Use of Antisense Vectors and Oligodeoxynucleotides in Neuro-Oncology

Introduction

The discovery that complementary fragments of DNA can cause the transcription arrest of selected genes [1, 2] has launched a new field of drug development in which early clinical trials are now proceeding [3-7]. The idea of using antisense-mediated gene inhibition as an alternative to conventional chemotherapy is particularly exciting for malignant brain tumors, since results with standard chemotherapy have been disappointing. The term ‘antisense’ refers to the fact that the nucleic acids synthesized are complementary (in an antiparallel orientation) to the coding (i.e. ‘sense’) genetic sequence of the target mRNA [4, 6, 8]. Two main types of antisense treatment have been employed to date; (1) transfection of cells with antisense cDNA, and (2) treatment of cells with antisense oligodeoxynucleotides (ODNs). Antisense constructs are also used in the laboratory as probes for the detection of specific mRNA sequences in cells or tissue specimens.

In order to be useful therapeutically, an antisense construct must: (1) exhibit stability in the physiologic environment; (2) be taken up and retained by the target cells; (3) specifically bind target mRNA; (4) successfully block expression of the target gene; (5) be free of unwanted toxic and nonspecific side effects, and (6) be easily synthesized in sufficient quantities to facilitate clinical use [4, 9-12]. Antisense therapy is attractive due to its theoretical specificity [12-15], and (to date) relative lack of known adverse effects, particularly when the vector or ODN is administered directly into the CNS [7, 16-19].

Antisense cDNA versus ODNs: Background and Considerations for Use in Neuro-Oncology

Antisense mRNA control was first demonstrated for CoE1, a bacterial DNA plasmid [8, 20]. Posttranscriptional regulation of gene expression using the antisense approach has now been extensively studied. Typically, exogenous antisense cDNA constructs are introduced into cultured cells by plasmid transfection or microinjection. The antisense sequence is then transiently transcribed within the cell from the inserted DNA expression vector. The antisense vector strategy has been successfully used in vitro against glioblastoma cells for gene targets including basic fibroblast growth factor (bFGF), vascular endothelial growth factor (VEGF), insulin-like growth factor 1 (IGF-1), protein kinase C, isotype a (PKCo), the urokinase receptor, transforming growth factor-b1, calmodulin and E2F-1 [21-29].

Often in such studies, antisense-treated and control tumor cells are then implanted subcutaneously or intracerebrally into experimental animals and the growth of antisense-treated tumors is compared to control tumor growth. In this way, antisense-treated cells have been shown to be less tumorigenic than control glioblastoma cells. For true in vivo studies of tumor treatment using an antisense vector, however, the target tumor cells would have to be infected with a replication-defective virus administered to the host animal. In comparison with the cDNA approach, antisense ODNs do not require a viral vector delivery system; they are also easier to synthesize.