Hyperammonemia in Hepatic Encephalopathy

A.R. Jayakumar^{*,†}, Michael D. Norenberg^{‡,§,¶}

^{*}General Medical Research, Neuropathology Section, R&D Service, Veterans Affairs Medical Center, Miami, FL 33125, United States, [†]South Florida VA Foundation for Research and Education Inc., Veterans Affairs Medical Center, Miami, FL 33125, United States, [‡]Department of Pathology, University of Miami School of Medicine, Miami, FL 33125, United States, [§]Department of Biochemistry & Molecular Biology, University of Miami School of Medicine, Miami, FL 33125, United States and [¶]Department of Neurology and Neurological Surgery, University of Miami School of Medicine, Miami, FL 33125, United States

The precise mechanism underlying the neurotoxicity of Hepatic Encephalopathy (HE) remains unclear. The dominant view has been that gut-derived nitrogenous toxins are not extracted by the diseased liver and thereby enter the brain. Among the various toxins proposed, the case for ammonia is most compelling. Events that lead to increased levels of blood or brain ammonia have been shown to worsen HE, whereas reducing blood ammonia levels alleviates HE. Clinical, pathological, and biochemical changes observed in HE can be reproduced by increasing blood or brain ammonia levels in experimental animals, while exposure of cultured astrocytes to ammonium salts reproduces the morphological and biochemical findings observed in HE. However, factors other than ammonia have recently been proposed to be involved in the development of HE, including cytokines and other blood and brain immune factors. Moreover, recent studies have questioned the critical role of ammonia in the pathogenesis of HE since blood ammonia levels do not always correlate with the level/severity of encephalopathy. This review summarizes the vital role of ammonia in the pathogenesis of HE in humans, as well as in experimental models of acute and chronic liver failure. It further emphasizes recent advances in the molecular mechanisms involved in the progression of neurological complications that occur in acute and chronic liver failure. (J CLIN EXP HEPATOL 2018;8:272–280)

Hepatic Encephalopathy (HE) is a major neurological disorder that is associated with severe liver disease which presents in acute and chronic forms. Acute HE (AHE) occurs following massive liver necrosis due to viral hepatitis, acetaminophen toxicity, or exposure to other hepatotoxins, in particular, alcohol.¹ Acutely, it presents with brain edema, increase in intracranial pressure and brain herniation^{2–4} resulting in a high mortality rate (55–70%).⁵ Chronic HE (CHE) is usually a consequence of cirrhosis of the liver, generally associated with hepatitis C infection and alcoholism, and presents with neuropsychiatric symptoms that profoundly impact on socio-economic aspects of the patient's life. These patients

exhibit a wide-range of neurological symptoms, including mood swings, disturbed sleep/wake cycles, changes in muscle tone, as well as severe cognitive deficits.^{6,7}

Chronic liver disease, generally associated with alcoholism or drug-induced hepatotoxicity (e.g., acetaminophen and other chemical toxins), or due to Viral Hepatitis B and C infection (HBV, HCV), is increasingly recognized as an important cause of morbidity and mortality.^{8–10} Long-term complications of alcoholism, drug-induced hepatotoxicity, or HBV and HCV-infection include cirrhosis of the liver leading to end-stage liver disease. Alcohol-induced cirrhosis of the liver, HBV, HCV and decompensated liver disease, constitute major events leading to the development of CHE.⁸ HE due to a fatty liver (steatosis) is also common, which can additionally accelerate the liver damage caused by hepatitis B or C infection. Autoimmune hepatitis, primary biliary cirrhosis, primary sclerosing cholangitis, hemochromatosis, Wilson's disease, and alpha-1 antitrypsin deficiency are also major causes of liver disease leading to CHE.

While the precise molecular basis for the neurological disorder associated with chronic liver failure remains elusive, the dominant view has been that gut-derived ammonia is not adequately eliminated by the diseased liver. Ammonia then enters the systemic circulation, and ultimately the brain, where it exerts deleterious effects. Blood, Cerebrospinal Fluid (CSF) and brain ammonia levels are elevated in human and experimental HE,^{11–13} which ultimately results in CHE. Currently, there is no candidate other than ammonia that can better explain all of the clinical, pathological and neurochemical features of CHE.

Keywords: ammonia, hepatic encephalopathy, inflammation, extracellular proteins, TDP-43 and tau proteinopathies

Received: 16 April 2018; **Accepted:** 10 June 2018; **Available online:** 20 June 2018

Address for correspondence: A.R. Jayakumar and M.D. Norenberg, General Medical Research, Neuropathology Section, Veterans Affairs Medical Center, 1201 NW 16th St, Res-151, Room 315, Miami, FL, United States. Tel.: +1 305 575 7000X4851/6920.

E-mails: avrj_2000@yahoo.com

Abbreviations: AHE: Acute Hepatic Encephalopathy; ALF: Acute Liver Failure; CHE: Chronic Hepatic Encephalopathy; CNS: Central Nervous System; CSF: Cerebrospinal Fluid; ECs: Endothelial Cells; HE: Hepatic Encephalopathy; IL: Interleukin; LPS: Lipopolysaccharide; MAPKs: Mitogen-Activated Protein Kinases; NCX: Sodium-Calcium Exchanger; NF- κ B: Nuclear Factor-kappaB; NHE: Sodium/Hydrogen Exchanger-1 or SLC9A1 (Solute Carrier Family 9A1); SUR1: The Sulfonylurea Receptor 1; TDP-43: TAR DNA-Binding Protein, 43 kDa; TLR: Toll-like Receptor; TNF- α : Tumor Necrosis Factor-Alpha; TSP-1: Thrombospondin-1

<https://doi.org/10.1016/j.jceh.2018.06.007>

This review summarizes the critical role of ammonia in the pathogenesis of AHE and CHE and the means by which ammonia contributes to the progression of neurological complications that ultimately occur in acute and chronic liver failure.

HYPERAMMONEMIA IN ACUTE AND CHRONIC LIVER FAILURE

While the precise toxin(s) involved in the pathogenesis of HE remains unclear, increased blood and brain ammonia levels have generally been considered to be the crucial factors in the pathogenesis of HE. The involvement of ammonia in HE pathogenesis arises from its coma-engendering effects in dogs with Eck fistulae, and in humans with liver disease.^{14,15} Further, very small alterations in the supply of exogenous ammonia were shown to precipitate episodes of hepatic coma in patients with altered hepatic circulation.¹⁶ These findings were further extended by the intravenous injection of ammonium chloride in various animal models of liver diseases. Additionally, a number of studies dating from the 1950s showed that increased blood ammonia levels were closely associated with the development, as well as the severity of HE.

