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TREATMENT OF EPISODIC HYPERAMMONEMIA IN CHILDREN WITH INBORN ERRORS OF UREA SYNTHESIS

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Abstract Although normal plasma ammonium levels can be maintained in children with inborn errors of ureagenesis, these children are vulnerable to episodic hyperammonemia often resulting in coma and death. To treat such episodes, we designed a therapeutic protocol that included prompt recognition of hyperammonemia, therapy with intravenous sodium benzoate, sodium phenylacetate, and arginine, and nitrogen-free intravenous alimentation. Dialysis was performed if the hyperammonemia was unresponsive to drug therapy. Twelve episodes of hyperammonemia in seven children deficient in carbamyl phosphate synthetase, ornithine transcarbamylase, or argininosuccinic acid synthetase were treated; one patient died and the others recovered. In two patients measurement of the distribution of urinary nitrogen revealed that hippurate nitrogen and phenylacetylglutamine nitrogen together accounted for 60 per cent of "effective" urinary waste nitrogen. Successful therapy of episodic hyperammonemia plays an important part in the long-term management of disorders of the urea cycle. (N Engl J Med 1984; 310:1630-4.)

INFANTS with inborn errors of ureagenesis maintain normal plasma levels of ammonium and may have normal growth and development. Therapy relies on restriction of nitrogen intake, an abundant energy supply, activation of alternative pathways for waste-nitrogen excretion, and dietary supplementation with arginine. The rationale, details, and results of long-term therapy have been reported previously.[1-3] The maintenance therapy of carbamyl phosphate synthetase and ornithine transcarbamylase deficiency has recently been modified; sodium phenylacetate has been added, and citrulline has been substituted for arginine. Sodium phenylacetate has also been added to the treatment of argininosuccinic acid synthetase deficiency.

Despite long-term therapy, all such infants are constantly vulnerable to episodes of hyperammonemia and coma. The early clinical manifestations of impending hyperammonemic coma are nonspecific and include anorexia, lethargy, and vomiting associated with plasma ammonium levels that are three to four times normal.

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Before the observation that intravenous benzoate might be useful in treating hyperammonemia,[2] therapy was directed at eliminating dietary nitrogen and suppressing endogenous protein breakdown by supplying non-nitrogenous nutrition, principally in the form of glucose. Thus, hyperammonemia associated with hepatic encephalopathy was shown to respond to 5 per cent glucose administration.[1] However, six infants with inborn errors of ureagenesis who survived neonatal hyperammonemic coma all died during episodes of intercurrent hyperammonemia, despite treatment with intravenous glucose[5] supplying as much as 60 kcal per kilogram of body weight per day.[6] With the introduction of new methods of stimulating waste-nitrogen synthesis and excretion in patients with these diseases, the mortality from hyperammonemic coma was reduced. Hyperammonemic episodes in patients with argininosuccinase (EC 4.3.2.1.) deficiency have been simply and successfully treated by eliminating dietary nitrogen and giving a priming dose of 840 mg of intravenous 10 per cent arginine hydrochloride per kilogram over one hour, followed by a sustaining infusion of 840 mg per kilogram over 24 hours. There have been no deaths among 25 patients so treated.[3] However, 3 of 11 patients with deficiencies of ornithine transcarbamylase (EC 2.1.3.3.) or carbamyl phosphate synthetase (EC 2.7.2.2.) died during an episode of hyperammonemia treated with benzoate, glucose, and arginine.

Because of the high mortality, a new intravenous

protocol for the therapy of intercurrent hyperammonemia occurring in patients with inborn errors of urea synthesis other than argininosuccinase deficiency. By virtue of its ability to conjugate glutamine, with the subsequent renal excretion of phenylacetylglutamine[2] (which contains two nitrogen atoms per mole), phenylacetate activates a pathway of waste-nitrogen synthesis and excretion that is twice as effective as that activated by benzoate. After the first trial of this drug (in which only single intravenous doses of phenylacetate and benzoate were used) a new protocol was designed (Table 1). We report here the results of therapy for 12 episodes of intercurrent hyperammonemia.

