T177

SMALL INTESTINAL MICROSPORIDIOSIS IN HIV INFECTION: A REPORT OF 22 CASES

Blanshard C, Dowell S, Ellis D S, Canning E U, Gazzard B G
The Westminster Hospital, the London School of Hygiene and Tropical Medicine, Imperial College, London.

We describe 22 cases of small intestinal infection with Enterocytozoon bieneusi of whom 21 were identified as part of a study of the causes of chronic diarrhoea in 117 AIDS/ARC patients. The other was one of 15 patients with diarrhoea and early HIV infection. Microsporidial infection has not previously been described in such a patient.

All except 4 patients had a CD4 lymphocyte count below 50/mm^3 at presentation (median 23/mm^3) and over half (13) had a previous AIDS diagnosis. They presented with a mean stool volume of 950ml/day in 2-15 (mean 6) liquid or semi-liquid motions. Thirteen (59%) had a Body Mass Index below 20 and 90% had abnormal xylose and/or Schilling tests. In 8 cases other enteric infections were found but the diarrhoea did not resolve after treating them. Six patients have died a median of 3 months (1-6) from the diagnosis. Sixteen remain alive but still infected at up to 15 months follow-up.

Electron microscopy and light microscopy of Haematoxylin and Eosin stained paraffin sections or Giemsa-stained smears had equal diagnostic sensitivities (95%) and duodenal and jejunal biopsies gave similar results.

Ten patients have been treated with albendazole and the diarrhoea has markedly improved in 7; 9 have been treated with metronidazole of which 4 had some improvement although 3 of these had other metronidazole-sensitive infections. No treatment eliminated the infection.

T179

CCK ANTAGONISES THE ANTITUMOUR EFFECTS OF SOMATOSTATIN ANALOGUE RC-160 ON PANCREATIC CANCER IN VIVO.

P Kapur, N.M.Davies, J.Gillespie, A.V.Schally*, P.J.Guillou and G.J.Poston. Academic Surgical Unit, St Mary's, London and Tulane Medical School, New Orleans, USA*

Somatostatin inhibits pancreatic cancer via a tyrosine phosphatase receptor which dephosphorylates the products of the tyrosine kinase growth stimulatory family of receptors. The purpose of these studies was to examine the action of the cholecystokinin analogue caerulein, acting via cGMP, on the activity of somatostatin analogue RC-160 on the growth of pancreatic cancer, compared to other peptides which act through tyrosine kinase. 48 Syrian golden hamsters were inoculated with H2T hamster pancreatic cancer and randomised to 8 equal groups: saline; RC-160 (100ug/ml);EGF (10ug/ml);EGF+RC-160;TGF-alpha (10ug/ml) TGF-a+RC-160; caerulein (10ug/ml);caer+RC-160, each by intraperitoneal injection 3 times daily for 7 weeks. In a second study 24 hamsters were inoculated with H2T and randomised to 4 equal groups: saline; RC-160; insulin-like growth factor-1 (10ug/ml); IGF-1+RC-160, using the same regimen as study 1 but for 4 weeks. Tumour areas were measured weekly and from week 3 of both studies, trends emerged which persisted until sacrifice, which were (mm^2):

Con	RC-160	EGF	EGF+RC	TGF-a	TGF-a+RC	caer	caer+RC
99±6	83±7	145±12*	85±7*	140±6*	86±7*	102±7	110±12

con	RC-160	IGF-1	IGF+RC
37±2	25±2*	48±2*	21±1*

(* p<0.05 compared to control by 2-way ANOVA)

This is the first in vivo study to show a tumour trophic effect of IGF-1. In addition, although tyrosine kinase mediated growth stimulation can be inhibited by somatostatin, this effect can be reversed in vivo by caerulein acting through cGMP. This important interaction has clinical implications for the therapeutic use of somatostatin analogues in cancer treatment, and raises the question of combination therapy using somatostatin analogues with CCK antagonists in the treatment of patients with pancreatic cancer.

Neoplasia T178–T186

T178

A PUTATIVE HUMAN SPASMOLYTIC POLYPEPTIDE (hSP) ANTIBODY AND ITS STAINING PATTERN IN GASTROINTESTINAL EPITHELIUM.

George Elia, Andrew Hanby, Richard Poulsom, Janusz Jankowski, and Nicholas Wright.
ICRF/RCS Histopathology unit Lincoln's Inn Fields, London.

