

Drug-induced hyperammonaemia

Loai Shakerdi,¹ Aidan Ryan ^{2,3}

¹National Centre for Inherited Metabolic Disorders, Mater Misericordiae University Hospital, Dublin, Ireland

²Chemical Pathology, Cork University Hospital Biochemistry Laboratory, Cork, Ireland

³Pathology, University College Cork College of Medicine and Health, Cork, Ireland

Correspondence to

Dr Aidan Ryan, Chemical Pathology, Cork University Hospital Biochemistry Laboratory, Cork, Ireland; aidan.ryan1@hse.ie

Received 23 November 2022

Accepted 28 April 2023

Published Online First

10 May 2023

ABSTRACT

Hyperammonaemia (HA) as a consequence of numerous primary or secondary causes, gives rise to clinical manifestations due to its toxic effects on the brain. The neurological consequences broadly reflect the ammonia level, duration and age, with paediatric patients being more susceptible. Drug-induced HA may arise due to either decreased ammonia elimination or increased production. This is associated most frequently with use of valproate and presents a dilemma between ongoing therapeutic need, toxicity and the possibility of an alternative cause. As there is no specific test for drug-induced HA, prompt discussion with a metabolic physician is recommended, as the neurotoxic effects are time-dependent. Specific guidelines for managing drug-induced HA have yet to be published and hence the treatment approach outlined in this review reflects that outlined in relevant urea cycle disorder guidelines.

INTRODUCTION

Hyperammonaemia (HA) ($>40 \mu\text{mol/L}$, $68 \mu\text{g/L}$ in adults, although method dependent) arises due to a number of causes and in general, clinical manifestations broadly reflect the degree of ammonia elevation, arising due to central nervous system toxicity.^{1,2} Ammonia is a highly potent neurotoxin and severe acute HA is a medical emergency. It may result in a rapidly progressive, often fatal, encephalopathy with brain damage. Chronic HA may cause progressive cognitive impairment, behavioural abnormalities and neuropsychiatric illness. Patients may present with encephalopathy, with confusion, agitation, ataxia, seizures or coma, particularly as values of plasma ammonia increase above $100\text{--}200 \mu\text{mol/L}$, however the threshold for clinical symptoms is highly variable.³ Clinical features are non-specific and can arise in more common conditions such as sepsis. As a consequence of this, HA is often missed as it is not part of the standard panel for encephalopathy in adult general medicine outside of hepatology. In adults, most cases arise due to liver failure, and although rare there is a growing appreciation that inherited metabolic disorders (IMDs) can present at any age.

An important source of adult-onset non-cirrhotic HA are IMDs which lead to primary or secondary urea cycle disorders (UCDs) and conditions associated with increased ammonia production such as urease-producing infections⁴ (see tables 1 and 2). Although the overall estimated incidence for all UCDs is rare at 1 in 35 000, there are some cohort data supporting the view that 50% present in adolescence/adulthood and of those presenting in adulthood 50% present acutely.^{5–7} Timely management of the first presentation is an important predictor of

neurological outcome with persistent values $>360 \mu\text{mol/L}$ in early onset and $>200 \mu\text{mol/L}$ in adult onset, being associated with poorer neurological outcomes.^{6–8} This is important and reinforces the need to consider checking ammonia in any patient of any age with encephalopathy, as early treatment can reverse the neurological deterioration.

This review provides a brief overview of the biochemistry of primary and secondary causes of HA and reviews in detail drug-induced HA and its management in adults. Much has been written about the preanalytical factors that can affect ammonia analysis and we assess this in a systematic fashion. A similar approach is used in describing relevant diagnostic testing and management, to avoid missing relevant primary disorders and treatment delays that may exacerbate neurological outcomes.

PATHOPHYSIOLOGY

Ammonia is a metabolic by-product of all cells. The major contributors to the body load are the gastrointestinal (GI) tract, skeletal muscle and kidney. Ammonia is produced in the GI tract from three sources: bacterial urease breakdown of circulating urea, bacterial deamination of luminal protein and glutaminase breakdown of glutamine from circulating glutamine.⁹ Large amounts of ammonia are generated in skeletal muscle, especially during activity/seizures mainly from deamidation of AMP, and to a lesser extent from amino acid catabolism. In kidneys it is released from glutamine entering the proximal tubular cells from the glomerular filtrate and the circulation.

Because of its neurotoxicity, arterial blood concentration is normally tightly regulated and does not vary much even after meals with the main sites of production being the GI tract, skeletal muscle and the kidney. At physiological pH in body fluids, ammonia (NH_3) is predominantly hydrogenated to ammonium (NH_4^+). At neutral pH NH_4^+ predominates, which is useful as this limits its perfusion across cell membranes. Throughout the text 'ammonia' refers to summated NH_3 and NH_4^+ concentrations, which is the common convention.

Ammonia is toxic to the brain but not to other tissues and readily crosses the blood-brain barrier.¹⁰ Average levels of brain ammonia are normally approximately twice those of blood. In experimental animals with acute liver failure, brain ammonia flux may be up to 45-fold higher than normal.¹¹

A number of theories have been proposed to explain HA-induced brain damage.^{12–18} It has been postulated that increased intracerebral ammonia levels may interfere with mitochondrial function, may disrupt inhibitory and excitatory neurotransmission, and lead to excessive glutamine glial accumulation leading to astrocyte swelling. One of the



© Author(s) (or their employer(s)) 2023. No commercial re-use. See rights and permissions. Published by BMJ.

To cite: Shakerdi L, Ryan A. *J Clin Pathol* 2023;**76**:501–509.

Table 1 Inherited metabolic disorders which may cause primary hyperammonaemia (HA)⁸

Urea cycle disorders	
Enzyme Deficiency	
1. Ornithine carbamoyl transferase deficiency	
2. Carbamoyl phosphate synthetase deficiency	
3. Citrullinaemia	
4. Argininosuccinic aciduria	
5. Argininaemia	
6. N-Acetylglutamate synthetase deficiency	
Transporter Defect	
1. Citrin deficiency-aspartate glutamate carrier	
2. Hyperornithinaemia, HA and homocitrullinuria (3 hour syndrome)-ornithine transport	

most concerning outcomes of this pathology is that severe HA can cause cerebral oedema with a rise in intracranial pressure that can lead to herniation and brainstem compression.

Large amounts of NH₃ are produced daily. NH₃ not recycled for biosynthetic processes must be excreted rapidly from the body. This is achieved by conversion to urea via the urea cycle in the liver, which is then excreted by the kidneys. The conversion of ammonia into urea requires the function of the six enzymes and two transporters of the urea cycle. The liver is the only organ that houses the complete urea cycle of 5five enzymes (two mitochondrial, three cytosolic), together with N-acetylglutamate synthetase (NAGS) which produces N-acetylglutamate (NAG). NAG acts as an activator of carbamoylphosphate synthetase (CPS1) (see [table 1](#) and [figure 1](#)).

In order to transfer NH₃ safely from the tissues to the liver via the circulation, it is combined with glutamate by glutamine

synthetase to produce non-toxic glutamine. Some is extracted by the kidneys and the immune system for biosynthesis. Most is taken up by the small intestinal mucosa. Here NH₃ is released by glutaminase and transported directly to the liver in the portal circulation.⁹

Cytosolic ornithine is transported into the mitochondria in exchange for intramitochondrial citrulline by the ornithine/citrulline carrier encoded by the *SLC25A15* gene. Citrin, a liver transporter encoded by the *SLC25A13* gene, exports aspartate from the mitochondria in exchange for glutamate that is used in the formation of NAG ([figure 1](#)). Two atoms of nitrogen are converted to urea for each turn of the urea cycle. One comes from ammonia and the other comes via the citrin carrier and the amino acid pool in the form of aspartate. The latter derives from the amino group of alanine, which is transferred to 2-oxoglutarate to form glutamate initially and then onto oxaloacetate to produce aspartate. NAGS produces NAG which is an essential cofactor for CPS1, the first and rate-limiting enzyme of the urea cycle.^{19 20}

DRUG-INDUCED HA

A number of medications have been associated with HA, with valproate being the most common. Confounding factors such as sepsis and liver disease, and lack of clear mechanisms for some drugs weaken the evidence for association.

