu, MD[4], Taeun Chang, MD[5], and Andrea Gropman, MD[5]

Abstract

Neonatal-onset urea cycle disorders (UCDs) may result in hyperammonemic (HA) encephalopathy presenting with several neurologic sequelae including seizures, coma, and death. However, no recommendations are given in how and when neurodiagnostic studies should be used to screen or assess for these neurologic complications. We present a case of carbamoyl phosphate synthetase 1 (CPS1) deficiency in a newborn female in which electroencephalogram monitoring to assess encephalopathy and seizures, and magnetic resonance imaging measurements of brain metabolites were used to guide care during her hyperammonemic crisis. Her neurologic course and response to treatment characterizes the significant neurologic impact of HA encephalopathy. Our group herein proposes a clinical neurodiagnostic pathway for managing acute HA encephalopathy.

Keywords

carbamoyl phosphate synthetase, hyperammonemia, newborn, eeg, mri, mrs, seizures, urea cycle disorder

Received September 6, 2020. Received revised November 14, 2020. Accepted for publication December 6, 2020.

Introduction

The urea cycle is the body's primary biochemical route of nitrogen excretion and mutations affecting this pathway lead to elevated levels of ammonia (NH_3) and glutamine. Severe enzymatic deficits may result in hyperammonemic (HA) encephalopathy with possible seizures, coma, and even death.[1] They may occur across the lifespan with the most compromised enzyme activity presenting in the newborn period.

In neonatal-onset urea cycle disorders (UCDs), there is no standard guideline for using electroencephalogram (EEG) monitoring and magnetic resonance (MR) imaging to guide treatment and predict prognosis as are available for neonatal hypoxic ischemic encephalopathy.[2,3] Several small studies have demonstrated the potential of neurodiagnostic tools to expedite treatment and assess prognosis in newborns with UCDs.[4-6] However, no one has proposed a clinical protocol for appropriate neuromonitoring or seizure management of neonates with UCDs.

The true incidence of seizures and their relation to absolute ammonia level and clinical outcomes in UCDs remains unknown, but recent increase in access to bedside EEG monitoring has shown that seizures are common in UCDs, including subclinical seizures.[7] Whether seizure burden promotes additional brain injury has not been examined.

We propose that use of neurodiagnostic studies can inform and guide the acute management of neonatal-onset UCDs, improve our understanding of the neurologic disease progression of these

[1] University of Illinois College of Medicine, Chicago, IL, USA
[2] Medical Genomics and Metabolic Genetics Branch, National Human Genome Research Institute, NIH, Bethesda, MD, USA
[3] Neuroradiology, Children's National Hospital, George Washington University School of Medicine, Washington, DC, USA
[4] Neonatology, Children's National Hospital, George Washington University School of Medicine, Washington, DC, USA
[5] Neurology, Children's National Hospital, George Washington University School of Medicine, Washington, DC, USA

Corresponding Author:
Andrea Gropman, MD, Department of Neurology, Children's National Hospital, Washington, DC 20010, USA.
Email: agropman@childrensnational.org

newborns, and may protect the brain from additional injury. We present a case of neonatal-onset carbamoyl phosphate synthetase 1 (CPS1) deficiency co-managed between Genetics, Neonatal Neurocritical Care, and Neonatology after instating a neurodiagnostic pathway including prolonged continuous EEG monitoring and advance MR imaging to examine potential early proximal biomarkers of brain injury in UCD.

Case Presentation

This 5-day-old female infant born at 36 and 5/7 weeks gestational age was transferred to our neonatal intensive care unit (NICU) in hyperammonemic crisis. She was born via cesarean section to a mother with a history of polyhydramnios, pre-eclampsia, and diabetes mellitus and had a prenatal diagnosis of fetal ventriculoseptal defect. She remained in the nursery for conservative management of desaturations and hypoglycemia, and was discharged after resolution on day 3 of life. She presented to the outside emergency department the next day with lethargy, hypothermia, and decreased responsiveness and was found to have clinical seizures after admission. Her blood gases were normal, but her plasma ammonia level was exceedingly high, >925 μmol/L (normal <100 μmol/L), and so emergently transferred to our tertiary care NICU for management.