The most notable findings strongly supporting the critical role of hyperammonemia in the neurological/psychiatric symptoms and electroencephalographic abnormalities following HE were that these findings were improved by lowering blood ammonia levels in these patients,^{17–22} as well as arterial or venous ammonia levels that were found to closely predict mortality rate with 75–80% sensitivity and specificity, and an 80% diagnostic accuracy.^{23–44}

The severity of encephalopathy was also shown to correlate well with blood and brain ammonia levels in animal models of HE.^{45–47} However, some studies showed that the correlation between blood ammonia level and the severity of HE were inconsistent. While the reason for the differences in ammonia level in these studies is unclear, it is likely that the time of ammonia estimates, the patient ammonia metabolism capability, the accuracy of ammonia assays and the analytical methods used, as well as the site of blood sampling varied in these studies. One study further noted differences in the grading and managing of patients with HE, as well as in the measurement of blood ammonia between accurately trained, and untrained practitioners.⁴⁸ Collectively, these findings suggest that a more comprehensive methodology should be considered for patients with HE.

FACTORS OTHER THAN AMMONIA IN THE PATHOGENESIS OF HE

Factors besides ammonia have also been implicated in the pathogenesis of HE, including infections, Central Nervous System (CNS) as well as systemic inflammation,

inflammatory cytokines (49,50 and references therein), increased cerebral blood flow, vasoparalysis and hyperemia, hyperthermia, hyponatremia, substances derived from the necrotic liver, lactic acid, TSPO and neurosteroids, glutamate/glutamine (for review, see 51), and more recently, the accumulation of cholesterol.⁵² Currently, data on these factors are limited and contradictory.

MECHANISMS OF AMMONIA-INDUCED CNS TOXICITY AFTER LIVER FAILURE

The mechanisms by which ammonia ultimately exerts its neurotoxicity still remain poorly defined. Traditional views have included impaired bioenergetics, electrophysiological effects, changes in intracellular pH and calcium, altered neurotransmission and excitotoxicity (53 and references therein). More recently, several additional factors have emerged that also appear to play major roles in the mechanism by which ammonia impacts the CNS in general, and astrocytes in particular. Among these mechanisms, include oxidative/nitrative stress, the mitochondrial permeability transition, Mitogen-Activated Protein Kinases (MAPK), activation of the Nuclear Factor-KappaB (NF-κB) and p53 (53 and references therein), the ion transporters NKCC1, The Sulfonylurea Receptor 1 (SUR1), Sodium/Hydrogen Exchanger-1 or SLC9A1 (Solute Carrier Family 9A1) (NHE1), and Sodium-Calcium Exchanger (NCX) (54,55 and references therein), AQP4,^{56,57} inactivation of the transcription factor STAT3,⁵⁸ activation of endothelial TLR4,^{59–61} matrix metalloproteinase-2 and 9,^{62–64} altered NO/cGMP pathway activity and γ(+)LAT2-mediated exchange of extracellular glutamine for intracellular arginine,^{65–67} increased alpha-1 antichymotrypsin,⁶⁸ multidrug resistance-associated protein 4,⁶⁹ senescence,⁷⁰ downregulation of the gap-junction channel connexin 43 (Cx43), the water channel aquaporin 4 (Aqp4) gene,⁷¹ and the astrocytic inward-rectifying potassium channel (Kir) genes Kir4.1 and Kir5.1,^{71,72} increased translocator protein, the 18 kDa (TSPO)/neurosteroids,^{73,74} increased O-GlcNAcylation,⁷⁵ upregulation of the Ephrin/Ephrin receptor,⁷⁶ cholesterol accumulation,⁵² increased brain levels of TGF-β in chronic liver failure,⁷⁷ decreased TGF-β in ammonia (with 5 mM) treated cultured astrocytes,⁷⁸ increased heat shock protein-25,⁷⁷ among others. Recently, the role of astrocyte-specific growth factor abnormalities, including thrombospondin-1, hevin and glypicans, have been strongly implicated in the pathogenesis of neuronal injury associated with HE (see following section).

A. AHE: ROLE OF AMMONIA

The major neuropathological finding in Acute Liver Failure (ALF) are swollen astrocytes, which largely contribute

to the development of increased intracranial pressure, coma and death. These occur within hours to a few days depending upon the precipitating factors (e.g., viral infection, alcohol consumption or drug overdose). While the precise etiological factor involved in the development of HE in ALF remains unclear, the role of ammonia is most compelling as studies with hyperammonemic primates, dogs, rats, mice,⁵¹ the exposure of organotypic brain slices to ammonia,⁷⁹ as well as in vitro models of hyperammonemia,⁵¹ have all been shown to cause astrocyte swelling.

While the molecular mechanisms involved in the development of astrocyte swelling/brain edema by ammonia are incompletely understood, the involvement of oxidative stress, the mitochondrial permeability transition, energy impairment, MAPK,⁵¹ activation of the NF- κ B and p53 (53 and references therein),⁸⁰ ion transporters, including NKCC1, SUR1, NHE1, NCX (54,55,81 and references therein), as well as the inactivation of the transcription factor STAT3^{53,82} have all been strongly implicated in such astrocytic changes. Collectively, there is compelling evidence for a major role of ammonia in the development of astrocyte swelling/brain edema in AHE.

Brain AQP4 and ammonia toxicity after liver failure

In addition to the above mentioned signaling pathways, increased plasma membrane AQP4 levels were observed after exposure of cultured astrocytes to ammonia or treatment of mice or rats with the liver toxin, thioacetamide (TAA).^{56,57,83} The authors further reported that treatment of mice with TAA that had received whole body AQP4 knock-down, exhibited a reduction in brain edema as compared to the effect of TAA alone.⁵⁷ These findings strongly suggest that increased plasma membrane AQP4 also contributes to the astrocyte swelling/brain edema in AHE. However, in contrast to the findings by Rama Rao et al.,^{56,57,83} Jalan et al., and Butterworth et al. have shown decreased plasma membrane AQP4 post-ALF.^{84,85} Noteworthy, the latter research group also showed increased plasma membrane AQP4 levels in patients who subsequently died with AHE.⁸⁶ The reason for these contradictory findings are unclear.

