A Dynamic Study of Rectally Absorbed Ammonia in Liver Cirrhosis Using [^{13}N]Ammonia and a Positron Camera

HIROFUMI KOEN, MD, KUNIO OKUDA, MD, HIROTAKA MUSHA, MD, YUKIO TATENO, MD, NOBUO FUKUDA, MD, TORU MATSUMOTO, MD, FUMIO SHISIDO, MD, TOMOYUKI RIKITAKE, MD, TAKESHI IINUMA, PhD, AKIRA KURISU, MD, and NOBORU ARIMIZU, MD

[^{13}N]Ammonia produced by the cyclotron was instilled intrarectally in patients with cirrhosis and other liver diseases to study the turnover of rectally absorbed [^{13}N]ammonia. A positron camera connected to an on-line computer system was used for the measurement of sequential changes of ^{13}N activity in blood and for coincidence positron imaging of the liver and heart. ^{13}N activity over the head was also recorded. Chromatographic analysis of ^{13}N-labeled substances in blood was carried out using a Dowex 50Wx8 column at varying times after the administration. In the control, [^{13}N]ammonia was absorbed quickly and visualized the liver, whereas in patients with cirrhosis, the lungs and heart were first visualized, and ^{13}N activity over the head was also higher. It was suggested that a large proportion of absorbed [^{13}N]ammonia bypassed hepatocytes and reached peripheral tissues in cirrhosis. The heart/liver ratio of ^{13}N and ^{13}N over the head were correlated with various indices of portal hypertension. The relative proportion of nonammonia ^{13}N metabolites in blood was lower at 5 and 15 min after administration in cirrhosis, suggesting a reduced capacity of the liver to remove and metabolize ammonia.

Ammonia produced in the gastrointestinal tract is absorbed into the portal circulation and transported to the liver. Most of the absorbed ammonia is removed from the portal blood and converted to urea and glutamine by the liver, keeping the blood ammonia level low in peripheral blood (1). In liver disease, particularly in cirrhosis, some of the abosrbed ammonia may reach the systemic circulation as a result of reduced effective hepatic blood flow (intrahepatic shunting), development of portasystemic (extrahepatic) shunts, and impaired metabolism of ammonia in the liver (2, 3). Thus, blood ammonia concentrations are frequently elevated in hepatic coma and in advanced cirrhosis (4).

Portal circulation and portasystemic shunts have been investigated with various radiological techniques, such as portography (5–8) and intrasplenic or intrarectal injection of radionuclides (9–11). These procedures are of value for the evaluation of portal hemodynamics but not of metabolic function of the liver. The ammonia tolerance test is not very satisfactory for the assessment of the latter, because it is influenced by hemodynamic changes of portal blood flow.

In this study, [^{13}N]ammonia produced by the cyclotron was administered intrarectally in patients with cirrhosis and other liver diseases, and dynamic transport of ^{13}N to the liver and other organs was

Manuscript received March 10, 1980; revised manuscript received May 19, 1980; accepted May 27, 1980.

From the Departments of Radiology and Medicine, Chiba University School of Medicine, and National Institute of Radiological Sciences, Chiba, Japan.

Address for reprint requests: Dr. Kunio Okuda, Department of Medicine, Chiba University Hospital, Chiba (280), Japan.

0163-2116/80/1100-0842$03.00/1

investigated by coincidence positron imaging, measurement of organ radioactivity, and biochemical analysis of blood ^{13}N.

MATERIALS AND METHODS

The 34 patients studied consisted of 19 with liver cirrhosis, 4 with chronic hepatitis, 3 with acute hepatitis in convalescence, 2 with extrahepatic portal hypertension, 1 with idiopathic portal hypertension, and 5 control patients who had recovered from a mild nonhepatic disorder. None of the patients with cirrhosis had encephalopathy at the time of the study. Informed consent to the procedure was obtained from each patient before the study. Diagnosis of liver disease was made by routine liver function tests, laparoscopy, scintiscan, and liver biopsy. The presence of esophageal varices was diagnosed by barium esophagography and esophagoscopy. The size of the spleen was measured on the posterior view of liver scintigraphy, and when the longest axis of the spleen was greater than 13 cm, it was interpreted as "splenomegaly" (12).

[^{13}N]Ammonia was produced by the $^{16}O(p,\alpha)^{13}N$ reaction of H_2O in the cyclotron at the National Institute of Radiological Sciences (NIRS) and subsequent reduction of the generated ^{13}N-labeled nitric acid. [^{13}N]Ammonia was immediately isolated by distillation into a vessel containing 5 ml of 0.9% $(NH_4)_2CO_3$ (13). ^{13}N is a short-lived positron-emitting isotope ($T^1/_2$ 10 min) that gives rise to annihilation gamma ray photons with an energy of 511 keV. Therefore, a positron camera recently developed by NIRS was used for the imaging of the liver and heart. It consisted of a focal detector of hexagonal multicrystal array and a conventional Anger-type gamma camera (Toshiba, GCA-202, delayed line type), connecting with an on-line computer system (TOSBAC-3400 model 31).

