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NEW STRATEGIES FOR GENE THERAPY OF A.I.D.S.

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We are developing HIV-based retroviral vectors for gene therapy of A.I.D.S. In comparison with other retrovirus vectors mainly based on the Moloney murine leukemia virus, the use of HIV-1 vectors for the delivery of antiviral sequences has several distinct advantages: (i) the cell targeting, regulated by gp120-CD4 interaction, and (ii) the HIV-specific inducibility of the HIV long terminal repeat. In order to achieve high levels of the transducible gene in the target cells, a number of HIV-1 based vectors have been designed, carrying additional *gag* sequences that might increase packaging specificities. In the packaging system used, the HIV LTR have been replaced by the immediate early promoter of CMV in order to avoid the possibility of helper virus formation. A selectable marker located upstream or downstream of the major splice donor is also included, as well as the *rev-responsive element* (RRE). In addition to being studied for their transduction ability, these vectors are under investigation for their possible selective interference with HIV by production of dominant negative factors.

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Gene Therapy Of Viral Infections: The Use Of Retroviral Vectors Expressing Catalytic And Antisense RNA_S.

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Infections caused by HIV and HBV are affecting millions of people throughout the world and are characterized by viral persistance and chronic clinical progression. The possibility of treating such infections may only rest on a therapeutic approach that is capable of inhibiting viral replication in de novo and chronically infected cells. Since gene therapy could work at both levels, it might represent a therapeutic and prophylactic system to be adopted in healty subjects as well as in already infected individuals. Our attempts have been devoted to the development of retroviral vectors based on the Moloney provirus. Therapeutic genes have been cloned under the control of the vector LTR or within the context of a RNA pol III promoter-terminator unit. The therapeutic genes consisted of antisense and catalytic RNA (hammerhead ribozyme) directed against HIV env, tat and rev sequences and against an HBV DNA pol region wich is required for encapsidation of pregenomic RNA_S into subviral core particles. Constructs were transduced into human T cells that were exposed to HIV challenge, or into hepatoblastoma human cells that support the raplication of HBV DNA and intact virus particles. Different levels of antiviral activity were produced by many of the constructs. Results are considered in the light of the target-effector sequence structure and the strength of the promoter used. A more rational design of therapeutic genes can be envisaged.