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6(iii): Therapeutic Targets in Cancer

6(iv): Clinical Research in Cancer Immunology

Session Chair
Dr. Mahin Khatami
National Cancer Institute, USA

Session Co-Chair
Dr. Roman Kischel
Micromet AG, Germany

Session Introduction

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Title: Clinical pharmacokinetics of cisplatin in patients with malignant tumor of limb (mal) by Hyperthermic Antiblastic Perfusion (HAP) treatment

Dr. Jianshi Lou, Tianjin Medical University P. R. China

Title: Impaired cytolytic function of natural killer (NK) cells obtained from patients with head and neck cancer can be partially restored by the triggering of toll-like receptor 3 (TLR3) expressed on NK cells

Dr. Miroslaw J. Szczepanski, Poznan University of Medical Sciences, Poland

Title: Targeted and image-guided cancer treatment using Theranostic nanoparticles

Dr. Lily Yang, Emory University, USA

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Targeting superoxide dismutase 1 for chemosensitization of platinum resistant ovarian cancer cells

Mu Wang

Department of Biochemistry and Molecular Biology, Indiana University School of Medicine, USA

Platinum-based chemotherapy, such as cisplatin, is the primary treatment for ovarian cancer. However, drug resistance has become a major impediment to the successful treatment of ovarian cancer. To date, the molecular mechanisms of resistance to platinum-based chemotherapy remain unclear. In our previous study using a proteomic approach, more than 90 proteins showed significant expression changes when two pairs of ovarian cancer cell lines, A2780/A2780-CP (cisplatin-sensitive/cisplatin-resistant) and 2008/2008-C13*5.25 (cisplatin-sensitive/cisplatin-resistant), were compared. Bioinformatics analysis suggested several potential pathways that may be involved in platinum resistance. Among these potential pathways, a redox regulated pathway involving superoxide dismutase 1 (SOD1) was targeted in order to further explore its involvement in drug resistance. Inhibition of SOD1 activity in the resistant cells by either small-molecule inhibitors or siRNA enabled partial reversal of platinum resistance. Our data suggest that targeting SOD1 can potentially lead to sensitization of platinum-resistant ovarian cancer cells, and SOD1 may be used as a therapeutic target for chemosensitization of ovarian cancer.

Biography

Dr. Wang is the Director of Proteomics and Associate Professor of Biochemistry and Molecular Biology at Indiana University School of Medicine. He received his PhD in Bio-organic Chemistry from Washington University in St. Louis, Missouri, USA and was an NIH postdoctoral fellow studying mechanism of DNA repair in mammalian system. He has published more than 60 peer-reviewed articles in biochemistry and proteomics related journals. His own research involves mechanistic study of drug resistance in ovarian cancer and DNA repair mechanisms in mammalian systems in response to genomic stresses. In his recent study in searching for biomarkers of cisplatin resistance in human ovarian cancer using a proteomic approach, he identified multiple pathways that are involved in cisplatin resistance. His preliminary data suggests that SOD1 is a key determinant of drug resistance. Through inhibition of SOD1 activity, the cisplatin resistant ovarian cancer cells can be sensitized. He is in the process of developing a chemosensitizer to overcome platinum resistance in ovarian cancer. Dr. Wang was a recipient of the HUPO (Human Proteome Organization) 2004 Young Investigator Award.

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Unresolved inflammation: Immune dynamics of aging process and tumorigenesis

Mahin Khatami
National Cancer Institute (NCI), National Institutes of Health (Retired)

