Correlation between Ammonia Levels and the Severity of Hepatic Encephalopathy

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PURPOSE: Because the correlation between ammonia levels and the severity of hepatic encephalopathy remains controversial, we prospectively evaluated the correlation in 121 consecutive patients with cirrhosis.

METHODS: The diagnosis of hepatic encephalopathy was based on clinical criteria, and the severity of hepatic encephalopathy was based on the West Haven Criteria for grading of mental status. Arterial and venous blood samples were obtained from each patient. Four types of ammonia measurements were analyzed: arterial and venous total ammonia, and arterial and venous partial pressure of ammonia. Spearman rank correlations (r_s) were calculated.

RESULTS: Of the 121 patients, 30 (25%) had grade 0 encephalopathy (no signs or symptoms), 27 (22%) had grade 1, 23 (19%) had grade 2, 28 (23%) had grade 3, and 13 (11%) had grade 4 (the most severe signs and symptoms). Each of the four measures of ammonia increased with the severity of hepatic encephalopathy: arterial total ammonia ($r_s = 0.61$, $P \leq 0.001$), venous total ammonia ($r_s = 0.56$, $P \leq 0.001$), arterial partial pressure of ammonia ($r_s = 0.55$, $P \leq 0.001$), and venous partial pressure of ammonia ($r_s = 0.52$, $P \leq 0.001$).

CONCLUSION: Ammonia levels correlate with the severity of hepatic encephalopathy. Venous sampling is adequate for ammonia measurement. There appears to be no additional advantage of measuring the partial pressure of ammonia compared with total ammonia levels. **Am J Med. 2003;114:188–193.**

The pathogenesis of hepatic encephalopathy in chronic liver dysfunction is widely accepted to be due to the failure of hepatic clearance of toxic products from the gut (1). The exact toxins involved remain controversial, but ammonia is thought to be an important factor (2). However, the correlation between plasma ammonia levels and the severity of hepatic encephalopathy is not consistent (3–7), perhaps because previous researchers have used suboptimal blood sampling sites (e.g., venous instead of arterial), and older, less reliable ammonia assays (6,8,9). Recently, the partial pressure of ammonia has been reported to correlate with the severity of hepatic encephalopathy more closely than does total ammonia (7). The partial pressure of ammonia represents the nonionized form of ammonia, which diffuses freely across the blood-brain barrier and thus may reflect brain exposure to ammonia in patients with hepatic encephalopathy (10,11).

Ammonia levels are used widely in the diagnosis of hepatic encephalopathy in cirrhotic patients with altered mental status, and standard textbooks recommend obtaining ammonia levels when hepatic encephalopathy is suspected, both for diagnosis and as a guide to treatment (12). Often, an elevated ammonia level is used as an indication to institute therapy for hepatic encephalopathy.

The aim of this prospective study was to examine the correlation between plasma ammonia levels and hepatic encephalopathy. Because arterial ammonia is thought to be superior to venous ammonia and partial pressure of ammonia to be superior to total ammonia, we evaluated four types of ammonia measurements: arterial total ammonia, venous total ammonia, arterial partial pressure of ammonia, and venous partial pressure of ammonia.

METHODS

Patients

We recruited consecutive patients with cirrhosis who were admitted to our institution between September 1998 and December 1999. Patients were enrolled within 24 hours of admission, regardless of their mental status. A diagnosis of cirrhosis was determined by biopsy (41% [50/121] of patients) or by signs of portal hypertension, such as ascites, gastroesophageal varices, or previous variceal bleeding. We obtained informed consent from each patient or from an immediate family member if the patient was unable to give consent. Patients were ex-

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Table 1. West Haven Criteria for Grading of Mental Status

Grade 0	No signs or sumptoms
Grade 1	Trivial lack of awareness
	Euphoria or anxiety
	Shortened attention span
	Impaired performance of addition
Grade 2	Lethargy or apathy
	Minimal disorientation for time or place
	Subtle personality change
	Inappropriate behavior
	Impaired performance of subtraction
Grade 3	Somnolence to semistupor, but responsive to verbal stimuli
	Confusion
	Gross disorientation
Grade 4	Coma (unresponsive to verbal or noxious stimuli)

cluded from the study if they were younger than 18 years, had used alcohol within the previous month, had mental status changes due to a cause other than hepatic encephalopathy, or were undergoing hemodialysis, or if informed consent could not be obtained. This study was reviewed and approved by the institutional review board.