The trefoil peptides are a novel group of peptides with a characteristic cloverleaf motif held in place by three disulphide bonds. The most well known members of this group in gastrointestinal disease are pS2 and hSP which are of particular interest because of their distribution in the novel reparative cell lineage the Ulcer associated cell lineage (UACL).

Both peptides have been investigated by the distributions of their mRNA's using in-situ hybridisation. It has only been possible to investigate the pS2 using immunohistochemical techniques.Here we present our preliminary findings on a putative antibody to hSP.

These mouse monoclonal antibodies were prepared by subcutaneous injections of a 16 amino-acid hSP peptide with and without Freund's adjuvant followed by intraperitoneal then intravenous injections of this peptide; the immune spleen was then fused with the NS1 myeloma cell line.

Screening for positive clones was undertaken by employing the supernatent upon duodenal sections in conjuction with an ABC/peroxidase technique.

The most promising clones stained the epithelial compartments in the gut which mirror the known distribution of hSP mRNA. In particular antibody A5 co-localises with pS2 staining in the gastric foveolar epithelium but not in the Ulcer Associated Cell Lineage (UACL) as with hSP mRNA, hSP is present in the acinar but not upper ductular and surface components.

The co-localisation of hSP mRNA with A5 antibody provides good evidence that this antibody truly recognises this trefoil-peptide and as such will provide a valuable research tool.

T180

ROLE OF RECTAL EPITHELIAL PROLIFERATION IN PREDICTING COLORECTAL ADENOMAS RECURRENCE.

M Anti, G Marra, F Armelao, R Ficarelli, A Percesepe, M Ponz de Leon*
Ist. di Clinica Medica, Universita' Cattolica di Roma; *Ist. di Patologia Medica, Universita' di Modena, Italy.

Surveillance colonoscopy is recommended in patients with sporadic colorectal adenomas after initial polypectomy. However, criteria to predict the level of risk for subsequent adenomas have not been well identified yet. AIMS: 1)to determine whether the proliferation status of flat rectal mucosa in initial colonoscopy can affect the discovery of colorectal metachronous polyps; 2) to evaluate the changes, over time, of proliferative pattern in subsequent surveillance colonoscopy.

METHODS: After a "clearing" polipectomy, 55 patients with adenomas were followed up for 18 months with a complete colonoscopy every 6 months. 3H-Thymidine autoradiography was used to evaluate the epithelial cell proliferation on endoscopic biopsies of flat rectal mucosa at the 1st colonoscopy. The kinetic pattern was reevaluated in 38 patients at one of the 3 subsequent surveillance examinations. At least 30 longitudinal hemicrypts (i.e. each column of well oriented crypt unit) were evaluated for each study. The ratio of 3H- thymidine labeled cells (S-phase cells) to the total number of hemicrypt cells was expressed as Total Labeling Index (tLI). In addition, each hemicrypt was conventionally divided into 5 longitudinal compartments from the base (compartment 1) to the mouth of the gland (compartment 4+5) and the percentage of labeled cells for each compartment was calculated in order to evaluate the height of its distribution. Paired or unpaired t-test was used as appropriate.

RESULTS: one or more metachronous adenomas were found in 21 patients, while no recurrence was observed in 34 (tLI=12.7 ± 0.9 vs 10.4 ± 0.6, P=0.08, respectively). A statistically significant difference was observed in labeling index for the compartment 4+5 (5.0 ± 0.6 vs 2.2 ± 0.4, P=0.002, relapsing vs non-relapsing). Labeling indexes did not change over time in the group with repeated kinetic measures (tLI=10.0 ± 0.8 vs 10.6 ± 0.9; LI for comp. 4+5= 2.8 ± 0.6 vs 2.7 ± 0.7, 1st vs 2nd kinetic evaluation) both in relapsing and in non-relapsing patients.

CONCLUSIONS: 1)the unregulated compartimentalization of replicating cells (upward shift of the proliferative zone), which is considered a reliable marker for colon cancer risk, could be included among the criteria indicated to establish the surveillance intervals in patients with adenomas. 2)The reproducibility of this proliferative abnormality over a long period of time is reassuring.