For over 150 years increasing publications reported on circumstantial association between injuries/inflammation and cancer. However, until recently no data were available on a direct link between inflammation and tumor development. In 1980's, we established experimental models of acute and chronic inflammatory diseases in conjunctival associated lymphoid tissues (CALTs) in guinea pigs by topical application of fluoresceinyl ovalbumin (FLOA) for up to 30 months. Analyses of series of clinical and immunopathological findings demonstrated at least three distinct developmental phases of immune responses: a) Acute phase, involving IgE-Fc receptor aggregation and mast cell degranulation, histamine and prostaglandin release and vascular hyperpermeability; b) Intermediate phase, involving desensitization phenomenon, loss/exhaustion of mast cells function, infiltration of inflammatory cells (e.g., eosinophils) into subepithelium and goblet cells and neovascularization; c) Chronic phase, induction of massive lymphoid hyperplasia, follicular formation with germinal centers, increased swollen goblet cells, increased degranulation of mast cells ('leaky'), increased activity of macrophages, extensive epithelial thickening and thinning, changes in local antibody profiles (IgG1/IgG2 ratios) and angiogenesis.

The results are suggestive of a first evidence for direct association between inflammation and development of tumor-like lesions in lymphoid tissues, extensive changes in adjacent epithelium and angiogenesis. Mast cells are effector cells within innate immunity and play important roles, being 'tumoricidal' in their granulated (mature-resting) status during acute inflammation, while they possess 'tumorigenic' properties when partially granulated ('leaky') under persistent inflammation.

Designs of Clinical Trials and Drug Development: Unresolved inflammation is loss of balance between 'tumoricidal' vs. 'tumorigenic' (pleiotropy or 'Yin' and 'Yang') properties of acute inflammation. Promotion of innate and adaptive immune cells plays key roles in tumor surveillance ability of host tissues. Designs of suitable clinical trials and drug development will be discussed based on a concept that chronic inflammation is a common denominator in the genesis and progression of nearly all age-associated chronic diseases including cancer.

Recent Selected References:

Khatami, M: Developmental phases of inflammation-induced massive lymphoid hyperplasia and extensive changes in epithelium in an experimental model of allergy: Implications for a direct link between inflammation and carcinogenesis. Am. J. Ther. 12: 117-120, 2005.

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IPMN of the pancreas - Evaluation of pathohistological subtypes and clinical outcome

Marius Distler

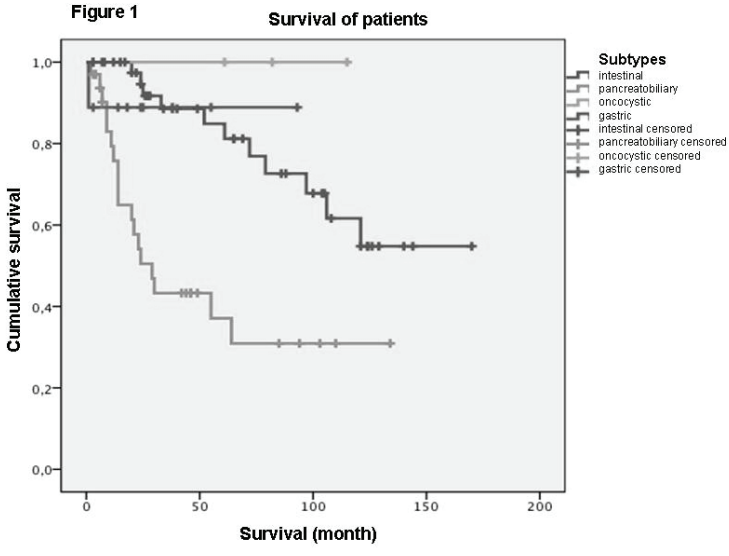
Department of General-, Thoracic- and Vascular Surgery, University hospital Carl Gustav Carus, Germany

In recent years papillary mucinous neoplasms of the pancreas (IPMN) have been increasingly recognized in clinical practice. IPMNs are estimated to have a better prognosis than pancreatic ductal adenocarcinomas. In addition to the different growth types (main duct vs. branch duct), the histological subtypes of IPMN (intestinal, pancreatobiliary, gastric and oncocystic type) became prognostically relevant. These subtypes can be characterized by different expression patterns of MUC using immunohistochemistry. In this study we analyzed the IPMNs of two pancreatic centers regarding to MUC expression and subtypes as well as the clinical outcome.