Some characteristics of the positron camera are: (1) maximum attainable high counting rate of about 2.5–5 kcps, (2) detection efficiency of 0.6–0.9 dots/sec/Ci, (3) spatial resolution of 6–9 mm (FWHM), depending on the distance from the collimator surface of the gamma camera to the source, and (4) area of uniform sensitivity of about 20 cm in dameter at the depth of 20 cm from collimator surface (14).

No dietary restrictions were imposed on patients, and the rectum was cleansed by enema before the procedure. A catheter was inserted approximately 15 cm into the rectum, and 15–30 mCi of [^{13}N]ammonia in 5 ml of 0.9% $(NH_4)_2CO_3$ was instilled. The catheter was immediately flushed with 10 ml of 0.15 M bicarbonate buffer of pH 8.1 (15). Sequential images of the liver and heart were recorded on polaroid films, stored as 32 sec frames of digitalized data and transferred in the magnetic tape in a 32 × 32 matrix form per one frame for a period of 40 min. The localization or area of the liver and heart had previously been determined by ^{99m}Tc-phytate imaging and ^{99m}Tc-phytate angiography, respectively. ^{13}N activity was calculated per unit area in a single cross-sectional view of the liver and heart at 5 cm from the anterior skin surface.

Since it was not possible to use the positron camera for the measurement of brain uptake while the liver and heart were monitored, a detector used in renography with a rate meter (Toshiba, RDU-501) and 2-channel recorder (Yokogawa Type-3047) were positioned over the left temporal region without the rectangular collimator and lead sheld, and total activity under the fixed machine condition was measured at 30 min, and the measurement was expressed as kcps per mCi administered. Although inaccurate, it was thought that such measurements might provide additional information regarding extrahepatic turnover of absorbed [^{13}N]ammonia.

Blood samples were obtained from an antecubital vein at 5 and 15 min after rectal instillation. Four milliliters of heparinized plasma were passed through Dowex 50Wx8 (column size 1 × 20 cm), and eluted with 20 ml of a pH 6.4, 0.15 M bicarbonate buffer at a flow rate of 4 ml/5 min. Under these conditions, ammonia is adsorbed onto the column, but urea, glutamate, glutamine, and other metabolites of ammonia collectively called "metabolites" hereafter, are not adsorbed (16, 17). In a preliminary study using this column, which has an adsorption capacity of approximately 16 mEq, it was demonstrated that at a load of various amounts of NH_3 up to 5 mEq, 85–90% of NH_3 were adsorbed and 85–90% of nonbasic amino acids eluted in one elution procedure. Eluates from the Dowex 50 column were measured for ^{13}N-labeled metabolites and the proportion of ^{13}N-labeled metabolites in the eluate was expressed as a percent of the total activity (% ^{13}N metabolites). The term "metabolites" is not exact because basic amino acids were not included, and the proportion of the real ^{13}N-labeled metabolites should have been somewhat greater than the measurements.

RESULTS

Sequential Positron Imaging of the Liver and Heart. In the control subjects, [^{13}N]ammonia was absorbed quickly into the portal vein visualizing the liver shortly after administration. ^{13}N activity frequently appeared in the area corresponding to the portal vein, but rather transiently. The positron image of the liver was very much like the liver image obtained by ^{99m}Tc-phytate scintigraphy, while the heart image was very faint or hardly discernible (Figure 1A). The decay-corrected liver activity (counts/cm^2) reached a maximum in 10–15 min. The heart activity (counts/cm^2) was much lower and increased gradually (Figure 1B).

Figure 2 shows the corresponding imaging and activity curves obtained in a patient with liver cirrhosis who had prominent esophageal varices and marked splenomegaly. The wedged hepatic vein pressure was 370 mm H_2O and 15-min retention of intravenously administered indocyanine green (ICG) was 45% (normal, less than 10%). The radioactivity of the liver was much lower compared to that of the heart. At 5 min from rectal administration, images of the lung and heart became apparent,

