studies in humans, and a study in patients is ongoing. The data support observations of improved efficacy and reduced cardiotoxicity using this method of drug delivery, and suggest that in clinical use in metastatic breast cancer patients the liposomal formulation will provide an enhanced therapeutic index compared with conventional doxorubicin.

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Detection and characterisation of novel biliary metabolites of the anticancer agent ifosfamide using in-vivo and analytical ^{31}P MRS and mass spectrometry

G.S. Payne[1], A.S.K. Dzik-Jurasz[1], L. Mancini[1], B. Nutley[2], F. Raynaud[2], M.O. Leach[1]. [1] *Royal Marsden Hosp & Institute of Cancer Research, Cancer Research UK Clin Magnetic Resonance Group, SUTTON, United Kingdom;* [2] *The Institute of Cancer Research, Cancer Research UK Centre for Cancer Therapeutics, SUTTON, United Kingdom*

Introduction: Many drugs undergo biliary excretion, potentially affecting pharmacokinetics and toxicology. Conventional methods to study this are highly invasive. Here biliary excretion of metabolites of the alkylating agent Ifosfamide (IF) is demonstrated using *in vivo* ^{31}P Magnetic Resonance Spectroscopy (MRS). High resolution ^{31}P-MRS and analytical mass spectrometry enabled provisional assignment of the major biliary metabolite to the glutathione conjugate of IF. The conjugate represents a previously unreported metabolite of IF.

In vivo studies: Ten male Dunkin-Hartley guinea pigs (900 ± 20 g) were cannulated, anaesthetised and placed prone over a 5cm ^{1}H/^{31}P coil system[1] in a 1.5T Siemens Vision MR scanner. A peak at the IF frequency appears within 20 minutes of administration of 500mg/kg IF (Fig 1). Localised data (Fig 2) show that IF signal arises from the liver and gall bladder.

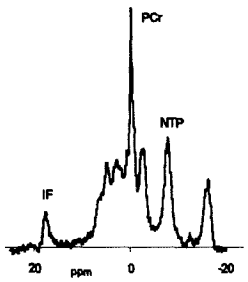

Fig. 1. Unlocalised ^{31}P MRS in vivo after 500 mg/kg IF

High Field ^{31}P MRS Studies of Bile: Spectra of extracted bile were acquired at 11.74 T (Fig 3). Spiking identified Peak 5 as IF. Published data from measurements in urine[2] suggest Peak 2 is carboxy IF, while Peak 3 is 2-dechloroethyl IF or 2,3-dechloroethyl IF. Peak 4 has not previously been reported. If gall bladder volume is 4 cm^3 then approximately 1.9% of injected IF is present as IF and its metabolites in the bile.

Identification of the 16.02 ppm peak using Liquid Chromatography Mass Spectrometry (LCMS): LC analytes were ionised and their masses measured using an ion trap mass spectrometer. The most intense peak detected not present in control bile had a molecular weight of 531, consistent with formation of a conjugate of IF where one Cl atom has been replaced with GSH. Comparative MSMS fragmentation of GSH, IF and the putative GS-IF conjugate showed patterns consistent with this.

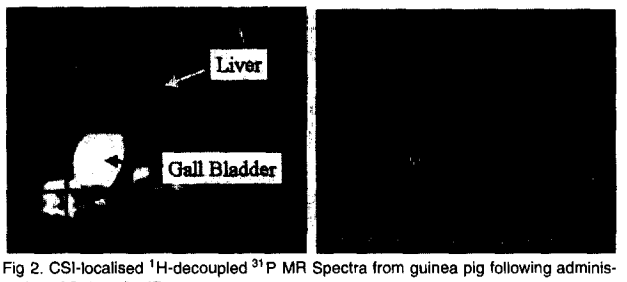

Fig 2. CSI-localised ^{1}H-decoupled ^{31}P MR Spectra from guinea pig following administration of 500 mg/kg IF

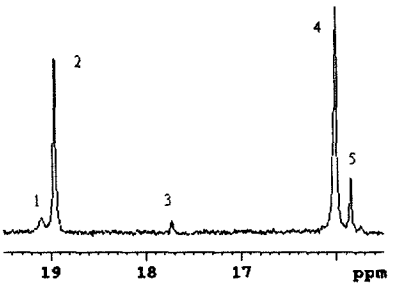

Fig 3. ^{31}P MR Spectrum from Bile at 11.74 T

Table 1. Metabolite concentrations in bile (N=10; mean ± s.d.)

