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Häberle, J

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Clinical practice

The management of hyperammonemia

Johannes Häberle

Abstract Hyperammonemia is a life-threatening condition which can affect patients at any age. Elevations of ammonia in plasma indicate its increased production and/or decreased detoxification. The hepatic urea cycle is the main pathway to detoxify ammonia; it can be defective due to an inherited enzyme deficiency or secondary to accumulated toxic metabolites or substrate depletion. Clinical signs and symptoms in hyperammonemia are unspecific but they are mostly neurological. Thus, in any unexplained change in consciousness or in any unexplained encephalopathy, hyperammonemia must be excluded as fast as possible. Any delay in recognition and start of treatment of hyperammonemia may have deleterious consequences for the patient. Treatment largely depends on the underlying cause but is, at least in pediatric patients, mainly aimed at establishing anabolism to avoid endogenous protein breakdown and amino acid imbalances. In addition, pharmacological treatment options exist to improve urea cycle function or to remove nitrogen, but their use depend on the underlying disorder. To improve the prognosis of acute hyperammonemia, an increased awareness of this condition is probably more needed than anything else. Likewise, the immediate start of appropriate therapy is of utmost importance. This review focuses on a better understanding of factors leading to ammonia elevations and on practical aspects related to diagnosis and treatment in order to improve clinical management of hyperammonemia.

Introduction

Hyperammonemic disorders not due to general liver failure are rare and the symptoms non-specific. The clinical presentation varies depending on age of the patient and on type and severity of the underlying disorder. In all age groups, loss of appetite and then vomiting are early and reversible findings if treated. In newborns, first symptoms are poor feeding, vomiting, seizures, unstable body temperature, respiratory distress or poor peripheral blood circulation leading to an initial suspicion of intracranial bleeding, septicemia or meningitis; in infants, vomiting may evoke pyloric stenosis, cow milk intolerance or infectious enteritis; in children, adolescents and adults, vomiting, ataxia, confusion, disorientation, hallucinations or abnormal behavior point to central nervous system or psychiatric disorders. In all age groups, the change of consciousness should shift the search to intoxications, encephalitis or metabolic disorders. Since more common disorders are considered first, valuable time is often lost when hyperammonemia has already reached levels above 400–500 μmol/L thus increasing the risk of irreversible brain damage, of neurodevelopmental retardation or even death.

The first goal of this review is to stimulate the reader to consider the rare metabolic disorders in presence of non-specific symptoms in order to rule out hyperammonemia and—if present—to prevent irreversible damage to the brain by timely action. The second goal is to outline the principles of the available treatments and necessary controls in order to empower the pediatrician to follow and guide

J. Häberle (✉)
Kinderspital Zurich, Division of Metabolism,
University Children's Hospital Zurich,
Steinwiesstrasse 75,
8032 Zurich, Switzerland
e-mail: Johannes.Haeberle@kispi.uzh.ch

the patient when the disorder has been diagnosed. An increased awareness of all medical professionals towards the possibility of hyperammonemia is needed for an improved prognosis of affected patients.

Background

Physiology

About 90% of nitrogen-containing compounds are normally excreted as urea. They originate from the obligate oxidation of amino acids and from excess waste nitrogen, mainly from amino acids not used for protein neo-synthesis, from cell turnover of hydrolyzed body protein and from protein intake. The bulk of protein mass is in the skeletal and visceral musculature. The rate of protein breakdown is relatively constant while the synthesis is regulated e.g. by hormones, cytokines and available substrates. The capacity of urea formation adapts normally within a few days to changes of the amount of protein intake.

Ammonia (NH_3) is a constituent of all human body fluids and at neutral pH present mostly (>98%) in its ionized form, ammonium (NH_4^+) [8]. This is physiologically advantageous because ammonium, in comparison to ammonia, barely permeates cell membranes. Mainly for convention, "ammonia" is used in this review although "ammonium" would be the correct biochemical term. The concentration of ammonia in human plasma is micromolar and varies in venous, arterial or capillary blood as well as depending on the time and mode of sampling (see below). Tissue ammonia concentrations are higher and ammonia is trapped as ammonium in compartments with lower pH such as in lysosomes and renal tubules [3]. Plasma ammonia concentrations depend on the age of the patient and assay method used but it should be noted that well-defined reference limits for ammonia do not exist (limits for use in clinical practice are depicted in Table 1 [18]).

Hyperammonemia indicates an elevation of ammonia in blood and tissues by its increased production and/or decreased detoxification and is a strong indicator of

Table 1 Plasma ammonia concentrations depending on age of patients (adapted from [18])

	μmol/l	μg/dl
Newborns (arterial cord blood)	50–159	85–271
Infants and children	24–48	41–82
Adults (female)	11–48	19–82
Adults (male)	15–55	26–94

Conversion μg/dl×0.5872=μmol/l. The levels given are decision limits which should be interpreted together with the clinical situation

abnormal nitrogen homeostasis. Since in clinical practice ammonia can be determined very fast but also because of the associated neurotoxicity, ammonia belongs to the core parameters of metabolic medicine together with blood gases, glucose, lactate and ketone bodies. However, the clinical condition of a patient should guide the management rather than solely ammonia concentrations because they can be fluctuating and may not entirely correlate with already impaired brain function.

In mammals, skeletal muscle and intestinal mucosa are mainly responsible for ammonia production (Fig. 1a). Many of the reactions of amino acid metabolism take place in skeletal muscle, where protein is broken down and where single amino acids are transaminated for new protein synthesis or to form glutamine from glutamate and/or alanine from pyruvate [28]. Glutamine is not only the most abundant amino acid in the human organism, the temporary storage form of waste nitrogen and the main transport form of amino groups between organs but also a major source of ammonia if deaminated by glutaminase [48]. Also in skeletal muscle, deamination of adenosine monophosphate, particularly during physical exercise, results in ammonia production. In intestinal mucosa, ammonia is produced after uptake of amino acids as a result of glutamine deamination. In colon and bladder, microorganisms expressing enzymes enabling protein and urea degradation, respectively, can lead to hyperammonemia [40, 41, 66, 67]. About 25% of endogenous ammonia is derived from intestinal production [46, 63].