Methods

All methods were similar to those reported previously.[3] All patients were presumed to have complete or nearly complete enzyme deficiencies because they either presented with neonatal hyperammonemic coma or had a sibling who did. Phenylacetate and phenylacetylglutamine were measured in plasma and urine according to a liquid-chromatographic method described earlier, but the column effluent was monitored at 220 nm. Phenylacetylglutamine for use in standard solutions was prepared as described by Thierfelder and Sherwin.[7]

Patients with plasma ammonium levels three times normal or higher were given a priming infusion over one to two hours. It contained sodium benzoate and sodium phenylacetate, each in a dose of 250 mg per kilogram, given in 25 to 35 ml (per kilogram) of 10 per cent glucose with added 10 per cent arginine hydrochloride; the dose of arginine was 210 mg per kilogram for deficiencies of carbamyl phosphate synthetase and ornithine transcarbamylase, and 840 mg per kilogram for argininosuccinic acid synthetase (EC 6.3.4.5.) deficiency. The priming infusion was immediately followed by a 24-hour sustaining infusion containing 250 to 500 mg (per kilogram) of sodium benzoate and of sodium phenylacetate, which was reduced to 250 mg per kilogram of each drug after the plasma ammonium level decreased. The arginine was continued in a dose of 210 mg per kilogram for patients with deficiencies of ornithine transcarbamylase and carbamyl phosphate synthetase and 840 mg for patients deficient in argininosuccinase. These drugs were given in the estimated daily requirement of maintenance fluid containing 10 per cent glucose, to provide at least 40 kcal per kilogram per day. Nitrogen intake was eliminated. Failure of drug therapy, as manifested by lack of change in the plasma ammonium level or by a rebound increase after it had decreased, was established as an indication for either peritoneal dialysis or hemodialysis. Maintenance oral therapy was resumed after the plasma ammonium level stabilized at normal or nearly normal levels.

These studies were approved by the institutional review boards of each participating hospital, and informed consent was obtained from the parents of the patients. Intravenous sodium benzoate and phenylacetate are investigational new drugs approved for human use by the Food and Drug Administration.

Table 1. Protocol for the Management of Intercurrent Hyperammonemia in Patients with Inborn Errors of Ureagenesis.

1. Early diagnosis (plasma ammonium ≤ 200 μM).
2. Elimination of dietary nitrogen.
3. Priming infusion (over 1–2 hr), in 30 ml of 10 per cent glucose per kilogram of body weight, containing sodium benzoate (250 mg/kg) and sodium phenylacetate (250 mg/kg) (both these drugs are omitted in patients with argininosuccinase deficiency), and arginine hydrochloride (210 mg/kg; 840 in patients with argininosuccinic acid synthetase and argininosuccinase deficiencies).
4. Sustaining infusion containing sodium benzoate (500 mg/kg/24 hr, reduced to 250 if plasma ammonium decreases) and sodium phenylacetate (500 mg/kg/24 hr, reduced to 250 if plasma ammonium decreases) (both these drugs are omitted in patients with argininosuccinase deficiency), arginine hydrochloride (210 mg/kg/24 hr, 840 in patients with argininosuccinic acid synthetase and argininosuccinase deficiencies), and non-nitrogenous intravenous alimentation providing more than 40 kcal per kilogram per day.
5. Hemodialysis if there is no response to above steps.

Results

The first trial of intravenous phenylacetate was conducted in a 12-month-old boy with carbamyl phosphate synthetase deficiency (Patient 1) who had been anorexic and lethargic for 24 hours. Upon admission the plasma level of ammonium was 111 μM, glutamine 1550 μM, glycine 218 μM, and arginine 34 μM. One hour later the plasma ammonium level rose to 145 μM. Figure 1 shows the course of the plasma and urinary laboratory values. Two hours after admission the patient was given intravenous sodium benzoate and phenylacetate (each in a dose of 250 mg per kilogram) and arginine hydrochloride (210 mg per kilogram) over a one-hour period. Three hours after the infusion was completed the ammonium level decreased to 79 μM. Maintenance oral therapy, begun 5½ hours later, consisted of sodium benzoate (500 mg per kilogram) and citrulline (175 mg per kilogram), given orally in three divided doses over six hours.

After 19 hours of therapy the plasma ammonium level was within normal limits, at 33 μM. The plasma glutamine level fell from 1500 to 990 μM. Arginine levels were unchanged, but the glycine level fell from 230 to 89 μM.