Peak	ppm	Conc (mM)
1	19.09	0.49 ± 0.25
2	18.96	2.04 ± 1.04
3	17.74	0.16 ± 0.07
4	16.02	4.05 ± 2.38
5	15.86	1.19 ± 1.47

Conclusions: ^{31}P MRS signals *in vivo* of IF and its metabolites arise predominantly from liver and gall bladder. Biliary excretion of IF or its metabolites has not previously been reported. High-resolution ^{31}P MRS and mass spectrometry show the main metabolite present to be GSH conjugate of IF. These or other biliary metabolites may be implicated in previously described oxazophosphorine related cholecystitis[3-5].

Acknowledgements: This work was funded by Cancer Research UK (C1060/A808/G7643)

References

[1] DWJ Klomp. Magn. Reson. Imag. 19: 755 (2001).
[2] R Martino. J Pharmacol. Exp. Ther. 260: 1133 (1992).
[3] CR Swanepoel Br Med J. 286: 251.
[4] C Collins. Cancer 63: 228 (1989)
[5] Kuttah Ann Oncol. 6: 302 (1995).

Paediatric oncology

717 ORAL

Risk adapted treatment for childhood hepatoblastoma (HB): final report of the second study of the International Society of Paediatric Oncology' SIOPEL 2

G. Perilongo[1], E. Shafford[2], L. Brugieres[3], P. Brook[4], R. Maibach[5], P. Czauderna[6], G. Mackinlay[7], D. Aroson[8], M. Childs[9], J. Plaschkes[10]. [1] *Division of Haematology-Oncology, Department of Pediatrics, Padova, Italy;* [2] *Pediatric Oncology Unit, St. Bartholomew's Hospital, London, United Kingdom;* [3] *Service d'Oncologie Pediatrique, Institut Gustave Roussy, Paris, France;* [4] *Paediatric Oncology Unit, The Hospital for Sick Children, London, United Kingdom;* [6] *Department of Pediatric Surgery, Medical University, Gdansk, Poland;* [7] *Pediatric Surgical Unit, Royal Hospital for Sick Children, Edinburg, United Kingdom;* [8] *Pediatric Surgical Unit, AMC, Amsterdam, The Netherlands;* [10] *Pediatric Surgical Unit, Children's Hospital, Berne, Switzerland*

Background: SIOPEL2 was a co-operative, international pilot study aiming to test effectiveness - in terms of response, resection rate (RR, RsR), progression-free (PFS), overall survival (OS) - and toxicity of two chemotherapy regimens, incorporated into a therapeutic strategy based on

pre-operative chemotherapy (CT), for so called standard risk (SR) and high-risk (HR) HB, respectively. SR-HB was defined as tumour confined to the liver, involving at the most 3 hepatic sectors, and HR-HB as tumour extending to all 4 sectors and/or with intra-abdominal and/or distant extrahepatic disease.

Material and methods: SR-HB patients were treated with Cisplatin (CDDP) alone (80mg/m2 in 24hours continuous infusion -c.i.-) every 14 days X four, delayed surgery, then two more courses of CDDP. HR-HB patients were given CDDP(as above) alternating every 14 days with carboplatin (500mg/m2) and doxorubicin (60mg/m2 48hour c.i.). Three CDDP and four carboplatin/doxorubicin were given pre surgery and two carboplatin/doxorubicin and one CDDP post operatively.

