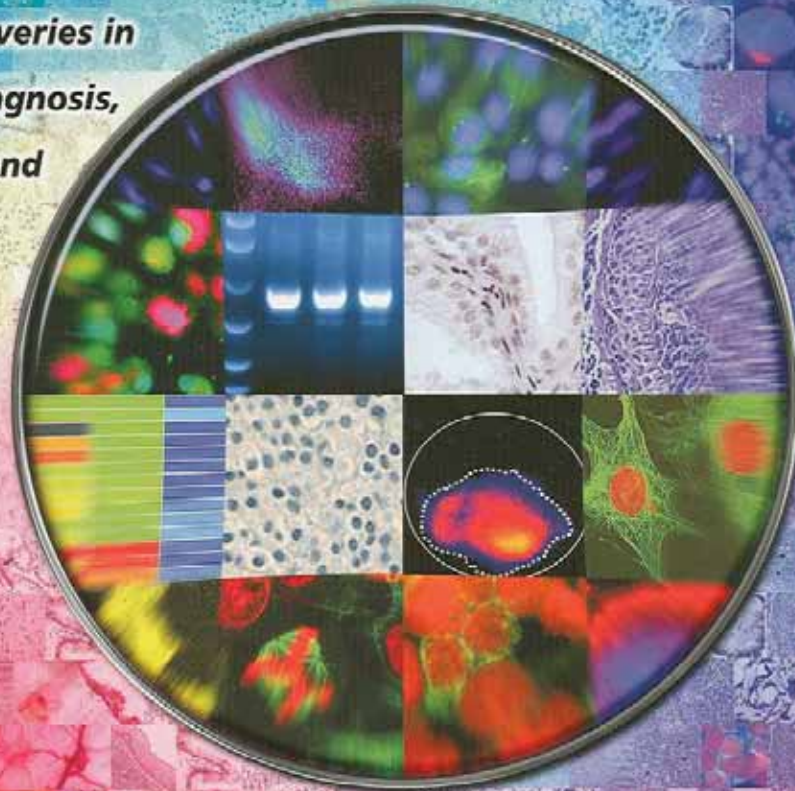


ACR *American Association
for Cancer Research*

**Featuring the
Latest Discoveries in
Etiology, Diagnosis,
Treatment, and
Prevention**



96th Annual Meeting 2005

CME jointly sponsored
by the University of
Minnesota and the
American Association
for Cancer Research

April 16-20, 2005 • Anaheim Convention Center • Anaheim, Orange County, CA

Proceedings Supplement

96th Annual Meeting Proceedings Supplement

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This volume contains abstracts that were accepted for presentation in the Late-Breaking Research Sessions. Abstracts are numbered from LB-1 through LB-322; however, several numbers may be omitted in the sequence. Abstracts were received directly from the authors by electronic submission. Every effort has been made to reproduce the content of the abstracts according to the electronic version submitted, except in certain instances where changes were made to comply with AACR style. The AACR does not assume any responsibility for proofreading or correcting any scientific, grammatical, or typographical errors, nor does the AACR assume responsibility for errors in the conversion of customized software, newly released software, or special characters. No responsibility is assumed by the AACR, publisher and copyright owner of the Proceedings Supplement; by the University of Minnesota; by Database Publishing Group, providers of abstract management software and services; or by the meeting organizers for any injury and/or damage to persons or property as a matter of products liability, negligence or otherwise, or for any use or operation of any methods, products, instructions, or ideas contained in the material herein. Independent verification of diagnoses and drug dosages should be made by readers or users of this information.

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Accreditation Statement

This activity has been planned and implemented in accordance with the Essential Areas and Policies of the Accreditation Council for Continuing Medical Education (ACCME) through the joint sponsorship of the University of Minnesota and the American Association for Cancer Research (AACR). The University of Minnesota is accredited by the ACCME to provide continuing medical education activities for physicians.

Credit Designation Statement

The University of Minnesota designates this educational activity for a maximum of 51 Category 1 credits toward the AMA Physician's Recognition Award. Each physician should claim only those credits that he/she actually spent in the activity.