In a search of the WHO adverse drug reaction database (Vigibase), for the period 1967 to 8 May 2019, 73 drugs were reported to be suspected of causing HA. [table 3](#) lists those most frequently recorded; 63% were associated with valproate, 11% with fluorouracil, 5% topiramate and the rest accounting less than 5% each of overall cases.²¹ Vigibase is a retrospective observational database used to record all adverse drug reactions from over 130 countries, and the table below lists the most frequently recorded.

The following were excluded: drugs reported less than three times and drugs used to treat HA or hepatic encephalopathy. There are some obvious limitations to using this as a source as it relies on spontaneous notification, retrospectively, so delay may affect recall, incomplete information, lack of direct inspection of laboratory data and undernotification. For instance, the case numbers with liver failure are low compared with the estimated population prevalence and this may represent the fact that clinicians attributed the HA (in those cases excluded) to liver disease rather than to a particular drug.

Part of the challenge in dealing with such HA is teasing out confounders such as liver disease, sepsis, malnutrition, IMD or a drug effect. Although for the drugs we have tabulated in [table 3](#) there is some mechanistic linking the drug to HA, for some medications this is not the case. In addition, this is hampered by the absence of a specific test to rule in or rule out the drug-associated HA. To strengthen the evidence for association, Weiss *et al* extended their Vigibase study by checking potential causation checking time of onset of HA to discontinuation and whether this correlated with the product characteristics documented from the European Medicines Agency and the Federal Drug Authority.^{21 22} Of interest is the fact that 74% of the cases were reported in the last decade of the search period; a median time of onset of HA following drug commencement was 13 days (IQR 2–59 days).²² Of the 73 drugs, 10 described the association with HA in the drug product characteristics (valproic acid (VPA), valpromide (VPA prodrug), topiramate, asparaginase, fluorouracil, haloperidol, pegaspargase, zonisamide, deferasirox, amphotericin B). [table 3](#) lists the most frequently recorded drugs

Table 2 Differential diagnosis and mechanism of primary and secondary hyperammonaemia (HA)^{11 79 81}

Increased ammonia production	
Condition	Suggested mechanism
Exercise/seizures/trauma	AMP deamination
Gastric bypass/starvation	Increased protein catabolism
GI haemorrhage Total parenteral nutrition	Excess protein/nitrogen load
Infections with urease-producing bacteria	Urinary tract infection, with relevant organisms splitting urea to form ammonia
Decreased ammonia elimination	
Acute or chronic liver disease	Reduced urea cycle, glutamine synthesis; portosystemic shunt
Urea cycle disorder	Enzyme block or substrate transport affecting urea cycle
Fatty acid disorder of oxidation	Lack of acetyl-CoA leading to reduced CPS1 activity
Organic acidaemia	NAGS inhibition by relevant increased acid
Carbonic anhydrase Va deficiency	Lack of bicarb leading to reduced CPS1
Mitochondrial disorders	Impaired ATP production/substrate
Ornithine aminotransferase deficiency	Lack of ornithine affecting OTC, urea cycle defects
Glutamine synthase deficiency	Decreased glutamine and hence ammonia clearance
Lysinuric protein intolerance	Lack of urea cycle ornithine and arginine

CPS1, carbamoylphosphate synthetase; GI, gastrointestinal; NAGS, N-acetylglutamate synthetase; OTC, ornithine transcarbamylase.

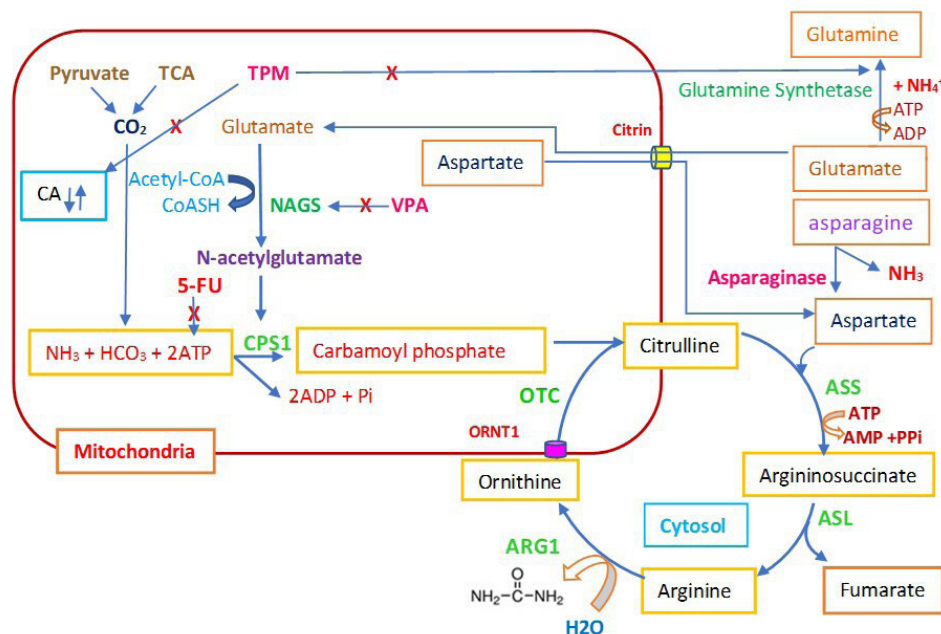


Figure 1 Urea cycle diagram including normal enzymes/transporters and where each of the drugs is purported to block. Asparaginase catalyses the formation of aspartic acid and ammonia from asparagine. ADP, adenosine diphosphate; AMP, adenosine monophosphate; ATP, adenosine triphosphate; ARG1, arginase 1; ASL, argininosuccinate lyase; ASS, argininosuccinate synthetase; CA, carbonic anhydrase; CoASH, coenzyme A; citrin, mitochondrial aspartate/glutamate carrier; CPS1, carbamoylphosphate synthetase 1; CO₂, carbon dioxide; H₂O, water; HCO₃, bicarbonate; NAGS, N-acetylglutamate synthase; ORNT1, ornithine/citrulline antiporter; OTC, ornithine transcarbamylase; Pi, phosphate; PPI, pyrophosphate; TPM, topiramate; TCA, tricarboxylic acid cycle; VPA, valproic acid; 5-FU, 5-fluorouracil; X, denotes the site of medication inhibition.

associated with HA. The frequency of HA was uncommon for asparaginase (between 1/100 and 1/1000 cases), rare (between 1/1000 and 1/10000) for VPA, valpromide, topiramate and undetermined for the other six drugs. Of the remaining 61 drugs, 6 were published in reference databases/books, 45 were described in case reports/series and 10 had never been published. This might reflect greater awareness of HA as a drug complication and/or a higher incidence with introduction of new drugs (see reference²² for a comprehensive list).

VALPROATE

VPA (2-n-propylpentanoic acid) is a C8-branch chain fatty acid that has been used for many years to treat epilepsy, more recently as mood stabiliser for a number of psychiatric conditions and migraine. In a recent audit of general hospitalised adults who received VPA, 20.4% developed HA.²³ This audit included those patients admitted over a 1-year period in one hospital, age >18

years, receiving at least one dose of VPA. Patients with cirrhosis were excluded from the audit. HA was defined as above the institutional reference range of >23.6 µmol/L, 162 patients were included and ammonia ranged from 12 µmol/L to 75 µmol/L. Epilepsy and psychiatric condition were the most frequent indication for VPA and the mean plasma ammonia in symptomatic patients was 39.5 µmol/L. A limit for this paper is that it was retrospective, duration of VPA use was not recorded and neither was dose. However, the strengths of the study include the large sample size and general hospital location, rather than a specific medical discipline.