Because our patients had little or no capacity to synthesize urea (except from arginine), we used the concept of "effective" waste nitrogen.[3] It excludes all urinary urea nitrogen, half the urinary argininosuccinate nitrogen, and two thirds of the citrulline nitrogen; the source of the two nitrogen atoms of these molecules is presumed to be arginine derived from exogenous sources or proteolysis. In Patient 1, urinary hippurate nitrogen (5.1 to 6.8 mg of nitrogen per milligram of creatinine) accounted for 27 to 57 per cent of effective waste nitrogen, and urinary phenylacetylglutamine nitrogen (2.6 to 6.2 mg of nitrogen per milligram of creatinine) accounted for 21 to 30 per cent; together (9.1 to 12.8 mg of nitrogen per milligram of creatinine) they accounted for 48 to 79 per cent of the effective urinary waste nitrogen (Fig. 1).

The single-dose pharmacokinetics of phenylacetate and benzoate are also shown in Figure 1. Although the peak levels occurred at the same time, phenylacetate levels were initially higher than benzoate levels and remained so over the study period. Their reaction products behaved differently in that hippurate levels reached a peak earlier and the peak was higher than that of phenylacetylglutamine. However, the levels of the latter remained high for a longer period, notwithstanding the additional oral dose of benzoate.

Figure 2 shows the course of a five-month-old boy

tient 2) who also had a 24-hour history of anorexia and lethargy. Upon admission the plasma level of ammonium was 333 μM, glutamine 1211 μM, glycine 349 μM, and arginine 43 μM. One hour later the treatment protocol was begun. The plasma ammonium level decreased to 129 μM within 12 hours of therapy; from this level it slowly decreased to 74 μM 20 hours later. Plasma levels of glutamine and glycine decreased to 600 μM and 110 μM, respectively, within 10 hours of therapy. Arginine levels remained unchanged, except for the final value.

Urinary hippurate nitrogen (8 to 18 mg of nitrogen per milligram of creatinine) accounted for 18 to 34 per cent of effective waste nitrogen, and urinary phenylacetylglutamine nitrogen (4 to 26 mg of nitrogen per milligram of creatinine) accounted for 15 to 53 per cent; together (14.5 to 36.7 mg of nitrogen per milligram of creatinine) they accounted for 45 to 66 per cent of effective urinary waste nitrogen (Fig. 2).

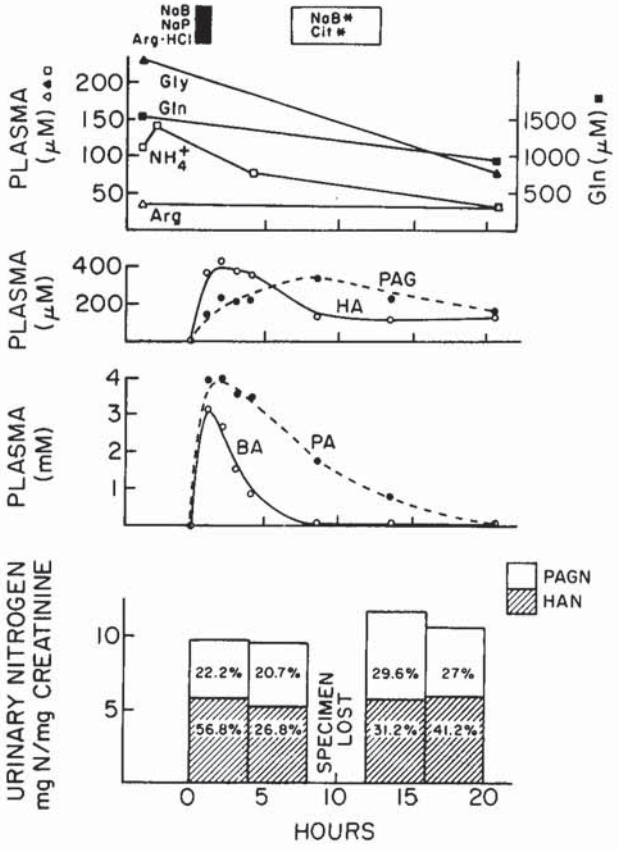

Figure 1. Effect of Intravenous Sodium Benzoate (NaB), Sodium Phenylacetate (NaP), and Arginine (Arg-HCl) for Carbamyl Phosphate Synthetase Deficiency — Patient 1.