Statement of Educational Need, Target Audience, and Learning Objectives

New technology and the resulting explosion in biological information have led to a significantly improved understanding of cancer development and progression, with major implications for cancer prevention, diagnosis, and treatment. By participating in the AACR 96th Annual Meeting, clinical investigators and physicians will better understand the contributions of laboratory research to patient care, the design and conduct of clinical research protocols, the epidemiological implications of cancer

incidence, and the translation of the findings of laboratory research immediately for the benefit and care of the cancer patient. Through this participation of clinical investigators and physicians, laboratory researchers will obtain a better understanding of the wider context of their research and of the particular clinical problems that regularly confront physicians. This activity is presented for scientists and clinicians engaged in all aspects of clinical investigations pertaining to human cancer as well as the scientific disciplines of cellular and molecular biology; tumor biology; carcinogenesis; chemistry; clinical research; endocrinology; epidemiology; experimental and molecular therapeutics; immunology; and prevention research. At the conclusion of this educational activity, each participant should be able to:

- Understand the latest research findings in all areas of cancer research;
- Understand the importance of integrating information from basic and clinical research to conquer cancer;
- Understand the epidemiological implications of cancer incidence;
- Understand how technological advances can be used to accelerate discovery; and
- Use these findings to expedite the development of detection, prevention; and treatment strategies to reduce cancer incidence and mortality.

Disclosure Statement

It is the policy of University of Minnesota that the information presented at University of Minnesota-sponsored CME activities will be unbiased and based on scientific evidence. To help participants make judgments about the presence of bias, the University of Minnesota and the AACR provide information that speakers have disclosed about financial relationships they have with commercial entities that produce or market products or services related to the content of this CME activity. This disclosure information is available in the back of the Proceedings of the AACR. Disclosures received after the Proceedings went to press are listed in the back of this publication.

Claiming Credit

Individuals wishing to receive Category 1 credit for their attendance at this meeting should obtain a Documentation of Attendance and Request for Credit form from the CME booth located in the registration area. This form must be completed and returned to the CME booth to obtain a Certificate of Attendance.

Acknowledgment of Commercial Support

Commercial entities that provide unrestricted educational grants in support of this educational activity will be listed in the Annual Meeting Program.

Next Annual Meeting: April 1-5, 2006, Washington, DC

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activating K⁺ channels in the vascular wall, without affecting healthy cells (Circulation Research, 2004). We hypothesized that DCA might be a novel treatment for human cancer. Three unrelated human cancer cell lines (A549-non-small lung carcinoma, MO59K-glioblastoma, MCF7-breast carcinoma) were found to have hyperpolarized mitochondria ($\Delta \psi$ studied with TMRM confocal microscopy) and smaller K⁺ current density (whole-cell patch clamping), compared to normal epithelial and smooth muscle cell lines. Exposure to DCA for 48 hours (500 μ M) depolarized $\Delta \psi$ by ~50%, increased H₂O₂ by ~30%, increased K⁺ current density by ~50% and increased mRNA and protein levels of Kv1.5 in all cancer cell-types, but had no effects in normal cells. Acutely, catalase, rotenone (ETC complex I inhibitor) and 4-aminopyridine (Kv inhibitor), but not TTFB (complex II inhibitor), prevented the activation of the K⁺ current, suggesting that it was due to the opening of Kv channel(s) by complex-I produced H₂O₂. Compared to the untreated cells, DCA increased TUNEL 6-fold and decreased proliferating cell nuclear antigen by 50%; furthermore, DCA caused mitochondrial release of cytochrome c to the cytoplasm, translocation of apoptosis-inducing factor into the nucleus and activation of caspase 3 and 9. Athymic nude rats injected with 3x10⁶ A549 cells subcutaneously, rapidly developed tumors and received DCA (75mg/L in the drinking water) either on day 1 or day 10, followed for 4 weeks. In both protocols DCA decreased the size of the tumor by 50%, without any evidence of liver, kidney or blood toxicity. In vivo, DCA increased TUNEL 5-fold and decreased PCNA by ~70%. Metabolic modulators might be a new class of orally available and well-tolerated anticancer drugs; the mitochondria-K⁺ channel axis needs to be further explored in cancer therapeutics.

