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Suicide Gene Therapy for Bladder Tumors

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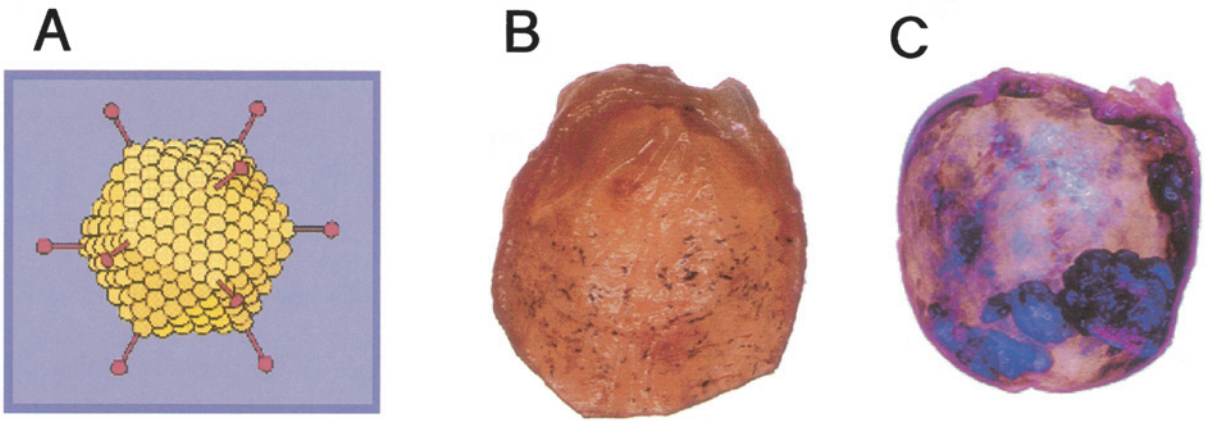

Fig. 1

Cancer is now an important target for gene therapy. Most animal experiments were done with the use of transplanted tumors produced by implantation of cell lines, an approach that ignores histologic and anatomic specificity of organs. Chemically induced orthotopic models are much superior to the subcutaneous transplanted models with respect to the evaluation of the efficiency of gene transfer and gene therapy. BBN (N-butyl-N-(4-hydroxybutyl)nitrosamine-induced bladder tumors are known to be biologically and histologically very similar to human transitional cell carcinomas and are thought to be an excellent model for gene therapy of bladder cancer.

We prepared a recombinant adenoviral vector containing the bacterial LacZ gene (Adex 1 CalacZ) and transurethrally instilled into the bladder with BBN induced tumors (**Fig. 1**)[1]. The normal bladder mucosa was highly resistent to adenoviral infection probably because glucosaminoglycan layers blocked viral entry to the normal cells. This tumor specificity suggests that adenoviral mediated suicide gene therapy may be applicable for treatment of bladder cancer.

The therapeutic protocol thus entailed instillation of an adenoviral vector containing the HSV-tk suicide gene (Ad. CAGTK) into rat bladder followed by a regimen of intraperitoneal ganciclovir (GCV) injections. Histological examination after a short-term GCV regimen(3 days)revealed marked vacuolization of the tumor cells (**Fig. 2A, 2B**). Moreover, TUNEL assays showed that the cytotoxic reaction was partly mediated by apoptosis(**Fig. 2 C, 2D**). In a long-term experiment, Ad. CAGTK or Adex1CALacZ (a control adenoviral vector containing the lacZ gene)was instilled 25 weeks after the start of BBN administration and GCV was administered for 14 days. In untreated (**Fig. 3A**) and Adex1CALacZ transduced (**Fig. 3B**) bladders, multiple papillomas developed which almost filled the bladder cavity. In contrast, significant inhibition of tumor growth was observed after transduction with Ad. CAGTK followed by GCV treatment (**Fig. 3C**)[2].

This is the first report demonstrating the efficacy of in vivo suicide gene therapy in a chemically-induced bladder tumors. Intravesical instillation is already a well established clinical technique. Our findings strongly encourage the consideration of gene therapy in the treatment of human bladder cancer.