Inflammation and ammonia toxicity after liver failure

In addition to ammonia, studies have suggested a potential role of inflammation in the pathogenesis of HE.^{49,50,87} Blood levels of Tumor Necrosis Factor-Alpha (TNF- α), Interleukin (IL)-1 β and IL-6 were found to be elevated in patients following AHE (for review see, 49). Additionally, the induction of endotoxemia was shown to exacerbate the brain edema in an experimental model of hyperammonemia (for review see, 49). Further, microglial activation was identified in brains post-AHE,^{88,89} while some

studies have shown no change in microglial activation in AHE, as well as in CHE (for review, see 49). The Butterworth research group also reported that treatment of mice with the TNF- α inhibitor etanercept prevented the brain edema induced by the liver toxin azoxymethane (AOM).⁹⁰ This research group further noted that treatment of IL-1 or TNF- α knock-out (KO) mice with the liver toxin AOM resulted in a reduction in brain water content.⁹¹ Additionally, exposure of cultured astrocytes to ammonia was shown to increase cell swelling, and such swelling was potentiated by co-treatment with cytokines (TNF- α , IL-1 β and IL-6),⁹² suggesting the possibility of an exacerbation of astrocyte swelling/brain edema by cytokines in the presence of sepsis and inflammation.

While microglia are traditionally considered as the major inflammatory cell in the CNS, we and others have shown that exposure of astrocytes to ammonia altered the activities of inflammatory factors, including NADPH oxidase, constitutive nitric oxide synthase, phospholipase A2, cyclooxygenase-2, p53, NF- κ B, TLR-4 and TGF- β .^{49,51,53,60,78,93-95} In aggregate, these findings suggest that inflammation in the CNS following HE is a complex event as cells other than microglia are also involved in the neuroinflammatory response in HE (see below for the role of brain microvessel Endothelial Cells (ECs) and inflammatory response in AHE).

EFFECT OF AMMONIA ON BRAIN MICROVESSEL ECS AND MICROGLIA AND THEIR CONTRIBUTION TO ASTROCYTE SWELLING

ECs

Since systemic inflammation has recently been proposed to be involved in the pathogenesis of HE, and ECs are the first brain cells exposed to blood-borne "noxious agents" (i.e., ammonia, cytokines, immune cells, lipopolysaccharide (LPS), and others), we recently examined whether ammonia, cytokines or LPS have any effect on brain ECs that may also contribute to the astrocyte swelling/brain edema in AHE. We therefore examined the effect of conditioned media (CM) from ammonia, LPS and cytokine-treated cultured ECs on cell swelling in cultured astrocytes.⁹⁶ CM from ECs treated with these agents when added to cultured astrocytes caused significant cell swelling, and such swelling was exacerbated when astrocytes were exposed to CM from ECs treated with a combination of ammonia, LPS and CKs. We found an additive effect when astrocytes were exposed to ammonia along with CM from ammonia-treated ECs,⁹⁶ suggesting that in addition to the direct effect of ammonia on astrocytes, ammonia also impact brain ECs, likely by the activation of the Toll-Like Receptor-4 (TLR)⁶⁰ and the subsequent stimulation of inflammatory factors (e.g., intracellular calcium,

inducible nitric oxide synthase, NADPH oxidase, phospholipase A2, cyclooxygenase-2 and NF- κ B), ultimately resulting in the release of swelling factors (e.g., arachidonic acid, reactive oxygen/nitrogen species, prostaglandins and cytokines) that likely contributed to the astrocyte swelling in ALF.^{93,97} We further identified the release of cytokines when ECs were exposed to ammonia.⁶⁰ Subsequently, a reduction in brain edema was observed when TLR4-KO mice were treated with the liver toxin TAA.⁶⁰ These observations strongly support the findings of the Jalan research group, who showed the prevention of brain edema when TLR4-KO mice were only treated with the liver toxin acetaminophen,⁹⁸ or when the brain edema was inhibited in mice that were pretreated with a novel TLR4 antagonist, STM28 in ALF.⁹⁸ Collectively, these findings suggest the important role of brain ECs-mediated inflammation in the mechanism of brain edema in ALF.

Microglia

Similar to the effect of ammonia on brain ECs, a recent study documented that ammonia also impact microglia resulting in an inflammatory response that contribute to the development of brain edema.⁹⁹ Briefly, CM derived from ammonia-treated cultured microglia when added to cultured astrocytes resulted in significant cell swelling. Such swelling was synergistically increased when astrocytes were additionally treated with ammonia. Such swelling likely occurred through the release of oxy-radicals and nitric oxide into the CM. Further, CM from ammonia-treated microglia containing antioxidants (Tempol or uric acid), when added to astrocytes resulted in a marked reduction in cell swelling, suggesting that, similar to ECs, microglia also contribute to the astrocyte swelling.⁹⁹ These findings collectively suggest that ammonia acts on ECs and microglia resulting in the release of inflammatory agents that ultimately exacerbate the astrocyte swelling in AHE.

B. CHE: ROLE OF AMMONIA

As noted above, the potential role of ammonia in the pathogenesis of CHE is well established and the severity of the encephalopathy in CHE was shown to correlate well with blood and brain ammonia levels in patients with CHE, as well as in various animal models of CHE. The major neuropathological finding in CHE is the presence of Alzheimer type II astrocytosis^{78,100,101} while neuronal death has never been reported in CHE, or in ammonia-treated cultured neurons.^{78,102} Further, adding ammonia directly to neurons for prolonged periods of time (12–96 h) does not alter levels of critical neuronal proteins (e.g., synaptophysin, PSD95, synaptotagmin, NMDA-nr1),⁷⁸ leading to the suggestion that CHE largely represents a primary astroglialopathy.

Astrocytes are well-known to play a critical role in many aspects of CNS physiology, including ammonia metabolism,^{103,104} the uptake of neurotransmitters; free radical scavenging; release of neurotrophic factors (NGF, GDNF, basic FGF, among many others).¹⁰⁵ Astrocytes are also involved in synapse formation during development, as well as in their maintenance in adults.¹⁰⁵ Further, astrocytic dysfunction in CHE has been postulated to also to largely contribute to the neurobehavioral abnormalities associated with CHE.^{106–111}

Among their many biological roles, astrocytes are also important for neuronal development, synaptic transmission, homeostasis, and neuroprotection.¹⁰⁵ While the Alzheimer type II astrocyte is the only histopathological change observed in brains of patients with chronic liver failure and is thought to significantly contribute to the encephalopathy in HE,^{100–104} its precise role remains unclear. Alzheimer type II astrocytosis has been associated with cellular, nuclear and nucleolar enlargement.⁷⁸ When cultured astrocytes are exposed to ammonia they transform into Alzheimer type II astrocytes.^{112,113} While these abnormal astrocytes are a characteristic feature of CHE, their involvement in the pathogenesis of HE still remains to be determined.