The upper three panels show plasma ammonium (NH_4^+), glutamine (Gln), glycine (Gly), arginine (Arg), hippurate (HA), phenylacetylglutamine (PAG), benzoate (BA), and phenylacetate (PA). The bottom panel shows the urinary excretion of hippurate nitrogen (HAN) and phenylacetylglutamine nitrogen (PAGN) and the percentages of urinary effective waste nitrogen contributed by these compounds. Cit denotes citrulline, and asterisks denote oral therapy. Normal plasma values are <35 μM for ammonium, 62±9 μM for arginine, 483±86 μM for glutamine, and 213±25 μM for glycine (mean ±S.D.).

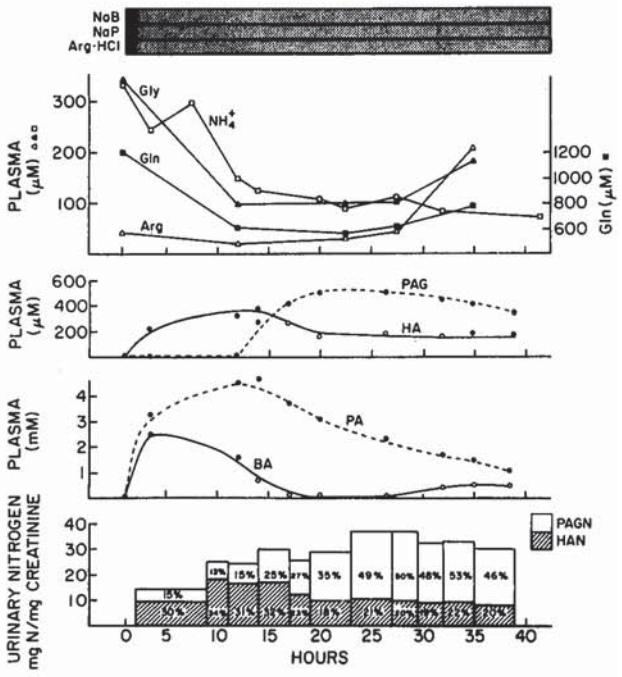

Figure 2. Effect of Therapy in Patient 2.

The solid and stippled areas denote the periods of priming and sustaining infusions, respectively. For explanation of abbreviations, see Figure 1.

The pharmacokinetics in Patient 2 were similar to those in Patient 1 in that peak plasma phenylacetate levels were higher than benzoate levels. Plasma hippurate levels reached a peak much earlier than phenylacetylglutamine levels, which, over the first 12 hours, were below the limits of detectability in plasma despite the appearance of phenylacetylglutamine in the urine. Subsequently, plasma phenylacetylglutamine levels were twice hippurate levels.

The results of therapy in six other episodes of hyperammonemia in four of the patients treated according to our protocol are shown in Figure 3. Apart from Patient 3, these patients either had had no symptoms or had had lethargy and vomiting for less than 48 hours, and had pretreatment plasma ammonium levels ranging from 101 to 287 μM. After the therapeutic protocol was followed, the plasma ammonium levels returned to normal within 48 hours and the symptoms abated.

Patient 1 had three such episodes; one episode involved rebound hyperammonemia, which occurred after the patient had responded to therapy for an earlier episode. This rebound hyperammonemia at 26 hours was successfully treated with a second priming infusion rather than with peritoneal dialysis, which would have been performed if there had not been a prompt response to drug therapy.

Patient 3 was admitted to the hospital with a plasma ammonium level of 483 μM and a three-day histo-

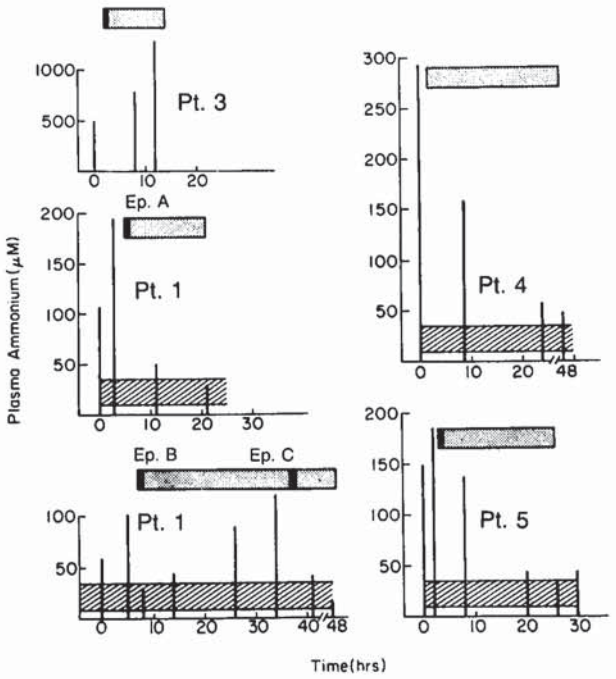

Figure 3. Plasma Ammonium during Six Episodes of Hyperammonemic Coma Treated by Infusion.

