
# Sodium Phenylacetate and Sodium Benzoate in the Treatment of Neonatal Hyperammonemia

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## Introduction

Ammonia is present in all body fluids and exists primarily as ammonium ion at physiologic pH. Hyperammonemia is defined as a blood ammonia concentration greater than about 100 mcmol/L in neonates or 50 mcmol/L in children and adults (precise cut-offs vary, depending on individual laboratory normative ranges). The concentration of ammonia is 10 times higher in tissue than in blood. A 5- to 10-fold increase in blood ammonia concentration usually is toxic to the nervous system.

Hyperammonemia in the neonatal period, especially when due to inborn errors of metabolism, can progress rapidly and cause severe neurologic damage or early death. Hyperammonemia can be caused by inborn errors of metabolism as well as by a variety of acquired conditions (Tables 1 and 2). Urgent treatment is required because of the potential for irreversible neurologic sequelae that can, in many cases, be prevented by prompt diagnosis and institution of therapy.

The combination of sodium phenylacetate (NAPA) and sodium benzoate (NABZ) in a 10%/10% solution is an intravenously administered United States Food and Drug Administration (FDA)-approved drug used as adjunctive therapy for the treatment of acute hyperammonemia and associated encephalopathy in patients who have urea cycle disorders (UCDs). Its concomitant use with protein restriction, provision of adequate calories to prevent catabolism, arginine hydrochloride, and hemodialysis in treating neonatal hyperammonemia helps prevent the reaccumulation of ammonia by increasing waste nitrogen excretion. The purpose of this article is to review the pharmacology and use of NAPA/NABZ in the treatment of neonatal hyperammonemia.

## Neonatal Hyperammonemia

Because the inheritance of most inborn errors of metabolism that cause neonatal hyperammonemia is autosomal recessive (exceptions include ornithine transcarbamylase [OTC] deficiency, which is X-linked, and hyperinsulinism/hyperammonemia syndrome, which is autosomal dominant), family history may offer no information of note or may reveal unexplained neonatal deaths.

UCDs are the most common cause of neonatal hyperammonemia and typically present with symptoms of poor feeding, lethargy, hypotonia, irritability, seizures, respiratory distress, grunting, and hyperventilation. Patients may have a bulging fontanelle if intracranial pressure is increased. Because the clinical presentation of hyperammonemia is nonspecific, other disorders common in neonates, such as sepsis, cardiac failure, and intracranial hemorrhage, are included in the differential diagnosis. Therefore, blood ammonia concentrations should be measured

### Abbreviations

| | |
|---|---|
| AL: | argininosuccinic acid lyase |
| ASA: | argininosuccinic acid |
| ATP: | adenosine triphosphate |
| BZ: | benzoate |
| CoA: | coenzyme A |
| HIP: | hippurate |
| NABZ: | sodium benzoate |
| NAPA: | sodium phenylacerate |
| OTC: | ornithine transcarbamylase |
| PA: | phenylacetic acid |
| PAGN: | phenylacetylglutamine |
| PD: | peritoneal dialysis |
| UCD: | urea cycle disorder |

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**Table 1. Inborn Errors of Metabolism Associated With Hyperammonemia**

Urea cycle defects
- *N*-acetylglutamate synthetase deficiency
- Carbamyl phosphate synthetase deficiency
- Ornithine transcarbamylase deficiency
- Argininosuccinate synthetase deficiency (citrullinemia)
- Argininosuccinate lyase deficiency
- Arginase deficiency

Amino acid transporter deficiencies
- Hyperornithinemia-hyperammonemia-homocitrullinuria (HHH) syndrome
- Lysinuric protein intolerance
- Citrin deficiency (citrullinemia type II)

Organic acidemias
- Methylmalonic acidemia
- Propionic acidemia
- Isovaleric acidemia
- Multiple carboxylase deficiency
- Multiple acyl-CoA dehydrogenase deficiency
- 3-Hydroxymethylglutaryl-CoA dehydrogenase deficiency
- 3-Methylcrotonyl-CoA carboxylase deficiency
- 3-Oxothiolase deficiency
- L-2-Hydroxyglutaric acidemia
- 3-Methylglutaconyl-CoA hydratase deficiency