On admission in the NICU, she was encephalopathic, intubated, and her ammonia level was 1,677 μmol/L and glutamine level was 3,989 μmol/L. She was started on hemodialysis, intravenous sodium benzoate/sodium phenylacetate, and a continuous video EEG was placed (Figure 1). She required dopamine support for hypotension throughout the night, thus, arginine was not started. Arginine infusions can lead to hypotension. Since the child already had hypotension and required dopamine, arginine was withheld. By the next day, ammonia level decreased to 569 μmol/L and so she was transitioned to continuous renal replacement therapy.

On hospital day 3 (HD3), she experienced 10 multifocal electrographic seizures arising from the left central, right central, and left temporal regions and subsequently received a phenobarbital loading dose. Subsequent urine organic acids and plasma amino acid analysis revealed an absence of orotic acid and citrulline, respectively. She was later confirmed to have a homozygous deletion within *CPS1*, including exon 32. The ammonia continued to decrease to normal levels, and eventually she was transitioned to oral scavenger therapy.

On HD4, she had several refractory electrographic seizures and was started on fosphenytoin. The next day, her ammonia level rapidly increased to 1,293 μmol/L, with a concomitant increase in glutamine concentration to 620 μmol/L; she continued to have refractory electrographic only seizures. Hemodialysis and intravenous ammonia scavengers were resumed, and a midazolam infusion was initiated on HD6 to regain control of her seizures. Although her ammonia level improved with dialysis, her seizures persisted even with the addition of a fourth seizure medication, levetiracetam.

At 2 weeks of age, the patient underwent neuroimaging. MRI demonstrated diffuse white matter signal changes with reduced diffusion involving the corpus callosum, sagittal stratum, internal capsules, and frontal white matter and facilitated diffusion diffusely throughout the remainder of the cerebral white matter, extensive bilateral cerebral laminar necrosis, and heterogeneous cerebral blood flow consistent with a urea cycle disorder (Figure 2A-E). ^{1}H magnetic resonance spectroscopy (^{1}HMRS) demonstrated elevated lactate suggesting anaerobic metabolism, elevated glutamine (+glutamate, Glx), decreased myo-inositol, and decreased N-acetylaspartate (NAA). An abnormal peak was also noted at 2.8 ppm, most likely representing elevated aspartate, a breakdown product of NAA (Figure 2F-G). A repeat MRI/MRS was performed on day of life 18 with improved perfusion and spectroscopy indicating smaller Glx peaks (Figure 2H).

Despite stable ammonia levels (15-110 μmol/L) in the following weeks, she continued to have electrographic only seizures despite maximized phenobarbital, levetiracetam, and midazolam infusion. Because these breakthrough seizures likely represented ongoing cortical injury despite normalized ammonia levels rather than a primary seizure condition, anti-epileptic drugs (AEDs) were slowly weaned along with sodium benzoate/sodium phenylacetate. Her last electrographic seizure was on day of life 21, and she continues to receive maintenance doses of topiramate and levetiracetam while awaiting liver transplantation for definitive enzymatic correction.

Discussion

Hyperammonemia can be the first clinical presentation of an inborn error of metabolism including a urea cycle disorder or organic acidemia and can provoke irreversible damage to the developing central nervous system (CNS), leading to cognitive impairment, seizures and cerebral palsy.[8] Hyperammonemic neonates and infants develop cortical atrophy with ventricular enlargement.[8-10] The extent of the irreversible damages depends upon the maturation of the brain and on the magnitude and duration of the ammonia exposure.[11] Irreversibility mainly occurs in case of prolonged hyperammonemic crises and/or when blood ammonia reaches levels between 200 and 500 μM, during the 2 first years of life.[12-17]