The solid and stippled areas denote the periods of priming and sustaining infusions, respectively. (Patient 4 did not receive a priming infusion). The hatched area denotes the normal range.

Patient 1 was a boy with carbamyl phosphate synthetase deficiency, which led to one episode of coma at the age of 19 months (Ep. A) and two episodes at 22 months (Ep. B and Ep. C); Patient 3 was a 9-month-old boy with ornithine transcarbamylase deficiency; Patient 4 was a 26-month-old boy with carbamyl phosphate synthetase deficiency; and Patient 5 was a 3-month-old boy with ornithine transcarbamylase deficiency.

ry of irritability, lethargy, and vomiting. Despite therapy the plasma ammonium level rose to 754 μM at 8 hours and to 1278 μM at 11 hours. Shortly thereafter hypotension and Cheyne–Stokes respiration developed, and the patient died 14 hours after admission. Dialysis had been offered and refused.

Two other patients who had rebound hyperammonemia within seven hours of their initial response to therapy were considered to require peritoneal dialysis (Fig. 4). Patient 6 received a second priming infusion followed by peritoneal dialysis shortly after the plasma ammonium level exceeded 100 μM at 15 hours after the first priming infusion. Ammonium levels decreased to nearly normal within eight hours after the second priming infusion and six hours of dialysis. Patient 7 underwent dialysis because the plasma ammonium level, after decreasing to normal limits after therapy, increased to 105 μM at 19 hours and to 70 μM at 21 hours after the priming infusion and during a sustaining infusion. However, after five hours of peritoneal dialysis the plasma ammonium returned to nearly normal values.

Six of the 12 hyperammonemic episodes were preceded by a clinically identifiable illness (e.g., upper respiratory infection, otitis media, and fever). Seven of 12 episodes involved anorexia and vomiting, and all but 1 patient had a history of lethargy and irritability. No dietary alterations were made before the hyperammonemic episodes.

The only side effect of therapy was the association of nausea and vomiting with the priming infusion. An increased anion gap may be noted during therapy as a consequence of the accumulation of benzoate, phenylacetate, and their conjugation products in plasma, the sum of which may attain levels of 7 mM (Fig. 1 and 2).

Discussion

Seven children with inborn errors of urea synthesis were treated for 12 episodes of hyperammonemia (i.e., a plasma ammonium level more than three times normal). The plasma ammonium level decreased to normal or nearly normal levels in all patients but one.

Three patients had rebound hyperammonemia within 18 hours after the initial response. One of these patients (Fig. 3, Patient 1, Episode A) responded to a second priming infusion with a decrease in plasma ammonium and did not require peritoneal dialysis. Retrospective analysis of the course of Patient 6 (Fig. 4) suggests that she may also have responded to the second priming infusion of benzoate, phenylacetate,

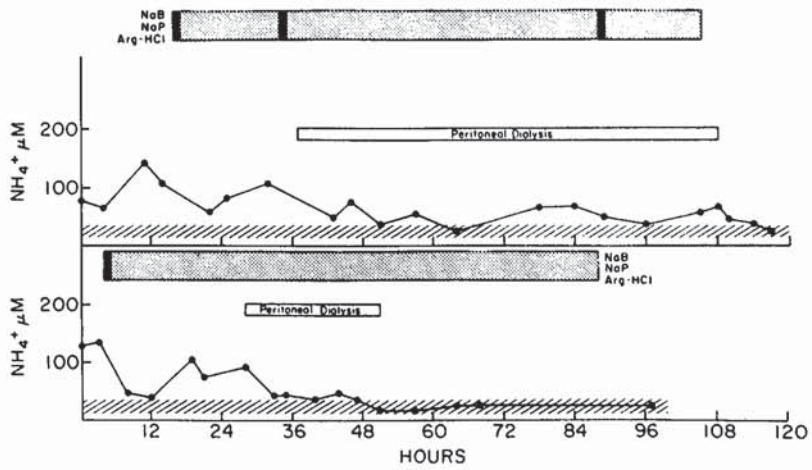

Figure 4. Plasma Ammonium during Episodes of Hyperammonemia — Patients 6 (Upper Panel) and 7 (Lower Panel).