T181

STABILITY OF K-RAS MUTATIONS THROUGHOUT THE NATURAL HISTORY OF HUMAN COLORECTAL CANCER

L Losi, J Benhattar, L Roncucci, M Ponz de Leon, and J Costa
Istituto di Anatomia Patologica and Istituto di Patologia Medica, University of Modena, via del Pozzo 71, 41100 Modena, Italy; Institut Universitaire de Pathologie, Lausanne, Switzerland

We have used a rapid, nonradioactive and sensitive method for the identification of K-ras mutations in archival tissues of colorectal carcinomas. Our strategy to detect single base substitutions in the K-ras gene was based on mutation-specific amplification - or allele-specific polymerase chain reaction - exploiting the lack of 3'-5' proofreading of Taq-polymerase enzyme. The purpose of the study was to determine whether or not K-ras mutation provides, when present, a tumor marker throughout the natural history of the disease. We have studied 35 patients who developed metastases or local recurrences. In 71% of these patients a ras mutation in codons 12 or 13 was observed in the primary tumor. For each of these cases an identical ras mutation was found in the DNA from the local or distant recurrence. In the 29% of cases where no ras mutation was observed in the primary tumor, no newly acquired ras mutation appeared in the recurrent tumor. The time interval between primary tumors and recurrences varied from 3 to 60 months. Our results indicate that K-ras mutation provides a stable tumor marker throughout the natural history of colorectal cancer.

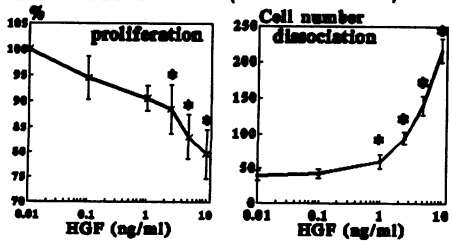
T182

HEPATOCYTE GROWTH FACTOR INHIBITS PROLIFERATION BUT INCREASES SPREADING OF COLON CANCER CELLS

WG JIANG AND MCA PUNTIS
University Department of Surgery, University of Wales College of Medicine, Cardiff

Hepatocyte growth factor(HGF), also known as scatter factor is a newly defined cytokine which has major effects on hepatocyte proliferation in primary culture. We studied the effects of HGF colon cancer cells.

A human colon cancer cell line HT115 was used. Cells were cultured with or without HGF at different concentrations for 3 days and proliferation was quantified by using a dimethyl-thiazol-di-phenyl-tetrazolium bromide assay. Cell spreading was assayed using colony scattering and microcarrier dissociation assay. Results were shown as percentage of control (proliferation) or number of cells dissociated from microcarrier beads(dissociation).



* p<0.05 vs control cultures

HGF showed a dose dependent inhibition of HT115 proliferation but also a dose-dependent dissociations of the cells. The colony scattering was observed with HGF over 1.0ng/ml.

We conclude that hepatocyte growth factor has bi-directional effects on the colon cancer cell-HT115, namely both inhibition of proliferation and causing increased spreading.

T183

ASPARTIC PROTEINASES AND GASTRIN IN THE DIAGNOSIS OF GASTRIC CANCER.

F.Farinati, M.Plebani, F.Di Mario, F.Valiante, G.Della Libera, MC Fanton, M. De Boni, R.Cielo, R.Naccarato, Catt. Mal. App. Dig., Ist. Med. Lab., Padua University and OC. Feltre, Italy.

Several studies in the past decade have focused on the role of aspartic proteinases and gastrin in the diagnosis of gastric cancer and promising results have been obtained with the determination of pepsinogen A (PGA) and PGA/pepsinogen C ratio (PGA/PGC). We studied 276 patients undergoing endoscopy for dyspepsia: 40 had gastric ulcer (GU), 25 duodenal ulcer (DU), 84 chronic atrophic gastritis (CAG), 23 epithelial dysplasia (ED), 51 gastric cancer (GC) and 53 lacking significant endoscopic or hystologic changes, defined as "controls"(CO). After endoscopy, serum samples were obtained upon the patients' informed consent. Serum level of PGA, PGC and gastrin (G) were determined by RIA methods; we then calculated PGA/PGC ratio and a new index number (PGAxG). Results (means):

DGN.	No	PGA	PGC	G	PGA/PGC	PGAxG
CO	53	78.7	20.4	44.8	6.4	3248
DU	25	57.9	51.5	68.8	1.2	4288
GU	40	81.2	45.5	71.2	3.0	5898
GCA	84	96.5	31.7	85.2	4.2	5860
ED	23	80.6	27.6	39.0	4.0	3791
GC	51	20.3	18.8	47.1	1.9	773

PGA and PGA/PGC levels were significantly lower in gastric cancer pts ($p<0.001$). The index number PGAxG was also sharply reduced in gastric cancer pts ($p<0.001$):using an arbitrarily chosen cut-off (1100), this marker showed higher sensitivity (92%), specificity (94%) and overall accuracy (72%, by Youden J test) than PGA or PGA/PGC (sensitivity 70% and 68%, specificity 93% and 53% respectively). If these results will be confirmed in populations at different risk for gastric cancer, PGAXG could become an useful screening tool for the tumor.

