distributed throughout acini. Interestingly, hE2 immunoreactivity is also associated with small particles found in the acinar lumen, which is in agreement with the presence of small membrane particles containing Prominin-1 in seminal plasma. The analysis of several prostate cancer samples revealed a down-regulation of the hE2 immunoreactivity in the tumor region, independent of their Gleason score (5–10). In those tissues however we found that the hE2 immunoreactivity is up-regulated in luminal cells in the vicinity of the tumor, especially in the areas of inflammation or intensive proliferation of basal cells.
Conclusions: These data showed that the overall expression of Prominin-1 in prostate is not limited to the basal stem cells as assumed from a previous study with mAb AC133, but only the Prominin-1 molecule carrying the AC133 epitope appears to label these stem cells. With regard to the prostate diseases, our pilot screen shows that Prominin-1, as detected by hE2 immunoreactivity, is down- and up-regulated in the tumor and inflammatory regions, respectively. Further studies are needed to determine the potential application of Prominin-1–containing particles as a novel biomarker for human prostate cancer.

879 POSTER

Value of the innovated agarose cell block technique in improving the diagnostic sensitivity of urine cytology in cancer bladder cases

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Background: Proper handling and processing of urine sample have great impact on improving diagnostic sensitivity. Agarose cell block (ACB) technique is an innovated technique by Mansy (2004) based on the use of melted agarose gel as an embedding media, for the processing of the sediment of urine sample in block manner.
Objective: The aim of this work is to investigate the validity of ACB technique in processing urine samples simultaneously for light and electron microscopic (EM) examination with the prospect to enhance the quality of diagnosis.
Material and methods: The material of this study consisted of 45 voided urine samples collected from 30 patients (Pt) with bladder carcinoma, 14 Pt with non specific cystitis and one Pt who underwent transurethral resection of the primary tumour (TUR-T) followed by adjuvant immunotherapy with BCG. The sediment of the collected urine from each case was processed for the performance of Papanicolaou (Pap) stained smear and the preparation of ACB. The solidified agarose block was divided longitudinally into two halves. One half was processed for paraffin, hematoxylin and eosin (H&E) stained sections and the other half for EM examination.
Results: Significant increase in the number of sedimented urothelial cells in the ACB paraffin prepared sections versus the corresponding Pap stained smear was noticed. Moreover, the diagnostic sensitivity of urine cytology was improved with the application of ACB technique. Out of the 30 malignant cases confirmed by histopathology of bladder biopsies (two grade I Ta, one grade I T1, two grade II Ta, five grade II T1, eleven grade II T2, two grade III T2, three grade III T3a, four grade III T3b), 70% were diagnosed by Pap stained urine smear versus 90% by ACB paraffin H&E stained sections and 100% by ACB EM processed samples. Furthermore, simultaneous processing of the same sample for light and EM examination allowed proper study, facilitated the discrimination between normal, dysplastic and malignant urothelial cells, the identification of type of malignancy and the accurate diagnosis of controversial cases specially those revealing severe urothelial dysplastic changes and patient under adjuvant immunotherapy after TUR-T.
Conclusion: Thus, ACB technique could be considered a useful technique which helps in increasing the sensitivity of urine cytology and opens a new prospect for cytomorphological study.

880 POSTER

Clonal origin of multifocal renal cell carcinoma as determined by microsatellite analysis

