cine vectors are able to do. Therefore, a strategy that facilitates the spread of antigen to other APCs may significantly enhance the potency of naked DNA vaccines delivered intradermally. We have recently enhanced the potency of DNA vaccines using herpes simplex virus (HSV-1) VP22, an HSV-1 tegument protein that has demonstrated the remarkable property of intercellular transport and is capable of distributing protein to many surrounding cells. We showed that HSV-1 VP22 (HVP22) was capable of enhancing intercellular spreading of linked protein, such as E7. Furthermore, we demonstrated that mice vaccinated intradermally with HSV-1 VP22/E7 DNA generated a significantly greater number of E7 specific CD8+ T cell precursors and stronger antitumor effect than mice vaccinated with wild-type E7 DNA. The impressive pre-clinical data based on these strategies have led to several nucleic acid vaccine trials tentatively scheduled to begin in 2003.

Wednesday 20 November

WORKSHOP

Novel targets and radiation response

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COX-inhibitors and radiation

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Cyclooxygenase-2 (COX-2), the enzyme that converts arachidonic acid to prostaglandins, overexpressed in a variety of different tumors, including colon, pancreatic, prostate, lung and head and neck cancers. COX-2 is also observed within human tumor neovasculature, suggesting that COX-2 derived prostaglandins contribute to tumor growth by inducing formation of new blood vessels

Angiogenesis is the process by which new capillaries are formed from preexisting vessel networks. Angiogenesis into a newly growing tumor provides a pathway for escape and systemic dissemination via the blood or lymph system and represents an important therapeutic target. Tumors that demonstrate intense immunostaining for COX-2 also demonstrate co-localization for cytokines involved in angiogenesis.

Angiogenesis has been considered as a potential target for the treatment of cancer, either by the inhibition of endothelial cell proliferation and migration or by inhibition of the production of angiogenic factors by tumors. In contrast to tumor cells, endothelial cells, derived from the host, are genetically stable and have a low mutation rate. Kerbel has suggested that antiangiogenic therapy may be a strategy to bypass drug resistance. Celecoxib and rofecoxib, have been shown to posses potent inhibitor of angiogenesis and tumor growth.

A growing tumor requires a blood supply; and, thus, it secretes numerous angiogenic compounds that induce host endothelial cells to proliferate, migrate, and differentiate into patent vessels. It is proposed that by inhibiting the angiogenic processes of endothelial cells, tumor growth and metastasis will be inhibited as well. It is unclear at the point whether the target for selective inhibitors of COX-2 is tumor, tumor associated neovasculature or both. Clinical trials will help elucidate the role of this interesting class of agents in combination with cytotoxic therapy for the treatment of cancer. The use of COX-2 inhibitors in cancer therapy may complement current strategies while potentially minimizing the impact on quality of life.

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Combination therapy with anti-angiogenic agents and radiotherapy

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Radiation therapy has been used for over 100 years in the treatment of cancer. The conventional explanation for the mechanism of the -radiation effect- against tumor cells is that DNA damage caused by ionizing radiation results in reproductive cell death. However, what if, damage to the DNA of tumor cells is not the primary target? What if the primary target is actually the supporting endothelial cell?

The oxygen effect on the radio-resistance of hypoxic tumor cells has been well demonstrated. It may be counterintuitive to some that the use of anti-angiogenic therapy may augment local control with radiotherapy. Classic dogma teaches that if the tumor bed is rendered more hypoxic with anti-angiogenic therapy, then the tumor cells must be less radiosensitive. Generating hypoxia with anti-angiogenic therapies may also select cancer cells that have acquired hypoxia-resistance and have a higher metastatic and invasive potential. Fortunately, three pre-clinical studies have shown that the treatment of tumors with anti-angiogenic drugs actually increases the tumor pO2. In 1992, Teicher published the first paper using a combination of anti-angiogenic therapy and radiotherapy against a primary tumor. She demonstrated, in a tumor growth delay study, that the combination of minocycline (a weak metalloproteinase inhibitor), TNP-470 and radiotherapy was synergistic against Lewis lung carcinoma cells in mice. These results triggered a paradigm shift in the rationale for combining anti-angiogenic therapy with radiotherapy.

Since this landmark study, radiotherapy has been used in combination with numerous anti-angiogenic agents including angiostatin, endostatin, anti-VEGF therapy, thalidomide, as well as numerous other agents, in pre-clinical models. Numerous clinical trials are also ongoing with many newer combinations in the planning phase.

It is the objective of this session to review the data concerning anti-angiogenic therapy and its effects on tumor vasculature and to describe the potential use of anti-angiogenic therapy from the point of view of the radiation oncologist. We will then explore the promising evidence and rationale for combining anti-angiogenic drugs and radiotherapy to enhance local control.

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Protein expression and tumor hypoxia

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There is strong evidence that poor oxygenation (hypoxia) influences important physiological and pathological conditions. This includes development, ischemia, stroke and cancer. In cancer, hypoxia is a negative prognostic factor and is implicated in carcinogenesis, metastasis, angiogenesis and therapy resistance. The influence of hypoxia is due in large part to changes in gene expression. We are investigating the both the mechanisms of gene regulation during hypoxic stress and attempting to identify hypoxia responsive genes. Most of the known hypoxia-induced genes are regulated at the level of transcription through HIF-1. We have found that a second important mode of gene regulation during hypoxic stress occurs through inhibition of mRNA translation. The eukaryotic initiation factor 2-alpha (eIF2-alpha) becomes phosphorylated at Ser51 within 1 hr of hypoxia, resulting in a rapid decrease in protein synthesis. At higher oxygen concentrations (0.05% - 1%), the phosphorylation occurs later, and is less pronounced. This effect is specific to hypoxia and is independent of HIF-1alpha. Prolonged hypoxia causes further inhibition of translation through inhibition of the mRNA cap binding protein eIF4E. The eIF4E binding proteins (4E-BPs) become dephosphorylated, resulting in increased binding to eIF4E. Concomitantly there is loss of association of eIF4E with the scaffolding protein eIF4G, which brings eIF4E, the mRNA and the ribosomal subunit together. Finally, the eIF4E transporter (4E-T) also becomes dephosphorylated and both proteins relocalise to the cell nucleus. These changes demonstrate that cells respond to hypoxia by a rapid, co-ordinated and persistent down regulation of protein synthesis that has important implications for understanding protein expression in hypoxic tumors.

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P53 differential radiosensitizing mechanism of a PKC-inhibitor (PKC412)

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The cellular response to ionising radiation (IR) is complex and includes a great number of intra- and extracellular targets. Increasing the tumor specific cell kill of IR with pharmacological sensitizers is an attractive goal. But so far only few genes, growth receptors and signaling proteins are known to have a key function for tumor selective radiosensitization. Based on recent data it seems unlikely that single target radiosensitizers will have a major impact for radiocurability in advanced solid human tumors. However, targeting entire survival signaling pathways with known molecular aberrations in tumor cells is an attractive concept. P53 mutations are common in locally advanced solid human tumors and might confer a radioresistant phenotype in some tumors. Growth stimulatory protein kinase C (PKC) antagonizes IR-induced cell death. Likewise activation of the phosphatidylinositol 3-kinase/Akt survival pathway suppresses pro-apoptotic signals. Compounds