T184

THE MANAGEMENT AND OUTCOME FOR EARLY GASTRIC CANCER (EGC); THE WEST MIDLANDS EXPERIENCE.

M C Winslet, Y Mohsen, M Hallissey, J W L Fielding
Department of Surgery, Queen Elizabeth Hospital, Birmingham.

Between 1985 and 1991, twenty one patients (m = 13, f = 8, median age 73 (56-86)) have presented with EGC. All were symptomatic. Initial endoscopy was diagnostic in 10. Repeat endoscopy (median = 2 (2 -5)) was associated with type IIb antral lesions. The lesion (type I = 4, type II = 13, type III = 4) was situated in the antrum = 12, body = 7 and cardia = 2. The median maximal diameter was 2(0.3 - 6.0) cm and was unrelated to site, type, depth of invasion or stage (I = 15, III = 5, unknown = 1).

Twenty patients underwent gastrectomy (R1 = 13 (total = 6, Partial = 7), R2 = 7 (total = 3, partial = 4)). Histology revealed adenocarcinoma alone = 6, plus carcinoid = 1, plus lymphoma = 1, lymphoma alone = 2. Two patients died (pancreatitis = 1, MI = 1). Actuarial survival = 90% at median follow up of 29 months. One patient underwent endoscopic resection and died 24 months later of metachronous disease.

In this series the good prognosis for EGC was unrelated to site, size, type, degree of differentiation stage or type of resection even though an R2 resection is theorectically desirable. In view of the survival rates following resection, endoscopic management should be confined to the poor risk patient or those refusing surgery.

T185

ORNITHINE DECARBOXYLASE ACTIVITY IN CACO-2 CELLS: EFFECT OF NORMAL AND POLYPOSIS BILE.

SE Patchett, L Trzeciak, EM Alstead, K Nugent, RKS Phillips, MJG Farthing. St Bartholomew's & St Mark's Hospitals, London, U.K.

The clustering of duodenal adenomas and cancers around the ampulla of Vater in patients with familial adenomatous polyposis (FAP) suggests that bile is important in the development of these tumours. Several reports indicate that this may be due to the greater carcinogenic potential of bile from polyposis patients. The aim of this study was to examine the effect of bile from polyposis patients (n=5) and from control patients (n=5) on the induction of ornithine decarboxylase (ODC) activity in a colorectal cancer cell line (Caco-2). Caco-2 cells were incubated for 4 hours in medium with and without FAP bile or normal bile at a concentration of 1:500(vol/vol). Gastrin (50μg/ml), a known inducer of ODC in Caco-2 cells was used as a positive control. Following incubation, medium was decanted and cells scraped and sonicated. ODC activity was measured using a [^{14}C]-ornithine bioassay and results expressed in pmol/mg protein/h.

Results

	n	Median	Range
Caco-2 control	12	87	25-121
Caco-2 + normal bile	12	243	55-532
Caco-2 + FAP bile	12	169	97-629
Caco-2 + gastrin	12	440	350-665

FAP bile and normal bile significantly increase ODC activity when compared to control ($p<0.02$). However, no significant difference between FAP bile and normal bile was observed ($p=0.4$). Induction of ODC with bile was significantly less than that observed with gastrin ($p<0.01$). These results suggest that the ability of bile to stimulate polyamine production is not further enhanced by bile from polyposis patients.

T186

MANIPULATION OF THE RETICULOENDOTHELIAL SYSTEM:- EFFECTS ON THE GROWTH OF LIVER TUMOUR.

N. Davies, H. Kynaston, J. Yates, S.A. Jenkins, B.A. Taylor.

University of Liverpool Dept of Surgery, Liverpool, U.K.

The prognosis for patients with liver metastases remains poor. We have developed an animal model of liver metastases using a cell line derived from a colonic carcinoma. Using this model we have shown that Octreotide, a long acting somatostatin analogue is effective in inhibiting tumour growth. Octreotide is a potent stimulator of hepatic reticuloendothelial (RES) activity and this may be the mechanism of action. Gadolinium chloride (GAD) is a well known RES toxin, which effectively blocks hepatic RES activity. We have used GAD in combination with octreotide to assess the effect of RES blockade.