Over a period of 10 years we revaluated all pancreatic resections due to a cystic tumor in two German university hospitals. Cases with IPMN were screened and subtypes were defined by histopathological analysis including immunohistochemical analysis of MUC ($MUC1^+$, $MUC1^-$, $MUC2^-$, $MUC5AC^+$) expression. Furthermore we determined clinical and follow up data as well as patients outcome.

A total of 128 IPMN were detected. In 98 cases histopathological subtype classification was possible: intestinal type n=45 (46%), pancreatobiliary type n=38 (39%), gastric type n=11 (11%) and oncocystic type n=4 (4%). We performed the following types of resections: pancreatic head resections in 76.4%, left resections in 14.2%, total pancreatectomies in 5.5% and pancreatic segment resections in 4% of the cases. Median survival of intestinal IPMN is significantly better than pancreatobiliary IPMN (60 vs. 21 month) (Figure 1). Clinical data of the IPMN subtypes showed no differences. Common preoperative clinical symptoms were dorsalgia, abdominal pain and obstructive jaundice.

Figure 1



Evaluation of IPMN subtypes supports the postoperative prediction of the patient's prognosis. Therefore, it could lead to improvement in clinical management. Potentially identification of subgroups with the need for adjuvant therapy is possible.

Biography

M. Distler started to study medicine at LMU Munich, Germany and earned his medical degree in 2004 at TU-Dresden, Germany. Till 2004 he works as a general surgeon at the department of general-, thoracic- and vascular surgery at the university hospital of Dresden, Germany. His research activity is focused on carcinogenesis and treatment of pancreatic cancer. He presented and published research results on several national and international meetings or journals.

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Using GlycoExpress for production of highly active antibodies directed against novel and existing targets

Steffen Goletz
Glycotope GmbH, Germany

Glycosylation is the major post-translational modifications of biotherapeutics that depends on the cell line used for production. By establishment of the GlycoExpress Toolbox we have generated a set of glycoengineered human cell lines to optimize the human glycosylation of biotherapeutics. PankoMab-GEX™ is a novel glycooptimized humanized monoclonal antibody produced in GlycoExpress. It recognizes a unique carbohydrate-induced conformational epitope (TA-MUC1). This epitope is tumor-distinctive and is present in the majority of cases of a variety of carcinomas. PankoMab-GEX™ is currently in late Phase I trial. Tumors carrying target molecules like Her2/neu, EGFR, CD20 and others are currently challenged by antibody therapeutics like Herceptin, Erbitux and Rituxan. However, clinical data shows that the success of the therapy depends on the Fc RIIIa allotype present within the treated patient. By Using GlycoExpress existing antibodies can be optimized in respect to manifold improvement of anti-cancer activity enabling clinicians to treat patients carrying the low affinity Fc RIIIa allotype and thus broadening the patient spectra. The activity of the antibodies was improved several hundred fold when measured by means of an ADCC assay. Furthermore the antibodies are improved with respect to half-life elongation and removal of immunogenic components. Two biobetter antibodies are currently in Phase I clinical trials.

Biography

Dr. Steffen Goletz, CEO, CSO and founder of the biotech company GLYCOTOPE, studied biology, biochemistry and molecular biology at the universities in Heidelberg, Kaiserslautern, Manchester (UK) and Berlin and holds a Ph.D. in biochemistry. During his research, Dr. Goletz has focused on glycobiology, tumor immunology, antibody engineering and cellular engineering. As CSO, Steffen is responsible for the development of GLYCOTOPEs product pipeline of glycooptimized biotherapeutics with four products currently in clinical trials.

