resistance to several of anticancer drugs with differing mechanisms of action.
Finally, the role of Bid as a lipid transfer protein [4] and its relationship to the pro-apoptotic function of Bid will be discussed.

References

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Roles of oncogenes and tumor suppressor genes in apoptosis

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p53 was initially identified as the "guardian of the genome" based on its ability to mediate a G1 arrest following DNA damage. However, p53 can participate in many processes involved in maintaining cellular integrity following stress, including cell-cycle checkpoints, DNA repair, senescence, apoptosis, angiogenesis, and the surveillance of genomic integrity. The relative contribution of each of these processes to tumor suppression is not known. We hypothesized that complete ablation of crucial p53 effector functions may produce tumors that are phenotypically identical to those with p53 mutations yet retain wild-type p53. To this end, we examined the ability of dominant-acting genes that completely disable apoptosis downstream of p53 to phenocopy the effects of p53 mutations during tumor development and treatment responses in the Em-myc transgenic mouse. This system provides an ideal setting in which to study p53 action during tumor development an therapy, since loss of p53 function dramatically accelerates tumor development and produces profound drug resistance to conventional chemotherapy. Using this system, we show that disruption of apoptosis provides the sole advantage to developing lymphoma cells that lose p53 function, whereas disruption of cell cycle checkpoints and anueploidy are mere byproducts of p53 loss. In contrast, disruption of both apoptosis and cellular senescence account for the impact of p53 loss on drug resistance. The implications of these results for understanding p53 action and the heterogeneity of treatment responses in human tumors will be discussed, as well as new insights into the program of cellular senescence and its role in treatment outcome.

Wednesday 20 November

PLENARY SESSION 2

Proffered Papers 1

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Increased ERCC1 expression predicts for improved survival in resected patients with non-small cell lung cancer (NSCLC)

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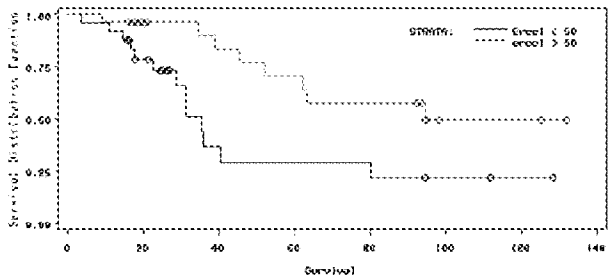
ERCC1 expression has been previously reported to predict for cisplatin resistance in patients with gastric carcinomas and NSCLCs (Metzger et al JCO 1998, Pg 309). Consequently gastric and NSCLCs treated with a cisplatin based chemotherapy had superior overall survival if their ERCC1 expressions were low. We evaluated the effect of ERCC1 expression on overall survival in 49 patients with Stage IA to IIIB NSCLC who underwent surgical resection. One of the 49 patients received postoperative adjuvant chemotherapy and radiation therapy. Five patients received post-operative adjuvant radiation therapy alone. Forty-three patients received no adjuvant therapy. Tissue specimens from these patients were collected and immediately frozen in liquid nitrogen. Total RNA was extracted, reverse transcribed, and used for real-time quantitative PCR (ABI Prism 7700). Gene expression was normalized using 18S rRNA as reference. ERCC1 expression ranged from 4.96 to 13,160.20. Median value for the entire group was 54.76. When we used 50 as the cut off there was a statistically significant difference in survival for patients with ERCC1 expression more than 50 (94.6 months) vs. less than 50 (35.5 months)(P=.01)(Wicoxon Rank Sum Test). See Graph 1.



Graph 1

Additionally, when we divided the entire cohort on the basis or ERCC1 expression to <30, 30 to 100 and >100. There was again a statistical significant survival between the three groups. Median Survival was 94.6 months for >100, 62.1 months for 30 to 100 and 35.5 months for <30. These differences were statistically significant (P value = 0.03). On the basis of our results we conclude that patients with an efficient DNA repair mechanism (High ERCC1 expression (>50)) have a better survival than patients in whom this mechanism is impaired (Low ERCC1 Expression (<50)). However patients with high ERCC1 expression also respond poorly to chemotherapy. Since patients with Low ERCC1 expression have a poorer prognosis but respond better to chemotherapy, they are likely to benefit from chemotherapy. Hence we propose that future adjuvant and neoadjuvant chemotherapy trials, should stratify patients according to their ERCC1 status. Updated results and multivariate analyses will be presented at the meeting.

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Mechanisms of cisplatin resistance - role of yeast SKY1 and its human homologue SRPK1

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The therapeutic potential of cisplatin, one of the most active and widely used anticancer drugs, is severely limited by the occurrence of cellular resistance. We used the budding yeast Saccharomyces cerevisiae as a model organism to identify and characterize novel genes involved in cisplatin-induced cell kill, and found several candidate players. Most strikingly, we identified SKY1 (serine/arginine-rich-protein-specific kinase from budding yeast) as a cisplatin sensitivity gene, whose disruption conferred a 4-fold cisplatin resistance. In cross-resistance studies, we observed resistance of yeast sky1del cells (i.e., cells from which the SKY1 gene had been disrupted) to cisplatin, carboplatin (but not oxaliplatin), doxorubicin and daunorubicin, and hypersensitivity to cadmium chloride and 5-fluorouracil. Furthermore, these cells did not display reduced platinum accumulation, DNA platination or doxorubicin accumulation, indicating that the resistance is unrelated to decreased drug import or increased drug export. Based on the modification of the anticancer drug sensitivity profile and our finding that sky1del cells display a mutator phenotype, we propose that the Sky1p protein might play a significant role in specific repair and/or tolerance pathways. Heterologous expression of the human SKY1 homologue SRPK1 (SR-protein-specific kinase) in yeast sky1del cells restored cisplatin sensitivity, suggesting that SRPK1 is also a cisplatin sensitivity gene, inactivation of which could lead to cisplatin resistance. Treatment of human ovarian carcinoma A2780 cells with antisense oligodeoxynucleotides directed against the translation initiation site of SRPK1, led to downregulation of SRPK1 protein and conferred a 4-fold resistance to cisplatin. Our findings strongly suggest that SRPK1 is involved in cisplatin-induced cell kill and indicate that SRPK1 might potentially be of importance for studying clinical drug resistance. Therefore, we have recently set out to screen clinical samples for SRPK1 expression and correlation with responsiveness to platinum-based chemotherapy.
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References

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