Commentary

Salmonella typhimurium as a novel RNA interference vector for cancer gene therapy

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The use of bacteria in cancer therapy dates back at least to the 1890s, when William B. Coley observed that acquired streptococcal infections (erysipelas) correlated with regression of some soft tissue sarcomas. He reported the efficacy of inducing erysipelas in a series of ten sarcoma patients, providing the foundation for subsequent historical efforts to develop microbiobased anti-cancer therapies. Currently, however, Bacillus Calmette-Guerin (BCG) is the only example of such a therapy in standard clinical use in its application for bladder carcinomas.

The continued observation that bacteria selectively accumulate in tumor tissues has sustained an interest in their potential as anticancer agents. The capacity of anaerobes to concentrate in necrotic tumor tissues is well established and has been used for therapy in mouse models with Bifidobacterium4, Clostridia species5 and Corynebacterium parvum.6 Targeting tumors in this manner has been proposed as one mechanism to overcome the chemotherapy and radiation resistance in hypoxic tumor micro-domains. The potential for facultative organisms to colonize both hypoxic and well-perfused components of tumors may confer them an advantage over obligate anaerobes. Tumor colonization using facultative anaerobes is best studied to date using E. coli,7 Vibrio cholera8 and Salmonella species. Among these, attenuated Salmonella typhimurium has generated the greatest overall interest and is noted for its selective intra-tumor accumulation and growth9 when administered in mouse models. Well known for its ability to persist within epithelial cells and macrophages, S. typhimurium produces a sustained infection during which it can exert anti-tumor effects. Salmonella organisms bearing auxotrophic mutations may survive more selectively in the nutrient environment of tumors with high metabolic activity or necrotic material. Compromised host defenses in the tumor microenvironment likely also contribute to this tumor specific localization.

One attenuated S. typhimurium strain, VP20009, appears to have a favorable safety profile in humans, having been administered systemically to colon cancer and melanoma patients in phase I trials with minimal side effects.10,11 This strain bears attenuating mutations that reduce the toxicity of its lipopolysaccharide and create a requirement for an external source of adenine. Efforts to then enhance S. typhimurium innate tumoricidal activity have included engineering it to express cytosine deaminase,10,12 which converts administered nontoxic 5-fluorocytosine to the active chemotherapeutic agent 5-fluorouracil. Other recent strategies have assessed S. typhimurium as a potential immunotherapy vector through engineered expression of human tumor antigens13 or cytokines.14 S. typhimurium has additional versatility because of its capacity to transfer eukaryotic expression plasmids to mammalian cells. It has therefore been proposed for use as both a vehicle for DNA tumor vaccines15 and an anti-tumor gene therapy vector.16

In this issue of Cancer Biology & Therapy, Yang et al. exploit these diverse properties of S. typhimurium to test an anti-cancer gene therapy approach in a mouse model of malignant melanoma. Their system uses S. typhimurium to transduce a eukaryotic expression vector for anti-bcl-2 shRNA into B16-F10 melanoma cells. This cell line shares the property of overexpressing bcl-2 with most human malignant melanomas and is known to become sensitized to apoptosis by bcl-2 antisense oligonucleotides. S. typhimurium is shown to be an effective vehicle to establish anti-bcl-2 shRNA expression and mediate apoptosis in B16-F10 cells. Accordingly, incorporating this shRNA vector enhanced the intrinsic capacity of S. typhimurium to retard tumor growth and prolong survival in mice with B16-F10 tumors. The study represents the first demonstrated use of S. typhimurium to mediate vector-based RNA interference in mammalian cells in vivo.

Salmonella species hold a potential advantage over recombinant viral cancer gene therapy approaches by allowing prolonged colonization of tumors while still allowing for immediate termination of treatment using antibiotics. However, the safety and efficiency of gene transfer and duration of expression are potential limitations for the S. typhimurium vehicle as for any other gene therapy approach. Highly stable eukaryotic expression plasmids are available for S. typhimurium,15 and such strategies may confer lasting potency to engineered tumor-colonizing organisms. Also, while mice in this study received a single bacterial dose and ultimately died of their tumors, the feasibility of eradicating established tumors using
S. typhimurium as monotherapy using multiple doses has been tested. An auxotrophic mutant of S. typhimurium lacking any further genetic modification recently achieved this outcome in a mouse xenograft model of human prostate cancer by using multiple bacterial doses. The basis of these direct antitumor effects remains poorly understood and is likely mediated by complex interaction with host innate immunity via production of nonspecific inflammatory mediators, effects on tumor vasculature, and production of toxic bacterial proteins. The successful future use of S. typhimurium in cancer therapy may be aided by finding the optimal synergy between these direct killing mechanisms and the engineered capacity to target molecular pathways that are critical to tumor survival.

References