xenographs. As an alternative approach we have applied the power of mouse genetics to produce animal models that recapitulate the genetics and pathobiology of human malignancies to study treatment responses of tumors treated at their natural site. One model we have focused on is the Em-myc transgenic mouse. These mice overexpress the c-myc oncogene in B-cells, and develop malignancies that resemble human Non-Hodgkin's lymphomas. Using methods for rapidly producing Eμ-myc lymphomas with compound genetic lesions and experimental strategies that parallel clinical trials, we have characterized biologic and genetic determinants of drug resistance in vivo. These studies have identified potential mechanisms of drug sensitivity and resistance, and highlight the relationship between tumor cell genotype and its response to cancer chemotherapy. They also suggest rational strategies to reverse drug resistance in some tumor types.

**13** INVITED

**Patient and cell line derived human tumor xenograft models – preclinical/clinical correlations**

H.H. Fiebig. *Oncotest GmbH, Institute for Experimental Oncology, Freiburg, Germany*

For all major solid human tumor types experimental models have been developed by engrafting patient tumors or permanent human cell lines into immunodeficient mice. Transferring the NCI 60-cell line panel – the largest cell line panel used for drug discovery – *in vivo* into nude mice resulted in sc growth in 47/58 cases (Fiebig et al 1989), or 49/60 cases (Plowmann et al. 1997), respectively. My group implanted more than 1600 solid tumors directly from patients, leading to more than 400 permanent xenograft models. The take rate was highest (38–51%) for melanomas, lung and colon cancers. 60 xenografts were characterized in detail by comparing histology, and 10 surface markers over 10 *in vivo* passages with the original patient tumor. More than 90% showed very similar histology and marker profiles. From 100 xenografts the histology, sensitivity to standard cytotoxic agents, 35 molecular drug targets and expression profiles of 34.000 genes (Affymetrix chips HU133) were determined and compared with the occurrence in the *in vivo* growing NCI-60-cell line panel. 85% of the NCI cell lines showed an undifferentiated or very poorly differentiated histology *in vivo* without the typical tumor architecture seen in e.g. adenocarcinomas of the colon or lung in contrast to the patient's and the patient derived xenograft models. Many cell line-derived xenografts grow faster than patient-derived ones. For colon xenografts, comparison of gene expression profiles showed some differences in cell line-derived models to patient derived xenografts in typical colon associated genes. Activity of standard cytotoxic agents (regression) was observed in cell line-derived xenografts only with alkylating agents, but not with Vinca-alkaloids, Adriamycin, VP16, 5-FU and Methotrexat. In the Freiburg xenograft panel we obtained regressions for 12 clinical active standard agents except 5-FU from which mice tolerate only 25% of the human dose. The response of the same tumors treated in the nude mouse and in the patient was investigated in 42 combinations and 38 single agent therapies. The xenografts showed a very similar response as the same tumor in the clinic, in 90% (19/21) for remission and in 97% (57/59) for resistance. The high correct predictivity validates the xenograft system for drug development. More recently, also target-directed compounds effected remissions and T/C values <10%, e.g. EGF-R-, VEGF- and HSP-90-inhibitors EMD72000, Erbitux, Avastin and 17-AAG. Modulation of the respective targets *in vivo* has been demonstrated. Patient derived xenograft models established in nude mice reflect very well the clinical situation except for metastasis and they are excellent models for tumor biology studies and for the discovery of target directed novel antitumor agents.

