British Society of Gastroenterology

W50

TOTAL URINARY NITROGEN MEASUREMENT BY NEAR INFRARED REFLECTOMETER, COMPARISON WITH CHEMICAL METHODS.
S.Callani, F.Ponti, L.Benini, M.T.Arntegani, G.Castellani, I.Ventini
Dept of Gastroenterology, University of Verona at Valeggio s/N, Verona, Italy.

Chemical methods for the measurement of total nitrogen in urine are complex and therefore rarely used; urea nitrogen is the most used alternative, but it often underestimates urinary losses. Aim of this study was to assess the analytical efficacy of near infrared reflectance (NIRA) in the measurement of total urinary nitrogen. This method provides the results in about one minute, simply injecting the sample in a terminated cell of the equipment. The comparison was made with both fresh urines (40 samples: 28 from GI patients, 8 of whom on enteral 5 on total parenteral nutrition; 4 operated, 4 neoplastic and 4 burned patients) and on thawed urines (70 samples from GI patients, 20 of whom in enteral, 8 in total parenteral nutrition). These had to be acidified and warmed to obtain a clear solution. Two different calibration curves had to be obtained for fresh and thawed urines. This was done by a multiparametric regression, comparing the results of the Kjeldahl method with those obtained with the two values at 10 degree of the wave. Results: we found a range of NIRA nitrogen concentration of 0.35-2.04% in fresh, of 0.1-1.7% in thawed urines. A coefficient of determination of 0.99 was obtained between the mineralometric and the NIRA nitrogen concentration in fresh and thawed urines respectively. An intrasay coefficient of variation of 4.6% and of 9.6% was found for fresh and thawed urines respectively (3 measurment measured 20 times). In fresh urines, a correlation coefficient of 0.97 was found between the NIRA total nitrogen and the urea nitrogen.

In conclusion, the near infrared reflectance analysis represents a quick and reliable alternative to the complex chemical methods for the day-by-day-study of the nitrogen balance.

W51

ETHANOL FLUSH FOR THE PREVENTION OF CATHETER OCCLUSION
D. A. Johnston, K. Walker, J. Richards & C. B. Pennington
Gastro-Intestinal Unit, Ninewells Hospital, Dundee, DD1 9SY, Scotland, UK.

Catheter occlusion by lipid material has been associated with the use of compounded nutrient solution containing lipid. We have studied 51 patients in whom either a salicylate containing intravenous lipid was used prior to a heparin lock in patients receiving parenteral nutrition with such solutions in order to determine improved catheter survival and the incidence of catheter occlusion could be achieved.

METHOD
Following overnight infusion of parenteral nutrition, the giving set was disconnected from the extension set and either 20 ml of isonic saline solution (n=22) or 10 ml aqueous solution of 20% ethanol (n=29) was flushed through the catheter. A siphon was placed on the extension set and a neoplastic lock of 3,000 units heparin was injected through the siphon. Catheter occlusion was recognised by increasing resistance during the flush or by activation of the occlusion alarm of the infusion pump. Catheters were removed if they were occluded, or if there was no further need for parenteral nutrition.

RESULTS
The incidence of catheter occlusion was significantly (p<0.01, Z test) lower in patients who received the ethanol flush (27.1%) compared with patients who received the saline flush (13/25). In addition catheter survival was significantly (p<0.1, logrank test) longer in patients who received the ethanol flush. In complications of the flush were observed in either group.

CONCLUSION
Ethanol flush is a simple, safe and effective method of reducing the incidence of catheter occlusion with compounded solutions.

Liver

W52-W59

FIBRINOlyTIC ACTIVITY IN CIRRHOSIS
Osman E. Hutton, P. Meinire N. Burroughs AK
Hepatobiliary and Liver Transplantation Unit, Ninewells Hospital and Haematology Unit, Ninewells Hospital and Haematology Unit, Royal Free Hospital and School of Medicine, Pond Street, London N3 2OG.