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Purpose: 3% of all carcinomas are renal cell carcinomas. The reported incidence of satellite tumor lesions in renal cell carcinoma (7% to 25%) suggests that there is a risk of local recurrence after nephron sparing surgery. It remains largely unknown whether small satellite tumors show malignant features and whether they are metastases from the primary tumor. Therefore, we determined the clonality of multifocal tumors by molecular genetic analysis.
Materials and methods: A total of 20 multifocal clear cell renal cell carcinomas were investigated by microsatellite analysis using 6 markers for chromosome 3p, namely D3S1560, D3S1289, D3S1766, D3S1300, D3S1566 and D3S1663. Polymerase chain reaction was performed according to standard protocols, followed by gel electrophoresis and automated analysis using an automated DNA sequencer (Li-Cor, Lincoln, Nebraska).
Results: All primary clear cell tumors were characterized by loss of heterozygosity on 3p. Multifocal tumors showed identical microsatellite alterations with at least 2 marker in all cases. 14 out of 20 matched completely in all 6 marker.
Conclusions: Identical loss of heterozygosity detected in different tumors in the same kidney strongly suggest that multifocal clear cell renal cell carcinomas have a common clonal origin in most cases. These findings indicate that satellite tumors are the result of intrarenal metastasis from the primary tumor. The clinical implications of these results need further investigation.

881 POSTER

The value of PTEN expression in smears of prostate cancer; correlation with prognostic factors and disease outcome

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The PTEN (phosphatase and tensin homolog deleted on chromosome 10) tumour suppressor gene is located on chromosome 10q23, a genomic region frequently lost in human cancers. Complete inactivation of the PTEN tumour suppressor gene is extremely common in advanced cancer, including prostate cancer (CaP). The aim of this study was to examine the expression of PTEN protein in prostate carcinoma cell samples and its association with clinicopathological parameters.
Materials and methods: eighty imprint smears were obtained at surgery and studied immunocytochemically using anti-PTEN antibody. Cases were considered positive when granular cytoplasmic staining was seen in all tumour cells, mixed when areas of both positive and negative tumour cell clones were seen, and negative when adjacent benign prostate tissue but not tumour tissue showed positive staining. The PTEN expression pattern was correlated with histopathological findings in the same samples. The results were correlated with postoperative Gleason score, preoperative Serum Prostate Antigen (PSA) and pathological stage.
Results: Thirteen smears (16.2%) of prostate cancer were positive, 50 (62.5%) were mixed, and seventeen (21.3%) were negative. Positive correlation between PTEN expression with Gleason score 7 or higher was observed ($p < 0.0001$). There was also significantly higher PTEN expression in smears from patients with PSA value $\geqslant 10$ ($p = 0.0003$) and poorly differentiated prostate carcinomas with Gleason score >7($p < 0.001$). Relationship was also observed between PTEN expression and disease outcome.
Conclusions: PTEN protein is correlated with pathological parameters of poor prognosis and could be a good marker for biological behaviour of prostate carcinomas.

882 POSTER

Expression Protein Kinase C in prostate hyperplasia and carcinomas in relationship with clinicopathological parameters

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Objective: Protein kinase C (PKC) comprises a family of serine/threonine kinases that plays a key role in the signal transduction pathways. It consists of at least 12 isoforms with different tissue expressions, substrate specificity, and subcellular localization that are related to specialized cell functions, including cell proliferation, differentiation, and apoptosis. Recent evidences prove that PKC isozymes play an important role in the transition from an androgen-dependent to an androgen-independent status The aim of this study was to investigate the PKC expression (PKC alpha and PKC delta) in smears of patients with benign hyperplasia or carcinomas in order to evaluate the malignant potential role of these diseases.
Methods: Sixty imprint smears (30 invasive carcinomas) and (30 hyperplastic prostates) were obtained at surgery and studied immunocytochemically using anti-PKC alpha and delta antibodies. The PKC expression was correlated with histopathological findings in the same samples.

Results: Differences between benign prostatic hyperplasia and prostatic carcinomas was observed in high level of expression of PKC isoforms alpha, and delta ($p < 0.001$). PKC isoforms alpha, and delta were elevated in prostate cancer (97%) and in poorly differentiated carcinomas (80%) and reduced to well differentiated prostatic carcinomas and prostate hiperplasia. The frequency of elevated PKC isoforms alpha, and delta expression was higher in tumours with Gleason score >5 ($p < 0.001$).
Conclusions: These results indicate that both PKC alpha and PKC 98delta may aid in prominence between benign and malignant prostatic diseases.