A similar study conducted on patients admitted to a psychiatric medicine unit had a prevalence of HA of 36%.²⁴ Symptoms of VPA-induced HA can vary from asymptomatic HA to VPA-induced hyperammonaemic life-threatening encephalopathy.²⁵

Various mechanisms have been implicated in the development of VPA-induced HA, mainly via effects on the liver and less so on the kidney. VPA β-oxidation leads to valproyl-CoA (valproyl Coenzyme A) which directly inhibits NAGS and induces secondary carnitine deficiency which leads to decreased acetyl-CoA that may also contribute to VPA-induced HA^{26 27} (figure 1). Hepatotoxicity (non-specific variable) may accompany this and a dose-dependent effect on plasma ammonia has been observed with increasing VPA doses and during use with other antiepileptic drugs (AEDs). Elevations in plasma ammonia levels, as high as 140 µmol/L, were well tolerated, and VPA dose reductions were not necessary.²⁸ However, in an audit of AED use, in the VPA only group, ammonia concentration ranges from 60 µmol/L to 400 µmol/L were recorded, with some being well above the treatment threshold (see below for treatment section).²⁹ Risk factors for hepatotoxicity associated with VPA include young age, polytherapy and developmental delay.³⁰ Although rare, coexisting UCD suggested by a sudden

Table 3 List of drugs commonly associated with HA²¹

Drug	Cases of HA	Serious events (%)	Fatal events (%)
1 Valproate	1260	768 (61)	54 (4)
2 Fluorouracil	221	213 (96)	20 (9)
3 Topiramate	96	65 (68)	0 (0)
4 Oxaliplatin	74	68 (92)	12 (16)
5 Asparaginase	71	50 (70)	6 (8)

Column 3 lists the absolute number of cases associated with HA for each drug. Columns 4 and 5 list the absolute number and % in brackets of serious events and fatal events. For serious events these were defined as severe case, death, life-threatening, caused or prolonged hospitalisation, disabling/incapacitating, congenital anomaly/birth defect, other medically important condition. For seriousness criteria it was possible to choose more than one. HA, hyperammonaemia.

development of hyperammonaemic crises should be considered. Hyperammonaemic crises may be triggered by catabolic events, protein overload, infection, GI bleeding or certain drugs.^{31 32} This needs to be considered when requesting relevant laboratory tests (see below laboratory investigations).

TOPIRAMATE

Topiramate (TPM) is a carbonic anhydrase inhibitor used to treat epilepsy and alcohol dependence, and prevent migraines. HA is an uncommon side effect of topiramate and is usually related to combined use with valproate and phenobarbital.³³ It is believed that the mechanism implicated in HA involves inhibition of carbonic anhydrase leading to reduced bicarbonate and reduction in activity of the cytosolic enzyme, glutamine synthetase. Reduced bicarbonate may decrease activity of CPS1, the rate-limiting enzyme in the urea cycle (figure 1).³⁴ The reduction in glutamine synthetase levels limits the incorporation of ammonia into glutamate, to form glutamine. Both of these mechanisms can lead to HA; however, it should be noted that both of these studies were in animal models and there are no human studies to date. Values of ammonia of up to 77 µmol/L in a single use of topiramate have been recorded and up to 146 µmol/L in combination with other AEDs as noted above.^{33–35} Addition of VPA to a patient already taking topiramate may increase the risk of HA.³⁶

FLUOROURACIL

5-fluorouracil (5-FU) is a common chemotherapy drug used in the treatment of head and neck, GI, and breast cancers. 5-FU is a pyrimidine analogue that acts principally as a thymidylate synthase inhibitor. After entering the cell, 5-FU is converted into several metabolites. The rate-limiting step of the catabolic process is the conversion of 5-FU to the inactive metabolite dihydrofluorouracil which is catalysed by the enzyme dihydropyrimidine dehydrogenase (DPD).³⁷ Decreased metabolism by DPD creates a number of potentially toxic metabolites and hence it is a standard practice to screen for DPD variants (associated with decreased metabolism) that increase this risk, in particular of severe neutropaenia.³⁸ To date there has been some suggestion that such variants in association with other factors contribute to HA. Boilère *et al* found an association with DPD deficiency in 27% and Milano *et al* found an association with neurotoxicity in 37%, but did not record ammonia values.^{39 40}

HA has been reported after treatment with 5-FU, with early symptoms being non-specific, such as nausea and vomiting progressing to encephalopathy.⁴¹ Large doses of 5-FU induce accumulation of fluoroacetate which can inhibit the enzyme *aconitase* and result in the impediment of citrate isomerisation. This in turn impairs Krebs cycle function reducing ATP³⁸ and oxaloacetate.⁴² Impairment of ATP-dependent urea cycle results in HA. These are two essential factors in the early proximal part of the urea cycle, with oxaloacetate providing the source of aspartate via aspartate transaminase (figure 1).

The reported incidence of encephalopathy has been around for 10% with HA being around 1.0%.^{40 43} Values of ammonia up to 522 µmol/L have been recorded, which is well above the therapeutic intervention threshold (see the Treatment section below).^{40 43} However, it should be borne in mind that often these patients may have multiple confounding factors such as sepsis, shock, sarcopenia and hepatic impairment, that need to be considered. The original reports described an acute cerebellar syndrome associated with HA and since then several case studies and cohort studies have been published.^{44–47}

For clinicians treating such patients the challenge is teasing out often what may not be multifactorial encephalopathy, due to the cancer, medications or malnutrition. A paraneoplastic encephalopathy is more likely to be subacute, however 5FU also interferes with the conversion of thiamine-to-thiamine pyrophosphate (its active form) which can present as Wernicke's encephalopathy. This is compounded by the fact that such a group is nutritionally vulnerable and may already be deficient in thiamine, among other vitamins.⁴⁸

OXALIPLATIN

Oxaliplatin a member of the platinum chemotherapy class, undergoes non-enzymatic conversion to active derivatives which preferentially bind guanine and cytosine bases in DNA leading to cross linking. This arrests DNA synthesis, with effects not being dependent on cell cycle, being particularly efficient on tumours with high cell turnover.^{49 50} It can be used to treat a variety of metastatic tumours but is commonly in combination with 5-FU to treat colorectal tumours. The drug causes dose-dependent toxicity on the haematopoietic system and dose-limiting effects on the nervous system causing acute or chronic peripheral neuropathy affecting multiple types of nerve fibres.⁵¹

HA has been uncommonly described, usually in combination with 5-FU and hepatic dysfunction, therefore may have synergistic effects, on the urea cycle and might increase the risk for HA.^{52 53} The exact mechanism however remains unclear but may be related to negative effects on the Krebs cycle impacting the proximal urea cycle. Values of ammonia of up to 200 µmol/L have been recorded comfortably above the treatment threshold (see the Treatment section).^{54 55}

ASPARAGINASE

Asparaginase has been a long-standing component of induction, consolidation and maintenance therapy for acute lymphoblastic leukaemia.^{56 57} The principal of this chemotherapy is that neoplastic cells have reduced capacity to synthesise asparagine and hence need this from the blood.