Although there is in vitro evidence for increased fibrinolytic activity in cirrhosis, in vivo evidence of fibrinolysis is indirect and controversial, because both pro and antifibrinolytic components need to be measured simultaneously, together with an index of thrombus degradation to distinguish primary from secondary fibrinolysis. We evaluated 51 cirrhotics (26 alcoholic, 13 PBC, 12 non-alcoholic non-biliary) measuring fibrinogen, activators of fibrinolysis (plasminogen activator inhibitor-TPA), which is solely endothelial derived, inhibitors of fibrinolysis, plasminogen activator inhibitor-PAI-1, also solely endothelial derived; anti-thrombin III-ATP II, antithrombin(II) as well as x-linked fibrin degradation products (XDP) - only present if thrombus degradation occurs and whole blood fibrinolysis (clot lysis index) by thromboelastography which has been used clinically as an index of fibrinolysis. TPA was elevated in 77% of alcoholics, 31% of PBC and 75% of the remainder, as was the PAI-1 in 58% alcoholics, 62% PBC, and 58% of the others, with an expected correlation (p<0.02). However the hepatic derived inhibitors, AT III and AT III were low in 90% and 88% of the total group but fibrinogen levels were low in only 25%. Clot lysis index was only abnormal in 8(16%), and it correlated with fibrinogen (p<0.02) and ATP (p<0.02), but not with TPA, PAI-1 or their ratio. In 4 of these XDP were not elevated suggesting primary fibrinolysis. XDP were raised in 18(33%) of the total, 13 of whom had raised TPA levels suggesting that these 13 had a degree of secondary fibrinolysis. This study shows that both primary and secondary fibrinolysis exist. Further investigation will clarify which tests are the most useful in a clinical setting such as bleeding, sepsis, ascitic recirculation or surgery.
S14

The effect of ursodeoxycholic acid (UDCA) on choledolithiasis, liver function and nutrition was assessed in 12 patients with cystic fibrosis related liver disease who were randomised to receive UDCA (20mg/kg/day) for 6 months or no treatment. UDCA treatment was associated with significant improvements in 5’Nucleotidase, gamma-glutyl transferase, aspartate aminotransferase and alanine aminotransferase levels which were not observed in the control group. After 6 months the hepatic clearance of Tc99 Dilo propophyllerylcarboxy-methyl-imido acetic acid (DISIDA) at 45 and 60 minutes as an index of biliary excretion and the plasma disappearance rate of indocyanine green as an index of hepatic function were unchanged in both groups. Body weight increased to a similar degree in both treatment and control groups, mean % change in body weight 7.6+/-.3-3% and 11.9+/-.3-4 % respectively (P=NS), an effect attributable to the intensive dietary advice and supervision given to all patients throughout the study period.

Although improvements in liver biochemistry may be anticipated with UDCA therapy further controlled studies are required to determine if long-term administration of UDCA influences hepatocellular function, biliary excretion or nutrition.


Kupffer cells are responsible for ingestion of portal endotoxin and secretion of potent cytokines. Their dysfunction has been implicated in the pathophysiology of cholestasis, cirrhosis, severe sepsis and multi-system organ failure. Our aim was to develop an accurate in vivo method of measuring Kupffer cell clearance and apply this system to an animal model of obstructive jaundice. Male Wistar rats (250-350g, n=25) were anesthetised and the liver was surgically isolated with cannulae in the portal and suprahepatic inferior vena cava (IVC). After ligation of the infrahepatic IVC the liver was perfused in situ with Krebs-Bassleit solvent for 10 minutes and then with buffer containing Fluoresbrite latex particles (0.7μm) by single pass. Samples of effluent perfusate were collected at 2 minute intervals for 10 minutes and fluorescence measured at 540nm (ex 44nm). Fluorescent particles appeared in the effluent perfusate in less than 1 minute and rapidly reached a steady state. Clearance was measured as percentage change in fluorescent effluent perfusate and expressed as percentage clearance. Clearance was normally distributed (Mean 34.1g 1.42 s.D 6.4). Electron microscopy confirmed exclusive ingestion of particles by Kupffer cells.

This system is both novel and reproducible and can be applied to established animal models to elucidate the role of the Kupffer cell population in the pathophysiology of disease. By measuring cytokine concentrations in the effluent perfusate secretory function can be measured synchronously with phagocytosis in the physiologically intact organ.

LIVER FUNCTION IN PATIENTS WITH ALPHA-1-ANTITRYPSIN DEFICIENCY AND SEVERE PULMONARY EMPHYSEMA. J.V. Schonfield, R. Zott, N. Breuer, H. Goebell (introduced by DL. Wingate).

Dept. of Gastroenterology, University of Essen, FRG.

Alpha-1-antitrypsin (AAT) deficiency is a hereditary disorder, which may cause pulmonary emphysema and liver disease. The aim of this study was to assess possible liver disease associated with AAT deficiency and severe pulmonary emphysema.

10 male and 6 female patients (mean age 46, range 26 - 59 years) with AAT deficiency (phenotype Pi ZZ) were studied. Patients gave a detailed medical history, they underwent complete physical examination and ultrasonography. The following parameters were measured using routine laboratory methods: bilirubin, AP, ALT, AST, g-GT, albumin, PTT and TTP. In a subgroup of 6 patients, quantitative liver function tests were performed after intravenous administration of galactose (500 mg/kg bw) and indocyaninegreen (0.5 mg/kg bw).