Fatty acid oxidation defects
- Carnitine transporter deficiency
- Carnitine palmitoyl transferase 2 deficiency
- Carnitine-acylcarnitine translocase deficiency
- Medium-chain acyl-CoA dehydrogenase deficiency
- Long-chain 3-hydroxyacyl-CoA dehydrogenase deficiency
- Very-long-chain acyl-CoA dehydrogenase deficiency

Pyruvate carboxylase deficiency

Mitochondrial disorders

Hyperinsulinism/hyperammonemia syndrome (glutamate dehydrogenase mutations)

Delta$^1$-pyrroline-5-carboxylate synthase deficiency

in all neonates presenting with nonspecific symptoms of distress. If the concentration is elevated, diagnostic evaluations and treatment should be started immediately (Tables 3 and 4).

## Early Efforts in Hyperammonemia Therapy

A number of different therapies aimed at removing accumulated ammonia in cases of hyperammonemic encephalopathy have been attempted, including administration of lactulose (reduces the production or absorption of the end products of bacterial nitrogen metabolism in the colon), (1) exchange transfusion, (2)(3)(4) peritoneal dialysis, (3)(4) hemodialysis, (3)(4)(5) and supplementation with nitrogen-free analogues of essential amino acids. (6)(7)(8) Although children treated with alpha-keto amino acid analogues showed some clinical improvement, such as improved seizure control, attention span, and weight gain, death in infancy remained common. (6)(7)(8) Exchange transfusions are ineffective in managing hyperammonemia. In 15 patients treated by exchange transfusion, the decrease in ammonia values immediately following the procedure was not statistically significant. (9) Peritoneal dialysis (PD) has shown variable efficacy in treating hyperammonemia. Seven neonates who had UCDs showed a significant decrease in plasma ammonia values (85%±6%, $P<0.001$) following PD for a mean duration of 60 hours, but PD was ineffective in 13 older children. (9) The early use of these treatments prolonged survival in some cases, but overall efficacy was disappointing, and the mortality and morbidity associated with UCDs continued to be high.

**Table 2. Causes of Acquired Hyperammonemia**

Sampling artifact

Cardiovascular
- Patent ductus venosus
- Portocaval shunt
- Hypovolemia
- Congestive heart failure

Perinatal asphyxia

Liver failure
- Infectious hepatitis (eg, herpes simplex virus)

Bacterial colonization (urease-positive organisms)
- Neurogenic bladder
- Prune belly syndrome
- Blind loop syndrome
- Ureterosigmoidostomy

Iatrogenic
- Valproate
- Arginine deficiency
- Total parenteral nutrition

**Table 3. Differential Diagnosis of Neonatal Hyperammonemia**

| Onset | Cause | Clues to Diagnosis |
|---|---|---|
| <24 h after birth | 1. THAN | • Preterm infant<br>• No acidosis/ketosis |
| <24 h or >24 h after birth | 2. Organic acidemias (eg, MMA, PA, IVA), defects of fatty acid oxidation, congenital lactic acidosis | • Term infant<br>• Acidosis (+/− ↑ lactate)<br>• +/− ketosis |
| >24 h after birth | 3. Urea cycle defects: | No acidosis, sometimes alkalosis |
| | a) CPS deficiency | • Low/absent citrulline<br>• Urine orotic acid low |
| | b) OTC deficiency | • Low/absent citrulline<br>• Urine orotic acid markedly elevated |
| | c) AS deficiency | • Citrulline markedly elevated (>1,000 mcmol)<br>• No ASA in urine<br>• Urine orotic acid elevated |
| | d) AL deficiency | • Citrulline moderately elevated (100 to 300 mcmol)<br>• ASA present in urine<br>• Urine orotic acid elevated |