It has previously shown that neonates with UCDs may present with seizures during HA.[7,18] However, most of the seizures are subclinical, and therefore only identified with prolonged continuous EEG. The mechanism of how hyperammonemia leads to the dysfunction of inhibitory neurotransmission has been explored in studies on astrocyte potassium buffering. It is proposed that the increase in brain glutamine and the resulting disturbed osmotic balance leads to impaired GABA-mediated neurotransmission.[19]

The pattern of metabolites in this patient in relation to seizure occurrence shown in Figure 1B is interesting to note several features. Seizures began after stable ammonia levels (72-130 μmol/L) were achieved with hemodialysis (HD 2-4), representing initial acute brain injury. Early re-emergence of

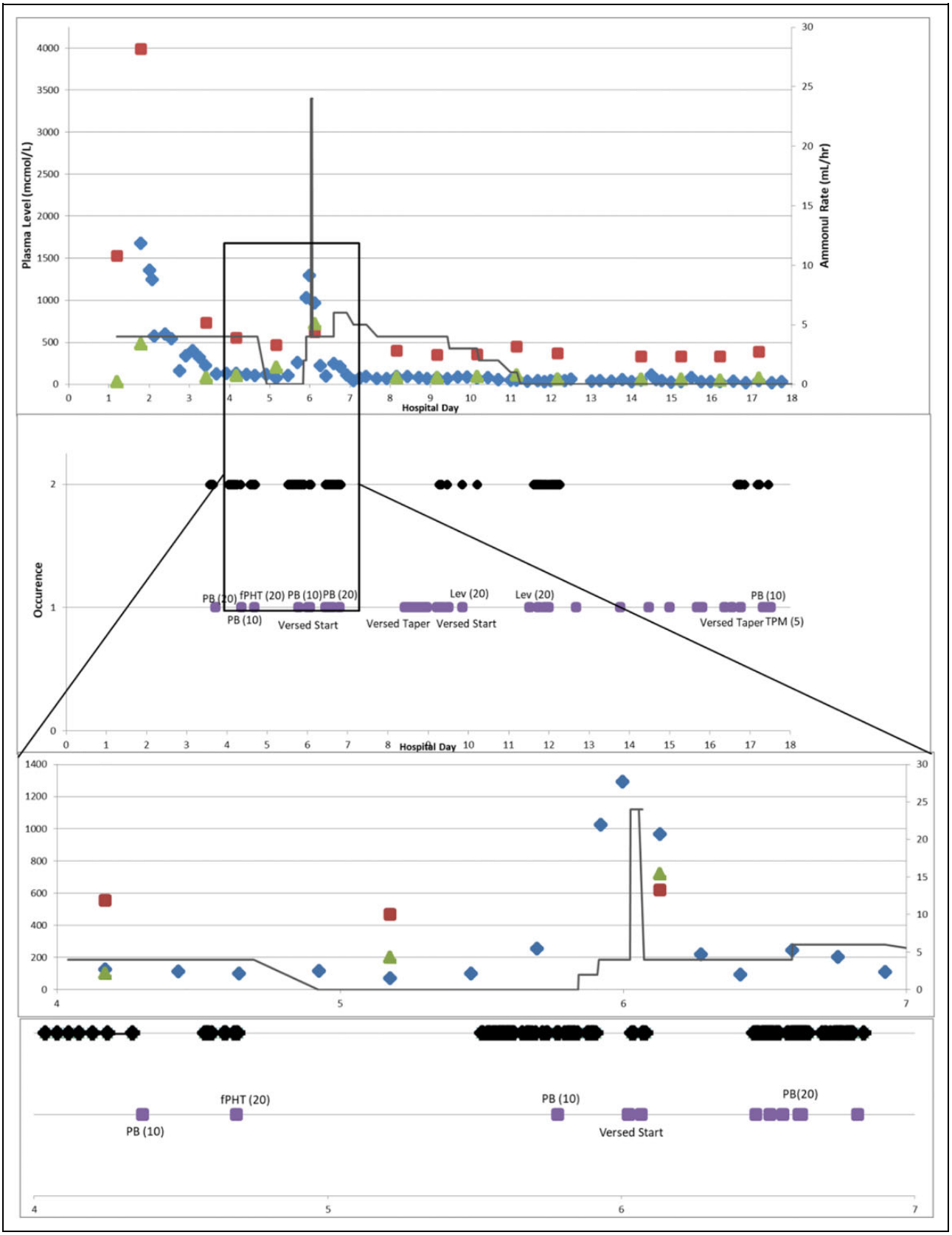

Figure 1. Top graph depicts ammonia (blue diamonds), glutamine (red squares), glutamate (green diamonds) levels (μmol/L) and the rate of an intravenous commercial preparation of ammonia scavengers and arginine (Ammonul) (gray line) over the first 18 days of hospitalization. Middle graph depicts the seizures (black diamonds) and anti-seizure medication (ASM) boluses or changes (purple squares) by day of hospital admission. The bottom graph is an enhancement of isochemical levels and seizure events preceding and during a relapse in hyperammonemia with the initial removal of intravenous scavengers.