For the final transformation of glutamine/glutamate to ammonia and for detoxification of the portal ammonia and export of ammonia, the liver and to a certain extent the kidney play the central role. In liver, the urea cycle is located in periportal hepatocytes and provides a high capacity for detoxification of the vast amount of surplus nitrogen while glutamine synthetase, expressed only in perivenous hepatocytes, serves as back-up system with high affinity (but low capacity) to ammonia (Fig. 1b) [36]. Accordingly, hyperammonemia can occur in many acquired and inherited hepatic disorders. In kidney, ammonia is formed from glutamine deamination. However, renal ammonia production mainly contributes to buffering H^+ ions while excretion of ammonia in urine plays only a minor role in overall ammonia detoxification (Fig. 1a and b show the key players of ammonia production and detoxification).

Practical points

➢ Ammonia is neurotoxic itself and indicates increased ammonia production or decreased ammonia detoxification

➢ Ammonia determination is crucial for many metabolic disorders

➢ The neurological condition of the patient should guide the clinical management rather than ammonia levels alone

Fig. 1 a, **b**. Illustration of the key players of ammonia production and detoxification. **a** Simplified graph showing the interplay of intestine and skeletal muscle (organs that produce ammonia and glutamine) with liver and kidney (organs that detoxify ammonia). Ammonia is detoxified in liver while glutamine is not. **b** This simplified graph focuses on periportal and perivenous hepatocytes as the two ammonia detoxifying compartments in liver. Ammonia is metabolized with high capacity but low affinity in the urea cycle which is solely expressed in periportal hepatocytes. As back-up, ammonia is detoxified by the action of glutamine synthetase that is solely expressed in perivenous hepatocytes and has a low capacity but high affinity towards ammonia. Urea and glutamine re-enter the circulation to be excreted in urine or further metabolized in the kidney, respectively. Urea cycle enzymes abbreviated: *NAGS* N-acetylglutamate synthase, *CPS1* carbamoylphosphate synthetase 1, *OTC* ornithine transcarbamylase, *ASS* argininosuccinate synthetase, *ASL* argininosuccinate lyase, *ARG1* arginase 1
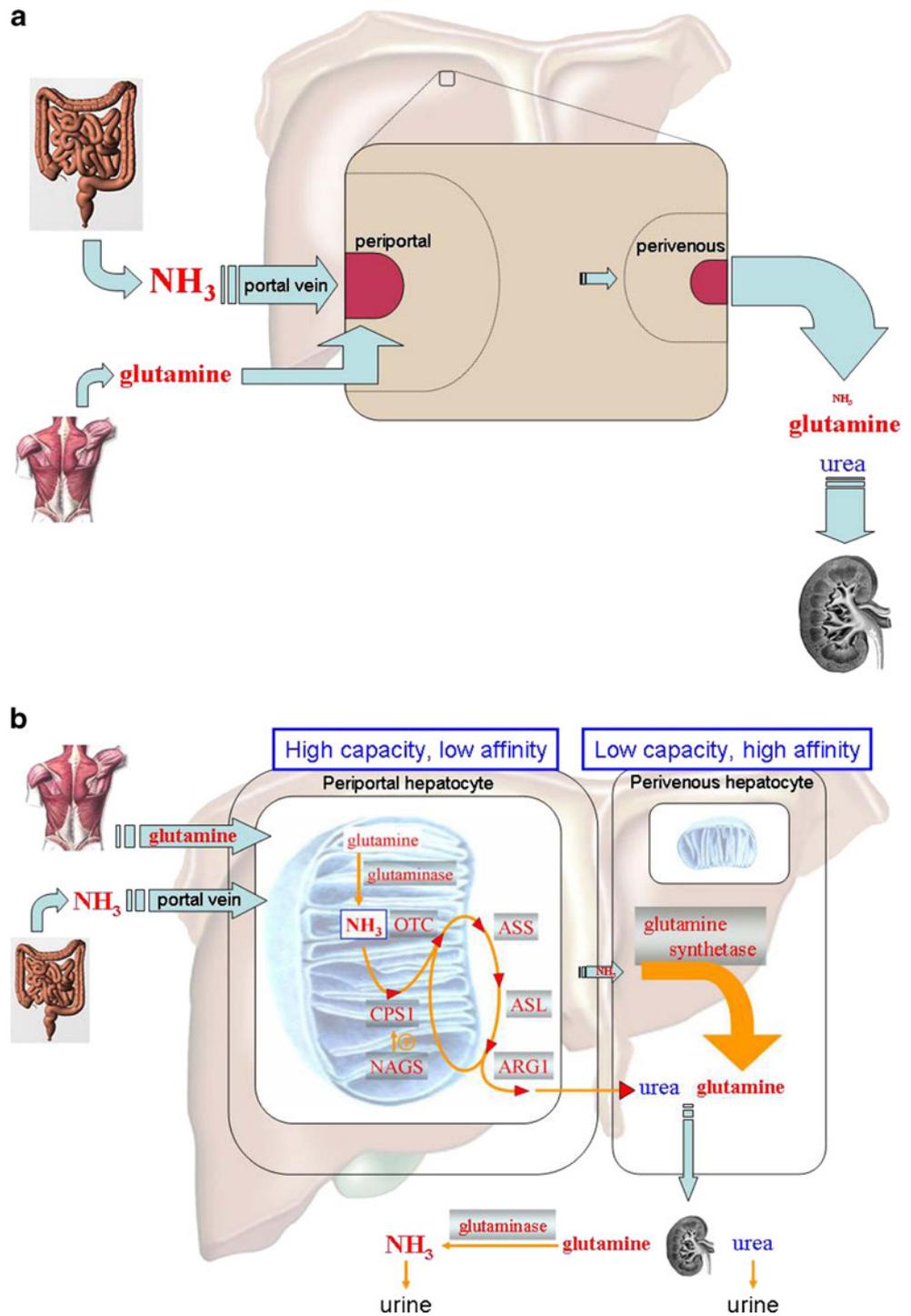
Neurotoxicity of ammonia

The brain is the main organ affected by hyperammonemia [21]. Ammonia enters the brain mainly by diffusion, but it is to a lesser extent also produced by brain metabolism [47]. A number of reversible and irreversible metabolic and neurotransmitter disturbances and ensuing morphologic changes add up to severe brain toxicity but the exact pathogenic mechanisms still need to be unraveled [4, 15, 29, 31]. Depending on age as well as duration and level of hyperammonemia severe cerebral edema, brain stem herniation and death can result. In acute hyperammonemia, astrocytes are swollen as observed by microscopy. One important factor in this pathology is the osmotic effect of newly synthesized brain glutamine on astrocytes which is taken up together with water [13, 69] but many other

mechanisms contributing to brain toxicity of ammonia have been suggested and the reader is referred to recent reviews [4, 10, 11, 14, 44, 62].

