GENE THERAPY

Principle, Strategies &Ethics related to Human Gene Therapy

INTRODUCTION

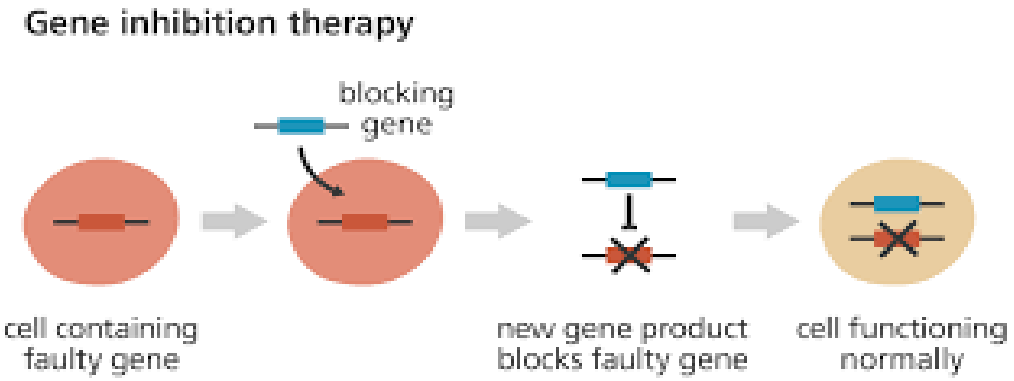
Progress in molecular biology technique has made possible the development of various strategies for the transfer of the exogenous gene to mammalian cells.

History: At approximately noon, on Sep. 14th,1990 history was made when a billion gene altered cells dripped down a plastic tube and into a vein of 4 year old girl named *Ashanti*. She had an inherited immune deficiency that left her unable to withstand microbial infection. Now, she is provided with the army of cells bearing the oritical genes she lacked. With that infusion of cells, researchers began the first federally approved use of gene therapy in a patient and by administrating the cells, medical practitioners took the first tentative steps into a new arena of DNA technology.

Definition: Gene therapy may be defined *as the introduction of a normal functional gene into cell of patient which contains the defective allele of concerned gene with the objective of correcting a genetic disorder or an acquired disorder.*

Or

in simple words, gene therapy may be defined as *the introduction of normal genes to the cell of a patient to replace defective gene failing to encode an essential protein and it is one of the most promising aspects of the use of gene technology in medicine.*



Ref. By: http://www.yourgenome.org/facts/what-is-gene-therapy

MECHANISM OF GENE THERAPY

The general mechanism of gene therapy involves basic steps -

1. The approximately 100000 genes in a cell encode the many proteins used by the body.

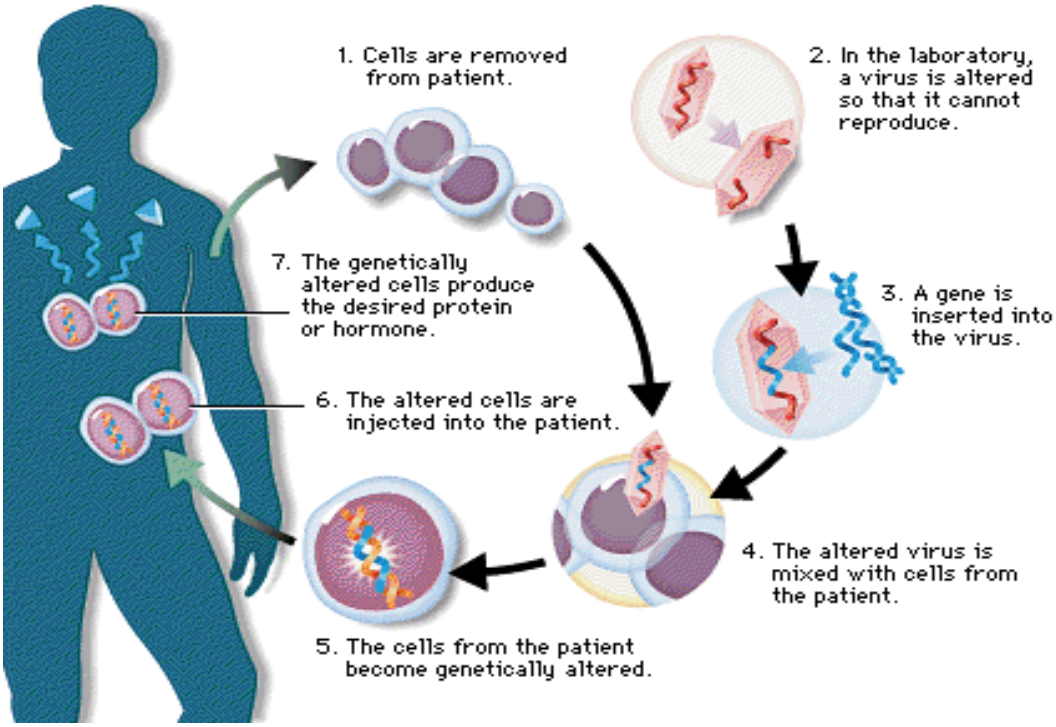
2. Each gene is a segment of the DNA molecule in a chromosome.