A number studies have additionally shown defective synaptic transmission in CHE following their exposure to ammonia.^{114–120} Such synaptic defect has been implicated in the development of neurobehavioral deficits observed in CHE.^{121–124} However, the means by which ammonia contributes to defective neuronal integrity and the subsequent neurobehavioral abnormalities in CHE remains unclear. Since defective synthesis and release of astrocytic factors have been shown to impair synaptic integrity in other neurological conditions,¹²⁵ we recently examined whether astrocytic Matricellular Proteins [(MCPs), including Thrombospondin-1 (TSP-1), glypicans 4 and 6 (Gly-4/6), hevin] play a role in the maintenance of synaptic integrity in CHE. We found decreased levels of TSP-1, Gly-4/6, as well as hevin, when cultured astrocytes were exposed to ammonia 10 days,⁷⁸ indicating that these cells are fundamentally impaired. Further, exposure of cultured neurons to conditioned media from ammonia-treated cultured astrocytes showed a decrease in synaptophysin, PSD95, synaptotagmin as well as GABA-A and NMDA-nr1 receptor levels.⁷⁸ We also found that conditioned media from TSP-1 over-expressing astrocytes that were treated with ammonia, when added to cultured neurons, reversed the decline in synaptic proteins, and that recombinant TSP-1 similarly reversed the decrease in synaptic protein levels. Moreover, metformin, an agent known to increase TSP-1 synthesis in other cell types, reversed the ammonia-induced TSP-1 reduction.⁷⁸ We further identified a significant decline in TSP-1 level in cortical astrocytes, as well as a reduction in synaptophysin content *in vivo* in a rat model of CHE.⁷⁸ In aggregate, these findings strongly suggest

that TSP-1 represents a potential therapeutic agent for patients with CHE.

Recent pathological evidence for TDP-43 and the tau proteinopathies in CHE

Hyper-phosphorylated tau has been strongly implicated in the development of neurobehavioral deficits in many neurological conditions (126,127 and references therein). Recent studies have further emphasized the presence of the Transactivating DNA-Binding Protein, molecular weight 43 kDa (TDP-43) inclusions consisting of hyper-phosphorylated, ubiquitinated and aggregated forms of TDP-43 in neurons (the TDP-43 proteinopathy) found in various neurological conditions, including Alzheimer's disease (AD), Parkinson's disease (PD), frontotemporal lobar degeneration (FTLD), amyotrophic lateral sclerosis (ALS), and chronic traumatic encephalopathy (CTE) (126,127 and references therein). Such inclusions have been strongly implicated in the development of the neurobehavioral defects associated with these conditions (126,127 and references therein).

Since CHE is a complex neuropsychiatric syndrome which resembles many of the events that occur in other neurological conditions, including AD, PD, CTE, FTLD and ALS, among others, we recently examined whether the TDP-43 and tau proteinopathies also occurs in CHE, and whether these proteinopathies contribute to the defective neuronal integrity and neurobehavioral deficits observed in CHE. In recent studies we have found increased levels of phosphorylated TDP-43 and tau in neurons from patients with CHE, as well as in experimental CHE. We also found a reduction in neuronal importin-beta, and an increase in casein kinase and JNK1/2, factors well-known to influence the development of the TDP-43 and tau proteinopathies in other conditions (127 and references therein) in neurons in humans with CHE, in experimental CHE, as well as after the exposure of cultured neurons to conditioned media from ammonia-treated astrocytes (these findings were presented at the 17th ISHEN meeting, India, 2017). These observations strongly suggest that the TDP-43 and tau proteinopathies are also involved in the neurobehavioral defects identified in humans with CHE.

CONCLUSIONS

While the precise etiological factor involved in the encephalopathy associated with acute and chronic liver failure remains unclear, the role of ammonia is most compelling as events that lead to increased levels of blood or brain ammonia have been shown to exacerbate HE, whereas reducing blood ammonia levels ameliorate HE. Further, clinical, pathological, and biochemical changes observed in HE can be reproduced by increasing blood or brain ammonia levels in experimental animals. While factors

other than ammonia have recently been proposed to be involved in the development of HE, including cytokines and other blood and brain immune factors, data on these factors are preliminary. In aggregate, our findings, as well as literature evidence strongly supports the view that ammonia is the primary factor responsible for development of HE, while factors other than ammonia (e.g., cytokines, infection, alcohol or drug overdose) may exacerbate this event in HE.

The mechanisms by which ammonia ultimately exerts its neurotoxicity still remains poorly defined. Impaired bioenergetics, electrophysiological changes, alterations in intracellular pH, glutamatergic/GABAergic abnormalities, involvement of TSPO and neurosteroids, oxidative stress, the mitochondrial permeability transition, mitogen-activated protein kinases, activation of the NF- κ B and p53, inactivation of STAT3, as well as the activation ion transporters and exchangers have all been strongly implicated in the mechanisms of ammonia toxicity in HE. More recently, a loss of astrocytic factors, including matricellular proteins and growth factors have also been strongly associated with the mechanisms of defective neuronal integrity in HE. In particular, decreased synthesis and release of astrocytic thrombospondin-1, hevin and glypicans have been shown to affect neuronal integrity (e.g., the loss of neuronal proteins) in HE. We further observed the TDP-43 and tau proteinopathies in humans and experimental animals with CHE is also a characteristic feature of other neurodegenerative disorders, including AD, PD and ALS.

CONFLICTS OF INTEREST

The authors have none to declare.

ACKNOWLEDGEMENTS

This work was supported by a Merit Review from the US Department of Veterans Affairs (MDN), AASLD/ALF and a Stanley J. Glaser grant (ARJ). The authors thank Alina Fernandez-Revuelta, for the preparation of cell cultures and Dr. Xiaoying Tong and Deepshikha Singh for technical assistance.

REFERENCES

1. Jones EA, Weissenborn K. Neurology and the liver. *J Neurol Neurosurg Psychiatry*. 1997;63:279–293.
2. Blei AT, Larsen FS. Pathophysiology of cerebral edema in fulminant hepatic failure. *J Hepatol*. 1999;31:771–776.
3. Blei AT. Brain edema in acute liver failure. *Crit Care Clin*. 2008;24:99–114.
4. Blei AT. The pathophysiology of brain edema in acute liver failure. *Neurochem Int*. 2005;47:71–77.
5. Capocaccia L, Angelico M. Fulminant hepatic failure. Clinical features, etiology, epidemiology, and current management. *Dig Dis Sci*. 1991;36:775–779.
6. Bajaj JS, Thacker LR, Heuman DM, et al. Cognitive performance as a predictor of hepatic encephalopathy in pretransplant