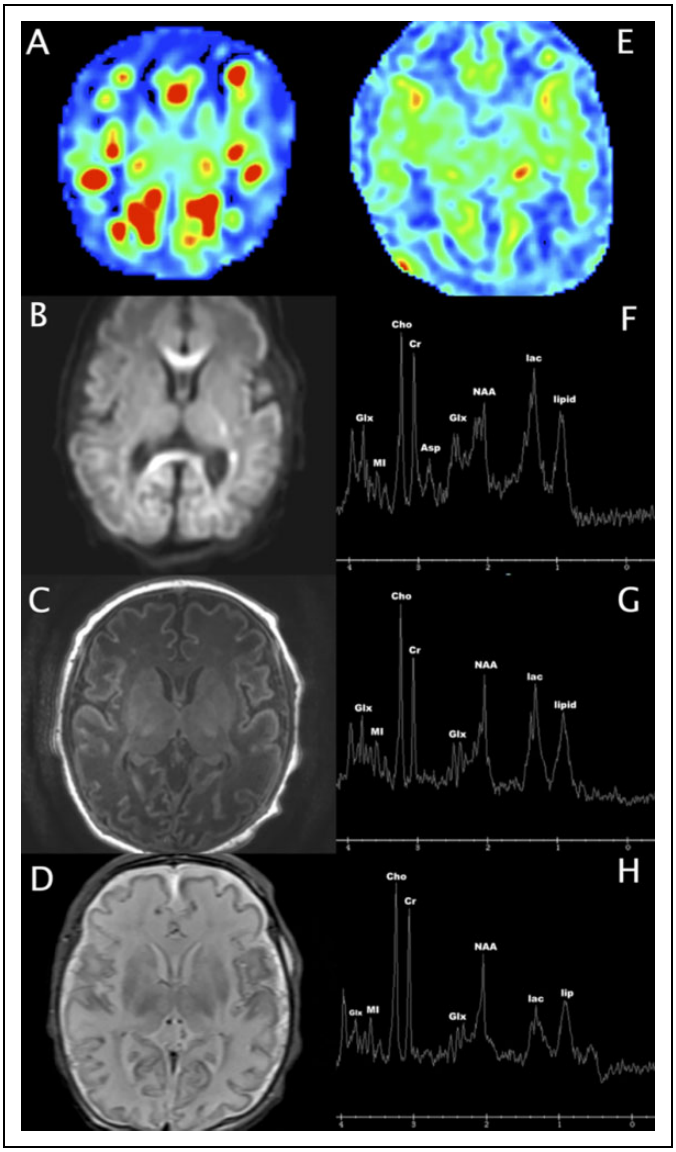

Figure 2. Selected axial brain MR images at the level of the basal ganglia at day of life 14 (a-d) and 18 (e). Heterogeneous cerebral hyperperfusion improves over time between exams (a and e). Reduced diffusion is present with hyperintense signal in the callosal splenium and genu, sagittal stratum, internal capsules, frontal white matter, and to a lesser extent (with partial pseudonormalization) in the cerebral cortex and deep gray nuclei in correlation with the apparent diffusion coefficient (ADC) map (not shown) (b). Hyperintensity on T1WI (c) and hypointensity onT2WI (d) is present extensively throughout most of the cerebral cortex and mild signal changes are present affecting the cerebral deep gray nuclei. The cortical signal changes on the T1 and T1WI represent laminar necrosis. The unmyelinated cerebral white matter demonstrates excessive T1 and T2 prolongation. There is mild diffuse cerebral volume loss with prominent sulci and ventricles. Single voxel proton MR spectroscopy (MRS) over the left basal ganglia at day 14 (f and g) and 18 (h). Initially, ultrashort TE MRS (STEAM; TR 1500 ms, TE 14 ms) and short TE MRS (PRESS; TR 1500ms, TE 35ms) reveal substantial metabolic alterations including elevated glutamine/glutamate (Glx) reflecting the urea cycle deficit and hyperammonemia induced glutamine synthetase activation, reduced myoinositol (MI) due to osmotic buffering, marked actate (lac) reflecting anaerobic metabolism, elevated lipid, and reduced NAA: Creatine and Choline:Creatine ratios. Aspartate (Asp) is also elevated, only visible with the ultrashort TE sequence (f). On follow-up, metabolic disturbances improved, with decreasing glutamine, lactate, and lipid and increasing myoinositol and creatine (h).

seizures was a proximal indicator of brain ammonia and glutamine re-elevation after the discontinuation of intravenous ammonia scavengers before detection remotely in plasma levels (HD4-5). Plasma glutamine remained within normal levels (376-819 μmol/L) for several days before, during, and after the second HA crisis despite ongoing refractory seizures. The persistent refractory seizures days after both plasma ammonia and glutamine levels were stable and the patient recovering suggests those seizures represent ongoing neuronal death rather than a primary seizure disorder or biochemical imbalance.

Although, the mechanism of ammonia neurotoxicity is still poorly understood and no specific treatment targeted for