Clinical and Laboratory Information

Clinical and laboratory information was collected at the time of admission. Clinical information included age, sex, race, etiology of cirrhosis, Child-Pugh score, source of admission, admitting diagnosis, prior portosystemic shunt placement, dominant precipitating factor for hepatic encephalopathy, and lactulose use. Laboratory data included albumin, total bilirubin, and creatinine levels, and the international normalized ratio (INR) of the prothrombin time.

Diagnosis of Hepatic Encephalopathy

Mental status was assessed and graded on admission by a single investigator (JPO) using the West Haven Criteria (13) for grading of mental status (Table 1). The diagnosis of hepatic encephalopathy was made when mental status was altered and appropriate laboratory and diagnostic testing excluded other causes of mental status changes. Two senior investigators (KDM and DK), who have extensive clinical experience in hepatic encephalopathy, confirmed all cases of hepatic encephalopathy by reviewing the clinical course of the patient during the hospitalization. This served as the "gold standard" for the diagnosis of hepatic encephalopathy. The investigators were blinded to the patients' ammonia levels. Patients whose clinical course was not consistent with hepatic encephalopathy were excluded.

Blood Ammonia Determination

Fasting arterial and venous blood samples were obtained immediately after mental status assessment. Samples were drawn into heparinized Vacutainer tubes (Becton, Dickinson and Company, Franklin Lakes, New Jersey), placed immediately on ice, and taken to the clinical laboratory where they were processed and analyzed within 30 minutes of having been obtained. Blood pH was determined using the Rapid Lab 800 series blood gas analyzer (Chiron Diagnostics, East Walpole, Massachusetts). Total ammonia levels were determined in venous and arterial plasma by the enzymatic method, using the glutamate dehydrogenase reaction with reagents obtained from Roche Diagnostics (Indianapolis, Indiana) according to the manufacturer's protocol on a Roche Diagnostics Hitachi 917 analyzer. (Total ammonia levels are routinely measured in most clinical laboratories.) The partial pressure of ammonia was calculated from the pH and total ammonia level using the formula described by Manning (11).

Statistical Analysis

We constructed box plots (median and interquartile range, and individual data values) for all four measures of ammonia (arterial total ammonia, venous total ammonia, arterial partial pressure of ammonia, and venous partial pressure of ammonia) by the severity of hepatic encephalopathy. The relation between ammonia levels and the severity of hepatic encephalopathy was analyzed with the Spearman rank correlation coefficients and 95% confidence intervals, along with the Jonckheere-Terpstra test for trend. All statistical tests were two-sided, and a P value ≤ 0.05 was considered statistically significant.

Table 2. Characteristics of 121 Patients with Cirrhosis

Characteristic	Number (%)
Male sex	77 (64)
Race	
White	94 (78)
Black	14 (12)
Other	13 (11)
Etiology of cirrhosis	
Alcohol	45 (37)
Hepatitis C	17 (14)
Cryptogenic	10 (8)
Hepatitis B	5 (4)
Primary biliary cirrhosis	5 (4)
Nonalcoholic steatohepatitis	5 (4)
Alcohol + other	20 (17)
Other	14 (12)
Child-Pugh class	
A	5 (4)
B	32 (26)
C	84 (69)
Prior portosystemic shunt	
Transjugular intrahepatic	14 (12)
Surgical	3 (2)
Admitted from emergency room	31 (26)
Lactulose use on admission	64 (53)