Fatty acid oxidation disorders, especially carnitine-acylcarnitine translocase deficiency, and organic acidemias may be difficult to distinguish from urea cycle defects in some instances. Metabolic acidosis, relatively high blood urea nitrogen (BUN) (urea cycle defects tend to be associated with a low BUN), and ketosis are more typical of organic acidemias. Hypoketotic hypoglycemia is suggestive of a fatty acid oxidation defect, but the level of ketosis is not always a reliable indicator in neonates. Prominent lactic acidosis may suggest pyruvate carboxylase deficiency or mitochondrial disorders. AL=argininosuccinate lysase, AS=argininosuccinate synthetase, ASA=argininosuccinic acid, CPS=carbamyl phosphate synthetase, IVA=isovaleric acidemia, MMA=methylmalonic acidemia, OTC=ornithine transcarbamylase; PA=propionic acidemia, THAN=transient hyperammonemia of the newborn

## Alternative Pathway Therapy

In 1914, Lewis demonstrated that NABZ could divert urea nitrogen to hippurate (HIP) nitrogen in two healthy subjects. (10) After ingestion of single 6- or 10-g aliquots of NABZ, blood urea nitrogen and ammonia levels fell, and urine HIP excretion showed a prominent rise, with little change in total urine nitrogen excretion. Shiple and Sherwin (11) later showed that oral administration of phenylacetic acid (PA) results in substitution of phenylacetylglutamine (PAGN) nitrogen for urea nitrogen in urine. Furthermore, coadministration of benzoate (BZ) and PA resulted in as much as 60% of urine nitrogen being excreted as HIP and PAGN. (11) Subsequently, the enzymes responsible for these reactions (acyl-CoA:glycine and acyl-CoA:glutamine *N*-acyltransferases) were identified and localized to both the kidney and liver in humans and primates. (12)(13)(14)(15) Synthesis of HIP (from conjugation of glycine with BZ) and PAGN (from conjugation of glutamine with PA) requires adenosine triphosphate (ATP) and coenzyme A (CoA). (16)

In 1979, Brusilow and associates (17) suggested that the use of endogenous biosynthetic pathways of non-urea waste nitrogen excretion could substitute for defective urea synthesis in patients who have UCDs. By promoting the synthesis of non-urea nitrogen-containing metabolites whose excretion rates are high or may be augmented, theoretically total body nitrogen load could be decreased despite the absence of normal urea cycle function. Two classes of such metabolites are: 1) urea cycle intermediates (citrulline and argininosuccinic acid) and 2) amino acid acylation products (HIP and PAGN). (18)

### Urea Cycle Intermediates

In argininosuccinic acid lyase (AL) deficiency, argininosuccinic acid (ASA) accumulates and is excreted in the urine. Because ASA contains two waste nitrogen atoms, production of this metabolite can be exploited to excrete waste nitrogen in AL deficiency, provided that an adequate amount of ornithine is present to supply the necessary carbon skeletons for ASA biosynthesis. (19) By administering pharmacologic doses of arginine, ornithine is synthesized by the action of arginase. Citrulline and ASA subsequently are produced by the sequential action of OTC and argininosuccinic acid synthetase. In AL deficiency, ASA cannot be metabolized further and is excreted in the urine, along with waste nitrogen (Fig. 1). (17)(18)(19)

Similarly, citrulline can serve as a

**Table 4. Management of Neonatal Hyperammonemia Caused by Urea Cycle Defects**

Laboratory studies
- Blood ammonia
- Anion gap
- Liver transaminases, alkaline phosphatase, bilirubin, prothrombin time
- Blood lactate and pyruvate
- Arterial blood gas
- Serum and urine amino acids
- Urine organic acids
- Urine quantitative orotic acid
- Plasma carnitine (total, free, and esterified)
- Plasma acylcarnitine profile

Treatment: Prevention of catabolism and ammonia accumulation
- Intravenous dextrose (20% or 25%)
- No exogenous protein for 24 to 48 h
- Use continuous insulin drip if hyperglycemic
- Intravenous lipid (once fatty acid oxidation disorders excluded)
- Provide total calories of approximately 100 to 120 kcal/kg per day

Treatment: Medications
- Sodium benzoate and sodium phenylacetate
- Arginine hydrochloride 10%*
- Lactulose 2.5 mL NG/PO tid prn
- Neomycin 50 mg/kg per day PR q 6 h†

Treatment: Other measures
- Central vascular access
- Correction of hypovolemia, anemia, and possible acidosis
- Treatment of underlying infection
- Intubation and ventilation (target $Paco_2$ of 30 to 35 mm Hg)
- Urinary catheter (for monitoring of urine output)
- Hemodialysis