- patients with cirrhosis receiving psychoactive medications: a prospective study. *Liver Transpl.* 2012;18:1179–1187.
7. Bajaj JS. Management options for minimal hepatic encephalopathy. *Expert Rev Gastroenterol Hepatol.* 2008;2:785–790.
 8. Beste LA, Ioannou GN. Prevalence and treatment of chronic hepatitis C virus infection in the US Department of Veterans Affairs. *Epidemiol Rev.* 2015;37(1):131–143.
 9. El-Serag HB. Epidemiology of viral hepatitis and hepatocellular carcinoma. *Gastroenterology.* 2012;142:1264–1273.e1.
 10. Kramer JR, Davila JA, Miller ED, et al. The validity of viral hepatitis and chronic liver disease diagnoses in Veterans Affairs administrative databases. *Aliment Pharmacol Ther.* 2008;27:274–282.
 11. Butterworth RF, Giguere JF, Michaud J, et al. Ammonia: key factor in the pathogenesis of hepatic encephalopathy. *Neurochem Pathol.* 1987;6:1–12.
 12. Norenberg MD. Astrocytic–ammonia interactions in hepatic encephalopathy. *Semin Liver Dis.* 1996;16:245–253.
 13. Weissenborn K, Ahl B, Fischer-Wasels D, et al. Correlations between magnetic resonance spectroscopy alterations and cerebral ammonia and glucose metabolism in cirrhotic patients with and without hepatic encephalopathy. *Gut.* 2007;56:1736–1742.
 14. Nencki M, Pawlow J, Zaleski J. Ueber den ammoniakgehalt des blutes unter der organe und die harnstoffbildung bei den saugethieren. *Arch Fuer Exp Pathol Pharmacol.* 1896;37:26–51.
 15. Van caulaert C, Deviller C, Halff M. Ueber den ammoniakgehalt des blutes unter der organe und die harnstoffbildung bei den saugethieren. *Arch Fuer Exp Pathol Pharmacol.* 1932;III:735.
 16. Seegmiller JE, Schwartz R, Davidson CS. The plasma ammonia and glutamine content in patients with hepatic coma. *J Clin Invest.* 1954;33:984–988.
 17. Sanjo K, Harihara Y, Kawasaki S, Umekita N, Idezuki Y. Effect of amino acid solutions on the blood ammonia level. *Nihon Geka Gakkai Zasshi.* 1985;86:1223–1226.
 18. Quero JC, Hartmann IJ, Meulstee J, Hop WC, Schalm SW. The diagnosis of subclinical hepatic encephalopathy in patients with cirrhosis using neuropsychological tests and automated electroencephalogram analysis. *Hepatology.* 1996;24:556–560.
 19. Gentile S, Guarino G, Romano M, et al. A randomized controlled trial of acarbose in hepatic encephalopathy. *Clin Gastroenterol Hepatol.* 2005;3:184–191.
 20. Jiang Q, Jiang G, Shi KQ, Cai H, Wang YX, Zheng MH. Oral acetyl-L-carnitine treatment in hepatic encephalopathy: view of evidence-based medicine. *Ann Hepatol.* 2013;12:803–809.
 21. Naderian M, Akbari H, Saeedi M, Sohrabpour AA. Polyethylene glycol and lactulose versus lactulose alone in the treatment of hepatic encephalopathy in patients with cirrhosis: a non-inferiority randomized controlled trial. *Middle East J Dig Dis.* 2017;9:12–19.
 22. Ikeda O, Inoue S, Tamura Y, et al. Shunt-preserving disconnection of the portal to systemic circulation in patients with hepatic encephalopathy. *Acta Radiol.* 2018;59:441–447.
 23. Lockwood AH, Yap EW, Wong WH. Cerebral ammonia metabolism in patients with severe liver disease and minimal hepatic encephalopathy. *J Cereb Blood Flow Metab.* 1991;11:337–341.
 24. Blei AT, Olafsson S, Therrien G, Butterworth RF. Ammonia-induced brain edema and intracranial hypertension in rats after portacaval anastomosis. *Hepatology.* 1994;19:1437–1444.
 25. Clemmesen JO, Larsen FS, Kondrup J, Hansen BA, Ott P. Cerebral herniation in patients with acute liver failure is correlated with arterial ammonia concentration. *Hepatology.* 1999;29:648–653.
 26. Kramer L, Tribl B, Gendo A, et al. Partial pressure of ammonia versus ammonia in hepatic encephalopathy. *Hepatology.* 2000;31:30–34.
 27. Ong JP, Aggarwal A, Krieger D, et al. Correlation between ammonia levels and the severity of hepatic encephalopathy. *Am J Med.* 2003;114:188–193.
 28. Kundra A, Jain A, Banga A, Bajaj G, Kar P. Evaluation of plasma ammonia levels in patients with acute liver failure and chronic liver disease and its correlation with the severity of hepatic encephalopathy and clinical features of raised intracranial tension. *Clin Biochem.* 2005;38:696–699.
 29. Bhatia V, Singh R, Acharya SK. Predictive value of arterial ammonia for complications and outcome in acute liver failure. *Gut.* 2006;55:98–104.
 30. Quero Guillén JC, Herrerías Gutiérrez JM. Diagnostic methods in hepatic encephalopathy. *Clin Chim Acta.* 2006;365:1–8.
 31. Fukuzawa T, Matsutani S, Maruyama H, Akiike T, Saisho H, Hattori T. Magnetic resonance images of the globus pallidus in patients with idiopathic portal hypertension: a quantitative analysis of the relationship between signal intensity and the grade of portosystemic shunt. *J Gastroenterol Hepatol.* 2006;21:902–907.
 32. Bernal W, Hall C, Karvellas CJ, Auzinger G, Sizer E, Wendon J. Arterial ammonia and clinical risk factors for encephalopathy and intracranial hypertension in acute liver failure. *Hepatology.* 2007;46:1844–1852.
 33. Mardini H, Saxby BK, Record CO. Computerized psychometric testing in minimal encephalopathy and modulation by nitrogen challenge and liver transplant. *Gastroenterology.* 2008;135:1582–1590.
 34. Tarantino G, Citro V, Esposito P, et al. *BMC Gastroenterol.* 2009;17(9):21.
 35. Kitzberger R, Funk GC, Holzinger U, et al. Severity of organ failure is an independent predictor of intracranial hypertension in acute liver failure. *Clin Gastroenterol Hepatol.* 2009;7:1000–1006.
 36. McKinney AM, Lohman BD, Sarikaya B, et al. Acute hepatic encephalopathy: diffusion-weighted and fluid-attenuated inversion recovery findings, and correlation with plasma ammonia level and clinical outcome. *AJNR Am J Neuroradiol.* 2010;31:1471–1479.
 37. Chojnacki C, Romanowski M, Winczyk K, Błasiak J, Chojnacki J. Melatonin levels in serum and ascitic fluid of patients with hepatic encephalopathy. *Gastroenterol Res Pract.* 2012;2012:510764.
 38. Chikamori F, Okamoto H, Kuniyoshi N. Relationships between splenorenal shunt/portal vein diameter ratio and systemic hemodynamics in patients with liver cirrhosis. *Digestion.* 2014;89:133–138.
 39. Qureshi MO, Khokhar N, Shafqat F. Ammonia levels and the severity of hepatic encephalopathy. *J Coll Physicians Surg Pak.* 2014;24:160–163.
 40. Jao T, Schröter M, Chen CL, et al. Functional brain network changes associated with clinical and biochemical measures of the severity of hepatic encephalopathy. *Neuroimage.* 2015;122:332–344.
 41. Malaguarnera G, Vacante M, Drago F, et al. Endozepine-4 levels are increased in hepatic coma. *World J Gastroenterol.* 2015;21:9103–9110.
 42. Zhang G, Cheng Y, Liu B. Abnormalities of voxel-based whole-brain functional connectivity patterns predict the progression of hepatic encephalopathy. *Brain Imaging Behav.* 2017;11:784–796.
 43. Chang CC, Lee WS, Chuang CL, et al. Effects of raloxifene on portal hypertension and hepatic encephalopathy in cirrhotic rats. *Eur J Pharmacol.* 2017;802:36–43.
 44. Zheng G, Lu H, Yu W, et al. Severity-specific alterations in CBF, OEF and CMRO2 in cirrhotic patients with hepatic encephalopathy. *Eur Radiol.* 2017;27:4699–4709.