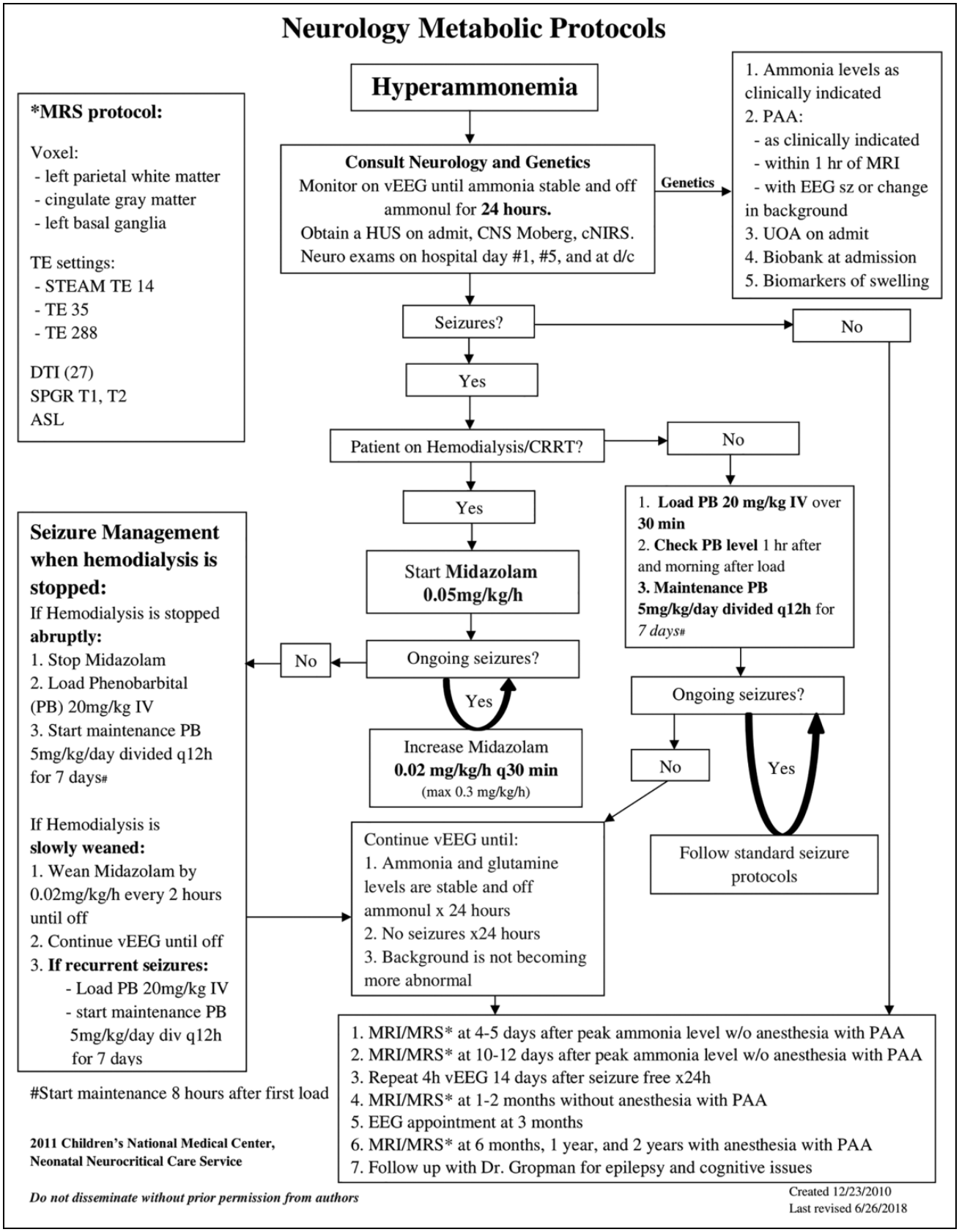

Figure 3. Clinical protocol used for the management of neonatal hyperammonemia by the neurology consult service. Abbreviations: Magnetic Resonance Spectrometry (MRS), Time Echo (TE), Diffusion Tensor Imaging (DTI), Spoiled Gradient Recall (SPGR), Arterial Spin Labeling (ASL), video Electroencephalography (vEEG), Head Ultrasound (HUS), continuous Near-Infrared Spectrometry (cNIRS), Plasma Amino Acids (PAA), Urine Organic Acid (UOA), Continuous Renal Replacement Therapy (CRRT).

ammonia neuroprotection is available, there have been frequent reports of seizures in patients with urea cycle disorders.[4,9,20] Accumulations of ammonia, glutamine, and glutamate have been shown to exert toxic effects upon the brain. In animal models, the HA state leads to excitotoxic cell death and, with prolonged exposure, to the loss of NMDA receptors. These same receptors are altered in the sparse fur (Spf) mouse model of ornithine transcarbamylase deficiency (OTCD).[14] The postulated effects of elevated ammonia and glutamine include astrocytic swelling, an increase in blood brain barrier permeability, and disruption of energy through depletion of intermediaries of metabolism including altered amino acid and neurotransmitter levels.[15-17,21]