*Acidosis may occur; arterial blood gases should be examined after loading dose.
†Only in neonates >2 days old.

vehicle for waste nitrogen excretion in AS deficiency (citrullinemia), as long as sufficient arginine is supplied (Fig. 1). (17)(18) However, citrulline contains only one waste nitrogen atom, and a high percentage of filtered citrulline is reabsorbed, so urine excretion is relatively poor. (18)

### Amino Acylation Products

HIP is an excellent metabolite for renal excretion because its renal clearance is five times the glomerular filtration rate. (17) HIP biosynthesis, by conjugation of BZ with glycine, is accomplished by the action of mitochondrial matrix enzymes (benzoyl thiokinase and a glycine-specific *N*-acyltransferase) (Figs. 1 and 2). (13)(16) Similarly, PAGN is formed by sequential action of phenylacetyl thiokinase and a glutamine-specific *N*-acyltransferase. (12)(13) Because PA has the ability to conjugate glutamine, forming PAGN (a compound that contains two nitrogen atoms), its nitrogen-scavenging ability was hypothesized to be twice as effective as BZ (which contains one nitrogen atom). (18) In 1979, Brusilow and colleagues (17) suggested using combined therapy with NAPA and NABZ for treating hyperammonemic coma.

### Initial Clinical Trials of NAPA and NABZ

The potential of alternative pathway therapy was demonstrated initially in 1980. A clinically stable 17-year-old girl who had carbamyl phosphate synthetase deficiency excreted significant amounts of HIP and PAGN in the urine after NABZ (6.25 g/d) or PA (6.4 g/d) was administered orally. Subsequent administration of NABZ (250 to 350 mg/kg, either orally or intravenously) in four patients who had UCD and were in hyperammonemic comas resulted in a prompt decrease in plasma ammonia concentrations and clinical improvement in each case. (20) In a further study, a single oral or intravenous dose of NABZ (250 to 500 mg/kg) lowered plasma ammonia concentrations in five of seven patients who had hyperammonemia (two of three neonates and four of five older children). (9) In another study, (21) 26 patients were treated with intravenous NABZ (250 mg/kg loading dose, followed by 250 to 500 mg/kg per day continuous infusion) and arginine hydrochloride (800 mg/kg loading dose, followed by 200 to 800 mg/kg per day) during acute neonatal hyperammonemia. PD was required during neonatal hyperammonemic coma episodes in 20 of 23 patients. There were three neonatal deaths. It was concluded that alternative pathway therapy (NABZ and arginine supplementation), combined with dietary restriction of protein and provision of supplemental calories in an amount no less than 100 kcal/kg per day, can prolong survival and improve clinical outcome in children who have UCDs.

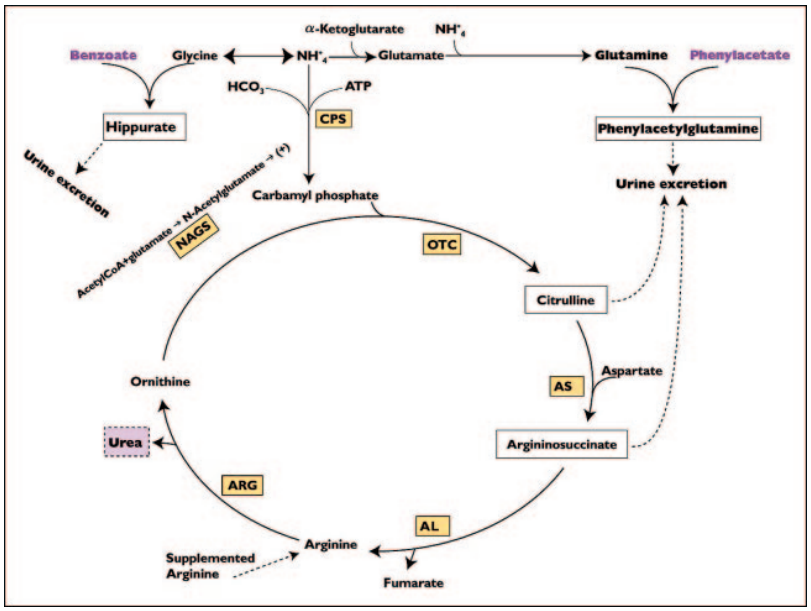


**Figure 1.** The urea cycle and alternative pathway therapy. AL=argininosuccinic acid lyase, ARG=arginase, AS=argininosuccinic acid synthetase, CPS=carbamyl phosphate synthetase, NAGS=*N*-acetylglutamate synthetase, OTC=ornithine transcarbamylase, ATP=adenosine triphosphate

