RELATION OF SERUM AMMONIA LEVEL WITH PORTO-SYSTEMIC ENCEPHALOPATHY AND ITS SHORT TERM PROGNOSTIC ROLE IN PATIENTS WITH LIVER CIRRHOSIS

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ABSTRACT

Background: Porto-systemic encephalopathy (PSE) and hepatic encephalopathy (HE) are the terms used synonymously to describe a central nervous system disturbance associated with acute or chronic failure of liver. Although PSE is supposed to arise due to hepatic failure to clear the toxic products from blood yet it has not been clearly established that which of those toxic products should be labeled as culprits in the etiology of hepatic or portosystemic encephalopathy.

Objectives: To study the relation of serum ammonia (NH_3) levels with portosystemic encephalopathy and its severity in patients with hepatic failure.

To assess the short term prognostic role of serum ammonia in hepatic encephalopathy.

Methodology: Total 100 patients having hepatic encephalopathy were selected for the study. All patients were evaluated by detailed history and examination for signs of chronic liver disease, grades of HE and serum ammonia levels along with other baseline investigations.

Results: This study included 70% male and 30% female patients with mean age of 50.89±12.81 years. Ninety seven percent of patients were positive for anti HCV while 3% for anti HCV and HbsAg both. Serial estimations of serum ammonia and evaluation of clinical grades of HE demonstrated a progressive decline in serum ammonia levels paralleled by improvement in grades of encephalopathy on days 1, 3, 5, 7 and 9.

Conclusion: Decrease in serum ammonia level was associated with decrease in grades of hepatic encephalopathy. This study showed a positive correlation between serum NH_3 levels and grades of hepatic encephalopathy. The study also revealed the short term prognostic role of NH_3 levels in patients with HE.

Key words: Serum ammonia (NH_3) level, hepatic encephalopahty, porto-systemic encephalopathy, liver cirrhosis.

INTRODUCTION

Porto-systemic encephalopathy or Hepatic encephalopathy (HE) is amongst one of the most debilitating complications of acute or chronic hepatic failure[1]. Hepatic encephalopathy can be defined as a disturbance in central nervous system function due to acute or chronic hepatic failure.

Hepatic encephalopathy is a cluster of neuropsychiatric manifestations which are present both in acute and chronic hepatic failure and are reversible by taking appropriate measures[2]. In HE, there is progressive decline in the level of consciousness i.e. from lethargy to somonolence to stupor and finally coma and hampering of intellectual capability i.e. reasoning, orientation and neurologic deficits like Asterixis or flapping tremors[3,4]. Hepatic encephalopathy must be differentiated from the other causes which lead to liver damage and the manifestation of similar symptoms. These other causes include brain and liver damage caused by alcohol and copper (Wilson's disease), neurologic deficits caused by bilirubin deposition in brain (Kernicterus), cerebral hemorrhage caused by disorders of coagulation due to liver damage and other abnormalities that are not associated with liver failure[5].

The pathogenesis of HE is quite complex and still it is not completely understood[6]. However it is clear that there is shunting of blood from portal to systemic circulation and it has got a crucial role in the etiopathogenesis of HE[5]. The toxins which are thought to be involved in the etiology of hepatic encephalopathy actually do so as a result of portosystemic shunting of blood and include; ammonia, aromatic amino acids, manganese, benzodiazepine like substances i.e. GABA, mercaptans and pro-inflammatory cytokines leading to changes in intestinal microbiota[1,7-10]. So lot of toxins have been nominated that can contribute to hepatic encephalopathy yet it is widely accepted that NH_3 plays a pivotal role in the pathogenesis of this disorder[4,11,12]. It is an established fact that all these substances are produced normally

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in the body and most of them are cleared by the liver on regular basis, so liver damage in a way which leads to shunting of blood from liver to systemic circulation facilitates these toxins and especially NH_3 to cross blood brain barrier leading to brain edema and HE.