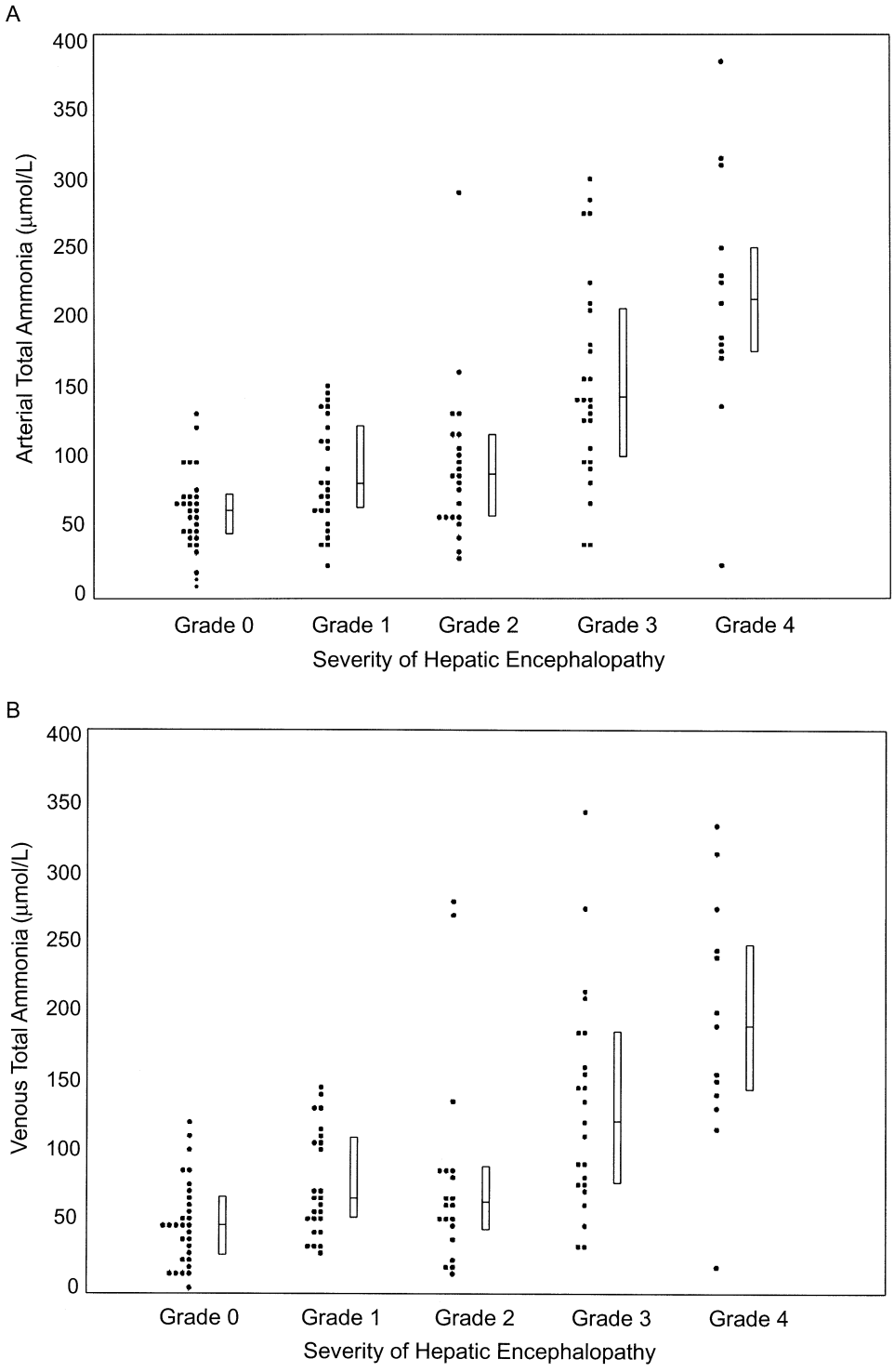