Metabolic crisis

Metabolic crises occur whenever the load of waste nitrogen exceeds the detoxification capacity. In the periportal liver (and to a lesser extent in the intestinal mucosa), the main part of ammonia from the gut is handled by urea synthesis [36]. All enzymes and membrane transporters needed are expressed in these cell systems [17, 48, 51]. In all other organs and cell systems including perivenous hepatocytes, amino groups and ammonia are detoxified by glutamine formation [37].

If the capacity for detoxification of ammonia is insufficient a vicious cycle can lead to crises. This occurs when the increasing systemic ammonia leads to loss of appetite and vomiting. Rapid intervention is needed to avoid a further increase of ammonia, i.e. when protein synthesis is reduced and catabolism prevails like during postpartum physiologic weight loss in neonates, infections (even minor ones) or increase of nutritional protein supply beyond the actual needs.

Therapeutic measures are initially non-specific in order to reduce hyperammonemia below 400–500 μmol/L or

optimally prevent its rise to such levels; a rapid diagnosis should be reached to allow the application of more efficient measures [6, 43]. Otherwise, the risk of irreversible damage to the brain is high.

Biochemical basis of primary hyperammonemia

In the mammalian organism, the major part of ammonia is detoxified by the urea cycle (Figs. 1a, b and 2). This cycle is fully expressed only in liver and intestinal mucosa and comprises six enzymatic steps of which three are intramitochondrial and three cytosolic [36]. The urea cycle has a second role—the synthesis of arginine—which is important for the treatment of hyperammonemia [12].

A defect in one of the six urea cycle enzymes and two membrane transporters results in so called primary hyperammonemia, while metabolic defects outside the urea cycle as well as side effects of drugs can lead to secondary hyperammonemia. This classification is by no means purely academic but is part of any differential diagnosis of unexplained hyperammonemia (Table 3).

The single most important of the urea cycle disorders (UCDs) is ornithine transcarbamylase (OTC) deficiency because it is the most common one and the only X-linked [57]. While male patients are often affected by severe neonatal metabolic decompensation, females with OTC
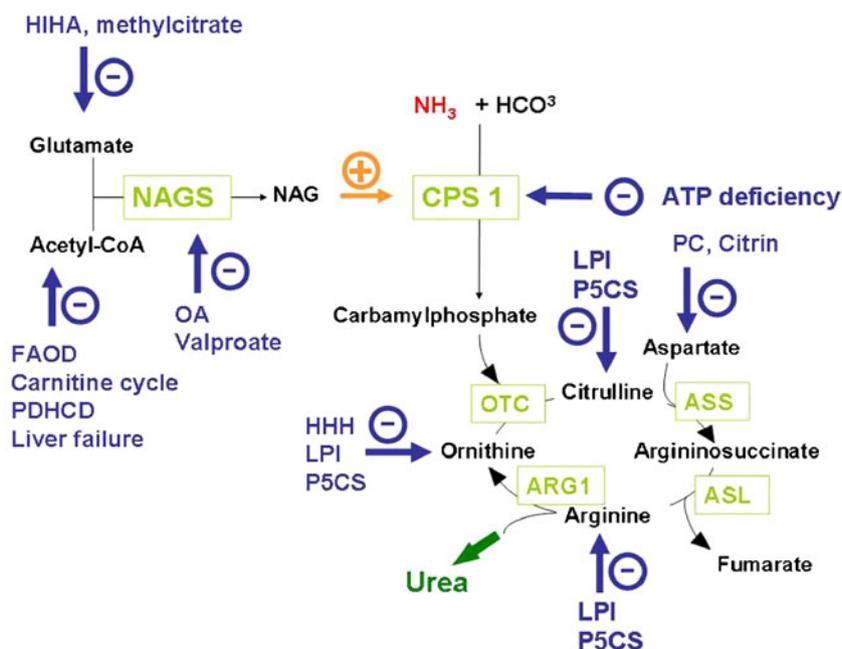

Fig. 2 Influence of metabolic disorders on function of urea cycle leading to secondary hyperammonemia. Figure showing sites of action of various compounds on urea cycle function by either leading to inhibition of enzymes (NAGS or CPS1) or to a decrease in intermediate substrates (both negative actions are depicted as ⊖). *HIHA* hyperinsulinism-hyperammonemia syndrome, *FAOD* fatty acid oxidation defects, *PDHCD* pyruvate dehydrogenase complex disor-

ders, *OA* organic acidemias, *HHH* hyperornithinemia-hyperammonemia-homocitrullinuria syndrome, *PC* pyruvate carboxylase defect, *Citrin* Citrullinemia type 2, *LPI* lysinuric protein intolerance, *P5CS* pyrroline-5-carboxylate synthetase defect. The site of action of valproate has also been added. ⊕ depicts the stimulatory effect of NAG on CPS1. Figure adapted from [53]

deficiency can present with a broad clinical picture [57, 59]. Depending on random X-inactivation, the resulting OTC phenotype is a continuum and affected females may remain asymptomatic but may also resemble hemizygous males [50]. All other UCDs are autosomal recessively inherited.