LB-286 Biochemical analyses and molecular modelling of two aminoacidic substitutions detected in two gastrointestinal stromal tumor (GIST) patients showing acquired resistance to imatinib. Elena Tamborini, Sabrina Priol, Tiziana Negri, Maria Stefania Lagonigro, Elisa Gabanti, Angela Greco, Gianpaolo Dagrada, Francesca Miselli, Marco Ferrone, Marco A. Pierotti, Silvana Pilotti. Istituto Nazionale per lo Studio e la Cura dei Tumori Milano, Milano, Italy, University of Trieste, Trieste, Italy, Istituto Nazionale per lo studio e la cura dei tumori milano, Milano, Italy.

Background: Imatinib acquired resistance in gastrointestinal stromal tumors (GIST) has becoming

a frequent event: patients initially responding to the drug and subsequently showing clinical and radiological symptoms of progressing disease have been described. Molecular mechanisms at the bases of this phenomenon are poorly understood and scarcely investigated. To date, only secondary point mutations affecting the ATP pocket of the KIT receptor have been reported in literature. **Materials and methods:** RNA and DNA were extracted from tumors using specific kits (QIAGEN). The sequencing was performed using an automated device. Protein extraction, immunoprecipitation experiments and Western Blotting were performed using 1 mg of total proteins and using the Ab-3 (Neomarkers Union City). Site-specific mutagenesis was performed using the Promega kit. **Modeling:** The molecular mechanics/Poisson Boltzmann Surface Area (MM/PBSA) computational techniques was applied to study the interactions of the wild-type and mutated receptors with imatinib at the molecular level. **Results:** Among a series of patients surgically treated in our Institute all showing clinical/radiological evidence of progressing disease despite of Imatinib treatment, 8 patients were molecularly and biochemically investigated for KIT and PDGFRA gene alterations. None of them showed mutations in PDGFRA gene, and FISH analysis reveal neither KIT or PDGFRA gene amplification. In one patient only chromosome 4 polysomy was observed. The sequencing of the whole coding sequence of KIT gene was performed in the tumoral specimens, revealing activating mutations in exon 11 in all patients. In two patients, two different adjunctive point mutations in KIT gene, one in exon 14 responsible for T670I substitution, and one in exon 13 causing the V654A substitution, were detected. Biochemical analyses showed KIT phosphorylation in cells transfected with vectors carrying the specific mutant genes and treated with different doses of imatinib. The modelling of the mutated receptors revealed that both substitutions affect imatinib binding-site, but with different mechanism. In the remaining 6 patients secondary resistance could be explained by alternative mechanism that exclude KIT/PDGFRA gene amplification or secondary point mutations. **Conclusions:** In the present series of patients with acquired resistance to imatinib, the only detected molecular mechanism responsible for this event is the presence of adjunctive point mutations affecting the ATP pocket of KIT receptor and it was demonstrated only in two out of 8 cases analyzed. By the application of molecular simulations we were able to quantify the interactions between the

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mutated receptors and imatinib, and to propose a molecular rationale for this type of drug resistance at a molecular level.

LB-287 Preclinical evaluation of an anti-CD33-maytansinoid immunoconjugate, huMy9-6-DM4 (AVE9633) for the targeted therapy of acute myeloid leukemia. Ravi V. J. Chari, Hongsheng Xie, Barbara A. Leece, Lisa Garrett, Michele Mayo, Kathleen Whiteman, Victor S. Goldmacher, John M. Lambert, Hichem Chakroun, Marie-Christine Bissery, Thierry Hercend, Robert J. Lutz. ImmunoGen, Inc., Cambridge, MA and sanofi-aventis, Vitry sur Seine, France.