Asparaginase catalyses the formation of aspartic acid and ammonia from asparagine and hence an increase in ammonia is expected as a consequence of its therapeutic effect. In a prospective cohort study, although 7/10 had HA as expected none were symptomatic and although peak levels increased up to seven times upper-normal limit 24 hours after dose, concentrations fell to normal by 2–3 days.⁵⁸

Symptomatic HA has previously been reported to be uncommon, however in a study where ammonia was checked regardless of symptoms, ammonia >100 µmol/L was found in 7 out of 10 patients being even higher in those where PEG-asparaginase was used, due to the prolonged half-life.⁵⁹ It is clear therefore that ammonia should be checked as a regular part of therapeutic management regardless of symptoms, otherwise this may be missed. Values of ammonia up to 400 µmol/L have been documented in setting of asparaginase use, well above the therapeutic threshold for intervention for HA (see the Treatment section). Other metabolic effects such as hypertriglyceridaemia due to antagonistic effect on lipoprotein lipase have also been described.

OTHER MEDICATIONS

Use of corticosteroids has been associated with increased skeletal muscle amino acid metabolism and thus has been believed to be due to increased ammonia production.⁶⁰ However, decrease in glutamine synthetase levels, carbamoyl-phosphate synthase 1,

ornithine transcarbamylase (OTC), arginosuccinate synthase 1 and arginosuccinate lyase have also been implicated.^{61 62} Imoto *et al* also looked at the case series of use of corticosteroids in those with OTC deficiency and found a high rate of mortality with mean ammonia 761 $\mu\text{mol/L}$ (ranging from 233 $\mu\text{mol/L}$ to 3039 $\mu\text{mol/L}$), well above the intervention threshold (see table 5).⁶² Due to the effect on a number of key UCD enzymes, the authors recommend early intervention with renal replacement therapy and certainly close monitoring of ammonia in this group post corticosteroid intervention.

Of historical interest is Reye's syndrome, a viral (varicella and influenza most common) induced acute liver failure due to mitochondrial injury; this was associated with high morbidity and mortality.⁶³ The risk was reported to be increased in those under 12 years when aspirin was used to control fever, which led in turn to recommendation not to use aspirin in those age groups during viral infection. Reye's syndrome peaked in the late 1970s, early 1980s and is now quite rare.⁶⁴ It is best viewed as multifactorial, including viruses, however some cases also had an underlying IMD, mainly UCD or fatty acid disorder of oxidation.⁶⁵

Combination therapy of VPA plus other AEDs (such as phenytoin, phenobarbital, carbamazepine and/or topiramate, zonisamide) has also been shown to be associated with increased risk of HA.^{55 56} While the mechanisms for HA are clear in some, others remain uncertain and so clinicians should be aware of the increased risk of HA in combination therapy particularly in those with symptoms associated with HA.^{58 59}

CLINICAL PRESENTATION

HA can present with a number of non-specific features with guidelines suggesting that checking of ammonia should be considered in any patient presenting with acute/intermittent neurological/psychiatric presentation, acute liver failure or in the differential of sepsis with a view to making a UCD diagnosis.⁶⁶ Most patients present after a catabolic trigger, such as intercurrent infection, postpartum, vomiting and diarrhoea, relevant medication or a high protein meal.

In all ages, loss of appetite and vomiting are common, but in adolescents/adults the encephalopathy, hallucinations or psychiatric symptoms or signs of tremor, seizures predominate.⁶⁶ Often these clinical features are mistaken for meningitis, brain tumour or intoxication, hence the heightened testing for checking ammonia in such clinical scenarios to avoid delay in diagnosis and adverse clinical outcomes. Around 50% of those over 16 years/adults with UCD can present acutely and mortality of up to 10% has been noted with approximately 90% requiring intensive care admission.^{7 67}

LABORATORY INVESTIGATIONS

Abnormal ammonia levels in adults should trigger further investigation with a particular focus on liver disease or a treatable IMD. Early engagement with a metabolic physician is recommended, where chronic liver disease is unlikely, in order to help expedite testing and facilitate acute management. First evaluations include: liver enzymes, arterial or venous acid-base status, renal function and electrolytes.

Low blood urea, normal glucose with a high ammonia and respiratory alkalosis increases the likelihood of UCD. The low urea could be due to UCD or protein restriction, however it should be noted that this is not a very sensitive marker and a normal urea should not be used to rule out UCD. In this context, urgent metabolic testing of urine organic acids, plasma amino acids and acylcarnitines, should be undertaken within 24 hours.

The urine organics acids are used to identify orotic acid, uracil in OTC deficiency, as well any evidence for an organic aciduria or dicarboxylic aciduria, glycines; in the case of a fatty acid disorder of oxidation. The plasma amino acids in the case of UCD are used to determine underlying UCD. In general, glutamine is elevated in UCD, and normal in organic aciduria and fatty acid disorder of oxidation. Citrulline would be low in OTC and proximal UCD, but elevated in those distal to OTC (see figure 1). A high anion gap metabolic acidosis is indicative of organic aciduria. (For more detailed discussion on interpreting relevant metabolic tests see reference 66.⁶⁶)

As there is no specific test to confirm that HA is secondary to a specific medication, ruling out an IMD with next-generation sequencing panel testing is usually undertaken as outlined in the relevant guideline and other sources.^{66 68 69} The sensitivity of genetic testing has improved with developments in sequencing technology, but particularly in OTC this is still around 90% and additional functional or enzymatic analysis maybe required.⁶⁹

ANALYSIS OF PLASMA AMMONIA

Although historically there have been a wide variety of laboratory methods, indirect enzymatic methods that measure either nicotinamide adenine dinucleotide phosphate (NADPH) or nicotinamide adenine dinucleotide (NADH) reduction at 340 nm are most frequently used. table 4 lists some of the relevant preanalytical factors including method and effect on ammonia. By far the most significant is the delay to transfer and separation from red cells as the concentration in red blood cells is approximately three times that of plasma and represents potential for ammonia contamination (table 4).⁷⁰ This is one of the main reasons for using plasma as the sample of choice rather than serum. Ice is used by some as a stopgap measure to reduce in vitro ammonia formation, in the absence of a suitable blood tube additive inhibitor. However, if sample handling or processing is prompt, the benefits of using ice are insignificant (table 4). The ammonia content range in 24-hour urine collection has been estimated at 4–24 mmol/L, however it is not widely available and so despite inaccuracies, underestimated use of urine/osmolal gap persists.⁷¹

There is a huge degree of variability between studies and different assays in terms of time cut-offs, compounded by a lack of a robust analytical performance specification. In the introduction we used the metbionet definition of HA ($>40 \mu\text{mol/L}$, $68.12 \mu\text{g/L}$). However, to illustrate the importance of considering the laboratory method, a recent Wales External Quality Assurance (WEQAS) ammonia return with an overall method mean of 41 $\mu\text{mol/L}$, had a method range of 23–50 $\mu\text{mol/L}$ which would impact on diagnosis.⁷² Similarly, another recent WEQAS ammonia EQA return with an overall mean of 79 $\mu\text{mol/L}$, had a method range of 61–102.1 $\mu\text{mol/L}$, which is also likely to impact on treatment choice.⁷²

One of the few studies to assess biological variation in healthy individuals showed a within-individual coefficient variation of approximately 14%, within-group coefficient of variation of approximately 17% and a reference change value of 43%.⁷³

TREATMENT

There are no evidence-based guidelines to deal with drug-induced HA and no specific tests. There is a variety of different approaches including the Australian guidelines for managing asparaginase-induced HA;⁷⁴ guidelines proposed by Boilève *et al* for 5-FU-induced toxicity⁴⁰ and expert panel recommendations on prevention and management of asparaginase/pegasparaginase-associated toxicities in adults and older adolescents.⁷⁵ These