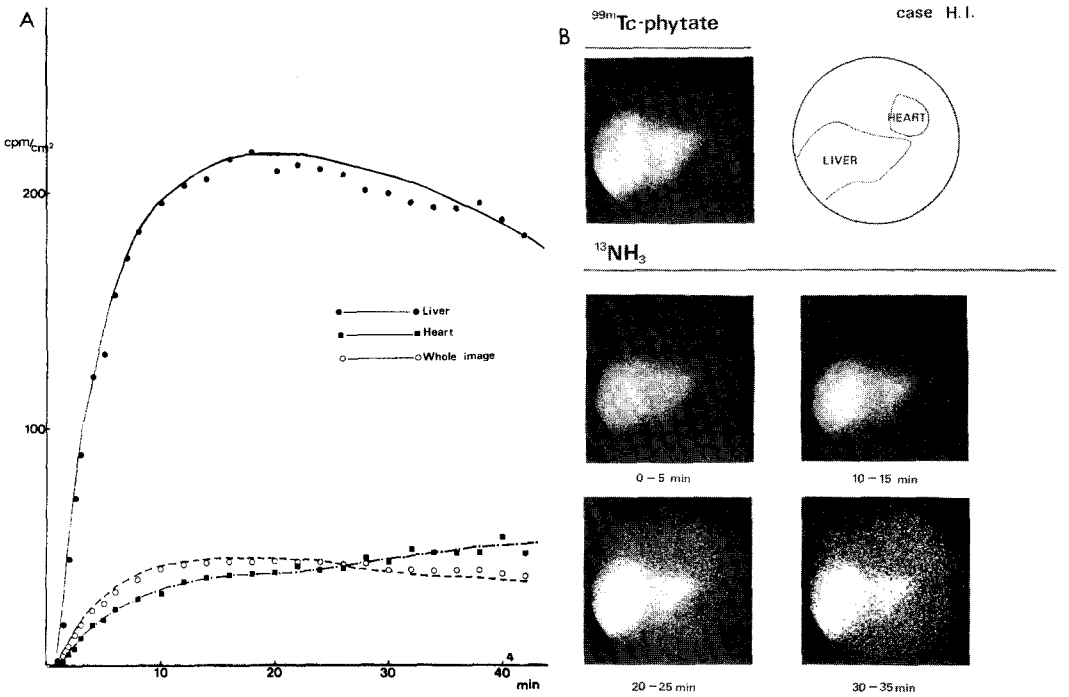

Fig 1. (A) Time course of counts over the liver, heart, and the whole image expressed per square centimeter. (B) Liver images obtained with ^{99m}Tc-phytate and with [^{13}N]ammonia in sequence in a control subject.

and the liver was barely discerned at 20 min. The heart took up considerable activity from 10 to 40 min, and myocardium was visualized to an extent similar to the myocardial image obtained by intravenous injection of [^{13}N]ammonia (Figure 2A). The heart activity increased, reaching a maximum in 10–15 min, in parallel with the liver activity (Figure 2B).

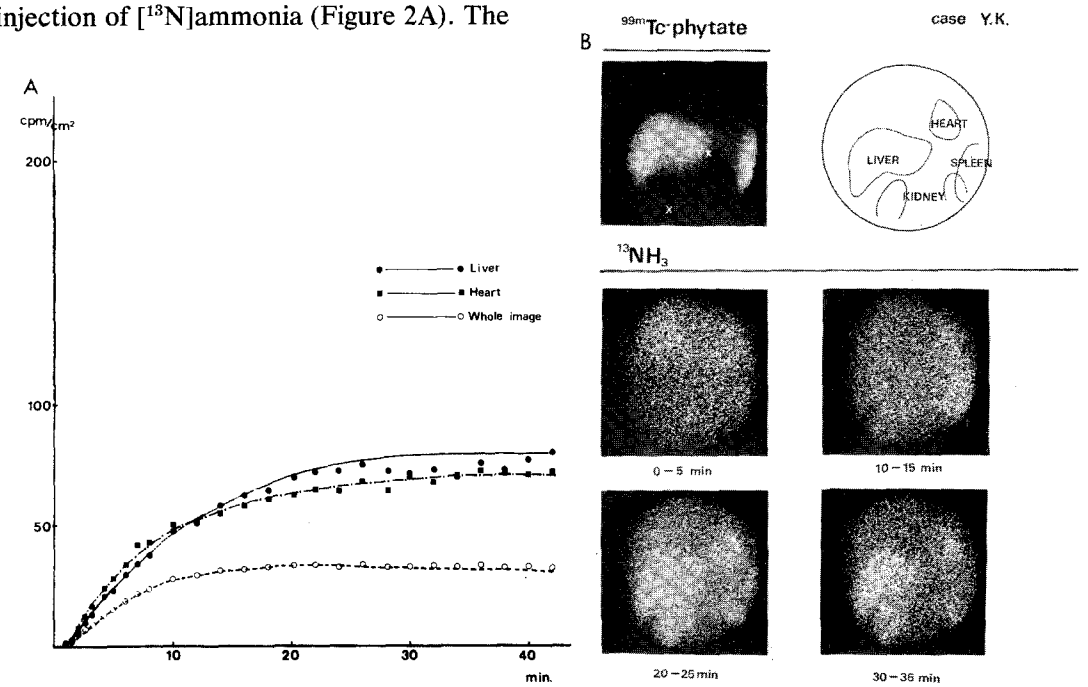

Fig 2. (A) Time course of counts over the liver, heart, and the whole image expressed per square centimeter. (B) Liver images obtained with ^{99m}Tc-phytate and with [^{13}N]ammonia in sequence in a cirrhotic patient.