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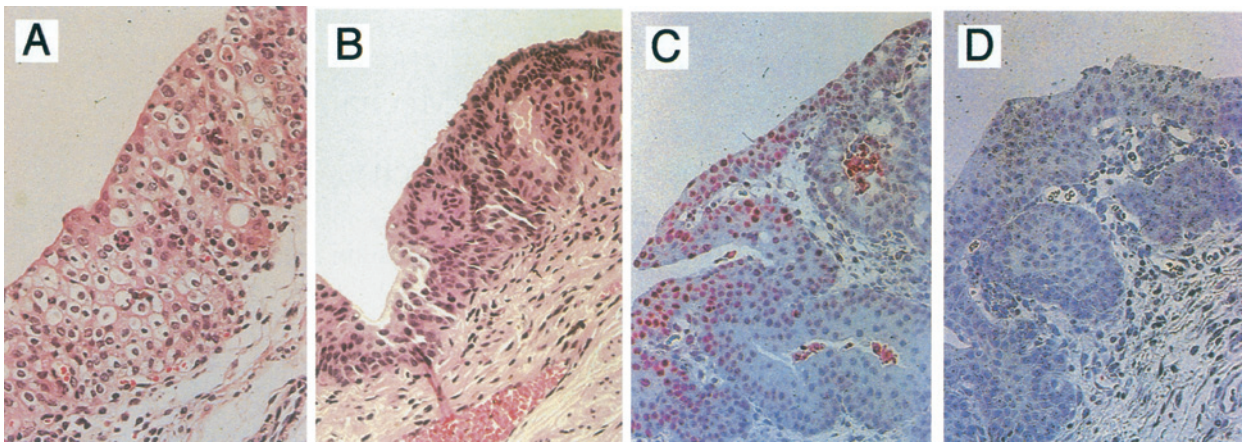

Fig. 2

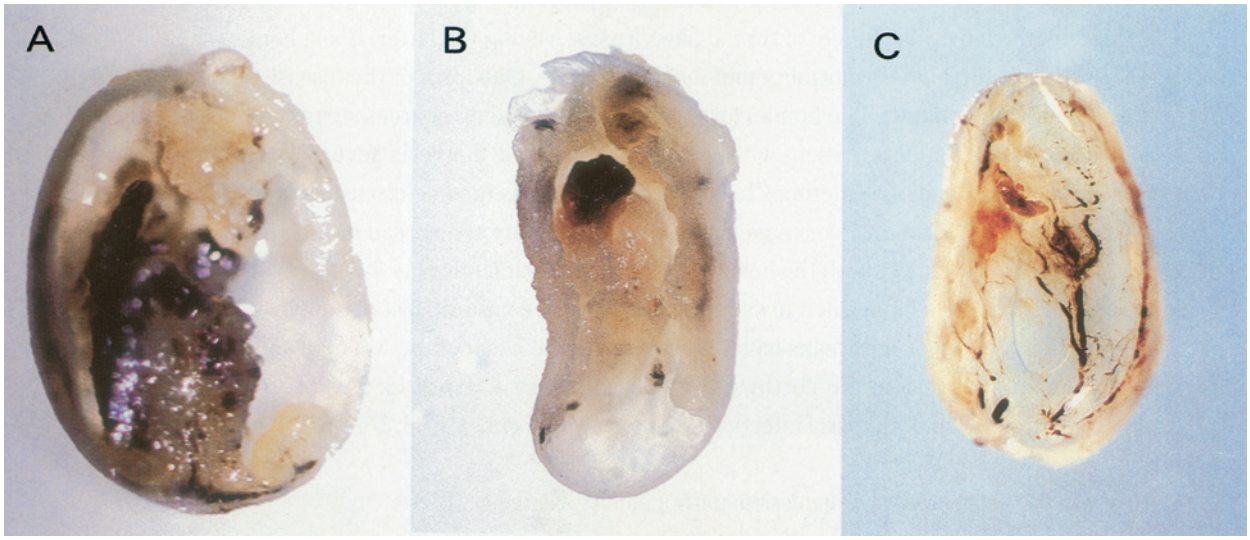

Fig. 3

Fig. 1 Adenoviral mediated gene transfer into BBN-induced rat bladder tumor. Adenoviral vectors (**A**) were transurethrally instilled into rat bladder with (**C**) or without (**B**) BBN induced tumor. Bladder tumor was preferentially transduced (blue stained).
Fig. 2 Histologic analysis of rat bladder immediately after suicide gene therapy. BBN-induced bladder tumor was transduced with Ad. CAGTK (**A**, **C**) or Adex1CALacZ (**B**, **D**) and subsequently treated with GCV. Hematoxylin eosine staining showed marked vacuolization in the tumor after suicide gene therapy (**A**), and TUNEL assays demonstrated that the cytotoxic reaction was partly mediated by apoptosis (red stained) (**C**).
Fig. 3 Bladder tumors two months after completion of suicide gene therapy. Bladders from an untreated rat (**A**), an Adex1CALacZ+GCV treated rat (**B**) and an Ad. CAGTK+GCV treated rat (**C**). Although multiple papillomas developed which almost filled the bladder cavity in control animals (A and B), significant tumor regression and growth inhibition were observed treated rat (**C**).

References

1. Shimizu H, Akasaka S, Suzuki S, Akimoto M, Shimada T: Preferential gene transfer to BBN-induced rat bladder tumor by simple instillation of adenoviral vector. Urology 2001; 57: 579–584.
2. Akasaka S, Suzuki S, Shimizu H, Igarashi T, Akimoto M, Shimada T: Suicide gene therapy for chemically induced rat bladder tumors entailing instillation of adenoviral vectors. Jpn J Cancer Res 2001; 92: 568–575.