Biochemical basis of secondary hyperammonemia

Many inborn errors of metabolism result in accumulation of toxic products which can lead to inhibition of other metabolic pathways (Fig. 2). This is also the case for the urea cycle which can loose its ammonia detoxifying capacity because accumulating metabolites can impair the synthesis of N-acetylglutamate (NAG), the obligate activator of CPS1. Furthermore, some organic acids (e.g. methylcitrate in case of propionic acidemia or methylmalonic acidemia) inhibit the mitochondrial Krebs cycle and thus the availability of α-ketoglutarate as ammonia acceptor for glutamate synthesis. In addition, deficiency of acetyl-CoA or glutamate in the mitochondria or of substrates required for normal urea cycle function can lead to secondary hyperammonemia (see also Table 3).

Increased ammonia production caused by bacterial overgrowth can occur in bladders, uretero-sigmoid shunts or within the intestine. This can be seen in intestinal hypomotility of any cause, e.g. postoperative, in diabetic gastroparesis or myotonic muscular dystrophy. Besides, hyperammonemia can result if ammonia does not reach the detoxifying hepatocytes, e.g. in open Ductus venosus or portocaval shunting, and this might also contribute to the unclear phenomenon of "transient hyperammonemia of the newborn" (THAN). In addition, drugs can lead to hyperammonemia which was mostly reported secondary to valproate but other antiepileptic agents, L-asparaginase, furosemide and salicylic acid are also possible triggers of severe ammonia elevations [4, 7, 35].

Key points

➢ Primary hyperammonemia results from a defect of one of the urea cycle enzymes or transporters of ornithine or aspartate/glutamate

➢ Secondary hyperammonemia is caused by a defect outside the urea cycle that indirectly affects urea cycle function via inhibition or substrate deficiency

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Clinical signs and symptoms

General aspects of signs and symptoms of hyperammonemia

Since ammonia is toxic mainly to the brain, most signs and symptoms of hyperammonemia are neurological. This is

even true for "vomiting" as one of the most common signs of hyperammonemia at all ages which is not a pure abdominal sign but also a neurological sign.

Although non-specific at all ages, the clinical presentation will now be discussed in relation to certain age groups.

Signs and symptoms in neonates

Neonates have long been regarded as the group of patients most affected by hyperammonemia. This is not true with regard to the proportion of patients beyond the neonatal period but still partly true in clinical practice. In primary and secondary defects of the urea cycle, the pregnancy and first days of life will be uneventful because the maternal urea cycle will clear off any surplus nitrogen from the fetus. Depending on the specific defect, postnatal catabolism can lead to a clinically relevant ammonia increase within days. In severe primary defects of the urea cycle, e.g. if the intramitochondrial enzymes are defective, the asymptomatic interval may be as short as 24 h. Milder variants might only decompensate during severe states of catabolism in later life. Up to 50% of urea cycle patients present with respiratory alkalosis. Since septicemia is the most common differential diagnosis in a sick neonate and is in general accompanied by metabolic acidosis, presence of respiratory alkalosis should alert the clinician to perform an immediate re-evaluation including ammonia determinations. Confirmed septicemia does not exclude a primary hyperammonemic defect, since the catabolism associated with infection can provoke the manifestation of the genetic defect.

Signs and symptoms in infants and young children

Despite an uneventful postnatal period, affected infants and young children can manifest during any catabolic state. Especially in late infancy, protein anabolism is decreasing when postnatal growth slows down. This can be estimated from levels of urea production which are very low during rapid growth but increase after late infancy [13]. Any imbalance in energy demands, e.g. during febrile illness when nutritional intake is decreased, will result in endogenous protein catabolism and risk for hyperammonemia.

Signs and symptoms in older children and adults

Hyperammonemia can manifest for the first time at any age. Even an uneventful history with many catabolic situations but no signs of metabolic decompensation must not be interpreted as an exclusion of a primary or secondary urea cycle dysfunction. A very early in life self-chosen vegetarian diet

is a striking finding in many patients with hyperammonemia. Along this, in any unexplained neurological symptoms and especially in any unexplained encephalopathy hyperammonemia should be excluded at the very beginning of the diagnostic evaluation.

Practical points

➢ Determine plasma ammonia in all neonates with suspected septicemia

➢ Be alert if respiratory alkalosis is present in neonatal septicemia

➢ Take a good history including information regarding a self-chosen vegetarian diet

➢ Be alert if loss of appetite or vomiting are accompanied by change in consciousness

➢ Exclude hyperammonemia in every unexplained encephalopathy

➢ Exclude hyperammonemia in every unexplained change in consciousness

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Signs and symptoms of acute and chronic hyperammonemia are, irrespective of the age of the patient, summarized in Table 2.

Differential diagnosis

The list of differential diagnoses of hyperammonemic disorders is long. The most important disorders can be grouped as listed in Table 3 and as shown in Fig. 2.

Laboratory work-up

The goal is to suspect and to rule in or out hyperammonemia without delay. If hyperammonemia is confirmed, other laboratory investigations should be done including blood gases, glucose, creatinine, electrolytes, plasma acylcarnitines and amino acids, coagulation factors, albumin, pre-albumin, AST, ALT, CRP, as well as spot urine analysis for organic acids and orotic acid. For reliable results the blood should ideally be taken from a central vessel, especially in patients with poor peripheral circulation or after seizures (see below). In a comatose patient, preservation of EDTA-blood for later DNA analysis is recommended. The list of investigations should be established together with the specialized center to which hyperammonemic patients will be sent. All samples collected should be transferred together with the patient to the metabolic center taking in charge the confirmation of diagnosis and treatment of the patient. Valuable time can be gained by such a procedure.

How to diagnose hyperammonemia?

In the emergency situation, blood should be drawn at once for a rapid ammonia determination but the following preanalytical aspects should still be taken into consideration [8, 23]:

■ Measurement of ammonia requires free flowing venous or arterial blood while capillary blood is only useful for excluding hyperammonemia [3].

■ Except in emergency situations, sampling should be done in fasting state or at least 4–6 h after a meal.

■ Struggling of the child or physical exercise before the blood is taken can lead to false high ammonia concentrations.

■ To prevent false high ammonia levels secondary to hemolysis blood must be collected with the use of an anticoagulant (EDTA or heparin) and preferably in a chilled tube.