45. Kanamori K, Ross BD, Chung JC, Kuo EL. Severity of hyperammonemic encephalopathy correlates with brain ammonia level and saturation of glutamine synthetase in vivo. *J Neurochem*. 1996;67:1584–1594.
46. Zwiener K, Thiel C, Thiel K, Morgalla MH, Königsrainer A, Schenk M. Extracellular brain ammonia levels in association with arterial ammonia, intracranial pressure and the use of albumin dialysis devices in pigs with acute liver failure. *Metab Brain Dis*. 2010;25:407–412.
47. Jayakumar AR, Valdes V, Norenberg MD. The Na-K-Cl cotransporter in the brain edema of acute liver failure. *J Hepatol*. 2011;54:272–278.
48. Reuter B, Walter K, Bissonnette J, et al. Assessment of the spectrum of hepatic encephalopathy: a multi-center study. *Liver Transpl*. 2018;19:.
49. Jayakumar AR, Rama Rao KV, Norenberg MD. Neuroinflammation in hepatic encephalopathy: mechanistic aspects. *J Clin Exp Hepatol*. 2015;5(suppl 1):S21–S28.
50. Butterworth RF. The concept of "the inflamed brain" in acute liver failure: mechanisms and new therapeutic opportunities. *Metab Brain Dis*. 2016;31:1283–1287.
51. Norenberg MD, Jayakumar AR, Rama Rao KV, Panickar KS. New concepts in the mechanism of ammonia-induced astrocyte swelling. *Metab Brain Dis*. 2007;22:219–234.
52. McMillin M, Grant S, Frampton G, et al. FXR-mediated cortical cholesterol accumulation contributes to the pathogenesis of type A hepatic encephalopathy. *Cell Mol Gastroenterol Hepatol*. 2018;6(1):47–63.
53. Norenberg MD, Rama Rao KV, Jayakumar AR. Signaling factors in the mechanism of ammonia neurotoxicity. *Metab Brain Dis*. 2009;24:103–117.
54. Jayakumar AR, Norenberg MD. The Na-K-Cl co-transporter in astrocyte swelling. *Metab Brain Dis*. 2010;25:31–38.
55. Jayakumar AR, Valdes V, Tong XY, Shamaladevi N, Gonzalez W, Norenberg MD. Sulfonylurea receptor 1 contributes to the astrocyte swelling and brain edema in acute liver failure. *Transl Stroke Res*. 2014;5:28–33.
56. Rama Rao KV, Jayakumar AR, Tong X, Curtis KM, Norenberg MD. Brain aquaporin-4 in experimental acute liver failure. *J Neuropathol Exp Neurol*. 2010;69:869–879.
57. Rama Rao KV, Verkman AS, Curtis KM, Norenberg MD. Aquaporin-4 deletion in mice reduces encephalopathy and brain edema in experimental acute liver failure. *Neurobiol Dis*. 2014;63:222–228.
58. Jayakumar AR, Curtis KM, Panickar KS, Shamaladevi N, Norenberg MD. Decreased STAT3 phosphorylation mediates cell swelling in ammonia-treated astrocyte cultures. *Biology (Basel)*. 2016;5: pii: E48.
59. Jayakumar AR, Norenberg MD. Endothelial–astrocytic interactions in acute liver failure. *Metab Brain Dis*. 2013;28:183–186.
60. Jayakumar AR, Tong XY, Curtis KM, Ruiz-Cordero R, Abreu MT, Norenberg MD. Increased toll-like receptor 4 in cerebral endothelial cells contributes to the astrocyte swelling and brain edema in acute hepatic encephalopathy. *J Neurochem*. 2014;128:890–903.
61. Shah N, Montes de Oca M, Jover-Cobos M, et al. Role of toll-like receptor 4 in mediating multiorgan dysfunction in mice with acetaminophen induced acute liver failure. *Liver Transpl*. 2013;19:751–761.
62. Skowrońska M, Zielińska M, Wójcik-Stanaszek L, et al. Ammonia increases paracellular permeability of rat brain endothelial cells by a mechanism encompassing oxidative/nitrosative stress and activation of matrix metalloproteinases. *J Neurochem*. 2012;121:125–134.
63. Chastre A, Bélanger M, Nguyen BN, Butterworth RF. Lipopolysaccharide precipitates hepatic encephalopathy and increases blood–brain barrier permeability in mice with acute liver failure. *Liver Int*. 2014;34:353–361.
64. McMillin MA, Frampton GA, Seiwel AP, Patel NS, Jacobs AN, DeMorrow S. TGFβ1 exacerbates blood–brain barrier permeability in a mouse model of hepatic encephalopathy via upregulation of MMP9 and downregulation of claudin-5. *Lab Invest*. 2015;95:903–913.
65. Hilgier W, Oja SS, Saransaari P, Albrecht J. A novel glycine site-specific N-methyl-D-aspartate receptor antagonist prevents activation of the NMDA/NO/cGMP pathway by ammonia. *Brain Res*. 2004;1015:186–188.
66. Zielińska M, Ruszkiewicz J, Hilgier W, Fręsko I, Albrecht J. Hyperammonemia increases the expression and activity of the glutamine/arginine transporter y+ LAT2 in rat cerebral cortex: implications for the nitric oxide/cGMP pathway. *Neurochem Int*. 2011;58:190–195.
67. Cabrera-Pastor A, Malaguarnera M, Taoro-Gonzalez L, Llansola M, Felipe V. Extracellular cGMP modulates learning biphasically by modulating glycine receptors, CaMKII and glutamate-nitric oxide-cGMP pathway. *Sci Rep*. 2016;6:33124.
68. Park J, Masaki T, Mezaki Y, et al. Alpha-1 antitrypsin is involved in astrocyte injury in concert with arginine-vasopressin during the development of acute hepatic encephalopathy. *PLoS ONE*. 2017;7(12):e0189346.
69. Jördens MS, Keitel V, Karababa A, et al. Multidrug resistance-associated protein 4 expression in ammonia-treated cultured rat astrocytes and cerebral cortex of cirrhotic patients with hepatic encephalopathy. *Glia*. 2015;23:.
70. Görg B, Karababa A, Shafiqullina A, Bidmon HJ, Häussinger D. Ammonia-induced senescence in cultured rat astrocytes and in human cerebral cortex in hepatic encephalopathy. *Glia*. 2015;63:37–50.
71. Lichter-Konecki U, Mangin JM, Gordish-Dressman H, Hoffman EP, Gallo V. Gene expression profiling of astrocytes from hyperammonemic mice reveals altered pathways for water and potassium homeostasis in vivo. *Glia*. 2008;56(4):365–367.
72. Obara-Michlewska M, Pannicke T, Karl A, et al. Down-regulation of Kir4.1 in the cerebral cortex of rats with liver failure and in cultured astrocytes treated with glutamine: implications for astrocytic dysfunction in hepatic encephalopathy. *J Neurosci Res*. 2011;89:2018–2027.
73. Ahboucha S, Butterworth RF. The neurosteroid system: an emerging therapeutic target for hepatic encephalopathy. *Metab Brain Dis*. 2007;22:291–308.
74. Panickar KS, Jayakumar AR, Rama Rao KV, Norenberg MD. Downregulation of the 18-kDa translocator protein: effects on the ammonia-induced mitochondrial permeability transition and cell swelling in cultured astrocytes. *Glia*. 2007;55:1720–1727.
75. Karababa A, Görg B, Schliess F, Häussinger D. O-GlcNAcylation as a novel ammonia-induced posttranslational protein modification in cultured rat astrocytes. *Metab Brain Dis*. 2014;29:975–982.
76. Sobczyk K, Jördens MS, Karababa A, Görg B, Häussinger D. Ephrin/Ephrin receptor expression in ammonia-treated rat astrocytes and in human cerebral cortex in hepatic encephalopathy. *Neurochem Res*. 2015;40:274–283.
77. Wright GA, Sharifi Y, Newman TA, et al. Characterisation of temporal microglia and astrocyte immune responses in bile duct-ligated rat models of cirrhosis. *Liver Int*. 2014;34:1184–1191.
78. Jayakumar AR, Tong XY, Curtis KM, et al. Decreased astrocytic thrombospondin-1 secretion after chronic ammonia treatment reduces the level of synaptic proteins: in vitro and in vivo studies. *J Neurochem*. 2014;131:333–347.
79. Back A, Tupper KY, Bai T, et al. Ammonia-induced brain swelling and neurotoxicity in an organotypic slice model. *Neurol Res*. 2011;33:1100–1108.