3. Researchers used a virus whose DNA naturally inserts into human DNA.

4. They splice a human gene into the viral genome.

5. Place the modified virus into the human respiratory tract.

6. The viruses invade the cells, carrying along the gene. The latter encodes proteins to provide relief from the disease.



Mechanism of Gene therapy, *Image Copyright Microsoft Encarta Online Concise Encyclopedia*

REQUIREMENT FOR GENE THERAPY

There are several requirements for a gene therapy protocol to be effective. Firstly, the gene defects itself will have been characterized and the gene cloned and available in a form suitable for use in a clinical program. Secondly, there must be a system available for getting a gene into a correct site in the patient. A large number of approaches have been tried for effective transfer of gene to appropriate target site.

These approaches fall into 4 categories:

1. Gene Modification- Gene modification can be accomplished by replacement theory or corrective gene therapy. Before giving any of the therapy, we should be very clear our goal replace or corrective. In replacement therapy, an effective gene is inserted somewhere in the genome so that its product could replace that of a defective gene.

On the other hand, corrective gene therapy requires replacement of a mutant gene or a part of it with a normal sequence. Another form of a corrective therapy involves the suspension of a particular mutation by a transfer RNA that is introduced into a cell.

2. Gene transfer/ Gene delivery- The gene transfer into cell can be brought about by physical, chemical, and biological methods. Gene delivery for gene therapy is achieved by one of the following two methods-

2.1. Viral transduction

2.2. Physical transduction.

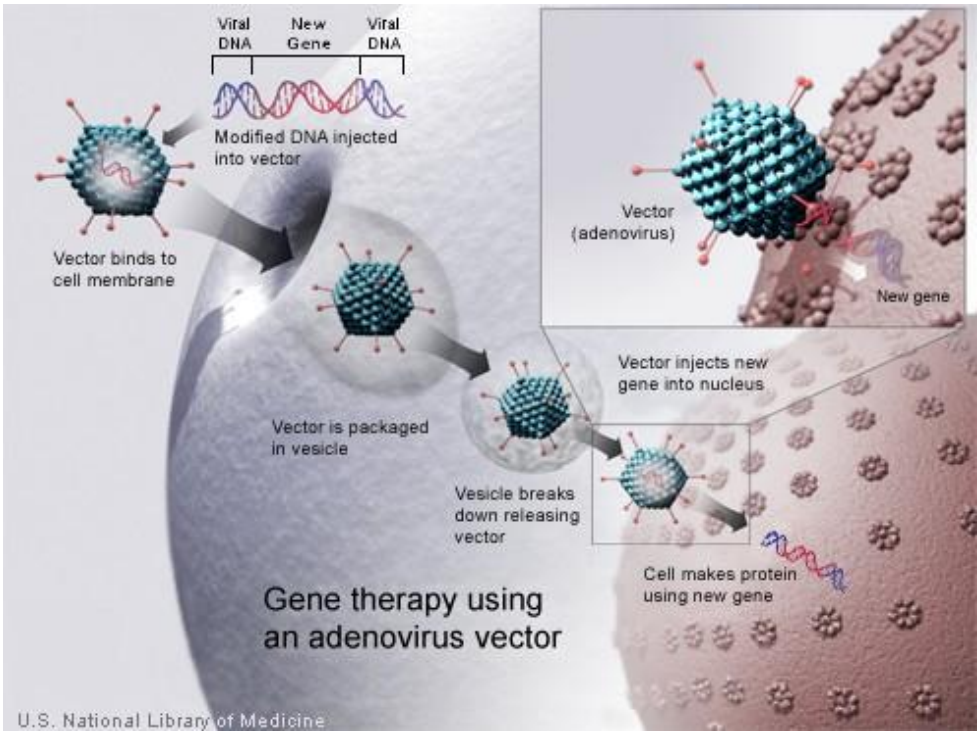
To delivered genes to somatic cell researchers use some type of carrier molecule called a vector. For example, in gene therapy the most commonly vectors used is a virus , a type of virus called retrovirus. Three main viral system have been developed for gene therapy protocols-