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BiTE Antibodies for Cancer Therapy

Patrick A. Baeuerle and Roman Kischel
Micromet Inc., USA

Bispecific antibodies can transiently link tumor cells with otherwise inactive cytotoxic T cells in patients for induction of potent redirected lysis of tumor cells. One example is blinatumomab (MT103), a CD19/-CD3-bispecific BiTE for the treatment of human B cell-derived malignancies. Blinatumomab and other BiTE antibodies were shown to activate T cells in a highly conditional manner that is strictly dependent on the presence of target cells. Blinatumomab has commenced a pivotal study for the treatment of adult patients with therapy-refractory acute lymphocytic leukemia (ALL). A phase 2 study in ALL patients has shown an 80% complete molecular response rate at a dose level of 15 micrograms/squaremeter per day. Blinatumomab has also shown high response rates in non-Hodgkin's lymphoma (NHL) patients with follicular and mantle cell lymphoma, and first signs of efficacy in patients with diffuse large B cell lymphoma. Centrally confirmed complete and partial responses according to Cheson criteria were seen in NHL patients treated at a dose of 60 micrograms/squaremeter/day. The presentation will update on the clinical development of blinatumomab in leukemia and lymphoma.

MT110 is a novel BiTE antibody recognizing the pan-carcinoma antigen EpCAM (CD326), which is expressed on a large variety of human adenocarcinoma, and on cancer-initiating or stem cells derived thereof. MT110 is a in phase 1 study with gastrointestinal, lung, breast, prostate, ovarian, and esophageal cancer patients. A murine EpCAM/CD3-specific version of the BiTE antibody, called muS110, has shown a robust therapeutic window in mice with no damage to EpCAM-expressing normal epithelia. A series of new BiTE antibodies for solid tumor treatment are being developed in collaboration with large biopharma partners, including MedImmune/AstraZeneca, Bayer Schering Pharma, Boehringer Ingelheim and Sanfi-aventis.

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Lactobacillus casei ssp.casei could induce the Th1 cytokine production and Natural Killer cells activity in BALB/c mice bearing invasive ductal carcinoma

Mohammad Mehdi Soltan Dallal[1], Mohammad Hossein Yazdi[1], Marzieh Holakuyee[2], Zuhair Mohammad Hassan[3], Abbas Mirshafiey[1] and Mehdi Mahdavi[3]

[1]Faculty of public health, Tehran University of medical science, Iran
[2]Department of immunology, Pasteur Institute of Iran, Tehran, Iran
[3]Faculty of medical sciences, Tarbiat Modares University, Iran

Lactic acid bacteria used as probiotics have ability to modulate immune responses. They have also been shown to affect the immune responses against solid tumors. In the present work, we proposed to study the effects of oral administration of *L.cacesi ssp casei* on the NK cytotoxicity and production of cytokines in spleen cells culture of BALB/c mice bearing invasive ductal carcinoma. Tow groups of female mice as test and control each containing 15 mice were used. 2 weeks before tumour transplantation test mice were orally administered by 0.5 ml of PBS containing 2.7×10^8 CFU/ml of *L.casei*. Administration was followed 3 weeks after transplantation with 3 days interval. Control mice received an equal volume of PBS in a same manner. Results showed that *L. casei* significantly increased the production of IL-12 and IFN-γ and increased the NK cytotoxicity. The growth rate of tumor in the test mice was decreased and survival rate of them significantly raised in comparison to the controls. Our findings suggested that daily intake of *L.casei* can improve the production of IL-12 and IFN-γ and motivate the NK cytotoxicity, but further studies are needed to investigate the other mechanisms of these effects.

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Clinical Pharmacokinetics of Cisplatin in Patients with Malignant Tumor of Limb (MAL) by Hyperthermic Antiblastic Perfusion (HAP) Treatment

Jianshi Lou, Jing Zhu and Zhiqing Cui

Dept. of Pharmacology, Tianjin Medical University, Tianjin 300070, P. R. China

Objective: To study the pharmacokinetics of cisplatin in MAL patients by the treatment of HAP, a quick and sensitive UV spectrometry was used to determine the cisplatin concentrations in plasma.

Methods: The patients were divided into 3 groups. I: The systemic blood cisplatin concentrations (SBCC) were determined after cisplatin (2mg/kg) iv gtt. II: The local and the SBCC were determined during HAP (3mg/kg). 200ml blood was discarded after HAP. The SBCC were determined for 72 hours. III: 200ml blood returned to systemic circulation after HAP. Other performances were same as II.