Table 4 Ammonia preanalytics, patient groups and laboratory method

Condition	Patients	Method	Effect on mean plasma ammonia (µmol/L)	Reference
Power grip variable intensity for 15 mins	Hospital	Seligson diffusion	No effect in controls until high intensity (96 µmol/L), effect on cirrhotics increased across all levels, 30 µmol/L to 87 µmol/L	91
Sweat	Healthy	Seligson diffusion	10–50 times blood ammonia-sweat ammonia increasing with decreasing sweat pH	92
Smoking tobacco	Hospital	Ion-exchange/phenol	After 1 hour an increase of 10 µmol/L	93
Capillary versus venous sample	Hospital	Roche Cobas Bio with Glutamate dehydrogenase (GLDH) Monotest (Boehringer Mannheim)	Increase in capillary of 74 µmol/L compared with venous plasma of 18 µmol/L	94
Platelet-rich versus platelet-poor sample	Hospital	Roche Cobas Bio with GLDH Monotest (Boehringer Mannheim)	Platelet-rich plasma ammonia of 34 µmol/L versus platelet-poor plasma ammonia of 21 µmol/L	94
Temperature over 90 min	Healthy	Enzymatic method	Mean rates of increase at 0°C, 20°C and 37°C were 3.9 µmol/L, 5.2 µmol/L and 25.2 µmol/L per hour, respectively	95
Platelet, erythrocyte, alanine transferase and gamma-glutamyl transferase	Healthy	Enzymatic method	Positive correlation	95
Tourniquet and clench	Hospital	Technicon RA-XT (Bayer Diagnostics, Basingstoke, UK) GLDH (Sigma diagnostics, Poole UK).	Increase of 60–75 µmol/L	96
Effect of hepatic dysfunction	Hospital	An enzymatic–Ultra violet kit from Thermo Electron Corporation (Infinity) was used on our AU640 analyser (Olympus UK Ltd, Hertfordshire) versus (Vitros 250, Ortho-Clinical Diagnostics, UK).	Due to one step enzymatic may over estimated ammonia due to NADH consumption, leading to a positive interference which can be up to threefold difference and likely to influence clinical practice	97
Haemolysis and Haemolysis Index cut-off	Unclear	Roche Integra 800	Haemolysis did not add anything over delay in time to separation Haemolysis Index at least 200 at ammonia 28 µmol/L	98
Temperature including ice, chill centrifuge versus room temperature	Healthy	AU2700 analyser (Beckman Coulter) using Randox Ammonia Reagent GLDH enzymatic (Randox, Crumlin, Co, Antrim, UK)	25.1 µmol/L versus 27.6 µmol/L for ice versus room temperature at 30 min, chilling centrifuge made no difference If there was further delay of 30 min then difference of 11 µmol/L seen between ice, chill and room temperature	99
Smoking cannabis	Hospital	Colorimetric assay kit (BioVision Inc., Milpitas, CA)	After 90 min an increase of 35 µmol/L was seen	100
EDTA versus Li-Hep versus oxalate	Healthy	Roche COBAS c501 GLDH	EDTA was more precise and stable once separated- 0.322 µmol/h at 4°C, 0.122 µmol/hour at –14°C, and negligible change at –70°C	89
Protein of 30 g and 60 g intake after 2–3 hours	Healthy	PocketChem BA blood ammonia, microdiffusion (Lancashire, United Kingdom)	Maximum mean increases of 54 µmol/L for 30 g at 2 hours and 71 µmol/L for 60 g at 3 hours	90

approaches include stopping, dose reduction or continuing with measures to deal with HA. If it is decided to continue the medication then close monitoring is required and testing for other causes should be undertaken, the urgency for which is dependent on the severity of the clinical symptoms and progression of ammonia.

Stopping the offending drug suspicious for HA if possible and supportive treatment is an essential first step in management. Some cohort studies have explored this using valproate dose reduction or withdrawal and appropriate AED substitution where required, and HA has resolved.^{76–77} It must be emphasised that treating other factors such as hepatic dysfunction, sepsis, nutrition and medication adjustment as well as HA are also essential.^{66–77–79} Management of hepatic dysfunction and sepsis will not be discussed here.⁷⁸ Also, although use of lactulose in the setting of cirrhosis has been shown in a meta-analysis of randomised clinical trial data to reduce mortality and morbidity (variceal bleed, hepatic encephalopathy, hepatorenal syndrome), its effects on lowering ammonia directly are modest; mean reduction in ammonia is 12 µmol/L (95% CI reduction of 21 µmol/L to 2 µmol/L).⁸⁰ Therefore, outside of cirrhosis, lactulose is not recommended for management of HA.⁶⁶

A metabolic physician should be consulted immediately once the diagnosis of HA has been confirmed. HA associated with symptoms must be treated as soon as possible. From the

authors' experience the initial steps will usually involve intravenous glucose plus ammonia scavengers (see tables 5 and 6). Any underlying dehydration will require 0.9% sodium chloride (normal saline) and if the patient is shocked or obviously unwell then admission to a high density care or intensive care unit should be arranged. Regular assessment with Glasgow coma scale (GCS), blood gas (pH, electrolytes), ammonia is routinely recommended (see tables 5 and 6). However a full updated clinical examination and blood tests is recommended if the patient becomes encephalopathic or ammonia is increasing despite initial therapy. These patients are also at risk of hyponatraemia and hypokalaemia.

With recent adult cohort data suggesting adverse neurological outcomes with ammonia values >200 µmol/L, this would suggest that more aggressive therapy (haemodiafiltration) should be considered earlier at this value.^{6–79–81} Ultimately however therapeutic interventions should be guided by both clinical presentation and ammonia level as occasionally even patients with ammonia >200 µmol/L maybe asymptomatic (tables 5 and 6). A coma for more than 3 days, raised intracranial pressure and ammonia >1000 µmol/L are particularly poor prognostic factors.⁶⁶

Sodium benzoate and sodium phenylbutyrate (the precursor of the active agent phenylacetate) promote ammonia removal from the body by combining with glycine to form hippurate; and with

Table 5 Levels of HA and suggested actions in case of symptomatic undiagnosed patients

Ammonia level (μmol/L)	Action	Investigations
Increased above normal limit but ≤100	Reduce/stop protein intake, give intravenous glucose 6 mg/kg/min±insulin*	Monitor ammonia blood levels every 3 hours
>100 and <250	Start intravenous L-arginine and sodium benzoate, carbamylglutamate, carnitine, vitamin B ₁₂ , biotin Correct electrolytes and phosphate	Monitor ammonia, electrolytes and phosphate
≥250	As above Avoid repetitive drug boluses Begin haemodiafiltration if no rapid drop of ammonia within 3–6 hours	As above Monitor supplement early especially during haemodialysis

Table is adapted from Häberle *et al.*⁶⁶

*If hyperglycaemia (glucose >10 mmol/L) occurs, start insulin infusion as per local diabetic protocol, with close monitoring.

glutamine to form phenylacetylglutamine which are excreted in urine. This alternative pathway reduces the amount of NH₃ presented to the urea cycle for detoxification. In theory each mole of benzoate removes one mole of ammonia and for phenylbutyrate this is two moles of ammonia, however due to variability in pharmacokinetics and pharmacodynamics, this is not achieved. However, such scavengers will not work adequately if hepatic function is compromised.

Dietary management of acute decompensation is crucial to prevent or stop catabolism and to correct biochemical abnormalities and ensure adequate nutritional intake. Protein intake should be reduced or paused completely. Adequate amounts of calories should be provided either enterally or parenterally (10%–20% dextrose and 20% fatty acids solutions with appropriate electrolytes: Na⁺, K⁺).^{82 83} This will require input from an experienced metabolic dietitian as prolonged restriction for greater than 48 hours can lead to catabolism and worsening in HA.