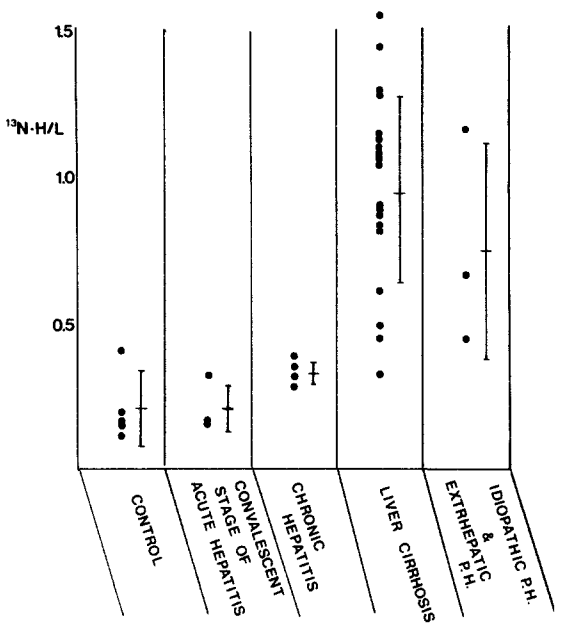

Fig 3. The ratio of ^{13}N-heart/liver (H/L) in various liver diseases. Vertical bars represent means and standard deviations.

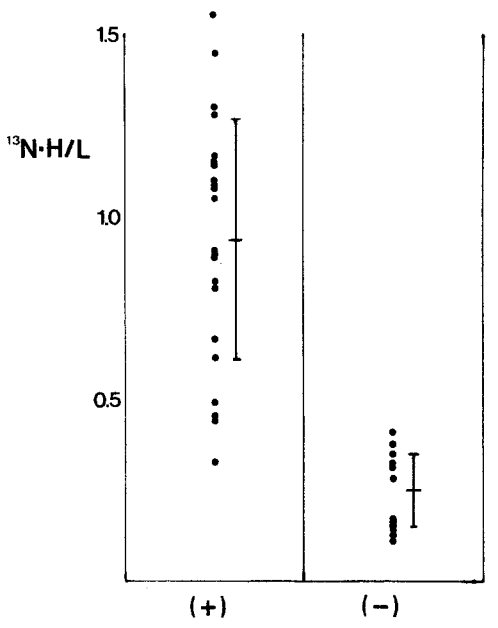

Fig 4. Correlation between ^{13}N-H/L and esophageal varices. Vertical bars represent means and standard deviations.

^{13}N-Heart/Liver Ratio. In control subjects, the liver activity was much higher compared with the heart activity, whereas in cirrhotic patients the heart activity was greater. Thus, the cardiac uptake relative to hepatic uptake would make a good indicator of the degree of shunt (and reduction in metabolic disposal of ammonia). We therefore determined the ^{13}N-heart/liver ratio at 15 min (^{13}N-H/L) after administration (Figure 3). The results demonstrated that ^{13}N-H/L was markedly increased in the cirrhotics and in patients with other types of portal hypertension—in control subjects, it was smaller than 0.45. It was suggested that most of the absorbed [^{13}N]ammonia would reach the systemic circulation bypassing the hepatocytes and enter the heart muscle. ^{13}N-H/L was increased in practically all subjects with demonstrable esophageal varices (Figure 4) and splenomegaly (Figure 5), and the differences from those without varices and splenomegaly were significant ($P < 0.01$). It was not closely correlated with 15-min ICG retention.

^{13}N Activity over the Brain. In 20 subjects studied, ^{13}N activity appeared over the head (brain) in about 1 min after administration and reached a maximum at 30 min. The brain activity at 30 min (B_{30}) was 1.24–2.7 kcps/mCi in the controls, whereas it was significantly higher in cirrhotic patients, ranging from 2.8 to 11.7 kcps/mCi. Although these measurements were not accurate, there was no overlap between the two groups. There was a close correlation between B_{30} and [^{13}N]H/L ($r = 0.861$, $P < 0.01$) (Fig 6).