■ It has become practice in many hospitals to keep the sample on ice directly after blood is drawn. However, it should be noted that even short-time storage on ice can

Table 2 Signs and symptoms of acute and chronic hyperammonemia	Acute hyperammonemia	Chronic hyperammonemia
	Lethargy	Protein aversion and self-chosen vegetarian diet
	Somnolence	Headaches and migraine
	Coma	Tremor, ataxia, dysarthria, and asterixis
	Vomiting (metabolic alkalosis)	Confusion, lethargy, and dizziness
	Seizures	Hyperactive, aggressive, or self-injurious behavior
	Peripheral circulatory failure (metabolic acidosis)	Cognitive deficits and learning disabilities
	Cerebral edema (respiratory alkalosis)	Abdominal pain and vomiting
	Liver failure	Failure to thrive
	Multiorgan failure	Elevated liver enzymes
	Post-partum psychosis	Seizures
	In neonates, sepsis-like picture	Psychiatric symptoms
	In neonates, respiratory distress	Stroke-like episodes
	In neonates, hypo/hyperthermia	Episodic character of signs and symptoms

Table 3 Differential diagnosis of hyperammonemic disorders

Primary hyperammonemia caused by defects of urea cycle enzymes or transporters

- *N*-acetylglutamate synthase deficiency
- Carbamoylphosphate synthetase 1 deficiency
- Ornithine transcarbamylase deficiency
- Argininosuccinate synthetase deficiency
- Argininosuccinate lyase deficiency
- Arginase 1 deficiency
- Hyperornithinemia-hyperammonemia-homocitrullinuria syndrome leading to intramitochondrial ornithine deficiency
- Citrin deficiency (citrullinemia type 2) leading to aspartate deficiency

Secondary hyperammonemia caused by inhibition of the urea cycle

- Propionic acidemia leading to glutamate deficiency
- Methylmalonic acidemias leading to production of methylcitrate
- 3-Hydroxy-3-methylglutaryl-CoA-lyase deficiency
- ATP deficiency of any cause leading to secondary CPS1 deficiency
- Valproate

Secondary hyperammonemia caused by deficiency of substrates

- Fatty acid oxidation defects leading to CoA deficiency
- Carnitine cycle disorders leading to CoA deficiency
- Pyruvate dehydrogenase complex disorders leading to CoA deficiency
- Acute or chronic liver failure leading to CoA deficiency
- Lysinuric protein intolerance leading to citrulline, arginine, and ornithine deficiency
- Hyperinsulinism-hyperammonemia syndrome leading to glutamate deficiency
- Pyruvate carboxylase deficiency leading to aspartate deficiency
- Pyrroline-5-carboxylate synthetase deficiency leading to citrulline, arginine, and ornithine deficiency

Liver bypass

- Open ductus venosus
- Vascular malformations resulting in portocaval shunting

Unclear

- Transient hyperammonemia of the newborn

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lead to partial freezing resulting in hemolysis; therefore, ice-water should be used instead.

- Plasma should be separated as soon as possible but certainly within 15–30 min.

Practical points

➢ Get an immediate ammonia analysis if hyperammonemia is suspected
➢ Obtain free flowing blood
➢ Use capillary blood only for exclusion of hyperammonemia
➢ Separate plasma within 15–30 min
➢ Use ice-water for transport of the sample instead of storage on ice

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Laboratory tests

There are different analytical methods of ammonia determination including titration, colorimetric/fluorimet- ric, electrode-based and enzymatic methods [8]. It should be noted that the normal values given by the manufacturers of some methods are unfortunately sometimes inaccurate. In clinical practice, a widely used method utilizing glutamate dehydrogenase (GLDH) determines ammonia concentrations by analyzing the decrease in absorption at 340 nm caused by oxidation of NADPH as shown in the following equation: α-ketoglutarate + NH_3 + NADPH $\xrightarrow{\text{GLDH}}$ glutamate + $NADP^+$.

Bedside tests

The major advantage of bedside ammonia determination is to get a rapid result and thus fast information on nitrogen homeostasis. If bedside tests are done correctly, i.e. from free flowing blood drops (no massage or squeezing of heels and fingertips) in warm heels or fingers picked in non-traumatized tissue, the result will be probably reliable and available within 3 min. However, the result will only allow to exclude hyperammonemia.

In addition, there are some concerns towards the use of ammonia bedside tests especially outside the hospital: one is related to the fact that due to contamination by intracellular fluid, capillary blood ammonia levels might be elevated. Another problem is the limited capability to precisely measure high ammonia levels (the upper limit of ammonia bedside tests is often below 300 μmol/L) [24] which might lead to underestimation and thus a dangerous delay. Furthermore, ammonia measurements at home can, opposite to their intention, lead to increased stress within the family caused by repetitive analyses. However, if used in a hospital setting, ammonia bedside tests can speed up the time to diagnosis in emergency situations both of known and not yet known patients.

Outlook: continuous monitoring of ammonia

Analyzing ammonia continuously instead of only a few times a day would allow for better understanding of fluctuating levels and closer regulation of therapies but this is not yet feasible. Recently, measurement of human breath ammonia using a liquid-film conductivity sensor was reported [56], but it remains to be seen whether this or yet another method for continuous monitoring will come into clinical practice.

What to do with slightly raised plasma ammonia concentrations?

If plasma ammonia is only slightly raised, it might be difficult to decide how to proceed. Considering the preanalytical pitfalls of ammonia determination, slight increases of ammonia might well be artificial. The following approach might be helpful in clinical practice:

- Repeat the plasma ammonia immediately.
- Find out the conditions under which the blood was collected, in particular, was the sample obtained after a meal and was the child struggling?
- Find out the conditions the sample was handled, in particular, how long was the time to analysis and storage?
- Review the clinical and family history very carefully. In particular, is there any evidence of previous episodes of encephalopathy—mild or severe?

Amino acids

Ammonia levels cannot replace close observation of plasma amino acid profiles including their careful interpretation [60]. Therefore, in any patient with hyperammonemia amino acid profiles are necessary not only for differential diagnosis in newly diagnosed patients but also for optimal treatment.

Thus, amino acids must be controlled and interpreted during the emergency phase at least daily and later in the course less frequently but at least every 3 months.