Three patients gave a history of liver disease (icterus and/or hepatitis B). Physical examination and ultrasonography did not suggest relevant liver disease in any patient. There were elevated serum activities of g-GT in 4, of ALT in 3 and of AST in 2 out of 16 patients. 10 out of 16 had normal results in all tests. Galactose elimination capacity was impaired in 5 out of 6 patients (4.6 - 6.2 mg/kg bw; normal > 6.6) and indocyaninegreen halftime was prolonged in 1 (9.2 min; normal < 6.3).

In conclusion, clinically relevant liver disease was not seen in any of the 16 patients with AAT deficiency and severe emphysema. Reduction of galactose elimination capacity could indicate impaired metabolic function of the liver. Since this metabolic pathway is oxygen dependent, it seems, however, more likely to be a consequence of the hypoxemia, present in all our patients. Our results suggest that severe lung and liver disease rarely coexist in the same patient.

PLASMA CATECHOLAMINE RESPONSES IN THE AUTONOMIC NEUROPATHY OF CIRRHOSIS. M.T. Hendrickse, D.R. Triger Department of Medicine and Pharmacology, University of Sheffield, U.K.

We have examined the catecholamine responses to alteration in posture in a group of cirrhotic patients with and without autonomic neuropathy. 14 cirrhotics (7 alcoholic, 3 primary biliary cirrhosis, 4 chronic active hepatitis; 13 Childs A and 1 B) and 7 age matched controls were evaluated using standard cardiovascular autonomic tests, and the hemodynamic and plasma noradrenaline response to 60 degrees passive head up tilt was then determined.

As reported previously, the noradrenaline response to tilting without vagal neuropathy (n=6) did not differ appreciably from controls (mean % increase at 5 mins: 64 % vs 100 % p > 0.3). In contrast, the presence of vagal neuropathy (n=8) was associated with a significantly impaired response (38 % vs 100 % p < 0.038). The blood pressure and heart rate changes in the three groups were similar. The % change in noradrenaline correlated with the valsalva ratio (r=0.48, p<0.03) and the lying standing ratio (r=0.2, p<0.005).

The impaired catecholamine response to tilting observed in cirrhotics with autonomic neuropathy could be explained by a defect in baroreflex function.
W58

**Gamma Linoleic Acid Increases Interferon-γ Inhibition of Human Hepatoma Cell PLC/PRF/5**

University Department of Surgery, University of Wales College of Medicine, Cardiff.

Gamma linoleic acid (GLA) has been reported to inhibit proliferation of malignant cells. We have investigated the effect of GLA on interferon-gamma (IFNγ) inhibition of hepatoma cells.

A human hepatoma cell line (PLC/PRF/5) was used in this study. Cells were co-cultured with different concentrations (three are shown here) of IFNγ alone or in the presence of GLA and cell proliferation was quantified by using a dimethyl-thiosulphate-tetrasulphide bromide assay. Results are shown as percentage proliferation compared with control cultures (100%). Statistics by Student T test.

<table>
<thead>
<tr>
<th>IFNγ (0.6IU/ml)</th>
<th>IFNγ (2.5IU/ml)</th>
<th>IFNγ (10IU/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>95.6±0.8%</td>
<td>89.42±4.8%</td>
</tr>
<tr>
<td>GLA 40µM</td>
<td>89.82±1.5%</td>
<td>86.84±2.0%</td>
</tr>
<tr>
<td>GLA 100µM</td>
<td>88.82±1.8%</td>
<td>85.12±1.9%</td>
</tr>
<tr>
<td>GLA 200µM</td>
<td>87.12±1.6%</td>
<td>83.02±3.4%</td>
</tr>
</tbody>
</table>

Analysis of the data shows a significant difference from the cultures without any IFNγ or GLA (medium only). Gamma linoleic acid itself also showed a dose dependent inhibition of PLC/PRF/5 proliferation (dose vs inhibition, r=−0.53, p<0.05) the peak value was 27.67±1.8% at 250 µM.

These data show that gamma linoleic acid, one of the n−6 fatty acids, increases hepatoma cell PLC/PRF/5 sensitivity to interferon gamma inhibition and GLA itself is a proliferation inhibitor to these cells.

W57

**Serum Tumor Markers for Diagnosis of Cholangiocarcinoma in Patients with Primary Sclerosing Cholangitis**

I.K. Ramsay, M.J. Farrant, R. Forn, R. Williams

Institute of Liver Studies, King's College Hospital, London SE5 9SR and *Royal Naval Hospital, Haslar, Gosport, Hants.

Cholangiocarcinoma (CC) is a frequent complication in patients with primary sclerosing cholangitis (PSC) and can be very difficult to detect. Diagnosis of CC pre-transplant is important because recurrence of the tumour is inevitable in the transplanted liver.