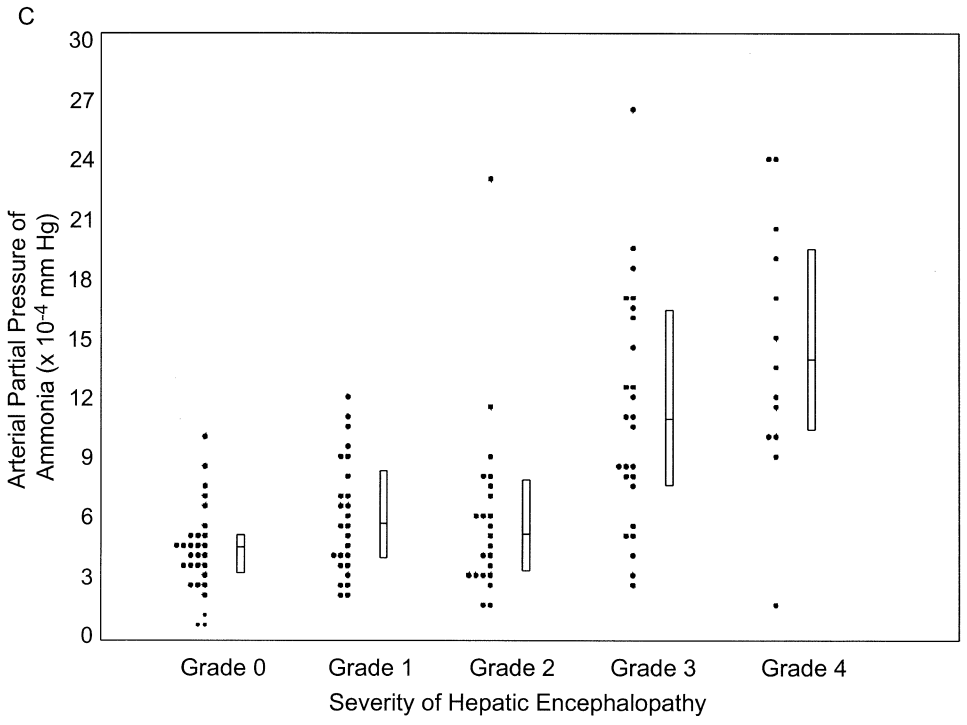