## Wednesday 29 September 08:00–09:45

**WORKSHOP 3**

# Mechanistic combinations

**14** INVITED

**Combined targeted agents with cytotoxic chemotherapy**

L. Gianni, G. Bianchi, G. Mariani, S. Cresta. *Istituto Nazionale Tumori, Division of Medical Oncology A, Milan, Italy*

The availability of targeted drugs interfering with signal transduction and/or intracellular signaling has led to the investigation of the potential for using them in combination with conventional chemotherapy that still is the leading therapeutic option for medical treatment of cancer patients. The possibility for combination stands on several considerations. The cellular targets of chemotherapeutic and of targeted drugs are different, supporting the concept that combinations would not lead to cross-resistance, while expected toxicity would not be overlapping. Indeed, perturbation of signals involved in regulation of growth, survival, invasion and metastasis could be associated with enhanced sensitivity to chemotherapeutic drugs and eventually lead to synergistic antitumor effects. The possibility of combining chemotherapy and targeted drugs has been extensively explored in several cellular and animal models. In most cases, the combination of cytotoxic agents and drugs targeting erbB receptors, farnesyl-transferase, m-TOR, PTEN, proteasome, VEGF and VEGF-receptors, PDGFR and many other signaling pathways including those involved in apoptosis has indicated at least additive and often synergistic results. Such experimental evidence has served as a basis for designing clinical studies for many such combinations. Most molecular targeted agents could be combined with most cytotoxic agents at full or nearly full doses. In most instances, continuous exposure to the targeted drugs was associated with concomitant delivery of monochemotherapy or classical combination chemotherapies. Less frequently, the targeted drugs were given sequentially after the delivery of the planned chemotherapy. Outcome of these trials have sometimes posed problems of enhanced or unexpected toxicity, although tolerability of the combinations has rarely been a limiting factor. The clinical antitumor activity and efficacy has offered mixed results. In some case, most notably that of combinations of anti-erbB2 drugs and chemotherapy, significantly improved efficacy and possibly syergism was documented. In other examples, the preclinical evidence supporting supra-additive effects was not confirmed in the clinic. The mixed success of the approach is possibly related to the emerging awareness that in most cases the presence of the target is not the only variable dictating the sensitivity to the targeted agent(s). The future success of combinations of cytotoxic agents and targeted drugs will likely depend on the clarification of what makes the target competent of the tumor survival, and in which cases. That clarification will restrict the applicability of targeted drugs, but, at the same time, expand the possibility of exploiting and measuring expected synergisms from the application of these new combinations in selected subgroups of patients.

**15** INVITED

**Radiation with targeted agents**

M. Brown. *Stanford University, Department of Radiation Oncology CCSR-South, Room 1255, CA Stanford, USA*

Two fundamental principles should guide the extrapolation of preclinical data with radiation and anticancer agents to the clinical situation as follows:

1. The mechanisms by which each agent produces its antitumor effect must be understood.
2. The extent to which the preclinical model mimics the human situation in those aspects relevant to the mechanisms of the agents must be known.

This presentation will review examples relevant to the combination of radiation with targeted agents with these two principles in mind. Specifically, we will review the extent to which the response of tumors to radiation is governed by the sensitivity of the tumor cells versus the sensitivity of the endothelial cells comprising the vasculature. The answer to this question will determine the efficacy and way in which radiation should be combined with antiangiogenic agents. Also of relevance to the combination of radiation with antiangiogenic drugs is the extent to which transplanted tumors reflect human spontaneous tumors in their reliance on neovasculature. We will show that preclinical models with transplanted tumors are likely to "overpredict" the efficacy of antiangiogenic therapy in the clinic because rapidly growing transplanted mouse tumors have a total reliance on the neovasculature whereas this is not the case with most human tumors. Second, we will review the extent to which preclinical models mimic the clinical situation when radiation is combined with hypoxic cell radiosensitizers or hypoxic cytotoxins. In this context many studies, particularly with hypoxic cell radiosensitizers, were conducted at doses of radiation and drugs that did reflect the clinical situation, and which let to inappropriate expectations in the clinic. Third, we will examine the extent to which short-term assays using apoptosis or tumor shrinkage are relevant to the response of human tumors to combinations of radiation with therapies designed to enhance tumor cell apoptosis.

**16** INVITED

**Targeted agent with targeted agents**

P. Houghton. *St Jude Children's Research Hospital, Molecular Pharmacology, Memphis, USA*

Dysregulation of signaling cascades involved in cellular proliferation and survival is a hallmark of many human cancers, and thus presents potentially selective targets for therapeutic intervention. Agents that target signaling pathways dysregulated during transformation and tumor progression are entering clinical trials. However, with the exception of imatinib, other