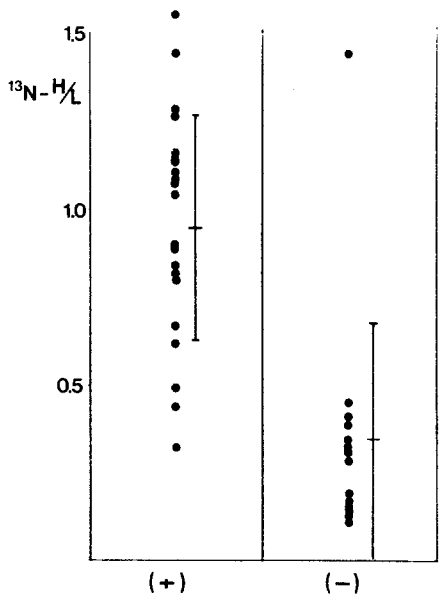

Fig 5. Correlation between ^{13}N-H/L and splenomegaly. Vertical bars represent means and standard deviations.

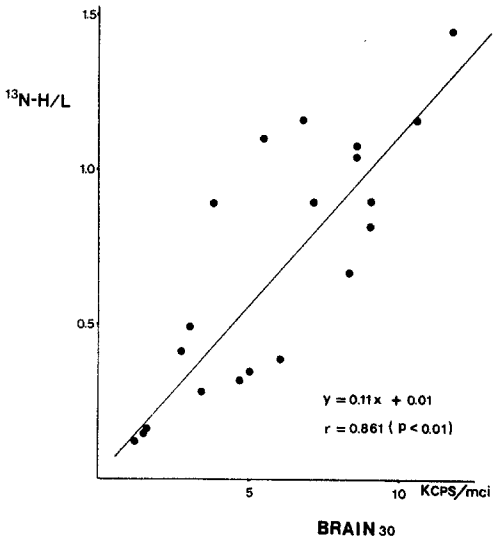

Fig 6. Correlation between ^{13}N-H/L and brain uptake of ^{13}N at 30 min ($Brain_{30}$).

Sequential Changes of ^{13}N Metabolites in the Blood: In all subjects, ^{13}N activity appeared in the blood during the first minute and increased rapidly, reaching a maximum at about 15 min. There was no remarkable difference in the sequential change of total ^{13}N-activity in the whole positron image between cirrhotic subjects and controls.

The blood should contain not only [^{13}N]ammonia but also other ^{13}N-labeled substances, because ammonia is quickly converted to urea and other nitrogenous compounds by hepatocytes. In fact, ^{13}N-labeled nonammonia substances were found in blood in considerable amounts as analyzed by chromatography. Figure 7 represents the percentages of ^{13}N metabolites to the total ^{13}N activity at 5 and 15 min (M_5 and M_{15}, respectively) after administration. In cirrhotic subjects, the percent of ^{13}N metabolites was comparatively low at both times, particularly at 5 min. Thus, ^{13}N-M_5 was significantly lower in the cirrhotic patients compared to the controls and other patients, and the percentage of ^{13}N metabolites increased thereafter more quickly in the cirrhotics. ^{13}N-M_{15}/M^5 correlated well with the presence and absence of esophageal varices ($P < 0.01$) (Figure 8) and with 15-min ICG retention ($r = +0.932$, $P < 0.01$) (Figure 9).

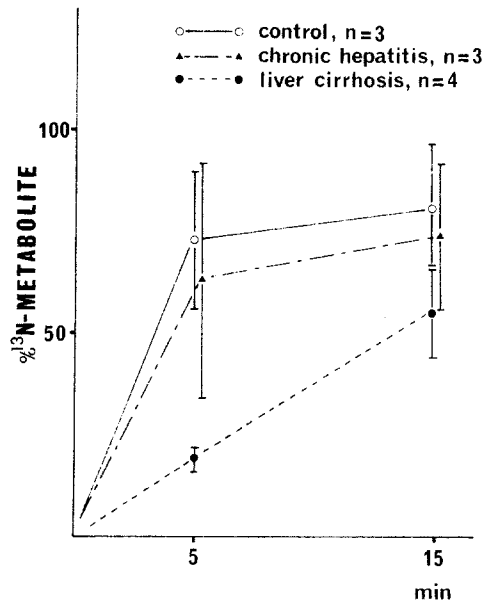

Fig 7. Proportion in percent of ^{13}N metabolites in blood 5 min and 15 min after rectal administration of [^{13}N]ammonia. Vertical bars represent means and standard deviations.