80. Dai H, Jia G, Wang W, et al. Genistein inhibited ammonia induced astrocyte swelling by inhibiting NF- κ B activation-mediated nitric oxide formation. *Metab Brain Dis.* 2017;32:841–848.
81. Hertz L, Peng L, Song D. Ammonia, like K(+), stimulates the Na(+), K(+), 2 Cl(–) cotransporter NKCC1 and the Na(+), K(+)ATPase and interacts with endogenous ouabain in astrocytes. *Neurochem Res.* 2015;40:241–257.
82. Hertz L, Song D, Peng L, Chen Y. Multifactorial effects on different types of brain cells contribute to ammonia toxicity. *Neurochem Res.* 2017;42:721–736.
83. Rama Rao KV, Chen M, Simard JM, Norenberg MD. Increased aquaporin-4 expression in ammonia-treated cultured astrocytes. *Neuroreport.* 2003;14:2379–2382.
84. Wright G, Soper R, Brooks HF, et al. Role of aquaporin-4 in the development of brain oedema in liver failure. *J Hepatol.* 2010;53:91–97.
85. Chastre A, Jiang W, Desjardins P, Butterworth RF. Ammonia and proinflammatory cytokines modify expression of genes coding for astrocytic proteins implicated in brain edema in acute liver failure. *Metab Brain Dis.* 2010;25:17–21.
86. Thumburu KK, Dhiman RK, Vasishta RK, et al. Expression of astrocytic genes coding for proteins implicated in neural excitation and brain edema is altered after acute liver failure. *J Neurochem.* 2014;617–627.
87. Wilkinson SP, Arroyo V, Gazzard BG, Moodie H, Williams R. Relation of renal impairment and haemorrhagic diathesis to endotoxaemia in fulminant hepatic failure. *Lancet.* 1974;1:521–524.
88. Jiang W, Desjardins P, Butterworth RF. Direct evidence for central proinflammatory mechanisms in rats with experimental acute liver failure: protective effect of hypothermia. *J Cereb Blood Flow Metab.* 2009;29:944–952.
89. Dennis CV, Sheahan PJ, Graeber MB, Sheedy DL, Kril JJ, Sutherland GT. Microglial proliferation in the brain of chronic alcoholics with hepatic encephalopathy. *Metab Brain Dis.* 2014;29:1027–1039.
90. Butterworth RF. Neuroinflammation in acute liver failure: mechanisms and novel therapeutic targets. *Neurochem Int.* 2011;59:830–836.
91. Bémeur C, Qu H, Desjardins P, Butterworth RF. IL-1 or TNF receptor gene deletion delays onset of encephalopathy and attenuates brain edema in experimental acute liver failure. *Neurochem Int.* 2010;56:213–215.
92. Rama Rao KV, Jayakumar AR, Tong X, Alvarez VM, Norenberg MD. Marked potentiation of cell swelling by cytokines in ammonia-sensitized cultured astrocytes. *J Neuroinflammation.* 2010;13(7):66.
93. Jayakumar AR, Rama Rao KV, Tong XY, Norenberg MD. Calcium in the mechanism of ammonia-induced astrocyte swelling. *J Neurochem.* 2009;109(suppl. 1):252–257.
94. Häussinger D, Görg B. Interaction of oxidative stress, astrocyte swelling and cerebral ammonia toxicity. *Curr Opin Clin Nutr Metab Care.* 2010;13:87–92.
95. Panickar KS, Jayakumar AR, Rao KV, Norenberg MD. Ammonia-induced activation of p53 in cultured astrocytes: role in cell swelling and glutamate uptake. *Neurochem Int.* 2009;55:98–105.
96. Jayakumar AR, Tong XY, Ospel J, Norenberg MD. Role of cerebral endothelial cells in the astrocyte swelling and brain edema associated with acute hepatic encephalopathy. *Neuroscience.* 2012;30(218):305–316.
97. Jayakumar AR, Norenberg MD. Endothelial–astrocytic interactions in acute liver failure. *Metab Brain Dis.* 2013;28:183–186.
98. Shah N, Montes de Oca M, Jover-Cobos M, et al. Role of toll-like receptor 4 in mediating multiorgan dysfunction in mice with acetaminophen induced acute liver failure. *Liver Transpl.* 2013;19:751–761.
99. Rao KV, Brahmabhatt M, Norenberg MD. Microglia contribute to ammonia-induced astrocyte swelling in culture. *Metab Brain Dis.* 2013;28:139–143.
100. Norenberg MD. Astrocytic–ammonia interactions in hepatic encephalopathy. *Semin Liver Dis.* 1996;16:245–253.
101. Norenberg MD. Astroglial dysfunction in hepatic encephalopathy. *Metab Brain Dis.* 1998;13:319–335.
102. Norenberg MD. The astrocyte in liver disease. *Adv Cell Neurobiol.* 1981;2:303–352.
103. Norenberg MD. A light and electron microscopic study of experimental portal-systemic (ammonia) encephalopathy. Progression and reversal of the disorder. *Lab Invest.* 1977;36:618–627.
104. Martinez-Hernandez A, Bell KP, Norenberg MD. Glutamine synthetase: glial localization in brain. *Science.* 1977;195:1356–1358.
105. Wang DD, Bordey A. The astrocyte odyssey. *Prog Neurobiol.* 2008;86:342–367.
106. Dezone RS, Stipursky J, Araujo AP, et al. Thyroid hormone treated astrocytes induce maturation of cerebral cortical neurons through modulation of proteoglycan levels. *Front Cell Neurosci.* 2013;7:125.
107. Schwartz JP, Nishiyama N. Neurotrophic factor gene expression in astrocytes during development and following injury. *Brain Res Bull.* 1994;35:403–407.
108. Vernadakis A. Glia-neuron intercommunications and synaptic plasticity. *Prog Neurobiol.* 1996;49:185214.
109. Slezak M, Priege FW, Soltys Z. Synaptic plasticity, astrocytes and morphological homeostasis. *J Physiol Paris.* 2006;99:84–91.
110. Ullian EM, Christopherson KS, Barres BA. Role for glia in synaptogenesis. *Glia.* 2004;47:209–216.
111. Norenberg MD, Neary JT, Bender AS, Dombro RS. Hepatic encephalopathy: a disorder in glial-neuronal communication. *Prog Brain Res.* 1992;94:261–269.
112. Gregorios JB, Mozes LW, Norenberg MD. Morphologic effects of ammonia on primary astrocyte cultures. II. Electron microscopic studies. *J Neuropathol Exp Neurol.* 1985;44:404–414.
113. Gregorios JB, Mozes LW, Norenberg LO, Norenberg MD. Morphologic effects of ammonia on primary astrocyte cultures. I. Light microscopic studies. *J Neuropathol Exp Neurol.* 1985;44:397–403.
114. Ott P, Vilstrup H. Cerebral effects of ammonia in liver disease: current hypotheses. *Metab Brain Dis.* 2014;29:901–911.
115. Monfort P, Muñoz MD, Felipe V. Molecular mechanisms of the alterations in NMDA receptor-dependent long-term potentiation in hyperammonemia. *Metab Brain Dis.* 2005;20:265–274.
116. Albrecht J, Zielińska M. The role of inhibitory amino acidergic neurotransmission in hepatic encephalopathy: a critical overview. *Metab Brain Dis.* 2002;17:283–294.
117. Basile AS, Jones EA. Ammonia and GABA-ergic neurotransmission: interrelated factors in the pathogenesis of hepatic encephalopathy. *Hepatology.* 1997;25:1303–1305.
118. Szerb JC, Butterworth RF. Effect of ammonium ions on synaptic transmission in the mammalian central nervous system. *Prog Neurobiol.* 1992;39:135–153.
119. Raabe W. Synaptic transmission in ammonia intoxication. *Neurochem Pathol.* 1987;6:145–166.
120. Cauli O, Rodrigo R, Llansola M, et al. Glutamatergic and gabaergic neurotransmission and neuronal circuits in hepatic encephalopathy. *Metab Brain Dis.* 2009;24:69–80.
121. Cauli O, Mili N, Llansola M, Felipe V. Motor activity is modulated via different neuronal circuits in rats with chronic liver failure than in normal rats. *Eur J Neurosci.* 2007;25:2112–2122.