Results: During HAP the cisplatin concentrations in local blood were 3-15 folds higher than that in systemic blood at the same time. After HAP SBCC of III were close to that of II at the same time. After HAP of 72 hours SBCC was still higher than that of 50% inhibiting cancer concentration in vitro. There was no significant deference in pharmacokinetic parameters within 3 groups. The toxicity in III was not increased.

Conclusion: The method without discarding blood after HAP is confirmed useful in clinic.

Biography

Prof. Jianshi Lou is the director of Dept. of Pharmacology, guided about 10 Ph D students and 40 MM students in his Lab. He has published more than 100 papers in repute journals and Chinese journals. Now he served as Vice-Chairmen of The Tianjin Pharmacological Society; Vice-Chairmen of The Tianjin Pharmaceutical Society; Vice-Chairmen, Branch of Mathematics Pharmacology, Chinese Pharmacological Society.

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Impaired cytolytic function of natural killer (NK) cells obtained from patients with head and neck cancer can be partially restored by the triggering of toll-like receptor 3 (TLR3) expressed on NK cells

Miroslaw J. Szczepanski
Poznan University of Medical Sciences, Poland

Background: Human natural killer (NK) cells play a critical role in innate immunity through their capacity to lyse malignant cells without prior antigen-specific priming. TLRs are expressed on inflammatory cells, including NK cells, and provide protection against infections benefiting the host. NK-cell function is impaired in patients with cancer. TLR3 is expressed on NK cells, but little is known about its role in NK-cell mediated activity in cancers. The aims of the study were: a) to analyze the frequency, phenotype and function of peripheral blood NK cells in patients with head and neck cancers (HNC) and b) to evaluate effects of TLR3 triggering on NK cell phenotype and function in these patients.

Materials and Methods: RT-PCR was used to evaluate the expression of TLR3 in NK cells TLR3 expression in NK cells was studied by RT-PCR. Multiparameter flow cytometry was used to evaluate the frequency of NK cells and expression of NK-cell activating receptors (NKp30, NKp46, NKG2D), CD69, interferon gamma, granzyme B, perforin, on NK cells isolated from the PBMC of normal controls (NC, n=10) and HNC (n=14). Lytic activity of NK cells stimulated or not with poly I:C, a ligand of TLR3, (50µg/mL) -/+ a constant dose of IL-2 (50 IU/mL) for 24h was tested in 51Cr-release assays against K562 targets. NF-kappaB translocation to nuclei and formation of conjugates with K562 cells after triggering of TLR3 was studied by confocal microscopy following immunostaining for a p65 subunit.

Results: Expression of CD69, activating receptors, granzyme B and perforin measured as mean fluorescence intensity (MFI) was significantly lower in NK cells of HNC patients vs NC ($p < 0.05$ for all) and correlated with the decreased function of NK cells (1170 vs. 1890 lytic units). TLR3 triggering on NK cell in HNC induced translocation of NF-kappaB, significantly increased lytic activity function and up-regulated expression of CD69 as well as IFN-gamma. However, it had no effect on the expression of activating receptors.

Conclusion: TLR3 expressed on NK cells is involved in the regulation of NK cell activity, and the impaired function of NK cells in HNC can be partially restored via TLR3 signaling using poly I:C.

Biography

Miroslaw J. Szczepanski, MD PhD graduated from Poznan University of Medical Sciences in Poznan, Poland in 2001 and completed his PhD in head and neck cancer immunology in 2010. He was a postdoctoral fellow at the University of Pittsburgh Cancer Institute from 2006-2009. After finishing his training in Pittsburgh he came back to Poland to complete his residency in Otolaryngology at the Department of Otolaryngology in Poznan. He has published 22 papers in reputed journal and has served as a reviewer for two scientific journals. His research interests are focused on cancer stem cells and on the role of toll-like receptors in head and neck cancer. He is also a principal investigator of two Polish Ministry of Sciences and Higher Education and Foundation for Polish Sciences grants on head and neck cancers.