Carbohydrate antigen 19-9 (CA19-9) is a tumour marker which is present in the serum of patients with pancreatic, biliary and other upper GI tumours. Carcinoembryonic antigen (CEA) is raised in many gut-related tumours. We aimed to measure CA19-9 and CEA levels in three groups of patient: A. 8 patients with PSC known to have CC in the explanted liver at transplantation. B. 16 patients with PSC having an explanted liver free of CC on sectioning. C. 20 patients with stable PSC.

Mean (and ranges) of serum CA19-9 (Units/ml) were: group A: 730 (24-2754); group B: 61.9 (5.5-284); group C: 42.1 (3.1-156). Mean and ranges of CEA (ng/ml) were: group A: 6.1(1.3-13.9); group B: 2.8(0.5-6.8); group C: 2.2 (0.5-7.0). For transplanted patients only (groups A and B, using 200 units/ml and 7 units/ml respectively as the cut-off for CA19-9 and CEA, and combining the tests, sensitivity for diagnosis of CC was 75% and specificity 87% with overall accuracy 83%. Three out of four cases in which the CC was occult prior to transplantation were correctly diagnosed by the combined test. All sera from patients with stable PSC (group C) were negative for both tests.

A combination of serum CA19-9 and CEA levels have reasonable accuracy for diagnosis of CC in patients with PSC and are at least as good as other available diagnostic measures.

W59

**Basement Membrane Components Increase Secretion of Type IV Collagenase/Gelatinase Activity by Cultured Human Hepatic Lipocytes.**

P.J. Kowalski-Taunders, *A. Strain, M.P. Arthur*

University of Southampton, UK. *The Liver Unit, Birmingham, UK.*

We have previously demonstrated that cultured human hepatic lipocytes synthesize and secrete 72KDa type IV collagenase/gelatinase (J Clin Invest, 100:54-71, 1997). This enzyme which can degrade the normal subendothelial liver matrix may contribute to liver injury and the early stages of liver fibrosis, but little is known about factors that regulate its biosynthesis or activity in liver. In this study we have investigated the effects of basement membrane components on release of 72KDa type IV collagenase/gelatinase activity by cultured human hepatic lipocytes.

Hepatic lipocytes were prepared from normal donor human liver (n=5) as previously described (Hepatology 12:908) and further purified by serial passage. All cells stained positively for the presence of desmin by immunofluorescence. Basement membrane proteins rich in laminin were extracted from the Engelbreth-Holm-Swarm murine sarcoma (EHS matrix). Paired cultures of lipocytes plated on plastic were studied in the presence or absence of soluble EHS matrix diluted 1:15 with culture medium.

By gelatin zymography, all lipocyte cultures secreted a gelatin-degrading activity of appropriate molecular size for 72KDa type IV collagenase/gelatinase. Quantitative analysis (mean ± standard mean of differences for paired data. CPM degraded/flask/24hrs) demonstrated a marked increase in secretion of this enzyme activity for lipocytes exposed to EHS matrix (179±5478) compared to paired controls (875±478; n=5, p<0.05, Wilcoxon).

These data indicate that an interaction between cultured lipocytes and components of EHS matrix leads to increased secretion of 72KDa type IV collagenase/gelatinase. The nature of this interaction is uncertain, but EHS contains matrix protein (eg laminin) degradation products which are known to increase synthesis of matrix metalloproteinases by other fibroblasts. Our findings suggest the possibility of a positive feedback loop for collagen degradation in liver.

W60

**Excessive Wasting of Blood Resources in Elective Colorectal Surgery**

R. Paroux, B. Jacques, J.K. Drury, I.S. Smith

Dept. of Surgery, Victoria Infirmary, Glasgow.

Current guidelines for cross-matching policy in relation to elective colorectal surgery suggest between 2 and 4 units of blood should be requested.

We have prospectively studied one hundred and six elective colorectal procedures in one six month period (94 for malignancy). The median patient age was 71 years (range 39 - 91 years). Twelve patients with a carcinoma of the right hemicolon were transfused preoperatively. Forty-two units of packed cells were reserved and subsequently used with no wastage. A median of 2 units (interquartile range 1-4) was cross-matched in the perioperative period. The median preoperative haemoglobin was 13.1 g/dL (9.8 - 16.4 g/dL) and the median postoperative haemoglobin was 12.1 g/dL (8.4 - 15.7 g/dL). Nine patients (8.5 %) required a transfusion (median 2 units) in the immediate post-operative period (< 48 hours). Four of the five patients receiving a transfusion within 24 hours of surgery had either undergone a subtotal colectomy or abdomino-perineal resection. One hundred and seventy nine of the two hundred and forty two units requested in the postoperative period were unused.

Perioperative cross-matching and reservation of blood for elective colorectal surgery is an unnecessary expense. The selective policy for blood resources being retained for grouping is justified.