Haemodialysis is the most effective therapy for HA and may serve as a rescue therapy. Intermittent haemodialysis achieves the highest ammonia clearance, however due to patient stability or raised intracranial pressure continuous veno-venous haemodialysis may be more suitable.⁸⁴ Most of the evidence around use of dialysis in HA is based on paediatric rather than adult based studies.

Liver transplantation is the definitive cure for cirrhosis and for urea cycle defects. It is considered the last resource for patients with recurrent decompensation and poor response to conventional therapies, but these are high-risk situations and require careful discussions if the patient's condition allows.⁸⁵

Table 6 Dosages of commonly used drugs in acute hyperammonaemic decompensations in undiagnosed adult patients

Drug	Initial dose (bolus)	Maintenance dose
Sodium benzoate*	250 mg/kg as bolus in 90–120 min	250–500 mg/kg/day >20 kg body weight: 5.5 g/m ² /day
Sodium PBA/sodium phenylacetate*	250 mg/kg as bolus in 90–120 min	250–500 mg/kg/day
L-arginine Hydrochloride*	250–400 mg/kg (1–2 mmol/kg) bolus in 90–120 min	250 mg/kg/day (1.2 mmol/kg/day)
N-carbamylglutamate†	100 mg/kg bolus per NG tube	25–62.5 mg/kg every 6 hours

Table is adapted from Häberle *et al.*⁶⁶

*Sodium benzoate, sodium phenylbutyrate (sodium PBA)/sodium phenylacetate, L-arginine hydrochloride to be given intravenously in glucose 10%. Nasogastric tube (NG).

†Only available as oral/enteral drug

There are a number of case reports/cohort studies suggesting the use of carnitine or L-arginine supplementation in the management of valproate-induced HA.^{72 86}

CONCLUSION

It is important to measure ammonia in any patient of any age with encephalopathy, as early treatment can reverse the neurological deterioration or prevent death. Clinicians treating patients with the medications listed in this paper should be aware of the possibility of drug-induced HA. A greater awareness by clinicians of the importance of early diagnosis of HA and late-onset UCDs should help reduce the risk of life-threatening complications.^{87 88}

Detailed medical history and laboratory tests are essential to establish diagnosis. The main role of laboratory testing is to identify liver disease and IMD. Eliciting relevant clinical factors should cover precipitants such as polypharmacy, coexistence of a relevant IMD, hypercatabolic state, poor nutrition and other comorbid conditions.^{89 90} Adjustment of dose or discontinuation of offending drug may be necessary in symptomatic patients with new-onset HA. However, in those with confirmed HA, urgent contact with a metabolic physician should be undertaken as more specific therapies as listed above maybe required.

Handling editor Patrick J Twomey.

Contributors AR and LS contributed equally to the drafting, writing and editing process.

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests None declared.

Patient consent for publication Not applicable.

Ethics approval Not applicable.

Provenance and peer review Commissioned; externally peer reviewed.

ORCID iD

Aidan Ryan <http://orcid.org/0000-0002-7443-689X>

REFERENCES

- 1 Network NMB. Guidelines for the investigation of hyperammonaemia. 2023. Available: <https://metbio.net/wp-content/uploads/MetBio-Guideline-PERE918546-10-12-2018.pdf>
- 2 Aitkenhead H. UK national audit of the measurement of ammonia. *Ann Clin Biochem* 2023;60:117–25.
- 3 Summar M, Tuchman M. Proceedings of a consensus conference for the management of patients with urea cycle disorders. *J Pediatr* 2001;138:S6–10.
- 4 Stergachis AB, Mogensen KM, Khoury CC, *et al.* A retrospective study of adult patients with noncirrhotic hyperammonemia. *J Inher Metab Dis* 2020;43:1165–72.
- 5 Waters D, Adeloye D, Woolham D, *et al.* Global birth prevalence and mortality from inborn errors of metabolism: a systematic analysis of the evidence. *J Glob Health* 2018;8:021102.