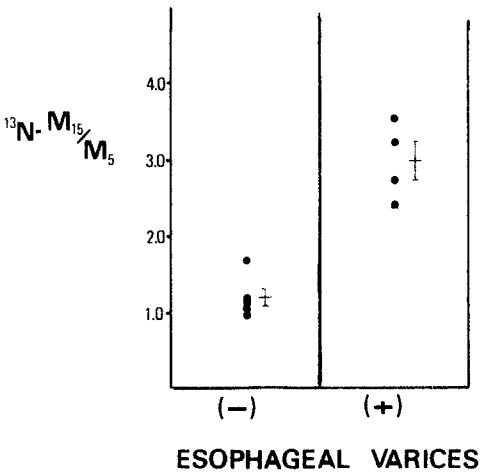

Fig 8. Ratio of percent ^{13}N metabolites in blood at 15 min (M_{15}) and at 5 min (M_5) in relation to esophageal varices.

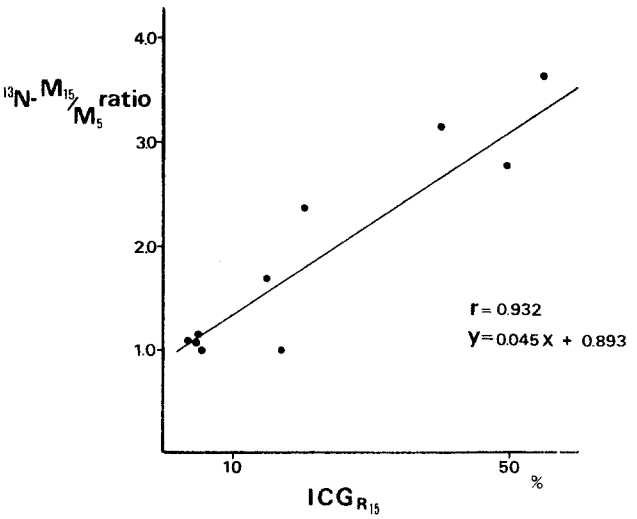

Fig 9. Correlation between ^{13}N-M_{15}/M_5 and 15-min retention of indocyanine green (ICG_{R15}).

DISCUSSION

Elevated blood ammonia levels are often associated with the state of reduced consciousness (3, 18), and elevation of ammonia occurs after ingestion of nitrogenous substances in advanced liver cirrhosis (19, 20). It is generally believed that portasystemic shunts are the primary cause of increased blood ammonia levels, and the magnitude of esophageal varices has been reported to correlate with ammonia tolerance test (21–23). Abnormal ammonia tolerance also reflects in some measure the hepatic blood flow and parenchymal function (24, 25).

[^{13}N]Ammonia, which is easily produced in the cyclotron, is virtually 100% pure with a high specific activity. With an external counting system suited for the 511-keV annihilation of ^{13}N, the dynamic behavior of absorbed [^{13}N]ammonia can be studied. [^{13}N]Ammonia also has a strong affinity for heart muscle, and has been used chiefly for myocardial perfusion imaging (26, 27). Hazenberg et al. have recently administered [^{13}N]ammonia intrarectally and studied sequential changes of the liver and heart images using an Anger-type gamma camera (28, 29). However, the quality of the image obtained by the gamma camera is severely limited, because lead collimation for high energy rays results in considerable image degradation, and the spatial resolution is inadequate for the determination of ^{13}N-H/L ratio. The coincidence imaging with a positron camera has the advantage of using electronic rather than mechanical collimation and having a much higher count rate (30). Such differences have clearly been demonstrated by Hoop et al, with their Massachusetts General Hospital positron camera (31). Using our NIRS positron system, a marked difference in the transport of rectally absorbed [^{13}N]ammonia was demonstrated between control and cirrhotic subjects. In the latter, with marked varices and liver dysfunction, the lungs and heart were visualized after rectal administration of [^{13}N]ammonia before the liver became visible. These abnormalities could be made more quantitative by the measurements of ^{13}N-H/L ratio which showed a close correlation with the presence or absence of varices and splenomegaly. The failure of Hazenberg et al (28) to demonstrate increased cardiac uptake of ^{13}N after administration in cirrhosis, which they could not explain, could have been due to several factors such as selection of patients with drug-induced cirrhosis, inadequate instrumentation for quantitation of radioactivity, and the use of a neutral to slightly acid [^{13}N]ammonia solution.

[^{13}N]Ammonia has in the past been mainly used in imaging of myocardium (27, 28, 30, 31). The initial blood clearance is very rapid, the liver being the principal site of uptake, and after 5 min most of the residual ^{13}N activity in blood is metabolically fixed nitrogen (32, 33). Myocardial uptake is also very rapid with approximately 90% extraction during a single pass, taking up 2–4% of the injected dose (32–34). [^{13}N]ammonia is also taken up by the lung, brain, muscle, kidney, and urinary bladder (34–36). Whereas the major mechanism for the removal of ammonia by nonhepatic organs, such as the heart, muscle, and brain, is glutamine formation via the glutamine synthetase pathway, the most important pathway in the liver is urea formation (37–39).