Figure. Box plot of the median and interquartile range and individual data values for (A) arterial total ammonia, (B) venous total ammonia, (C) arterial partial pressure of ammonia, and (D) venous partial pressure of ammonia, by severity of hepatic encephalopathy (from asymptomatic [grade 0] to most severe [grade 4]). The middle line within the box represents the median, the top line represents the 75th percentile, and the bottom line represents the 25th percentile. Data were missing for arterial values in 4 patients and for venous values in 10 patients. All four types of ammonia measurements correlated ($P \leq 0.001$) with the severity of hepatic encephalopathy: arterial total ammonia ($r_s = 0.61$; 95% confidence interval [CI]: 0.47 to 0.74), arterial partial pressure of ammonia ($r_s = 0.55$; 95% CI: 0.41 to 0.70), venous total ammonia ($r_s = 0.56$; 95% CI: 0.42 to 0.71), and venous partial pressure of ammonia ($r_s = 0.52$; 95% CI: 0.37 to 0.67).



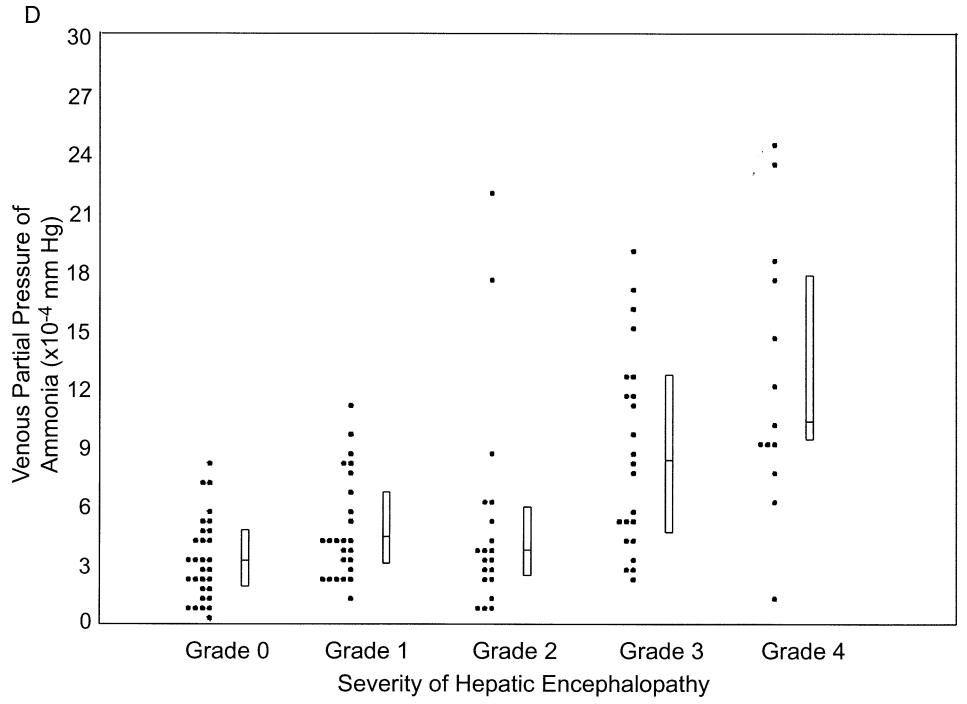


Figure. Continued.

In addition, the relations between the baseline clinical and laboratory variables and the severity of hepatic encephalopathy were evaluated. We used the Cochran-Armitage test for trend for dichotomous variables and the Spearman rank correlation coefficient and Jonckheere-Terpstra test for trend for continuous variables. Ordered logistic regression using the proportional odds model (14) was used to examine the relation between baseline characteristics and the severity of hepatic encephalopathy. Separate models, adjusted for ammonia levels, were fitted for each baseline variable that was associated significantly with the severity of hepatic encephalopathy. To assess the simultaneous effect of baseline characteristics, backward stepwise selection was used to select variables

for the multivariable model. Odds ratios and 95% confidence intervals were calculated. The odds ratio represents the increase in the odds of a 1-grade change in the severity of hepatic encephalopathy, per change in the predictor (independent) variable. All analyses were performed with SAS (SAS Institute, Cary, North Carolina).

RESULTS

Of the 153 patients whom we approached for the study, 16, including 10 with hepatic encephalopathy, refused to participate. Another 16 patients did not meet inclusion and exclusion criteria (2 did not have cirrhosis, 4 were on hemodialysis, 8 had recently been drinking alcohol, and 2 had altered mental status due to causes other than hepatic encephalopathy). The remaining 121 patients were included in the final analysis (Table 2). Their mean ($\pm$ SD) age was 54 $\pm$ 10 years (range, 29 to 75 years).

Presence and Severity of Hepatic Encephalopathy and Precipitating Factors

Of the 121 patients, 30 (25%) had grade 0 encephalopathy (no signs or symptoms), 27 (22%) had grade 1, 23 (19%) had grade 2, 28 (23%) had grade 3, and 13 (11%) had grade 4, the most severe form. The highest total ammonia levels, arterial or venous, were seen in patients with grade 3 or 4 encephalopathy (Figure); patients with grade 1 or 2 encephalopathy usually had ammonia levels $<$150 μmol/L. However, there was substantial overlap in the total ammonia levels by grade of hepatic encephalopathy. Of the 91 patients with hepatic encephalopathy (grade 1 or higher), the most common precipitating factors were azotemia (n = 25, 28%), infection (n = 19, 21%), gastrointestinal bleeding (n = 17, 19%), lactulose noncompliance (n = 5, 5%), and constipation (n = 5, 5%). Miscellaneous or unknown causes were responsible for 20 cases (22%).