Results: 77 LR and 58 HR-HB, registered from 10/1995 to 5/1998 are evaluable. 67 LR and all HR-HB were treated according to protocol. The epidemiological patients characteristics were as expected. Treatment outcome: positive RR RsR* 3-years PFS 3-years OS SR-HB 90% (80-96%) 96% (87-99%) 0.89 (0.11) 0.91 (0.07) HR-HB 78% (65-87%) 67% (54-79%) 0.48 (0.13) 0.53 (0.13) *Including liver transplantation. Time interval between courses, dose reduction, hospitalisation; organ and haematological toxicity analyses supported the finding of treatment feasibility (data not shown). No toxic deaths were reported but 2 HR and 2 SR died post-operatively.

Conclusions: Single agent CDDP and surgery seem to be effective for SR-HB treatment; despite therapy intensification, the survival data of HR-HB are only on the 50% range; short-term toxicity of SIOPEL2 regimens is acceptable. The question of CDDP-alone effectiveness is presently addressed by a prospective controlled trial (SIOPEL3)

718 ORAL

Treatment results in high risk hepatoblastoma: analysis of prognostic factors. Results from SIOPEL 2 and 3 trials.

J. Zsiros[1,2], R. Maibach[2], G. Perilongo[2], P. Brock[2], P. Czauderna[2], L. Brugieres[2], G. MacKinlay[2], A. Zimmermann[2], D. Roebuck[2], J. Plaschkes[2]. [1] *Academic Medical Center, Pediatric Oncology, Amsterdam, The Netherlands;* [2] *Childhood Liver Tumour Strategy Group, of the International Society of Pediatric Oncology (SIOPEL)*

Background: HR-HB patients are a heterogeneous group comprising of at least 4 subgroups: patients with local disease involving all four liver sectors (PRETEXT IV), pts. with extra-hepatic intra-abdominal disease, pts. with distant metastases and pts. with low AFP (<100 ug/L). The SIOPEL 2 and 3 international trials were conducted consecutively to test efficacy and toxicity of a new chemotherapy regimen given for these patients pre- and postoperatively.

Material and methods: Patients were given Cisplatin (80mg/m^2/24hour) alternating every 14 days with Carboplatin (500mg/m^2) and Doxorubicin (60mg/m^2/48 hour). In SIOPEL 3 four Cisplatin and three Carbo/Doxo courses were given pre-surgery and 1 and 2, respectively, post-operatively. In Siopel 2 only 3 Cisplatin courses were administered pre-surgery.

Results: Between 1994 and 2001, the SIOPEL 2 and 3 trials included 131 evaluable HR-HB patients from 21 different countries (60% male, age median 19 months range 0-14 years). Fourty-seven patients had PRETEXT IV disease, 38 had metastases and 20 both. Twenty patients had AFP<100ug/L. Partial response rate to pre-operative chemotherapy was 70%. Fifty-six patients (43%) achieved a complete macroscopic resection by partial hepatectomy. Eighteen children had complete hepatectomy followed by orthotopic liver transplantation. At 1.5 years the EFS was 54% (95%CI:45%-63%) and the OS was 62% (95%CI:53%-71%). AFP<100ug/L was associated with significantly shorter OS and EFS (p<0.0001). This is in line with a significantly lower response rate to pre-surgery chemotherapy in such patients. Metastatic disease was borderline associated with worse OS (p=0.05). No other patient characteristics were identified as prognostic factors.

Conclusions: Overall treatment results in HR-HB remain un satisfactory. The failure pattern suggests that patients could benefit from an intensified pre-operative chemotherapy regime. Chemotherapy for low AFP patients should be revised. A new treatment strategy for HR-HB patients with intensified use of Cisplatin is in preparation (SIOPEL 4).

719 ORAL

Hepatocellular carcinoma in children - results of the second prospective study of the international Society of Paediatric Oncology (SIOP) - siopel-2.

P. Czauderna[1], R. Maibach[2], J. Pritchard[2], D. Aronson[2], G. MacKinlay[2], E. Shafford[2], A. Zimmerman[2], J. Plaschkes[2], J.B. Otte[2], G. Perilongo[2]. [1] *Insurance Hospital Riyadh, Surgery, Riyadh, Saudi Arabia;* [2] *SIOPEL International Paediatric Liver Tumors Strategy Group, Leicester, UK*

Introduction: Hepatocellular carcinoma (HCC) is the second most common pediatric malignant liver neoplasm (after hepatoblastoma - HB).