Rebound hyperammonemia after initial therapy was treated with sodium benzoate, sodium phenylacetate, and peritoneal dialysis. The solid and stippled areas denote the periods of priming and sustaining infusions, respectively. The hatched area denotes the normal range.

Patient 6 was a 10-month-old girl with argininosuccinic acid synthetase deficiency, and Patient 7 a 19-month-old boy with ornithine transcarbamylase deficiency.

and arginine, as evidenced by a decrease in the plasma ammonium concentration within eight hours. Although she received dialysis during six of these eight hours, it is difficult to evaluate the relative contribution of dialysis and drugs. However, our previous report suggested that peritoneal dialysis in children two months old or older is not as effective as it is in neonates.[8] The relative effect of peritoneal dialysis and drugs on the plasma ammonium level in Patient 7 (Fig. 4) is also difficult to interpret. Nonetheless, peritoneal dialysis — or better yet, hemodialysis — may have an important role in the management of hyperammonemia that is unresponsive to drug therapy. We recommend hemodialysis[9] under such circumstances.

That nitrogen accumulation was decreased was indicated not only by the reduction in plasma levels of ammonium and glutamine but also by the appearance of hippurate and phenylacetylglutamine in plasma and urine. Figures 1 and 2 clearly demonstrate that most of the urinary nitrogen consisted of the amino acid acylation products hippurate and phenylacetylglutamine, the amounts of which were similar to the amount of urinary urea nitrogen found in fasting infants — 10 to 30 mg of nitrogen per milligram of creatinine.[10]

However, the rapidity and magnitude of the plasma changes permit speculation that the activation of the pathways of hippurate and phenylacetylglutamine synthesis may reduce the plasma concentrations of accumulated nitrogen products to a greater extent than might be expected from the amounts of hippurate and phenylacetylglutamine formed. For example, the rapidity of decline in the plasma ammonium levels shown in Figures 1 and 3 is not entirely compatible with the pharmacokinetics shown in Figure 1, in which the synthesis of hippurate and phenylacetylglutamine continues for as long as 8 and 12 hours, respectively, after a single dose of the drugs whereas the plasma ammonium concentration decreases within a period of 1 hour (Episode B), 4 hours (Episode C), and 5 hours (Fig. 1). Furthermore, assuming that the intracellular glutamine concentration (which is normally 33 times the plasma level[11]) increases in the same proportion as does the plasma level, the amount of accumulated nitrogen would be more than twice the amount that could be disposed of by the doses of benzoate and phenylacetate.

Therefore, we suggest that the rapid fall in the plasma concentrations of ammonium is in part a consequence of a new steady-state relationship between it and its precursors, induced by the flux of the precursors to hippurate and phenylacetylglutamine.

The reason for the failure of drug therapy to decrease the plasma ammonium level of Patient 3 is not clear. However, he differed from all the other patients in that he had the highest ammonium level and the longest delay between the onset of symptoms and therapy.

Although the precipitating causes of these hyperammonemic episodes cannot be completely defined, they must reflect an alteration in nitrogen balance. In some cases infection may have altered the steady state by causing net protein breakdown, interrupting drug ingestion, or both. The cause of hyperammonemia in the absence of documented infection is uncertain. Anorexia and vomiting may contribute to the development of hyperammonemia by interfering with caloric and drug intake, but they are also symptoms of hyperammonemia.

We conclude that prompt measurement of the plasma ammonium concentration in children with defective urea synthesis who have signs of infection or have vomiting, irritibility, anorexia, or lethargy may permit early intervention and thereby prevent progression to coma and death. The role of benzoate and phenylacetate in neonatal hyperammonemic coma or other hyperammonemic states (Reye's syndrome and liver failure) remains to be determined.

Note added in proof: Since submission of the manuscript, 42 additional episodes of hyperammonemia in 13 patients have been treated by this protocol. One episode occurred in a 14-month-old child who died 10 hours after presentation with a plasma ammonium level of 700 μM. All other episodes responded to drug therapy and did not require dialysis. A second death followed a priming infusion containing 2.5 g each of sodium benzoate and phenylacetate per kilogram — a 10-fold overdose.

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