1. Retrovirus

2. Adenovirus

3. Adeno associated virus

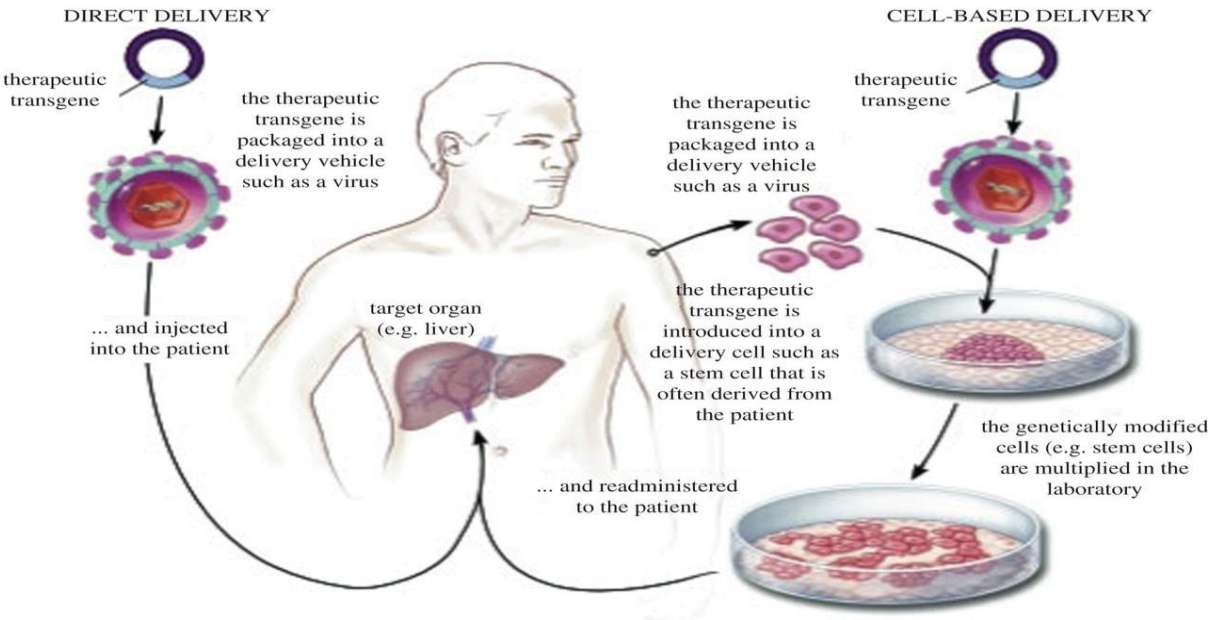
Other recombinant viral vector systems are also used like Herpes Symplex virus, Parovirus, Vaccinia virus.



