rate of 38% [11], placing these patients in danger of exhausting their treatment options [12]. Transmission of drug resistant HIV mutants is also an increasing problem. In a study among newly infected individuals, 14% of patients were infected with HIV that already had one or more key drug resistance mutations [13]. For these reasons, there is an increasing urgency to find a cure for HIV infection.

With the advent of the molecular and genetic age of medicine, research to create gene therapy for HIV has been on the rise. Since the 1980's, researchers have explored the possibility of using gene therapy to cure HIV-positive patients. In 1988, David Baltimore used the term 'intracellular immunization' to describe this treatment approach [14]. Initial *in vitro* experiments were successful and now scientists are applying some of these methods in clinical trials.

Strategies for inhibiting HIV

Figure 1 is a schematic representation of the life cycle of HIV showing the various stages at which genetic therapy could be applied. Therapy could also be aimed at any one of the many target cells for HIV infection *in vivo*, including immune cells such as $CD4^+$ and $CD8^+$ T cells, dendritic cells, monocytes, macrophages, hematopoietic stem cells (HSCs), brain cells, and other cells from the gastrointestinal tracts that could serve as host cells for HIV. Since T cells are the major cell population implicated in HIV infection and its progression to AIDS, making these cells immune to infection is a very important aspect of therapy. Even more desirable are the HSCs. These self-replicating progenitor cells give rise to all other members of the lymphoid and myeloid lineages and have the capability of repopulating the immune system with a potentially HIV-resistant phenotype.

A variety of viral or cellular components could serve as targets for anti-HIV gene therapy. Targeting viral factors is currently the most prevalent method. A major problem with this strategy is that HIV can quickly form resistant strains to these genetic modifications due to high mutation rates. Targeting cellular factors makes the occurrence of resistant strains less likely but raises the issues of adverse effects on normal cell function. HIV receptors and co-receptors are attractive targets, but there have been recent reports of liver toxicity with CCR5 antagonists in HIV-infected patients [15]. A summary of the main antiviral approaches is as follows:

Protein-based strategies

A. Introduction of suicide genes

Cells modified with suicide genes for negative selection. Generally, suicide genes code for enzymes that convert an inactive drug to a toxic form, allowing for the potential killing of the modified cells. The idea of using suicide genes was made popular as a potential approach for treating cancer. As an anti-HIV strategy this method was tried *in vitro* as proof-of-concept in a study that used a retrovirus to transduce autologous CD8+ cells from HIV-infected patients; the suicide gene was thymidine kinase expressed as a fusion protein with hygromycin phosphotransferase [16]. As with other gene therapy approaches, specific targeting of desired HIV-infected cell populations would present a challenge for this approach to be successful *in vivo*.

B. Transdominant negative proteins

Mutations in a specific gene expressed in a dominant fashion where the mutant protein can interfere with the function normally carried out by the parent gene product. If the protein is multimeric, the nonfunctional protein can multimerize with the normal protein and the resultant complex is functionally inactive. Thus, the dominant negative protein can have a strong inhibitory effect on the normal protein formation or function. Examples of this are HIV-1 Gag mutants [17] and Rev M10 [18]. Rev M10 was the first transdominant negative protein used in clinical trials; it is a dominant negative mutant capable of binding the Rev Responsive Element (RRE) and has the capability of forming multimers [18].

C. Chimeric receptors

CD4/CD3 chimeric receptor called CD4ζ. The CD4ζ has the extracellular and transmembrane domains of the CD4 antigen and the intracytoplasmic domain of the CD3 T cell receptor. The extracellular portion binds HIV, while the cytoplasmic portion initiates a signaling cascade similar to the one initiated by the normal T-cell receptor (TCR) binding with HIV [19]. Thus, engagement of CD4ζ with HIV results in the generation of an HIV-specific T cell response. CD4ζ-modified T lymphocytes inhibit viral replication in T cells and macrophages *in vitro* [20] and mediate killing of HIV-infected T cells [19].

D. Intracellular HIV-1 specific single chain antibodies (SFv)

These antibodies bind to viral proteins intracellularly and block their action, e.g. anti-Rev SFv [21] and anti-gp120 SFv [22].

RNA-based strategies

A. RNA decoys

Small RNA molecules containing essential *cis*-acting elements that bind *trans*-acting proteins. They function by luring away *trans*-acting proteins from their true target sequence. When expressed at high levels, they can successfully compete against viral *cis*-acting sequences that are indispensable for viral replication [23,24].