Of utmost importance is the concentration of glutamine which is less fluctuating than ammonia and most other amino acids and plays a central role in hyperammonemic neurotoxicity [1, 22]. The major part of glutamine in the CNS is newly synthesized within the brain while the transport system at the blood–brain barrier has an only low affinity for glutamine. At the same time, transaminations by glutaminase in endothelial cells [68] can diminish differences of arterio-venous glutamine concentrations even in presence of experimental hepatic coma. Thus, increased plasma glutamine levels are an indicator of the increased nitrogen load of the organism but are by no means a predictor or even precursor of brain glutamine.

In addition, essential amino acids need to be closely monitored with special emphasis on branched chain amino acids (BCAA) [3, 54, 55, 58]. These are a major nitrogen source for endogenous glutamine synthesis and might become depleted in hyperglutaminemia. Also, the nitrogen scavenger drug phenylacetate is known to aggravate BCAA deficiency because nitrogen molecules lost by excretion of phenylacetylglutamine originate in relevant amounts from BCAA [55].

Neuroimaging

To estimate the effect of acute or chronic ammonia on the brain, neuroimaging can provide additional information. Unfortunately, widely available techniques such as ultrasound (by scanning through the open fontanel in neonates and young infants) and computed tomography cannot detect acute or moderate changes of brain morphology except very severe brain edema. Color duplex sonography might help to detect brain edema but there are no studies on the sensitivity of this method in hyperammonemia. However, the role of neuroimaging in management of hyperammonemia has been evolving in recent years mainly using functional magnetic resonance imaging (fMRI), diffusion tensor imaging (DTI) and ^{1}H/^{13}C magnetic resonance spectroscopy (MRS) [33]. While ammonia cannot be followed directly by MRS, hyperammonemia leads to a number of secondary changes. One is the change in glutamine and myoinositol levels detected by MRS [20, 33]. Studies have shown that both acute and chronic hyperammonemia result in an increase of glutamine and at the same time in a decrease in myoinositol which functions as a compensatory organic osmolyte in astrocytes [32, 33, 69].

Although the role of neuroimaging needs further evaluation, it will hopefully become an important tool mainly to define the effect of subclinical hyperammonemic

episodes and of chronic moderate hyperammonemia and hyperglutaminemia on the brain.

Management of hyperammonemic crisis

Management of hyperammonemic crises is beyond the resources of non-specialized hospitals and transfer to a metabolic center is needed to allow efficient interventions.

How to proceed if hyperammonemia is suspected?

The result of plasma ammonia should be available within 30-(60) min at all times. Meanwhile, protein supply should be stopped and glucose supplied (see below and Fig. 3). Any somnolent or pre-comatose patient suspected to be

hyperammonemic should be transferred to a specialized center without delay, together with urine and plasma samples (kept on ice or frozen) and this will allow to gain time. The center should have the resources for ruling out or establishing the diagnosis within 12 h and for starting the specific therapy of a metabolic crisis.

Practical points
➢ Actively seek the result of plasma ammonia within 30–(60) min
➢ Stop protein supply
➢ Start glucose infusion±insulin (control blood lactate after 2 h)
➢ Contact metabolic center for advice on management
➢ Transfer any somnolent or pre-comatose patient (together with urine and plasma samples for further analyses) to the experienced center
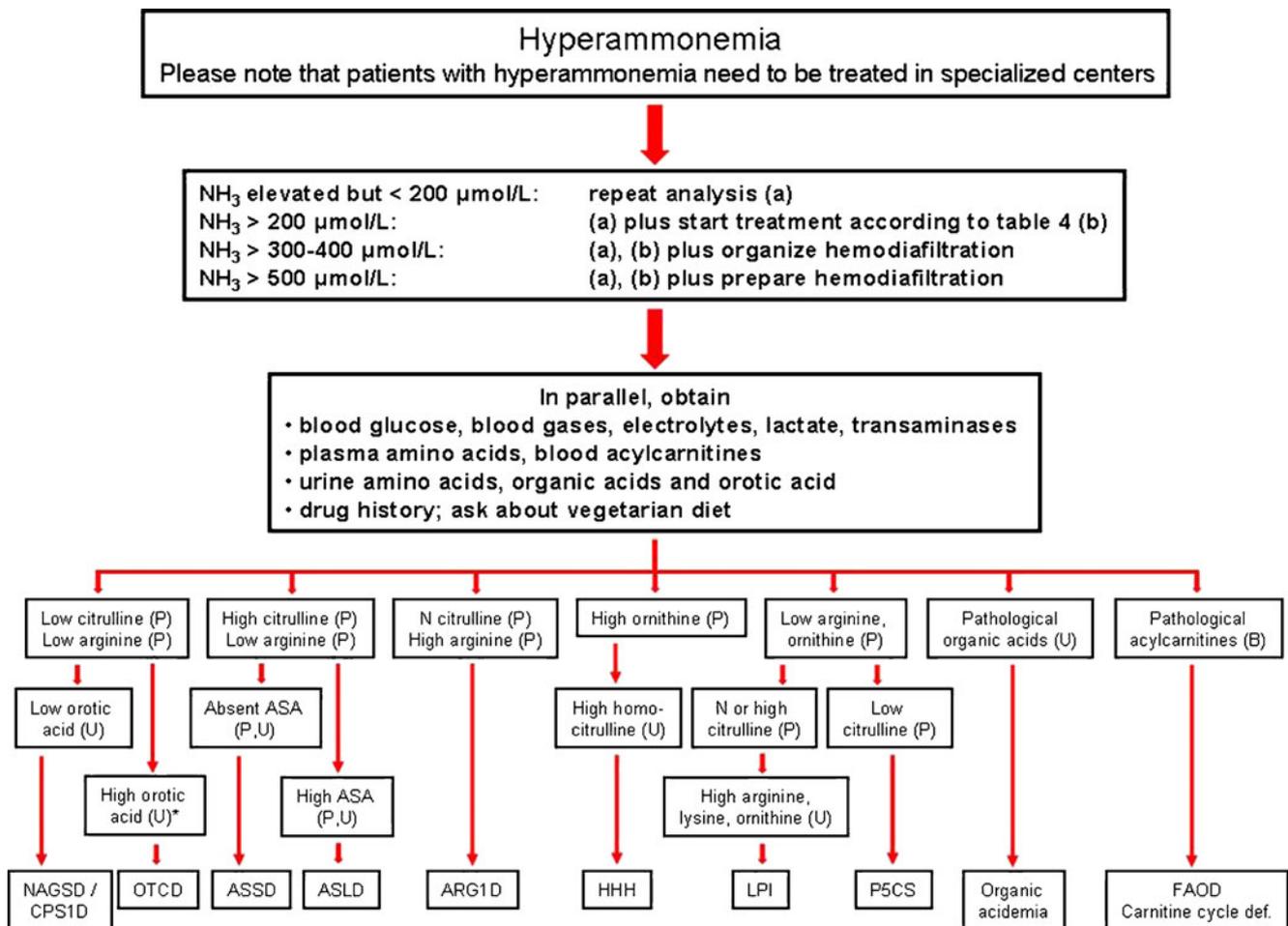