In liver cirrhosis, peripheral uptake of ammonia is increased in compensation for the reduced capacity of the liver to remove it from blood (40, 41), but such compensation is not sufficient, and blood ammonia concentration continues to be elevated (42). The turnover of ^{13}N metabolites may depend upon several factors other than liver function, and in this study, ^{13}N-M_{15}/M_5 was correlated both with the clinical indices of portal hypertension such as esophageal varices, and with 15-min ICG retention, an indicator of effective hepatic blood flow. Thus, analysis of individual positron images of the liver and heart, measurement of brain activity, and biochemical analysis of ^{13}N compounds in blood provide a clinically important, composite information regarding the degree of portasystemic shunt and metabolic function of the liver.

REFERENCES

1. Summerskill WHJ, Wolpert E: Ammonia metabolism in the gut. Am J Clin Nutr 23:633–639, 1970
2. Shaldon S, Chiandussi L, Guevara L, Caesar J, Sherlock S: The estimation of hepatic blood flow and intrahepatic shunted blood flow by colloidal heat denatured human serum albumin labelled with ^{131}I. J Clin Invest 40:1346–1354, 1961
3. McDermott WV Jr, Adams RD: Episodic stupor associated with an Eck fistula in the human with particular reference to the metabolism of ammonia. J Clin Invest 33:1–9, 1954
4. Breen KJ, Schenker S: Hepatic coma: Present concepts of pathogenesis and therapy. *In* Progress in Liver Diseases, H Popper, F Schaffner (eds). New York, Grune & Stratton, 1972, pp 301–312
5. Turner MD, Sherlock S, Steiner RE: Intrasplenic pressure measurement and splenic venography in the clinical investigation of the portal circulation. Am J Med 23:846–859, 1957
6. Gonzeles Calbalhaes O: Hepatoportografia por via umbilical. Rev Sanid Mil 12:42–48, 1959
7. Lunderquist A, Vang J: Transhepatic catheterization and obliteration of the coronary vein in patients with portal hypertension and esophageal varices. N Engl J Med 291:646–649, 1974