Ammonia is generated by different tissues, the principal tissues being liver and intestine while the others include muscle and kidney. Intestinal ammonia is derived firstly by the action of the enzyme urease produced by colonic bacteria which degrades urea into ammonia and secondly by deamidation of glutamine (amino acid) in the small intestine to gluatamate and ammonia by the help of enzyme glutaminase[2]. Liver causes production of urea in an attempt to metabolize ammonia. Liver causes production of urea in an attempt to metabolize ammonia. Liver is the principal site of removal of excess ammonia from the body and it does so by converting it into urea in periportal hepatocytes and glutamine in centrovenular hepatocytes[5], the urea is then excreted by kidneys. When there is acute or chronic hepatic failure, there is portosytemic shunting as well as reduced detoxification of ammonia by liver so reduced clearance of ammonia leads to hyperammonemia. Hyperammonemia exerts injurious effects on brain by causing brain edema[5]. Under physiological conditions astrocytes of the brain also carry on the function of detoxification of ammonia. Astrocytes form one third portion of the cerebral cortex. Astrocytes perform detoxification of ammonia by causing amidation of glutamate(excitatory neurotransmitter) and ammonia to glutamine[5].

In case of liver failure the additional detoxification function is then performed by astrocytes, as a result they become swollen, leading to cerebral edema and altered mental function[3,13]. Recent studies have put light on the precise mechanism behind astrocyte swelling and dysfunction. In fact increased ammonia levels lead to oxidative stress (nitrosative stress) on astrocytes by causing production of reactive nitrogen species like nitric oxide ($NO^{\cdot}$) and others which eventually leads to astrocyte dysfunction, edema and HE[6]. Increased neuronal nitric oxide synthase (nNOS) activity was found in the brains of rats in which portacaval anastomosis was done.[14] Along with increased activity of nNOS there was increased production of nitric oxide (NO) as well in brains of portacaval shunted rats when they were given an infusion of NH_3[15]. It means that in portacaval shunting there is increased production of NO in brain (astrocytes) due to hepatic bypassing of the blood. Nitric oxide then being a free radical causes swelling and astrocyte dysfunction secondary to increased NH_3 levels.

A lot of research work points towards the fact that NH_3 plays a pivotal role in the pathogenesis of portosystemic encephalopathy[4,11,16,17]. The role of NH_3 as a crucial factor in pathogenesis of HE has been proposed on the basis of a rise in NH_3 levels of patients having cirrhosis and HE, increase in blood-brain barrier permeability to NH_3 in HE; and elevations of NH_3 levels associated with astrocyte dysfunction[12,16-19].

Measuring venous ammonia levels is helpful in the evaluation of liver disease as a cause of disturbance in consciousness when one is suspecting other causes for such disturbance[20]. Venous sampling is enough and appropriate for measurement of ammonia and there seems no extra advantage of measuring partial pressure of NH[17]. Adult NH_3 levels in plasma are 19-60 μg/dL or 15-35 μmol/L[21].

The present study was conducted in order to assess an association between hepatic encephalopahty and serum ammonia levels and it was intended that this would provide guidelines for improved prevention and management of hepatic encephalopathy.

METHODOLOGY

Sampling was carried out at Gastroenterology department of Shaikh Zayed Hospital, Lahore. It was a descriptive, cross sectional study. Sampling was done on Non-probability convenient sampling basis. Duration of study was from 12th October 2017 to 11th April 2018. A total of hundred patients with hepatic encephalopathy who sought admission in Gastroenterology ward, of Shaikh Zayed Hospital through OPD or emergency department and fulfilled the inclusion criteria, were selected for the study.

Adult patients with chronic liver disease, irrespective of gender, presenting with hepatic encephalopathy were included in the study.

All cases of chronic obstructive pulmonary disease, uremia, cardiovascular accident, fulminant hepatic failure, meningitis, hypoglycemia, encephalitis, Wernicke's encephalopathy, psychiatric disturbances, sedative intake and acute respiratory distress syndrome were excluded from the study.

All patients underwent a detailed history and examination for signs of chronic liver disease and grades of encephalopathy. Grading of encephalopathy was done according to West Haven grading.

Serum ammonia of all the patients was measured along with other baseline investigations through standard laboratory techniques. Blood samples were collected in appropriate vacutainers and were processed in pathology laboratory of the Shaikh Zayed Hospital. Venous blood was tested for serum ammonia, full blood count, liver function tests and renal function tests at day 1, 3 5, 7 and 9. The kit used for the estimation of Serum ammonia was Amon flex reagent cartridge Cat No. DF19. All data was collected on the pre-designed proforma.