Fig. 3 Algorithm for the management of hyperammonemia in a newly recognized patient. Algorithm for the management of hyper-ammonemia in a newly recognized patient and for interpretation of first laboratory results. The ammonia limits given are decision limits which should guide the next steps. With this algorithm, most of the differential diagnoses listed in Table 3 can be detected. Citrin deficiency (where high citrulline and arginine in plasma can be found) was not included in this figure because it is extremely rare outside Japan. *P* plasma, *U* urine, *B* blood, *N* normal, *D* behind the enzyme abbreviations stands for deficiency, *ASA* argininosuccinic acid (specific metabolite of ASLD). *Asterisk* Orotic acid in urine might initially be low or only slightly elevated in a male OTC newborn (personal communications, M. Lindner, T. Marquardt, B. Fowler)

How to proceed if hyperammonemia is confirmed?

Hyperammonemia is always an emergency situation. Treatment should start immediately after elevated plasma ammonia has been found and before any specific diagnosis is made (see also Fig. 3). Since hyperammonemia is the result of protein catabolism, this must be reversed as fast as possible. Exogenous protein should be stopped for 24–48 h but exogenous nitrogen sources make up for only a small proportion of surplus nitrogen if endogenous protein catabolism is ongoing. Thus, it is the combination of stopping exogenous protein, reversing endogenous protein catabolism by high doses of parenteral glucose sometimes requiring additional insulin and the use of detoxifying drugs that need to be initiated as fast as possible [13, 27]. If hyperammonemia exceeds levels 300–400 μmol/L hemodiafiltration as the preferred method or, if not available, hemodialysis or hemofiltration must be prepared rapidly [6, 43]. The detailed approach to this depends on the local conditions but, as mentioned above, patients with a metabolic crisis should always be treated in specialized centers. See also Table 4.

Management of primary hyperammonemia

Until the diagnosis is available (ideally within 12–18 h after detection of ammonia >200 μmol/L), stop of protein supply and continuous infusion of high dose parenteral glucose (in newborns at least 8–10 mg/kg/min) is maintained [6]. A tentative treatment with carbamylglutamate has recently been proposed in order to lower ammonia in known propionic and methylmalonic acidemia, known NAGS deficiency or even any unknown neonatal hyperammonemia [34, 61].

In primary UCDs, specific conservative treatment options comprise the use of urea cycle metabolites distal to the metabolic block. If hyperargininemia is excluded, L-arginine HCl (initially intravenously) is the substance most often used but citrulline can also be given in the mitochondrial UCDs. Use of L-arginine aims at restoring residual function of the urea cycle allowing for urinary excretion of nitrogen as intermediate urea cycle metabolites (citrulline or argininosuccinate) but also at avoiding arginine deficiency [13, 43].

In addition, nitrogen scavenger drugs have a large impact on treatment of hyperammonemia [9, 13, 26]. In Europe, there are only two substances available, one of which is a licensed drug (sodium phenylbutyrate) and the other a chemical agent (sodium benzoate). For acute intervention, sodium benzoate with or without sodium phenylacetate (which is the active metabolite of sodium phenylbutyrate) can be administered intravenously. Care should be taken to avoid intoxication by excessive dosage in newborns since their drug metabolizing enzymes need 4–5 days induction (Bachmann C, personal communication). Phenylacetate binds glutamine to form phenylacetylglutamine while benzoate binds glycine to form hippurate which are both excreted in urine. Both conjugations take place in liver and require the presence of coenzymeA (CoA). At start of treatment, a loading dose of L-arginine as well as of nitrogen scavenger drug(s) is given intravenously over 2 h followed by a maintenance continuous infusion (see Table 4). Repetition of loading doses is not recommended because of benzoate or phenylbutyrate toxicity [13]. If nitrogen scavengers are used, plasma amino acids need to be controlled daily. The benzoate dose must be reduced if glycine is below 100 μmol/L. If phenylacetylglutamine excretion is enhanced, leucine and isoleucine might become deficient within days resulting in ongoing catabolism and limitation of protein neo-synthesis unless they are substituted.

Energy should be provided as glucose and lipids covering 110% of the energy demand. Enteral feeding should be started as soon as possible on the basis of a low protein diet [25, 42]. Breast milk can be given in limited amounts (up to about 0.7 g protein/kg body weight/d) but

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Table 4 Management of acute hyperammonemia

■ Stop oral protein intake and intravenous amino acid infusions

■ Provide high-energy intake, either oral as 10–25% glucose polymer or intravenous as glucose 8–10 mg/kg b.w./min (dose for neonates/small infants) plus, in case of hyperglycemia, insulin

■ Transfer the patient to specialized center together with plasma and urine spot samples

■ Give intravenous L-arginine-HCl 2 mmol/kg b.w. as loading dose in 2 h, then 2 mmol/kg b.w. as continuous infusion in 24 h

■ Consider nitrogen scavengers (should not be given in secondary hyperammonemias): sodium benzoate 250–350 mg/kg b.w. as loading dose in 2 h, then 250–350 mg/kg b.w. as continuous infusion in 24 h, and/or sodium phenylacetate 250–350 mg/kg b.w. as loading dose in 2 h, then 250–350 mg/kg b.w. as continuous infusion in 24 h

■ Consider use of carbamylglutamate 200 mg/kg b.w. divided in three oral doses

■ Plan hemodiafiltration if initial ammonia is >500 μmol/L or if ammonia does not fall within 2 h after start of treatment

the protein tolerance depends on the severity of the defect and is highly variable between patients [43]. To treat and prevent deficiencies of essential amino acids (EAAs), special mixtures of EAAs which should be high in branched chain amino acids [54] and low in neurotransmitter precursors are given (as intravenous solution, e.g. Aminosteril Hepa®, Fresenius; as enteral feed, e.g. UCD®, Milupa or EAM®, SHS) [3]. The diet must be adapted daily during the first weeks of treatment. Hidden sources of protein supply such as blood or plasma transfusions should be avoided or taken into account.