The current option for patients with acute myeloid leukemia (AML) consists of treatment with a combination of cytotoxic drugs. While, complete remissions are often achieved, a majority of patients eventually relapse. Treatment of relapsed patients is much less successful, often associated with high toxicity, and long-term disease-free survival is limited. The use of monoclonal antibodies to deliver highly cytotoxic drugs specifically to tumor antigens provides a promising approach to improving response rates and potentially lowering treatment-related toxicity in AML. The CD33 antigen is expressed on the surface of myeloid blasts cells in about 90% of AML patients. The antigen is not expressed on lymphoid or non-hematopoietic cells, making it an attractive target for antibody-directed therapy. In fact, a humanized anti-CD33 monoclonal antibody-calicheamicin conjugate (Mylotarg) has been approved for the treatment of patients with CD33 positive AML in first relapse who are 60 years of age or older. Here we report the results from the preclinical evaluation of a targeted therapy approach, where the anti-CD33 antibody huMy9-6 is used to deliver the potent anti-mitotic maytansinoid DM4 to AML tumor xenografts. Despite its 100 to 1000-fold greater *in vitro* potency than conventional anti-cancer drugs, DM4 exhibits extremely low non-specific toxicity when linked to a target-specific monoclonal antibody. huMy9-6-DM4 maintains the specificity and binding affinity of the unmodified antibody for CD33, and shows potent antigen-specific cytotoxicity toward CD33-positive tumor cells *in vitro*. Treatment of SCID mice bearing established subcutaneous HL-60 tumor xenografts with huMy9-6-DM4 administered as a single injection (51.5 mg/kg conjugate) or multiple injection (conjugate dose of 8.6 mg/kg, qd x 5), resulted in complete tumor regression and cures. Importantly, no evidence of toxicity was observed in

either schedule. Treatment with the huMy9-6 antibody alone had no effect on HL-60 tumor growth compared to vehicle control. Similar curative efficacy was observed with huMy9-6-DM4 upon treatment of mice bearing subcutaneous THP-1 xenografts. The potent efficacy and favorable toxicity profile of huMy9-6-DM4 observed in these studies supports the clinical development of this antibody-DM4 conjugate for the treatment of AML.

LB-288 RNA interference targeting aurora kinase A suppresses tumor growth and enhances the taxane chemosensitivity in human pancreatic cancer cells. Tatsuo Hata, Toru Furukawa, Makoto Sunamura, Shinichi Egawa, Fuyuhiko Motoi, Noriyuki Ohmura, Tomotoshi Marumoto, Hideyuki Saya, Akira Horii. Tohoku University School of Medicine, Sendai, Japan, Tohoku University Hospital, Sendai, Japan, Kumamoto University Graduate School of Medical Sciences, Kumamoto, Japan.

AURKA/STK15/BTAK, the gene encoding AURORA A kinase that is involved in the regulation of centrosomes and segregation of chromosomes, is frequently amplified and overexpressed in various kinds of human cancers, including pancreatic cancer. To address its possibility as a therapeutic target for pancreatic cancer, we employed the RNA interference technique to knockdown *AURKA* expression and analyzed its phenotypes. We found that the specific knockdown of *AURKA* in cultured pancreatic cancer cells strongly suppressed *in vitro* cell growth and *in vivo* tumorigenicity. The knockdown induced the accumulation of cells in the G2/M phase and eventual apoptosis. Furthermore, we observed a synergistic enhancement of the cytotoxicity of taxanes, a group of chemotherapeutic agents impairing G2/M transition, by the RNA interference-mediated knockdown of *AURKA*. These results indicate that inhibition of *AURKA* expression can result in potent antitumor activity and chemosensitizing activity to taxanes in human pancreatic cancer.

LB-289 Engineered mammalian ribonucleases inhibit tumor growth *in vivo*. Laura E. Strong, John A. Kink, Tony A. Klink, Jennifer G. Reich, Ronald T. Raines. Quintessence Biosciences, Inc., Madison, WI and University of Wisconsin - Madison, Madison, WI.

Destroying RNA precludes protein biosynthesis and ultimately results in cell death. Ribonucleases are ubiquitous enzymes that catalyze the cleavage of RNA. Perhaps the most widely studied ribonuclease is bovine pancreatic ribonuclease (RNase A). RNase A is rendered inactive inside cells