SPSS 23 was used to derive different statistical parameters. The study variables were age, gender, Hemoglobin (Hb), Total Bilirubin, Alanine transaminase

(ALT), Aspartate transaminase (AST), Alkaline phosphatase (ALP), Serum creatinine, Blood urea nitrogen (BUN), Grades of hepatic encephalopahty and erum ammonia levels. One way anova test was applied to evaluate changes in serum ammonia levels on different days. Chi Square test was applied for association with decrease in serum ammonia levels and grades of HE. A p value of ≤0.05 at 95 % confidence interval was considered as significant.

RESULTS

Mean age of the patients was 50.89±12.81 years. Minimum age range was 21-30 years while maximum range was 71-80 years. Maximum numbers of patients were in age range of 51-60 years, (41%).

In present study 70 (70%) patients were males and 30 (30%) patients were females.

Mean Hemoglobin levels of the subjects (11.07 ± 2.46 mg/dl) were low, while Bilirubin levels were high (6.17 ± 7.47 mg/dl) but not high enough. Similarly mean ALT (120.41 ± 242.07 U/L) and AST (151.51 ± 205.53 U/L) levels were raised indicating liver injury along with raised levels of ALP (162.95 ± 115.87 U/L) indicating slight degree of obstruction consequent upon cirrhosis. The Creatinine levels were within normal range, while BUN levels were slightly raised but insignificant, indicating normal renal functions.

In this study Hepatitis C was the commonest cause of liver cirrhosis and consequent hepatic encephalopathy, while Hepatitis B alone was not found to cause the cirrhosis and its complications.

Higher ammonia levels were associated with higher grades of HE. Ammonia levels of 40.59±12.49 did not show any association with hepatic encephalopathy, ammonia levels of 67.87±29.20 μmol/L were associated with Grade I and Grade II encephalopathy while ammonia levels of 123.44±52.27 μmol/L were associated with Grade III HE and levels of 249.65±92.57 μmol/L were related to Grade IV encephalopathy.

Day wise correlation of mean serum NH_3 levels and hepatic encephalopathy indicated that higher levels of serum NH_3 were associated with higher grades of hepatic encephalopathy and the grades of hepatic encephalopathy kept on decreasing with decreasing levels of serum ammonia with the passage of time due to compensation. Mean ammonia levels at day 1 and 3 were associated with grade IV HE, while levels on day 5 were associated with grade III encephalopathy. Serum ammonia levels at day 7 corresponded to grade II and I encephalopathy while levels at day 9 were not associated with any grade of encephalopathy.

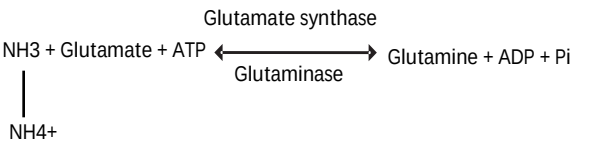

Figure 1: Conversion of NH3 and glutamate to glutamine and vice versa

Table 1: West Haven grading of Hepatic Encephalopathy(1)

Hepatic encephalopathy	Consciousness	Intellectual function	Personality	Behavior
Grade 0	No detectable changes	No detectable changes	No detectable changes	None
Grade I (mild)	Sleep disturbance, trivial lack of awareness	Shortened attention span, mildly impaired computations	Euphoria, depression, irritability	Muscular incoordination, impaired handwriting, Asterixis may be present
Grade II (moderate)	Lethargy, mild disorientation to time	Amnesia of recent events, grossly impaired computations	Overt change in personality, inappropriate behavior	Slurred speech, Asterixis, hypoactive reflexes, ataxia
Grade III (severe)	Somnolence, confusion, semistupor	Inability to compute, disorientation to place	Paranoia, bizarre behavior	Hyperactive reflexes, Nystagmus, (+) Babinski's sign, Clonus, rigidity
Grade IV (coma)	Stupor	None	None	Dilated pupils, Opisthotonus, coma, lack of verbal, eye & oral response