It has been hypothesized that brain MRI findings may reflect the differential distribution of brain injury involvement in UCD and may aid in assessing neurologic outcomes.[5,10,22] One previous report presented serial imaging in a patient with a UCD (OTCD) that show the progression of the disease.[22] Another study investigated the pattern of MRI findings in patients with neonatal UCDs as it relates to the severity of disease and neurodevelopmental outcomes at 2 years of age. It was concluded that cerebral involvement of injury on MRI and levels of biomarkers such as glutamine have the potential to be prognostic for outcome, although the limited data on this subject prevents imaging from being the sole determinate of decision-making.[21] MR spectroscopy allows us to determine that brain ammonia levels are elevated (by seeing elevations of the surrogate metabolite, glutamine) which may occur even with normal plasma ammonia and glutamine levels. Limited neuroimaging literature on CPS1 deficiency has previously demonstrated a territorial infarction, nonspecific widespread brain injury, and more specific patterns similar to what has been described with OTCD with involvement of the insular, deep perisylvian and basal ganglia regions.[6,23-26] The diffuse, severe involvement of most components of both cerebral hemispheres with associated laminar necrosis suggests that the brain sustained a substantial insult that is unlikely to be entirely reversible in our patient. The reduced diffusion in parts of the cerebral white matter discovered on the first MR at 2 weeks of age could represent any combination of pre-Wallerian degeneration associated with the cortical injury, intramyelinic edema, and/or demyelination in the setting of residual/ongoing myelin injury or recurrent injury. While MRS using PRESS sequences have been preferred due to the higher signal to noise ratio, STEAM MRS enables visualization of the shorter echo-time (TE) metabolites. Previous work by our group has shown elevated glutamine and glutamate representing the hyperammonemia-induced glutamine synthetase activation and reduced myoinositol due to loss of osmotic buffering (Figure 1). STEAM MRS allowed visualization of the elevated Aspartate (Asp), which can be inferred to be a breakdown product of N-acetylaspartate (NAA) and marker of neuronal loss.

Conclusion

Based on our experience with this and prior UCD newborns, we propose a clinical neurodiagnostic protocol that can be implemented to better understand the clinical and biochemical time course of neonatal-onset UCDs and lead to early recognition and treatment of their neurologic complications (Figure 3). Early and comprehensive neurologic evaluation throughout the patient's acute course may improve neurologic outcomes. We propose early prolonged continuous EEG monitoring with acute HA and monitoring during withdrawal of hemodialysis and intravenous scavengers to identify and treat acute seizures and to identify early relapse of HA. MRS TE settings should be adjusted to a lower level, from 35 ms to STEAM TE ultrashort (14 ms) sequences, in order to better understand the biochemical profile of proximal UCDs. We plan long term follow up of this infant with sequential MRI/MRS and neurocognitive screening to better characterize the pattern of brain injury and correlate with later functional issues and risk of developing of epilepsy.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: NIH U54 HD061221 (AG); O'Malley Family Foundation grant (AG).

ORCID iD

Andrea Gropman, MD https://orcid.org/0000-0002-2106-6776

References

1. Díez-Fernández C, Gallego J, Häberle J, Cervera J, Rubio V. The Study of carbamoyl phosphate synthetase 1 deficiency sheds light on the mechanism for switching on/off the urea cycle. *J Genet Genomics Yi Chuan Xue Bao*. 2015;42(5):249-260.
2. Wusthoff CJ, Clark CL, Glass HC, Shimotake TK, Schulman J, Bonifacio SL. Cooling in neonatal hypoxic-ischemic encephalopathy: practices and opinions on minimum standards in the state of California. *J Perinatol Off J Calif Perinat Assoc*. 2018;38(1): 54-58.
3. American Academy of Pediatrics. Neonatal encephalopathy and neurologic outcome, second edition report of the American College of obstetricians and gynecologists' task force on neonatal encephalopathy. *Pediatrics*. 2014;133(5):e1482-e1488.
4. Verma NP, Hart ZH, Kooi KA. Electroencephalographic findings in urea-cycle disorders. *Electroencephalogr Clin Neurophysiol*. 1984;57(2):105-112.
5. Bireley WR, Van Hove JLK, Gallagher RC, Fenton LZ. Urea cycle disorders: brain MRI and neurological outcome. *Pediatr Radiol*. 2012;42(4):455-462.
6. Takanashi J, Barkovich AJ, Cheng SF, et al. Brain MR imaging in neonatal hyperammonemic encephalopathy resulting from proximal urea cycle disorders. *AJNR Am J Neuroradiol*. 2003;24(6): 1184-1187.
7. Wiwattanadittakul N, Prust M, Gaillard WD, et al. The utility of EEG monitoring in neonates with hyperammonemia due to inborn errors of metabolism. *Mol Genet Metab*. 2018;125(3):235-240.