883 POSTER

Phase I trial of sorafenib (BAY 43–9006) in combination with interferon alpha-2a in patients with unresectable and/or metastatic renal cell carcinoma and malignant melanoma

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Background: Sorafenib (BAY 43–9006) is a novel, oral multi-kinase inhibitor that acts on both the tumour and the vasculature by targeting Raf kinase and the receptor tyrosine kinases VEGFR-2 and PDGFR-β. In Phase II/III trials, sorafenib significantly prolonged progression-free survival versus placebo, and had a favourable safety profile in patients with renal cell carcinoma (RCC). This Phase I, single-centre, open-label study was designed to determine the safety profile and maximum tolerated dose (MTD) of sorafenib in combination with interferon alpha-2a (IFN).
Patients and methods: Patients with metastatic RCC or malignant melanoma who were refractory to standard therapy were enrolled. Following a 2-week period of IFN alone, patients received 28-day cycles of continuous oral sorafenib 200 mg (cohort 1) or 400 mg bid (cohorts 2 and 3), with subcutaneous IFN 6 MIU (cohorts 1 and 2) or 9 MIU tiw (cohort 3). Patients continued on treatment until disease progression, unacceptable toxicity or death. Primary endpoints were the safety profile and MTD of combination therapy. Secondary endpoints included RECIST-evaluated best tumour response, changes in tumour vascularization by Doppler US, and various immunological parameters.
Results: Twelve patients with RCC and one patient with melanoma received treatment in cohorts 1 ($n = 4$), 2 ($n = 3$) and 3 ($n = 6$). Patients' characteristics were: median age 59 years (range 25–76); ECOG 0:1, 77%:23%; prior systemic anticancer therapy, 92%; prior IFN, 69%; $\geqslant 3$ metastatic sites, 92%. To date, no dose-limiting toxicities have been reported for patients in any cohort. Common grade 1 and 2 drug-related adverse events occurring in 10 evaluable patients during combination treatment were: fatigue (90% of patients); diarrhoea (80%); nausea (50%); dry skin, hand–foot skin reaction, pruritus and anorexia (40% each). One patient in cohort 2 experienced drug-related grade 3 asthenia; however, this decreased to grade 2 in Cycle 2. One patient in cohort 1 withdrew on Day 6 of Cycle 1 due to grade 2 asthenia and anorexia. No deaths have been reported. Of the nine evaluable patients, stable disease was achieved in five RCC and one melanoma patient, with tumour shrinkage in 5/6 clear-cell RCC patients.
Conclusions: This combination was safe and well tolerated. The recommended dose for Phase II trials is continuous oral sorafenib 400 mg bid and IFN 9 MIU tiw. Complete data will be updated at the meeting.

884 POSTER

The role of amifostine on late normal tissue damage induced by pelvic radiotherapy with concomitant gemcitabine: in vitro study