Table 2: Distribution of patients according to gender

Gender	Number	Percentage
Male	70	70 %
Female	30	30 %
Total	100	100 %

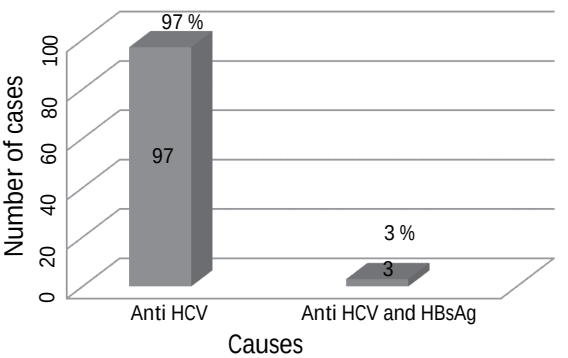

Figure 2: Distribution of patients according to causes of cirrhosis and HE

Table 3: Laboratory data of different parameters

Investigations	Mean ± SD
Hemoglobin (mg/dl)	11.07 ± 2.46
Total Bilirubin (mg/dl)	6.17 ± 7.47
ALT (U/L)	120.41 ± 242.07
AST (U/L)	151.51 ± 205.53
ALP (U/L)	162.95 ± 115.87
Serum Creatinine (mg/dl)	1.75 ± 1.22
BUN (mg/dl)	33.98 ± 25.41

Table 4: Correlation of mean serum NH3 levels and hepatic encephalopathy grades

HE grades	Serum ammonia level (μmol/L) (Mean ± SD)
Grade 0	40.59±12.49
Grade I & II	67.87±29.20
Grade III	123.44±52.27
Grade IV	249.65±92.57

DISCUSSION

Mean age of the patients in this study was 50.89±12.81 years. This finding was comparable with the study done by Queiroz et al[22] in which the mean age of the patients was 56.3 ± 10.8 years.

Table 5: Day wise correlation of mean serum NH3 levels and hepatic encephalopathy

Days	Serum ammonia level (μmol/L) (Mean ± SD)	HE grades	p value
Day 1	296.19±106.38	Grade IV	0.001
Day 3	203.11±78.77	Grade IV	0.001
Day 5	123.44±52.27	Grade III	0.001
Day 7	67.87±29.20	Grade I & II	0.001
Day 9	40.59±12.49	Grade 0	0.001

In present study 70% of the patients were males and 30% of the patients were females while in study conducted by Wazir Mohammad Khan et al[23], 63% were males and 37% were females, similarly in a study done by Abidullah khan et al[24], there were 81 males as compared to 34 females who had hyperammonemia and HE. In both studies the number of males and females was comparable to present study. Moreover the number of males was more as compared to females in all the studies.

The mean total bilirubin levels of the patients in study were 6.17±7.47 gm/dl. In a study conducted by Dasani and his co-workers[25] the total bilirubin of the subjects was 2.24±0.20 gm/dl. So the total bilirubin of the patients in this study was much higher than Dasani et al.

In present study the mean serum ALT levels were 120.41 ± 242.07 U/L, a value far higher than the mean value of 82.33 ± 43.2 U/L in a study conducted by Senthil Raj et al[26].

Out of 100 patients having HE, 97 (97%) were hepatitis C positive while 3 (3%) were both hepatitis B and C positive, and it was comparable to the study conducted by Wazir Mohammad Khan et[23] al in which out of a total 100 patients, 91 (91%)had hepatitis C while 9 (9%) patients were having hepatitis B.

The ammonia levels showed positive correlation with the grades of HE, higher the levels of serum ammonia, higher were the grades of HE. Patients with mean ammonia levels of 40.59±12.49 μmol/L had no sign of HE while the patients with mean levels of 249.65±92.57 μmol/L were having grade IV encephalopathy. The rest of the patients had grades in between. The fact that increasing levels of ammonia were associated with increasing grades of HE was consistent with the studies done by Abidullah khan et al [24]and Omar Qureshi et al[27].

In this study it was found that the levels of

ammonia kept on decreasing gradually along with the grades of encephalopathy during the stay of the patients in hospital that is there was a positive correlation between the decreasing NH_3 levels and the grades of encephalopathy. This finding points towards the fact that ammonia has got prognostic role in the treatment of hepatic encepaholopathy and it is in accordance with the study conducted by Shalimar et al[28]. Moreover it also strengthens the fact that ammonia levels and grades of HE are strongly associated with each other.

CONCLUSION

In present study it was found that NH_3 levels have strong positive correlation with the grades of hepatic encephalopathy, as the levels of NH_3 rise the severity of encephalopathy also increases and with decrease in serum NH_3 levels the severity of the disease also declines. Moreover it was also concluded that serum ammonia levels can be used as a prognostic indicator for the patients having hepatic encephalopathy during their hospital stay.

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