Correlation between Ammonia Levels, Clinical Characteristics, and the Severity of Hepatic Encephalopathy

All four measures of ammonia increased with the severity of hepatic encephalopathy (Figure; $P < 0.001$ for each measure). Although arterial total ammonia had the highest correlation coefficient ($r_s = 0.61$) with the grade of encephalopathy, there were no significant differences in the correlation coefficients among the four types of measurements, as can be seen in the very similar confidence intervals (Figure).

Lactulose use, INR values, and serum creatinine and bilirubin levels were associated with the severity of hepatic encephalopathy (all $P \leq 0.005$), whereas source of admission, major precipitating factor, prior portosystemic shunt placement, etiology of cirrhosis, and serum albumin levels were not associated with severity of encephalopathy.

In multivariable ordered logistic regression models, only ammonia levels (odds ratio [OR] = 1.55 per 20-μmol/L increase in arterial ammonia level; 95% confidence interval [CI]: 1.36 to 1.75) and INR values (OR = 2.0 per 0.5 increase; 95% CI: 1.4 to 2.9) were independently associated with severity of hepatic encephalopathy. (Arterial total ammonia level was selected for these analyses, rather than any of the other three measures, because it had the highest correlation with severity of encephalopathy.) There was no correlation between INR values and arterial total ammonia levels ($r_s = 0.15$, $P = 0.11$).

DISCUSSION

We found a strong correlation between ammonia levels and the severity of hepatic encephalopathy. To overcome some of the limitations of previous studies, we optimized the conditions under which the relation between hepatic encephalopathy and ammonia levels was examined. Ammonia measurement was performed using a newer and more reliable enzymatic method, and grading of encephalopathy was blinded to the ammonia measurements. Under these conditions, ammonia levels correlated strongly with the severity of hepatic encephalopathy. Nevertheless, there remains substantial overlap in ammonia levels by grade of hepatic encephalopathy, which may be explained by variability in ammonia levels throughout the day, a possible lag between elevation in ammonia level and hepatic encephalopathy in some patients, or the possibility that compounds other than ammonia are also involved in the pathogenesis of hepatic encephalopathy.

Because arterial blood has been thought to be the more appropriate sampling site for ammonia measurement (3), we compared arterial with venous ammonia measurements. We observed as strong a correlation between venous total ammonia and severity of encephalopathy as with arterial total ammonia, suggesting that venous blood may be adequate for ammonia measurement.

Total ammonia levels, either arterial or venous, are measured in routine clinical care. The partial pressure of ammonia (arterial or venous), which is calculated from the total ammonia levels and the pH, may reflect brain exposure to ammonia more closely than does total ammonia, because it represents the freely diffusible form of ammonia (7,10,11). In this study, however, the correlations between the partial pressure of ammonia and the severity of hepatic encephalopathy were no better than those for the total ammonia level. This finding is consistent with the results of Muting et al. (15). However, another study (7) reported that partial pressure of ammonia

was more useful, perhaps because the patients in that study were more homogeneous, as all of them had encephalopathy.

In our study, patients without evidence of hepatic encephalopathy (grade 0) frequently had ammonia levels above 47 μmol/L, the upper limit of normal set by our clinical laboratory; for example, 69% (20/29) of the patients without signs or symptoms of encephalopathy had total arterial ammonia levels greater than that threshold. Because of the substantial overlap in total ammonia levels and partial pressures between cirrhotic patients with and without hepatic encephalopathy, a single level has little clinical utility in the diagnosis of hepatic encephalopathy. A careful clinical evaluation in patients with cirrhosis presenting with altered mental status remains the "gold standard" for diagnosis.

In summary, we found that venous total ammonia levels correlated with the severity of hepatic encephalopathy as well as arterial total ammonia levels. Furthermore, we found that correlations between the partial pressure of ammonia and the severity of hepatic encephalopathy were not significantly better than those for total ammonia. Our results suggest that venous total ammonia levels may be adequate in studies of hepatic encephalopathy.

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