Objectives: To collect information on: biology, patients' characteristics, outcome and prognosis of pediatric HCC and compare it with HB experience.

Material: Twenty one patients diagnosed with hepatocellular carcinoma (HCC) were registered in the SIOPEL 2 study from 03.1994 to 05.1998 (17 were further analyzed). Metastases at diagnosis occurred in 18% children. Extrahepatic tumor extension and/or vascular invasion were found in 35% of patients. Multifocal tumors prevailed (53%). One patient died 17 days after diagnosis from massive GI bleeding, and never received treatment. Thirteen of the 16 treated patients received preoperative chemotherapy (SuperPLADO cisplatin / carboplatin and doxorubicin).

Results: Partial response to preop.CHT was observed in 6/13 cases (46%). Tumor resection was achieved in 8 patients (47%) (including 1 liver transplantation). Three of them underwent primary tumor excision. Six of the 8 operated pts received between 2 and 10 courses of postoperative chemotherapy. Nine cases (53%) never became operable. One patient was lost to follow-up just before planned surgery. Four of the resected patients were alive at a median follow up time of 53 months (35 to 73). Twelve pts. died due to progressive disease, one from surgical complications. The overall treatment results of HCC patients remain extremely poor (22% survival). These results show no improvement over the previous SIOPEL 1 study, in which the overall survival at 5 yrs was 28%, while event free survival was 17%.

Conclusions: 1. Survival for pediatric hepatocellular carcinoma patients remains significantly inferior to that for hepatoblastoma. Complete tumor excision remains the only realistic chance of cure. 2. Intensification of standard chemotherapy has not improved the patients' prognosis. 3. A new treatment approach is needed to increase HCC cure rate.

720 ORAL

Indefinite radiotherapy of anaplastic ependymomas and supratentorial PNET in babies and infants: results of the German HIT-SKK 87 and 92 trials.

B. Timmermann[1,2], R.D. Kortmann[1], J. Kühl[3], S. Rutkowski[3], C. Meisner[4], M. Bamberg[1]. [1] *University of Tübingen, Radiooncology, Tübingen, Germany;* [2] *Paul-Scherrer-Institute, Radiation Medicin, Villigen-PSI, Switzerland;* [3] *University of Würzburg, Children's Hospital, Würzburg, Germany;* [4] *University of Tübingen, Medical Information Processing, Tübingen, Germany*

Background: For infants and babies intensive chemotherapy was introduced in trials to delay or omit radiotherapy. We evaluate the outcome of infants and babies with supratentorial (st) PNET and anaplastic ependymoma after intensive postoperative chemotherapy and indefinite radiotherapy, and we identify factors predictive for survival.

Materials and methods: Since 1987 in Germany and Austria infants and babies with malignant brain tumours were enrolled in the HIT-SKK 87 trial. After surgery low risk patients received a maintenance chemotherapy consisting of PROC/VCR, MTX/VCR. In high risk patients PROC, IFO/VP-16, MTX, DDP/Ara-C was followed by maintenance chemotherapy until delayed radiotherapy. In the following HIT-SKK 92 the agents MTX/VCR/CPM, MTX/VCR, MTX/Carbo/VP-16 were applied. In children with complete response the therapy was finished. In case of tumour persistence salvage chemotherapy was added. Radiotherapy was administered only in non-responders.

Results: All children received chemotherapy. 29 children with st PNET were eligible (age 3.0 – 37.0 months); 3-years OS and PFS rates were 17.2% and 14.9%, respectively. The only significant predictive factor for both OS and PFS was the administration of radiotherapy. 34 patients with ependymomas were analysed (age 1.0-33.0 months); 3-years OS and PFS rates were 55.9% and 27.3% respectively. All failures occurred with local involvement. Positive impact on survival was observed in higher age, M-stage, complete resection, and applied radiotherapy.

Conclusions: In younger children delayed radiotherapy is reasonable to spare late effects. Omission of radiotherapy jeopardizes survival, even if intensive chemotherapy has been applied. Outcome of infants and babies