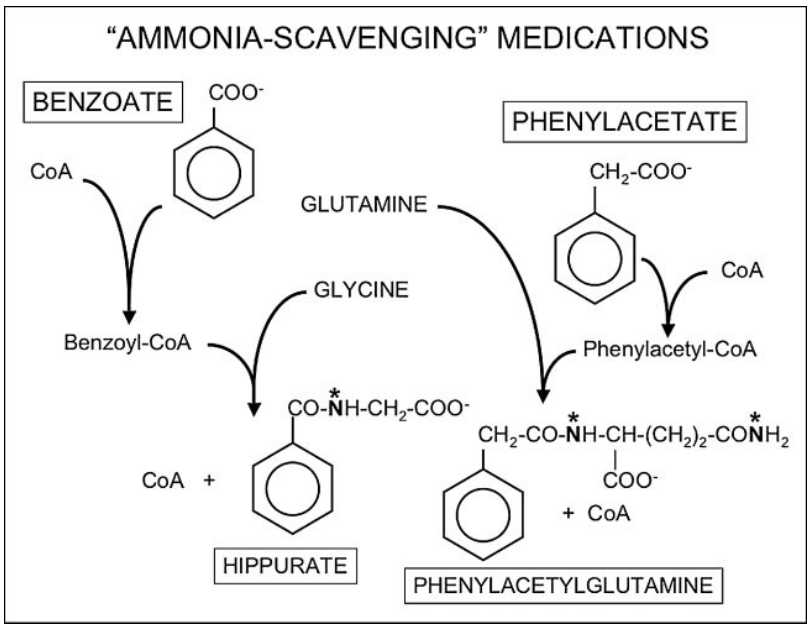


**Figure 2.** Mechanism of nitrogen scavenging by sodium benzoate and sodium phenylacetate. Hippurate and phenylacetylglutamine are formed by conjugation of benzoate with glycine and phenylacetate with glutamine, respectively. These reactions are performed by specific liver and kidney *N*-acyltransferases (see text). *=nitrogen atoms excreted

## NAPA/NABZ

In 1984, Brusilow and colleagues (22) reported the results of a therapeutic protocol for the treatment of hyperammonemia caused by UCDs using a combination of intravenous NAPA plus NABZ. The initial clinical trial of combined therapy involved 12 episodes of hyperammonemia in seven children ages 3 to 26 months who had a variety of UCDs. The plasma ammonia concentrations decreased to normal or nearly normal levels in all patients, except in a 9-month-old boy who had OTC deficiency and the highest pretreatment ammonia value and the longest delay between symptom onset and therapy.

The combination of NAPA (10%) and NABZ (10%) is an intravenously administered drug approved by the FDA as adjunctive therapy for the treatment of acute hyperammonemia and associated encephalopathy in patients who have urea cycle enzyme deficiencies. The concomitant use of NAPA/NABZ with protein restriction, high caloric nutrition, arginine hydrochloride, and hemodialysis in neonatal hyperammonemia helps to increase waste nitrogen excretion through the formation of HIP and PAGN by two different pathways (Figs. 1 and 2). Pharmacogenetic factors partly determine the activity of enzymes responsible for formation of HIP and PAGN and, therefore, play a role in determining the individual rate of nitrogen removal. Hemodialysis is recommended in cases of severe hyperammonemia or if ammonia concentrations are not significantly reduced within 4 to 8 hours after starting NAPA/NABZ therapy.

When a diagnosis of hyperammonemia is established in a neonate, NAPA/NABZ infusion should be started as soon as possible. A loading dose is administered over 90 minutes, followed by a similar mainte-

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