Currently used treatment regimens for advanced head and neck cancer utilize radiation, often combined with chemotherapy, resulting in a significant risk of toxicity to the patient.

In the search for safer alternatives, considerable interest has focused upon DNA repair pathways as potential targets for cancer therapy. The poly(ADP-ribose) polymerase (PARP) family are nuclear proteins that play a critical role in the DNA base excision repair (BER) pathway. BER is integral to the repair of single strand breaks (SSBs) that occur as part of normal cell replication. PARP inhibition impairs the BER system and results in a failure to repair these SSBs. In order to avoid replication fork collapse and death, the cell converts SSBs to double-strand breaks (DSBs).

The Mr11/Rad50/Nbs1 (MRN) protein complex has been identified as a critical component in the repair of DNA double strand breaks (DSBs). The MRN complex can effectively compensate for impaired PARP function. As a result, PARP inhibition alone is not effective.

We hypothesized that disruption of MRN DNA repair function through the use of a novel Rad50-disrupting adenovirus, combined with pharmacologic inhibition of PARP inhibition would impair the ability of HNSCC tumor cells to recover from physiologically induced DNA damage, triggering cell death without the need for chemotherapy or radiation.

RESULTS

Baseline PARP-1 and Rad50 expression. Expression of PARP-1 and Rad50 was determined in Western blot in human head and neck cancer cell lines. All lanes showed expression of the 116 kDa wild-type PARP-1 and the 153 kDa Rad50 protein. These are the tandem for the PARP inhibitor drug and the MRN-disrupting adenoviral vector, respectively.

Quantification of DNA DSBs. Neutral comet assay was used to quantify DNA DSB damage in the above groups at 12, 24, 36 and 48 hour time points. Images were captured digitally and analyzed with software to determine the mean tail moment (MTM), an indicator of DNA DSB quantity.

Cell proliferation assay. MTT assays were used to measure cell growth in each treatment group over 5 consecutive days. Cell density was assessed using a microplate reader.

CONCLUSIONS

Dual disruption of double strand break repair mechanisms, using Ad-Rad50, and single strand break repair, using the PARP inhibitor GPI-15427, resulted in:

- Significant inhibition of head and neck cancer cell growth with actual regression in cell number.
- This cytotoxic effect correlates with a significant increase in the number of unrepaired DNA double strand breaks.
- This treatment approach bypasses the need for chemotherapy or radiation by impairing the repair of physiologic DNA damage.

This novel treatment may be applicable to a broad range of malignancies and may dramatically reduce treatment-related morbidity in these patients while achieving adequate tumor kill.

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