- 6 Kido J, Matsumoto S, Häberle J, et al. Role of liver transplantation in urea cycle disorders: report from a nationwide study in Japan. *J Inherit Metab Dis* 2021;44:1311–22.
- 7 Toquet S, Spodenkiewicz M, Douillard C, et al. Adult-onset diagnosis of urea cycle disorders: results of a French cohort of 71 patients. *J Inherit Metab Dis* 2021;44:1199–214.
- 8 Häberle J. Primary hyperammonaemia: current diagnostic and therapeutic strategies. *J Mother Child* 2020;24:32–8.
- 9 Meijer AJ, Lamers WH, Chamuleau RA. Nitrogen metabolism and ornithine cycle function. *Physiol Rev* 1990;70:701–48.
- 10 Wright G, Noiret L, Olde Damink SWM, et al. Interorgan ammonia metabolism in liver failure: the basis of current and future therapies. *Liver Int* 2011;31:163–75.
- 11 Walker V. Severe hyperammonaemia in adults not explained by liver disease. *Ann Clin Biochem* 2012;49:214–28.
- 12 Dejong CH, Deutz NE, Soeters PB. Cerebral cortex ammonia and glutamine metabolism in two rat models of chronic liver insufficiency-induced hyperammonemia: influence of pair-feeding. *J Neurochem* 1993;60:1047–57.
- 13 Braissant O, McLin VA, Cudalbu C. Ammonia toxicity to the brain. *J Inherit Metab Dis* 2013;36:595–612.
- 14 Walker V. Ammonia metabolism and hyperammonemic disorders. *Adv Clin Chem* 2014;67:73–150.
- 15 Erceg S, Monfort P, Hernández-Viadel M, et al. Oral administration of sildenafil restores learning ability in rats with hyperammonemia and with portacaval shunts. *Hepatology* 2005;41:299–306.
- 16 Cooper AJ, Plum F. Biochemistry and physiology of brain ammonia. *Physiol Rev* 1987;67:440–519.
- 17 Rama Rao KV, Norenberg MD. Glutamine in the pathogenesis of hepatic encephalopathy: the Trojan horse hypothesis revisited. *Neurochem Res* 2014;39:593–8.
- 18 Sen K, Whitehead M, Castillo Pinto C, et al. Fifteen years of urea cycle disorders brain research: looking back, looking forward. *Anal Biochem* 2022;636:114343.
- 19 Shigesada K, Tatibana M. Enzymatic synthesis of acetylglutamate by mammalian liver preparations and its stimulation by arginine. *Biochem Biophys Res Commun* 1971;44:1117–24.
- 20 Moedas MF, Adam AAA, Farelo MA, et al. Advances in methods for characterization of hepatic urea cycle enzymatic activity in HepaRG cells using UPLC-MS/MS. *Anal Biochem* 2017;535:47–55.
- 21 Weiss N, Salem J-E, LeBrun-Vignes B, et al. P: 65 drug-induced hyperammonaemia: data from vigibase, the WHO database. *Am J Gastroenterol* 2019;114:532.
- 22 Balcerac A, Bihan K, Lebrun-Vignes B, et al. Drug-associated hyperammonaemia: a Bayesian analysis of the who pharmacovigilance database. *Ann Intensive Care* 2022;12:55.
- 23 McMorris T, Chu A, Vu L, et al. Hyperammonemia in patients receiving valproic acid in the hospital setting: a retrospective review. *Ment Health Clin* 2021;11:243–7.
- 24 Baddour E, Tewksbury A, Stauner N. Valproic acid-induced hyperammonemia: incidence, clinical significance, and treatment management. *Ment Health Clin* 2018;8:73–7.
- 25 Wu J, Li J, Jing W, et al. Valproic acid-induced encephalopathy: a review of clinical features, risk factors, diagnosis, and treatment. *Epilepsy & Behavior* 2021;120:107967.
- 26 Aires CCP, van Cruchten A, IJlst L, et al. New insights on the mechanisms of valproate-induced hyperammonemia: inhibition of hepatic N-acetylglutamate synthase activity by valproyl-CoA. *J Hepatol* 2011;55:426–34.
- 27 Silva MFB, Ruiter JPN, Overmars H, et al. Complete beta-oxidation of valproate: cleavage of 3-oxovalproyl-CoA by a mitochondrial 3-oxoacyl-CoA thiolase. *Biochem J* 2002;362:755–60.
- 28 Murphy JV, Marquardt K. Asymptomatic hyperammonemia in patients receiving valproic acid. *Archives of Neurology* 1982;39:591–2.
- 29 Yamada H, Shishido T, Mukai T, et al. Valproic acid-induced hyperammonemic encephalopathy in a patient receiving valproic acid monotherapy. *Rinsho Shinkeigaku* 2019;59:258–63.
- 30 Bryant AE, Dreifuss FE. Valproic acid hepatic fatalities. III. U.S. experience since 1986. *Neurology* 1996;46:465–9.
- 31 Felig DM, Brusilow SW, Boyer JL. Hyperammonemic coma due to parenteral nutrition in a woman with heterozygous ornithine transcarbamylase deficiency. *Gastroenterology* 1995;109:282–4.
- 32 Trivedi M, Zafar S, Spalding MJ, et al. Ornithine transcarbamylase deficiency unmasked because of gastrointestinal bleeding. *J Clin Gastroenterol* 2001;32:340–3.
- 33 Tantikittichaiikul S, Johnson J, Laengvejkal P, et al. Topiramate-induced hyperammonemic encephalopathy in a patient with mental retardation: a case report and review of the literature. *Epilepsy Behav Case Rep* 2015;4:84–5.
- 34 Häussinger D, Kaiser S, Stehle T, et al. Liver carbonic anhydrase and urea synthesis. The effect of diuretics. *Biochem Pharmacol* 1986;35:3317–22.
- 35 Gomez-Ibañez A, Urrestarazu-Bolumburu E, Viteri-Torres C. Hyperammonemic encephalopathy related to valproate, phenobarbital, and topiramate synergism. *Epilepsy Behav* 2011;21:480–2.
- 36 Deutsch SI, Burket JA, Rosse RB. Valproate-Induced hyperammonemic encephalopathy and normal liver functions. *Clin Neuropharmacol* 2009;32:350–2.
- 37 Longley DB, Harkin DP, Johnston PG. 5-Fluorouracil: mechanisms of action and clinical strategies. *Nat Rev Cancer* 2003;3:330–8.
- 38 van Kuilenburg ABP, De Abreu RA, van Genip AH. Pharmacogenetic and clinical aspects of dihydropyrimidine dehydrogenase deficiency. *Ann Clin Biochem* 2003;40:41–5.
- 39 Milano G, Etienne MC, Pierrefite V, et al. Dihydropyrimidine dehydrogenase deficiency and fluorouracil-related toxicity. *Br J Cancer* 1999;79:627–30.
- 40 Boilève A, Thomas L, Lillo-Le Louët A, et al. 5-Fluorouracil-Induced hyperammonaemic encephalopathy: a French national survey. *European Journal of Cancer* 2020;129:32–40.
- 41 Yi HJ, Hong KS, Moon N, et al. Acute hyperammonemic encephalopathy after 5-fluorouracil based chemotherapy. *Ann Surg Treat Res* 2016;90:179–82.
- 42 Koenig H, Patel A. Biochemical basis for fluorouracil neurotoxicity. The role of Krebs cycle inhibition by fluoroacetate. *Arch Neurol* 1970;23:155–60.
- 43 Mitani S, Kadowaki S, Komori A, et al. Acute hyperammonemic encephalopathy after fluoropyrimidine-based chemotherapy: a case series and review of the literature. *Medicine (Baltimore)* 2017;96:e6874.
- 44 Marella HK, Peravali R, Jain AL, et al. Hyperammonemic encephalopathy associated with 5-fluorouracil in a patient with previous orthotopic liver transplantation. *Proc (Bayl Univ Med Cent)* 2020;33:256–7.
- 45 Kim Y-A, Chung HC, Choi HJ, et al. Intermediate dose 5-fluorouracil-induced encephalopathy. *Jpn J Clin Oncol* 2006;36:55–9.
- 46 Acharya G, Cruz Carreras MT, Rice TW. 5-Fu-induced leukoencephalopathy with reversible lesion of splenium of corpus callosum in a patient with colorectal cancer. *BMJ Case Rep* 2017;2017:bcr2017222030.
- 47 Ithoriya H, Yamamoto H, Yamada T, et al. Hyperammonemic encephalopathy in a patient receiving fluorouracil/oxaliplatin chemotherapy. *Clin Case Rep* 2018;6:603–5.
- 48 Cho IJ, Chang HJ, Lee KE, et al. A case of Wernicke's encephalopathy following fluorouracil-based chemotherapy. *J Korean Med Sci* 2009;24:747–50.
- 49 Meyerhardt JA, Mayer RJ. Systemic therapy for colorectal cancer. *N Engl J Med* 2005;352:476–87.
- 50 Alcindor T, Beauger N. Oxaliplatin: a review in the era of molecularly targeted therapy. *Curr Oncol* 2011;18:18–25.
- 51 Gelibter AJ, Caponnetto S, Urbano F, et al. Adjuvant chemotherapy in resected colon cancer: when, how and how long? *Surg Oncol* 2019;30:100–7.
- 52 Leo M, Schmitt L-I, Küsterarent P, et al. Platinum-Based drugs cause mitochondrial dysfunction in cultured dorsal root ganglion neurons. *Int J Mol Sci* 2020;21:8636.
- 53 Wei G, Gu Z, Gu J, et al. Platinum accumulation in oxaliplatin-induced peripheral neuropathy. *J Peripher Nerv Syst* 2021;26:35–42.
- 54 Advani PP, Fakhri MG. 5-Fu-Induced hyperammonemic encephalopathy in a case of metastatic rectal adenocarcinoma successfully rechallenge with the fluoropyrimidine analog, capecitabine. *Anticancer Res* 2011;31:335–8.
- 55 Ogata T, Satake H, Ogata M, et al. Oxaliplatin-Induced hyperammonemic encephalopathy in a patient with metastatic pancreatic cancer: a case report. *Case Rep Oncol* 2017;10:885–9.
- 56 Egler RA, Ahuja SP, Matloub Y. L-Asparaginase in the treatment of patients with acute lymphoblastic leukemia. *J Pharmacol Pharmacother* 2016;7:62–71.
- 57 Müller HJ, Boos J. Use of L-asparaginase in childhood ALL. *Crit Rev Oncol Hematol* 1998;28:97–113.
- 58 Steiner M, Attarbaschi A, Kastner U, et al. Distinct fluctuations of ammonia levels during asparaginase therapy for childhood acute leukemia. *Pediatr Blood Cancer* 2007;49:640–2.
- 59 Heitink-Pollé KJM, Prinsen BHCMT, de Koning TJ, et al. High incidence of symptomatic hyperammonemia in children with acute lymphoblastic leukemia receiving PEGylated asparaginase. *JIMD Rep* 2013;7:103–8.
- 60 Löfberg E, Gutierrez A, Wernerman J, et al. Effects of high doses of glucocorticoids on free amino acids, ribosomes and protein turnover in human muscle. *Eur J Clin Invest* 2002;32:345–53.
- 61 Berry GT, Bridges ND, Nathanson KL, et al. Successful use of alternate waste nitrogen agents and hemodialysis in a patient with hyperammonemic coma after heart-lung transplantation. *Arch Neurol* 1999;56:481–4.
- 62 Imoto K, Tanaka M, Goya T, et al. Corticosteroid suppresses urea-cycle-related gene expressions in ornithine transcarbamylase deficiency. *BMC Gastroenterol* 2022;22:144.
- 63 REYE RD, MORGAN G, BARAL J. Encephalopathy and fatty degeneration of the viscera. A disease entity in childhood. *Lancet* 1963;2:749–52.
- 64 Belay ED, Bresee JS, Holman RC, et al. Reye's syndrome in the United States from 1981 through 1997. *N Engl J Med* 1999;340:1377–82.
- 65 Goetz V, Yang DD, Lacaille F, et al. What are the clues for an inherited metabolic disorder in Reye syndrome? A single centre study of 58 children. *Mol Genet Metab* 2022;135:320–6.
- 66 Häberle J, Burlina A, Chakrapani A, et al. Suggested guidelines for the diagnosis and management of urea cycle disorders: first revision. *J Inherit Metab Dis* 2019;42:1192–230.
- 67 Rüegger CM, Lindner M, Ballhausen D, et al. Cross-Sectional observational study of 208 patients with non-classical urea cycle disorders. *J Inherit Metab Dis* 2014;37:21–30.