8. Okuda K, Suzuki K, Musha H, Arimizu N: Percutaneous transhepatic catheterization of the portal vein for the study of portal hemodynamics and shunts. Gastroenterology 73:279-284, 1977
9. Kashiwagi T, Kameda T, Abe H: Dynamic studies on the portal hemodynamics by scintiphotosplenoportography: the visualization of portal venous system using ^{99m}T. Gastroenterology 67:668-673, 1974
10. Deterling RA, Powers SR, Bhonslay SB: The use of radioactive sodium in the evaluation of patency of portacaval shunts. Surg Forum 5:193-200, 1954
11. Castell DO, Grace ND, Wennar MH, Chalmers TC, Moore EW: Evaluation of portal circulation in hepatic cirrhosis. Gastroenterology 57:533-541, 1969
12. Triger DR, Boyer TD, Redeker AG, Reynolds TB, Waxman AD: Differences in intrahepatic portal-systemic shunting in alcoholic and nonalcoholic liver disease as assessed by liver scan, portal pressure, and *E. coli* antibodies. Dig Dis Sci 24:509-513, 1979
13. Gelbard AS, Clarke LP, McDonald JM, Monahan WG, Tilbury RS, Kuo TYT, Laughlin JS: Enzymatic synthesis and organ distribution studies with ^{13}N-labeled L-glutamine and L-glutamic acid. Radiology 116:127-132, 1975
14. Tomitani T, Tanaka E, Nohara N: A positron camera by use of a focal detector of hexagonal multi-crystal array. Radioisotopes 25:693-697, 1976
15. Wolpert E, Phillips SF, Summerskill WHJ: Ammonia production in the human colon. N Engl J Med 283:159-164, 1970
16. Dienst SG: An ion exchange method for plasma ammonia concentration. J Lab Clin Med 58:149-155, 1961
17. Moore S, Stein WH: Procedures for the chromatographic determination of amino acids on four percent cross-linked sulfonated polystyrene resins. J Biol Chem 211:893-906, 1954
18. Traeger HS, Gabuzda FJ Jr, Ballou AN, Davidson CS: Blood ammonia concentration in liver diseases and liver coma. Metabolism 3:99-109, 1954
19. Kirk E: Amino acid and ammonia metabolism in liver disease. Acta Med Scand (Suppl) 77:1-147, 1936
20. Phillips GB, Schwartz R, Gabuzda GJ, Davidson CS: The syndrome of impending hepatic coma in patients with cirrhosis of the liver given certain nitrogenous substances. N Engl J Med 247:239-246, 1952
21. Conn HO: Ammonia tolerance in the diagnosis of esophageal varices. A comparison of endoscopic, radiologic, and biochemical techniques. J Lab Clin Med 70:442-451, 1967
22. Castell DO: The ammonia tolerance test: An index of portal hypertension. Gastroenterology 49:539-543, 1965
23. Conn HO: Ammonia tolerance in assessing the patency of portacaval anastomoses. Arch Intern Med 13:221-226, 1973
24. Price JB, McCullough W, Peterson L, Britton RC, Voorhees AB: Determination of ammonia tolerance in animals with and without portal systemic shunting. Surgery 60:701-709, 1966
25. White LP, Phear AE, Summerskill WHJ, Sherlock S: Ammonia tolerance in liver diseases. Observations based on catheterization of the hepatic veins. J Clin Invest 34:158-168, 1955
26. Harper PV, Lathrop DA, Krizek H, Lembares N, Stark V, Hoffer PB: Clinical feasibility of myocardial imaging with $^{13}NH_3$. J Nucl Med 13:278-280, 1972
27. Walsh WF, Harper PV, Resnekov L, Fill H: Noninvasive evaluation of regional myocardial perfusion in 112 patients using a mobile scintillation camera and intravenous nitrogen-13-labelled ammonia. Circulation 54:266-275, 1976
28. Hazenberg HJA, Gips CH, Beekhuis H, Vaalburg W: Rectal administration of ^{13}N-ammonia in cirrhosis of the liver. Acta Hepato-Gastroenterol 23:308-318, 1976
29. Hazenberg HJA, Gips CH, Beekhuis H, Kruizinga K: Rectal ^{13}N-ammonia test (^{13}N-liver/heart ratio), hepatic sinusoidal pressure and prevailing portal flow direction in cirrhosis of the liver. Acta Hepato-Gastroenterol 23:319-327, 1976
30. Walsh WF, Fill HR, Harper PV: Nitrogen-13-labelled ammonia for myocardial imaging. Semin Nucl Med 7:59-66, 1977
31. Hoop B Jr, Smith TW, Burnham CA, Correll JE, Brownell GL, Sanders CA: Myocardial imaging with $^{13}NH_3$ and a multicrystal positron camera. J Nucl Med 14:181-183, 1973
32. Harper PV, Schwartz J, Beck RN, Lathrop KA, Lembares N, Krizek H, Gloria I, Dinwoodie R, Mclaughlin A, Stark V, Bekerman C, Hoffer PB, Gottschalk A, Resnekov L, Al-Sadir J, Mayorga A, Brooks HL: Clinical myocardial imaging with nitrogen-13-ammonia. Radiology 108:613-617, 1973
33. Carter CC, Lifton JF, Welch MJ: Organ uptake and blood pH and concentration effects of ammonia in dogs determined with ammonia labelled with 10 minute half lived nitrogen-13. Neurology 23:204-213, 1973
34. Monahan WG, Tilbury RS, Laughlin JS: Uptake of ^{13}N-labelled ammonia. J Nucl Med 13:274-277, 1972
35. Phelps ME, Hoffman EJ, Coleman RE, Welch MJ, Raichle MZ: Tomographic images of blood pool and perfusion in brain and heart. J Nucl Med 17:603-612, 1976
36. Lockwood AH, McDonald JN, Reiman RE, Gelbard AS, Laughlin JS, Duffy TE, Plum F: The dynamics of ammonia metabolism in man. J Clin Invest 63:449-460, 1979
37. Rodwell VM: Protein and amino acid metabolism. *In* Review of Physiological Chemistry, 16th ed., HA Harper (ed). Los Altos, Lange Med Publications, 1977, pp 337-368
38. Chamalau RAFM, Tager JM: Nitrogen metabolism in the perfused rat liver. Biochim Biophys Acta 222:109-118, 1970
39. Davidson S, Sonnenblick EH: Glutamine production by the isolated perfused rat heart during ammonium chloride perfusion. Cardiovasc. Res 9:295-301, 1975
40. Bessman SP, Bessman AN: The cerebral and peripheral uptake of ammonia in liver disease with an hypothesis for the mechanisms of hepatic coma. J Clin Invest 34:622-628, 1955
41. Bessman SP, Bradley JE: Uptake of ammonia by muscle: Its implications in ammoniagenic coma. N Engl J Med 253: 1143-1147, 1955
42. Egense J: Ammonium tolerance test. A diagnostic aid in liver disease. Acta Med Scand 167:53-59, 1960