Reference by:https://en.wikipedia.org/wiki/Gene_therapy

Gene delivery ways- There are two ways of gene delivery in somatic cell –

Exvivo* and *Invivo

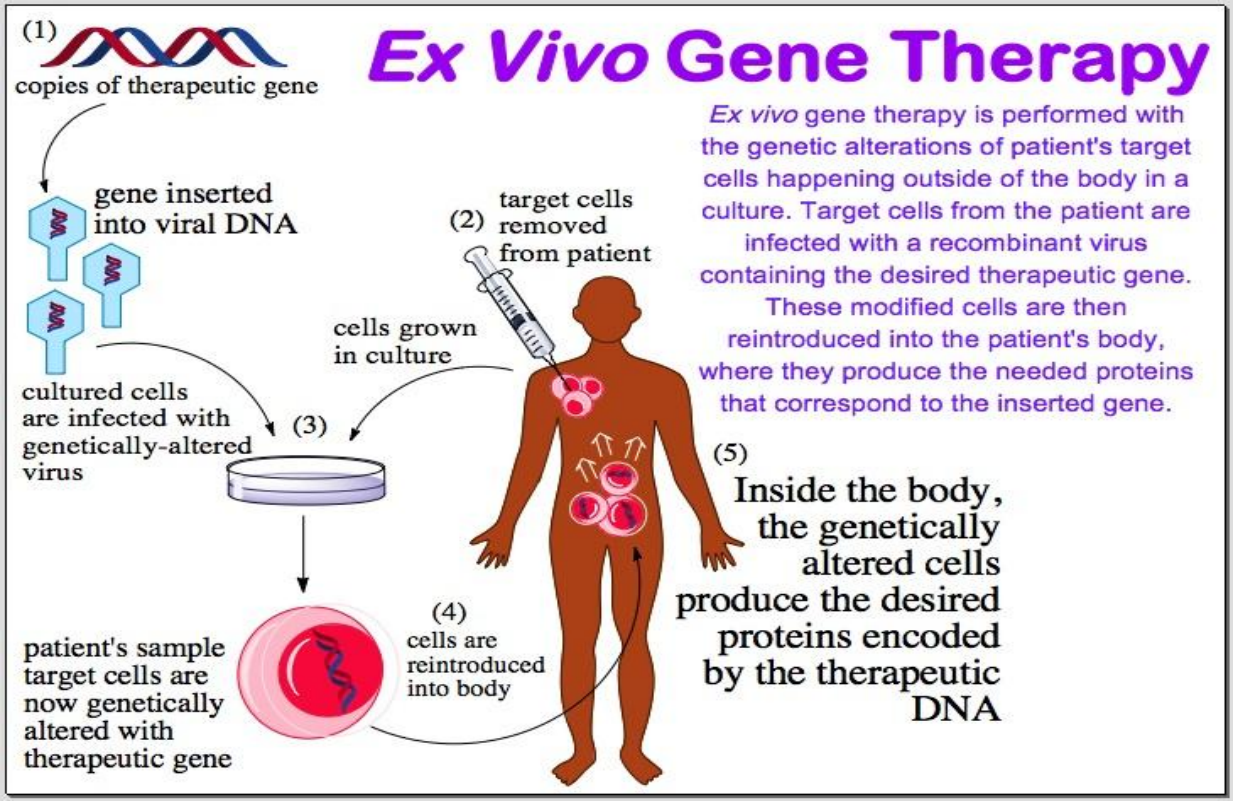


Ref. By: http://rspb.royalsocietypublishing.org/content/282/1821/20143003

Invivo(direct) and *Exvivo* (cell-based) gene therapy

1. *Exvivo*_–

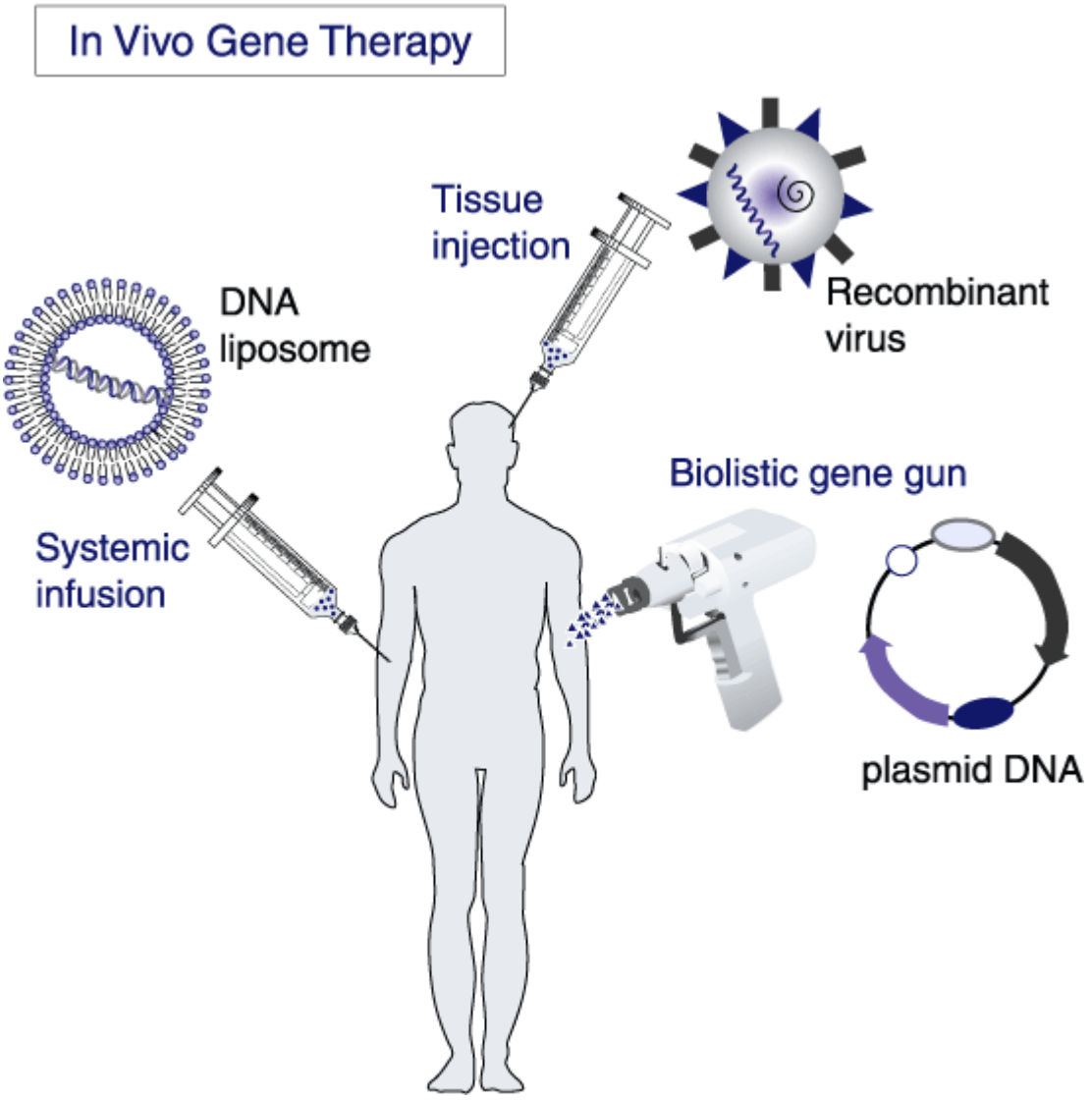
In this system, cells are removed from the patient and grown in culture medium. The transgenes is therefore introduced into the cell outside the body. Modified cell can be selected and amplified before injecting back into the patient.



Refd. By: http://gene-therapy.yolasite.com/process.php

2. *Invivo*-

Where genes are changed in cell still in the body. Recombination based approaches *in vivo* are especially uncommon because for most DNA constructs recombination has a very low probability.