- 68 Mitchell S, Ellingson C, Coyne T, *et al.* Genetic variation in the urea cycle: a model resource for investigating key candidate genes for common diseases. *Hum Mutat* 2009;30:56–60.
- 69 Makris G, Lauber M, Rüfenacht V, *et al.* Clinical and structural insights into potential dominant negative triggers of proximal urea cycle disorders. *Biochimie* 2021;183:89–99.
- 70 Conn HO. Studies on the origin and significance of blood ammonia. II. The distribution of ammonia in whole blood, plasma and erythrocytes of man. *Yale J Biol Med* 1966;39:38–53.
- 71 Bingham SA, Williams R, Cole TJ, *et al.* Reference values for analytes of 24-h urine collections known to be complete. *Ann Clin Biochem* 1988;25 (Pt 6):610–9.
- 72 Schrettl V, Felgenhauer N, Rabe C, *et al.* L-arginine in the treatment of valproate overdose-five clinical cases. *Clin Toxicol (Phila)* 2017;55:260–6.
- 73 Ucar F, Erden G, Ozdemir S, *et al.* First data on the biological variation and quality specifications for plasma ammonia concentrations in healthy subjects. *Clin Chem Lab Med* 2016;54:857–63.
- 74 Management of asparaginase therapy. 2021. Available: <https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/haematological/918-management-of-asparaginase-therapy> [Accessed 9 Apr 2023].
- 75 Stock W, Douer D, DeAngelo DJ, *et al.* Prevention and management of asparaginase/pegasparaginase-associated toxicities in adults and older adolescents: recommendations of an expert panel. *Leukemia & Lymphoma* 2011;52:2237–53.
- 76 Tseng Y-L, Huang C-R, Lin C-H, *et al.* Risk factors of hyperammonemia in patients with epilepsy under valproic acid therapy. *Medicine (Baltimore)* 2014;93:e66.
- 77 Vakrinou A, Murphy E, Sisodiya SM, *et al.* Risk factors and outcome of hyperammonaemia in people with epilepsy. *J Neurol* 2022;269:6395–405.
- 78 Frontera JA. Management of hepatic encephalopathy. *Curr Treat Options Neurol* 2014;16:297.
- 79 Stepien KM, Geberhiwot T, Hendriks CJ, *et al.* Challenges in diagnosing and managing adult patients with urea cycle disorders. *J Inher Metab Dis* 2019;42:1136–46.
- 80 Gluud LL, Vilstrup H, Morgan MY. Non-Absorbable disaccharides versus placebo/no intervention and lactulose versus lactitol for the prevention and treatment of hepatic encephalopathy in people with cirrhosis. *Cochrane Database Syst Rev* 2016;2016:CD003044.
- 81 Bélanger-Quintana A, Arrieta Blanco F, Barrio-Carreras D, *et al.* Recommendations for the diagnosis and therapeutic management of hyperammonaemia in paediatric and adult patients. *Nutrients* 2022;14:2755.
- 82 Singh RH. Nutritional management of patients with urea cycle disorders. *J of Inher Metab Dis* 2007;30:880–7.
- 83 Adam S, Almeida MF, Assoun M, *et al.* Dietary management of urea cycle disorders: European practice. *Molecular Genetics and Metabolism* 2013;110:439–45.
- 84 Naorungroj T, Yanase F, Eastwood GM, *et al.* Extracorporeal ammonia clearance for hyperammonemia in critically ill patients: a scoping review. *Blood Purif* 2021;50:453–61.
- 85 Machado MCC, Pinheiro da Silva F. Hyperammonemia due to urea cycle disorders: a potentially fatal condition in the intensive care setting. *J Intensive Care* 2014;2:22.
- 86 Lheureux PE, Penalzoza A, Zahir S, *et al.* Science review: carnitine in the treatment of valproic acid-induced toxicity-what is the evidence. *Crit Care* 2005;9:431.
- 87 Summar ML, Barr F, Dawling S, *et al.* Unmasked adult-onset urea cycle disorders in the critical care setting. *Critical Care Clinics* 2005;21:S1–8.
- 88 Häberle J. Clinical and biochemical aspects of primary and secondary hyperammonemic disorders. *Arch Biochem Biophys* 2013;536:101–8.
- 89 Goldstein BN, Wesler J, Nowacki AS, *et al.* Investigations of blood ammonia analysis: test matrices, storage, and stability. *Clin Biochem* 2017;50:537–9.
- 90 Spacek LA, Strzpek A, Saha S, *et al.* Repeated measures of blood and breath ammonia in response to control, moderate and high protein dose in healthy men. *Sci Rep* 2018;8:2554.
- 91 Allen SI, Conn HO. Observations on the effect of exercise on blood ammonia concentration in man. *Yale J Biol Med* 1960;33:133–44.
- 92 Brusilow SW, Gordes EH. Ammonia secretion in sweat. *Am J Physiol* 1968;214:513–7.
- 93 Gerron GG, Ansley JD, Isaacs JW, *et al.* Technical pitfalls in measurement of venous plasma NH₃ concentration. *Clin Chem* 1976;22:663–6.
- 94 Cowley DM, Nagle BA, Chalmers AH, *et al.* Effects of platelets on collection of specimens for assay of ammonia in plasma. *Clin Chem* 1985;31:332–3.
- 95 da Fonseca-Wollheim F. Preanalytical increase of ammonia in blood specimens from healthy subjects. *Clin Chem* 1990;36:1483–7.
- 96 Wassif WS, Sherman D, Salisbury JR, *et al.* Use of dynamic tests of muscle function and histomorphometry of quadriceps muscle biopsies in the investigation of patients with chronic alcohol misuse and chronic fatigue syndrome. *Ann Clin Biochem* 1994;31 (Pt 5):462–8.
- 97 Herrera DJ, Morris K, Johnston C, *et al.* Automated assay for plasma D-lactate by enzymatic spectrophotometric analysis with sample blank correction. *Ann Clin Biochem* 2008;45:177–83.
- 98 El-Khoury JM, Bunch DR, Wang S. Is the effect of hemolysis on plasma ammonia measurement overrated? *Arch Pathol Lab Med* 2012;136:471–2.
- 99 Nikolac N, Omazic J, Simundic AM. The evidence based practice for optimal sample quality for ammonia measurement. *Clinical Biochemistry* 2014;47:991–5.
- 100 Abulseoud OA, Zuccoli ML, Zhang L, *et al.* The acute effect of cannabis on plasma, liver and brain ammonia dynamics, a translational study. *Eur Neuropsychopharmacol* 2017;27:679–90.