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Targeted and image-guided cancer treatment using Theranostic nanoparticles

Lily Yang

Department of Surgery and Winship Cancer Institute, Emory University, USA

Recent advances in nanotechnology have opened an exciting frontier in developing and applying novel approaches for the detection and treatment of human cancer. The major challenges in clinical oncology are the selective delivery of large amounts of therapeutic agents into tumor cells, accurate evaluation of the drug delivery, timely assessment of the therapeutic response and effective treatment of drug resistant cancers. Nanomaterial is playing a pivotal role in cancer diagnostics and therapeutics due to their unique optical, electronic, and magnetic properties. Theranostic nanoparticles with the abilities to target tumors, carry therapeutic agents, and produce contrasts for tumor imaging offer a promising means for novel treatments of cancer patients. We have developed a multifunctional theranostic magnetic iron oxide nanoparticle (IONP) platform that utilizes receptor-targeted IONPs to carry single or multiple therapeutic agents for drug delivery and optical and magnetic resonance imaging (MRI). Our theranostic nanoparticles are designed to overcome physical and intrinsic barriers that reduce efficiency of drug delivery and confer drug resistance in human cancers. By targeting to cellular receptors that are highly expressed in tumor cells, angiogenic endothelial cells, and active tumor stromal cells, these IONPs allow the drug to overcome the physical barrier in stroma-rich tumors, such as pancreatic cancer and triple negative breast cancer (TNBC), by serving as carrier vehicles for passage through the tumor endothelial cell layer and stromal fibroblasts, thereby increasing the efficiency of delivery into tumors but not into normal tissues. Based on the surface functionalization of the IONPs and chemical properties of drug molecules, we developed approaches for encapsulating or conjugating drugs to the IONPs, resulting in theranostic IONPs which carry one or multiple therapeutic agents. Targeted delivery, drug release, tumor growth inhibition, and MRI of drug delivery and response have been demonstrated in orthotopic breast and pancreatic cancer animal models. Conjugation of a new near infrared dye with a lasting-signal to the theranostic nanoparticles provides an optical imaging modality that allows identifying and removal drug resistant residual tumors by image-guided surgery. Therefore, our theranostic IONPs have the potential to significantly improve the efficiency of cancer treatment, Current preclinical studies focus on the development of an integrated protocol for the treatment of locally advanced pancreatic and triple negative breast cancers using targeted neoadjuvant nanotherapy and image-guided surgery.

Biography

Dr. Yang is an Associate Professor of Surgery and Radiology and Nancy Panoz Chair of Surgery in Cancer Research at Emory University. Dr. Yang received her medical training in China at West China University of Medical Sciences and then in the Chinese Academy of Preventive Medicine. She received her PhD degree in Molecular and Cellular Biology at Brown University. She was a research fellow in gene therapy at the University of Southern California and Emory University before joining the Department of Surgery at Emory as an Assistant Professor. Dr. Yang's research has concerned liver stem cells and cancer, gene therapy, apoptosis, molecular targeted therapy, biomarker targeted drug delivery, and cancer nanotechnology. During the last several years, she leads a research team to develop targeted optical and magnetic resonance imaging (MRI) nanoparticle probes for early detection of breast and pancreatic cancers and for image-guided therapy and surgery. Her group has developed a theranostic magnetic iron oxide nanoparticle (IONP) platform that utilizes receptor-targeted IONPs to carry single or multiple therapeutic agents for drug delivery and multi-modality tumor imaging. Her current research also focuses on molecular targets and signal pathways that confer aggressive behavior, invasiveness and resistance to apoptosis in triple negative breast cancer. Dr. Yang is the PI of several research projects supported by NIH R01, NIH P50 Emory Molecular Imaging Center, and NIH U01 Cancer Nanotechnology Platform Partnership grants. Her research has resulted in several patent applications. Dr. Yang is a member of the editorial boards of Apoptosis and Breast Cancer-Targets and Therapy. She is a member of the NIH Developmental Therapeutics study section and has served in many other NIH study sections.