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Background: In this invitro study; we aimed to assay the role of radioprotective effect of amifostine on late normal tissue damage induced by pelvic radiotherapy with concomitant gemcitabine, by histopatologic and quantitative methods.
Material and methods: Fifty-six male Wistar albino rats were randomly divided into seven experimental groups (8 rats per group) (I) gemcitabine (25 mg/kg) alone (GM) (II) radiation+gemcitabine (25 mg/kg) (RT+GM) (III) radiation+gemcitabine (25 mg/kg)+amifostine (200 mg/kg) (RT+GM+AF) (IV) radiation+amifostine (200 mg/kg) (RT+AF) (V) sham radiation (S) (VI) amifostine (200 mg/kg) alone (AF) (VII) radiation alone (GM). Irradiation was given to the pelvic region with a dose of 20 Gy/5 fractions/5 days with Co60 gamma rays. A single dose of AF (200 mg/kg) was given intraperitoneally 30 minutes before the first day of irradiation. A single dose of GM (25 mg/kg) was injected intraperitoneally 24 hr before the first day of the radiotherapy. TGF-beta levels in plasma were assessed before the beginning of the treatment and 1 week after the treatment. All animals were sacrificed at the end of 4th month. Pathological examination was performed and the tissue collagen content was measured for bladder and rectal tissues.
Results: 52 animals that were alive at the end of the follow up period were analyzed. 35 animals (68.6%) revealed grade I-III late effect in histopathological examination and 5 of them were severe. We observed grade III colitis in 1, bladder fibrosis in 4 animals. In histopathological evaluation, bladder fibrosis and colitis was seen significantly higher in RT+GM groups than the other groups respectively ($p = 0.0027$, $p = 0.0005$). In groups that AF was used, collagen content of bladder and rectal tissue was lower than the other groups ($p = 0.02$ and $p = 0.04$). Although, the collagen contents of bladder and rectal tissues were lower in RT+GM+AF group than RT+GM group, this difference was not significant. The difference between pre-RT and post-RT levels of TGF-B1 was not significant in all groups.
Conclusion: By histopathological and quantitative methods we determined that, addition of amifostine to the pelvic radiotherapy with concomitant gemcitabine can reduce the late bladder and rectal damage. We couldn't show the relationship between plasma TGF-B levels and histopathological radiation injury in pelvic tissues.

Publication
Genitourinary cancer

885 PUBLICATION

Selective organ preservation in muscle-invasive TCC of the bladder: a biological approach

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Introduction: 1400 new cases of T2/T3 TCC bladder are diagnosed in the UK annually. Cystectomy alone is associated with 20–30% local failure rate and raises QoL issues, as reconstruction may not be available/ possible. Neo-adjuvant chemotherapy (neo-CT) has a 5% 5 year absolute survival benefit. (ABC Meta-analysis Collaboration 2003 Lancet;361:1927–34) and pathological response to treatment is associated with outcome (Splinter et al. 1992 J Urol;147:606–8). A pilot study of selective bladder preservation, giving radiation to patients with pathological down-staging after neo-CT is discussed.
Materials and Methods: Patients with T2/T3 TCC bladder received 3 cycles of neo-CT (accelerated MVAC) followed by rigid cystoscopy 2 weeks later. Patients down-staged to $\leqslant$pT1 received radical radiotherapy (64 Gy/32 fractions). Cystectomy was reserved for poor pathological responders ($\geqslant$pT2). Response and toxicity were evaluated.
Results: 24 patients were treated (2000–2004). pCR were seen in 12/25 patients (48%), and pTa/pT1 in a further 7/25 (28%). 21 (88%) patients underwent bladder preservation. After a median of 18 months follow up (8–34) 6 patients have died (metastatic bladder cancer 2, other causes 4) and 1 has required salvage cystectomy for invasive recurrence. 16 (67%) are alive in remission (3 after treatment for superficial disease). Of surviving patients; 15(83%) are alive with an intact bladder. Toxicity has been low with episode of grade 4 bowel toxicity reported.
Conclusion: Selective bladder preservation in patients with favourable pathological response to neo-CT represents a realistic option to cystectomy and merits further evaluation in a multi-centre study.

886 PUBLICATION

Impact of post-implant evaluation by different slice intervals using CT-based dosimetry in prostate brachytherapy

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Purpose: To compare the CT-based post-implant dosimetry by 1 mm slice intervals versus 5 mm slice intervals.
Material and Method: Twenty-one patients treated with permanent prostate brachytherapy were selected for this study.
The CT volume was based on each slice intervals calculated from the contours of the prostate on day 0. One radiologist randomly repeated the contouring and evaluation three times for each slice interval at weekly intervals. Post-implant dosimetry was performed and the DVH were calculated to report the reconstructed prostate volume (pvol), prostate