122. Méndez M, Méndez-López M, López L, Aller MA, Arias J, Arias JL. Working memory impairment and reduced hippocampal and prefrontal cortex c-Fos expression in a rat model of cirrhosis. *Physiol Behav.* 2008;20(95):302–307.
123. Solinas A, Piras MR, Deplano A. Cognitive dysfunction and hepatitis C virus infection. *World J Hepatol.* 2015;8(7):922–925.
124. Dhanda S, Sandhir R. Role of dopaminergic and serotonergic neurotransmitters in behavioral alterations observed in rodent model of hepatic encephalopathy. *Behav Brain Res.* 2015;286:222–235.
125. Kucukdereli H, Allen NJ, Lee AT, et al. Control of excitatory CNS synaptogenesis by astrocyte-secreted proteins Hevin and SPARC. *Proc Natl Acad Sci U S A.* 2011;9(108(32)):E440–E449.
126. Iqbal K, Liu F, Gong C-X. Tau and neurodegenerative disease: the story so far. *Nat Rev Neurol.* 2016;12:15–27.
127. Jayakumar AR, Tong XY, Shamaladevi N, et al. Defective synthesis and release of astrocytic thrombospondin-1 mediates the neuronal TDP-43 proteinopathy resulting in defects in neuronal integrity associated with chronic traumatic encephalopathy: in vitro studies. *J Neurochem.* 2017;140:531–535.