Twenty-four BDIX rats received an IV injection of GAD (5mg\Kg), and 24 animals an equivalent volume of saline. All animals then received an intraportal injection of $1x10^7$ K12\Tr cells. Half of the GAD treated rats received octreotide (2ug sc bd) for 4 weeks, and the remainder saline. At 4 weeks the amount of liver tumour present in each group was determined by % hepatic replacement: saline and saline(control) median 17.5% (range 5.7-24.2), saline and Octreotide 0.6% (0-2.5), GAD and saline 42% (21.2-68) and GAD and Octreotide 11.2% (1.9-32.4). These results show that RES blockade with GAD significantly (Mann Whitney U $p<0.01$) increases tumour growth compared to controls. Octreotide significantly reduces tumour growth but is more effective in the absence of GAD. These observations suggest that 1) RES activity is important in the growth of liver metastases and 2) the efficacy of octreotide in inhibiting the growth of hepatic tumour may be partly dependant on a functioning RES system, but other mechanisms are also involved.

T187

RANITIDINE BISMUTH CITRATE IN THE PREVENTION OF ASPIRIN – INDUCED GASTRIC MUCOSAL INJURY.

N Hudson, F E Murray, A T Cole, GM Turnbull*, S Lettis*, C J Hawkey.
Dept of Therapeutics, University Hospital, Nottingham NG7 2UH, UK and *Glaxo Group Research Ltd, Greenford, Middx. UK.

The aim of this study was to evaluate the ability of a new compound ranitidine bismuth citrate (RBC) to protect human gastric mucosa from aspirin – induced damage.

METHODS: Twenty – four male volunteers (mean age 22 years SD 2.6) participated in a randomised, double blind, placebo controlled, 3 way crossover trial in which they received 9 doses of placebo, and aspirin 900 mg bd both with or without RBC 800mg bd. Endoscopy was performed prior to each treatment, on day 1 and 5 and the number of gastric haemorrhagic and non haemorrhagic erosions counted. The stomach was washed and aspirated at each endoscopy and gastric microbleeding determined by the orthotolidine reaction.

RESULTS:

	Number of erosions Median* (IQR) Haemorrhagic	Non – haemorrhagic	Microbleeding(ul/10min) Median* (IQR)
Placebo	0 (0 – 2)	0 (0 – 0)	0.36 (– 0.28 – 0.9)
Aspirin	18 (12 – 30)	2 (0 – 4)	11.28 (4.89 – 20.7)
Aspirin + RBC	0 (0 – 1)	0 (0 – 3)	0.38 (– 0.29 – 0.79)

Aspirin vs aspirin + RBC $p<0.001$, $p=0.057$, $p=0.005$
* change from baseline endoscopy

CONCLUSIONS: RBC confers substantial mucosal protection against aspirin – induced gastric mucosal damage.

T188

ULTRASOUND IS A USEFUL INITIAL INVESTIGATION IN DYSPEPSIA

MA Mendall, AE Joseph, S Saverymuttu, TC Northfield and JD Maxwell.
St Georges Hospital and Medical School, London.

Abdominal ultrasound(US) can detect upper gastrointestinal pathology relevant to dyspepsia.We have examined its efficacy as an initial screening investigation.

Subjects:120 new subjects with dyspepsia (68F, mean age 46, range 16-95) had abdominal US performed on the same day prior to OGD. .

Results:OGD discovered 6 cases of oesophagitis, 3 gastric ulcers (GU), 2 gastric carcinomas (CA), and 9 duodenal ulcers (DU). US correctly identified all CAs and GUs and 2 DUs. One DUwas identified as an antral ulcer, 2 as duodenal thickening, 2 as antral thickening, one was normal on ultrasound and in one there was inadequate visualisation. 2 cases of oesophagitis were missed. OGD was said to be required if any gastro-duodenal mucosal abnormality was detected by US, 54/120 subjects(45%) of endoscopies would have been saved and 94% of pathology picked up (only one DU missed). In addition, 13 cases of gallstones, 4 cases of possible pancreatitis, and 1 renal calculus were detected on ultrasound (only one of whom had significant pathology on OGD).

Conclusion US contributed to the diagnosis of dyspepsia in 18/120 (15%) of cases (12 normal on OGD) and detected additional pathology (8 fatty livers, 1 ovarian cyst, 1 prostatic tumor, and 2 cases of renal cysts). US can accurately diagnose gastric lesions, but is less reliable at detecting DU. It can however detect duodenal pathology from duodenal wall thickening and/or associated gastric pathology. It may be a more rational and cost effective initial investigation in dyspepsia.