8. Enns GM. Neurologic damage and neurocognitive dysfunction in urea cycle disorders. *Semin Pediatr Neurol*. 2008;15(3):132-139.
9. Tuchman M, Lee B, Lichter-Konecki U, et al. Cross-sectional multicenter study of patients with urea cycle disorders in the United States. *Mol Genet Metab*. 2008;94(4):397-402.
10. Gropman AL, Summar M, Leonard JV. Neurological implications of urea cycle disorders. *J Inherit Metab Dis*. 2007;30(6):865-879.
11. Anderson A, Gropman A, Le Mons C, Stratakis C, Gandjbakhche A. Evaluation of neurocognitive function of prefrontal cortex in ornithine transcarbamylase deficiency. *Mol Genet Metab*. 2020 Mar;129:207-212.
12. Häberle J.Primary hyperammonaemia: current diagnostic and therapeutic strategies. *J Mother Child*. 2020;24(2):32-38.
13. Sen K, Whitehead MT, Gropman AL. Multimodal imaging in urea cycle-related neurological disease- What can imaging after hyperammonemia teach us? *Transl Sci Rare Dis*. 2020;5:87-95.
14. Qureshi IA, Rao KVR. Sparse-Fur (spf) Mouse as a model of hyperammonemia: alterations in the neurotransmitter systems. In: Felipo V, Grisolía S, eds *Advances in Cirrhosis, Hyperammonemia, and Hepatic Encephalopathy*; 1997:143-158.
15. Norenberg MD, Rao KVR, Jayakumar AR. Mechanisms of ammonia-induced astrocyte swelling. *Metab Brain Dis*. 2005; 20(4):303-318.
16. Butterworth RF. Effects of hyperammonaemia on brain function. *J Inherit Metab Dis*. 1998;21(1):6-20.
17. Watanabe A, Shiota T, Takei N, Nagashima H. Excitatory and inhibitory amino acid neurotransmitters and ammonia metabolism in hepatic failure rats. *Res Exp Med Z Gesamte Exp Med Einschl Exp Chir*. 1985;185(5):399-404.
18. Häberle J, Boddaert N, Burlina A, et al. Suggested guidelines for the diagnosis and management of urea cycle disorders. *Ophanet J Rare Dis*. 2012;7(1):32.
19. Thrane V, Thrane AS, Wang F, et al. Ammonia triggers neuronal disinhibition and seizures by impairing astrocyte potassium buffering. *Nat Med*. 2013;19(12):1643-1648.
20. Huemer M, Carvalho DR, Brum JM, et al. Clinical phenotype, biochemical profile, and treatment in 19 patients with arginase 1 deficiency. *J Inherit Metab Dis*. 2016;39(3): 331-340.
21. Bachmann C, Colombo JP. Increase of tryptophan and 5-hydroxyindole acetic acid in the brain of ornithine carbamoyltransferase deficient sparse-fur mice. *Pediatr Res*. 1984;18(4): 372-375.
22. Mourad M, Häberle J, Whitehead MT, Stricker T, Gropman AL. Brain biomarkers of long-term outcome of neonatal onset urea cycle disorder. *Int J Neonatal Screen*. 2016;2(4):10.
23. Sperl W, Felber S, Skladal D, Wermuth B. Metabolic stroke in carbamyl phosphate synthetase deficiency. *Neuropediatrics*. 1997;28(04):229-234.
24. Takeoka M, Soman TB, Shih VE, Cavinessn VS, Krishnamoorthy KS. Carbamyl phosphate synthetase 1 deficiency: a destructive encephalopathy. *Pediatr Neurol*. 2001;24(3):193-199.
25. Yang X, Shi J, Lei H, Xia B, Mu D. Neonatal-onset carbamoyl phosphate synthetase I deficiency: a case report. *Medicine (Baltimore)*. 2017;96(26):e7365.
26. Gunz A, Choong K, Potter M, Miller E. Magnetic resonance imaging findings and neurodevelopmental outcomes in neonates with urea-cycle defects. *Int Med Case Rep J*. 2013;6:41-48.