It should be noted, that excessive restriction of protein supply and the ensuing amino acid imbalance and BCAA deficiencies can cause ongoing catabolism resulting in chronic moderate hyperammonemia.

Besides, other treatment modalities are currently discussed including L-ornithine-L-aspartate to increase protein synthesis in skeletal muscle [39], hypothermia to decrease energy demands in brain [64], N-methyl-D-aspartate receptor antagonists to reduce glutamate toxicity [45] and others but these are not yet proven concepts.

Practical points

➢ Provide at least 110% of daily energy demand, either oral or intravenous
➢ Use L-arginine to restore urea cycle function
➢ Use nitrogen scavengers, but don't use repeated loading doses
➢ Consider use of carbamylglutamate
➢ Start enteral feeding as soon as possible based on a low protein diet
➢ Consider supplementation of nutrients (EAAs, vitamins, minerals, trace elements)

Treatment of secondary hyperammonemia

If hyperammonemia is secondary to inborn errors of metabolism other than UCDs, it can still be life threatening to the same extent as in primary hyperammonemia. Nevertheless, treatment is based on reversion of endogenous protein catabolism but is otherwise focused on the underlying disorder. In organic acidemias (including the iatrogenic hyperammonemia of valproate treatment) drugs used for increasing waste nitrogen excretion in UCDs are critical because they might result in toxic drug accumulation due to intramitochondrial CoA depletion. Likewise, amino acid mixtures high in BCAA should not be given. If bacterial overgrowth in intestine is considered to cause or contribute to hyperammonemia, eradication of urease producing bacteria with non-absorbable antibiotics (metronidazole, neomycin) should be aimed for. Also, acidification of the intestine with lactulose to shift NH_3 to NH_4^+ can contribute to decreased ammonia uptake [2, 19].

Practical points

➢ Define and treat the underlying disorder
➢ Provide at least 110% of daily energy demand, either oral or intravenous
➢ Be cautious with the use of nitrogen scavenger drugs in secondary hyperammonemia
➢ Treat and prevent intestinal bacterial overgrowth by non-absorbable antibiotics
➢ Consider acidification of intestinal tract with lactulose

Prognosis of hyperammonemia

For primary UCDs, the prognosis depends not only on the genotype but on many endogenous and exogenous factors. Patients without any residual enzyme activity generally have a poor prognosis and often die in the first few days of life. On the other extreme, females with OTC deficiency with a favorable mosaicism and high levels of residual in vitro activity usually have a good prognosis.

External causes like mild viral or bacterial infections with sudden predominant catabolism or events leading to prolonged vomiting can lead to excessive breakdown of proteins and hence to hyperammonemia. In addition, severe liver disease, trauma, extensive surgical interventions, excessive protein intake or the post-partum period can lead to the first metabolic decompensation well into adulthood. Still, the consequences can be lethal. Since these events cannot be predicted, any prognostic statement for an individual patient is almost impossible and survival statistics are of limited help.

Acute episodes

Acute episodes of hyperammonemia are always life threatening [26]. A retrospective study analyzing the outcome of neonatal onset UCDs showed that ammonia levels at presentation above 300 μmol/L carry a high risk of death or severe neurological sequelae and peak levels above 480 μmol/L at first hospitalization were never associated with normal outcome [5]. Although these data are historical, first manifestations of acute hyperammonemia carry a serious prognosis at all ages and unknown patients are at risk because any delay in start of treatment contributes to the poor outcome [65]. A recent cohort study revealed an up to 45% mortality of boys with late-onset OTC deficiency at the time of diagnosis irrespective of the age at onset [49].

Treatment modalities remained basically unchanged during the last decades and this is certainly one factor influencing the still poor prognosis of acute episodes of hyperammonemia. However, an even larger problem is the

delay in start of appropriate treatment often due to lack of awareness towards the rare differential diagnosis of an inborn error of metabolism. Improved prognosis does largely depend on shortening the time interval to diagnosis [52] and this might even be more effective for the overall cohort of patients than developing new treatment modalities although these are much needed.

Chronic hyperammonemia

There are no firm data on the relevance of moderate chronic hyperammonemia (which is a non-standard term used for ammonia levels of up to 200 μmol/L). Likewise, there is no safe upper level of ammonia concentration [4]. However, it should be noted that "unexplained" moderate hyperammonemia might be caused by excessive protein restriction or caused by therapy with nitrogen scavengers itself. Data from patients suffering from hepatic encephalopathy, an adult disorder associated with chronic hyperammonemia, point towards morphological changes of astrocytes, i.e. development of so called Alzheimer type II astrocytes as well as radical RNA damage and protein tyrosine nitration [15, 16, 38]. Recent results of spectroscopy studies performed in asymptomatic OTC carriers revealed changes in astrocytic osmolyte balance caused by chronic elevations of glutamine [30, 32]. Thus, there is cumulating evidence for a pathological role of chronic moderate hyperammonemia but this area needs further studies. For the time being, it appears to be advisable to strictly avoid any state of even moderate, seemingly asymptomatic hyperammonemia in order to avoid neurological damage.

Key points

➢ Good prognosis depends largely on shortening the time until start of therapy
➢ Subclinical hyperammonemic episodes probably have an impact on brain function
➢ Even moderate hyperammonemia should be avoided

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Additional note