Ref. by: http://istudy.pk/gene-therapy/

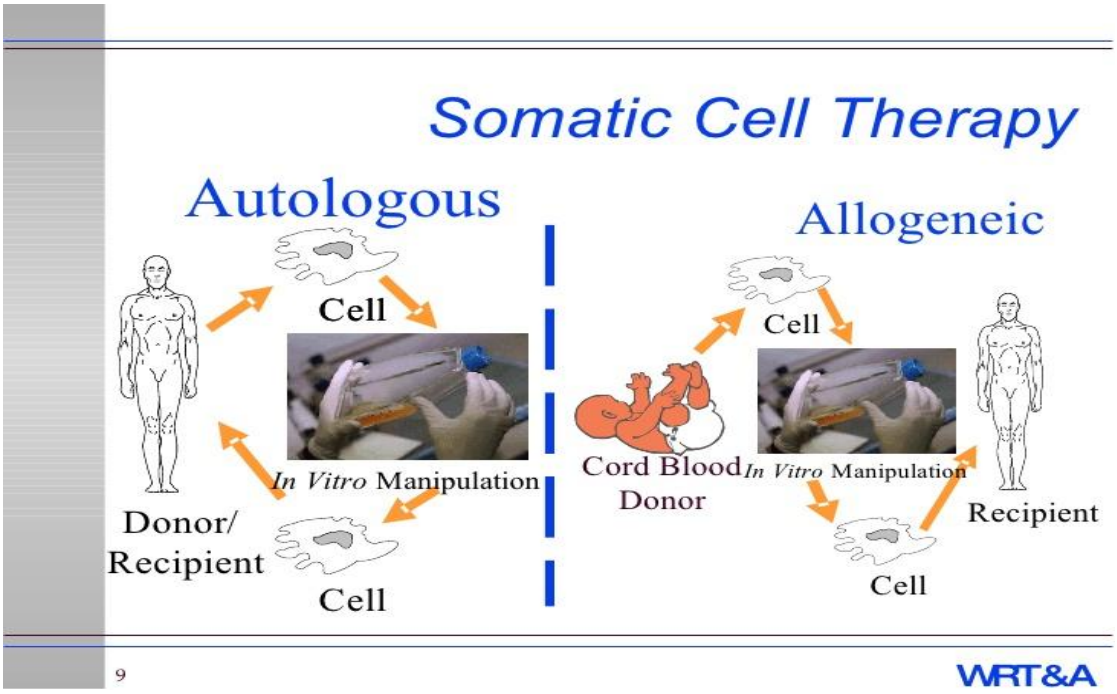
3.Gene therapy to specific cell lines –

Gene therapy can be classified into two specific types on the basis of cell line type-

1. Somatic cell gene therapy

2. Germ line gene therapy

1. Somatic cell gene therapy : which has emerged as a new approach for the treatment of a variety of genetic and acquired disease. In this gene is introduced only in somatic cells especially of that tissue in which expression of the concerned gene is critical for health.



Ref.By: https://www.slideshare.net/wrtolbert/gene-therapy-cell-therapy-stem-cells-regulations-for-the-quotnew-biologicsquot

The somatic cell gene therapy is again divided into 2 groups on the basis of end result of the process-

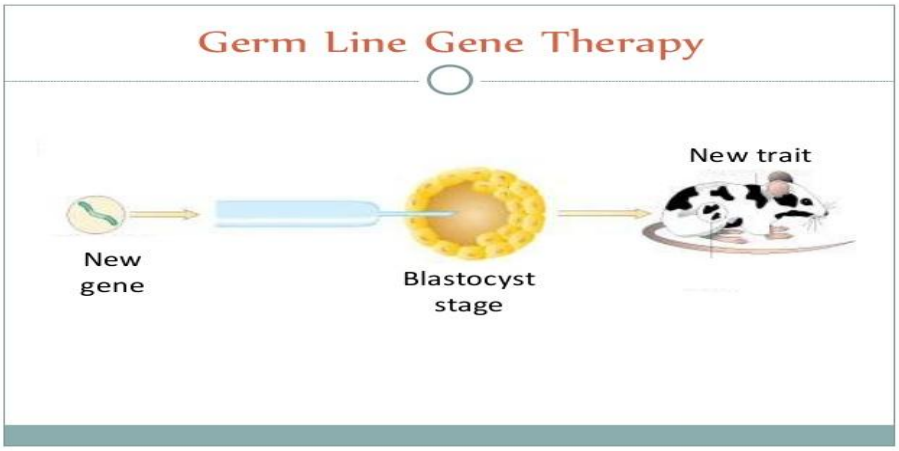
Augmentation gene therapy and Targeted gene therapy.

Augmentation gene therapy: In this gene therapy, the functional gene is introduced in addition to the defective gene endogenous to the cells. So the modified cell contains both functional & normal as well as the defective (endogenous) copies of gene.

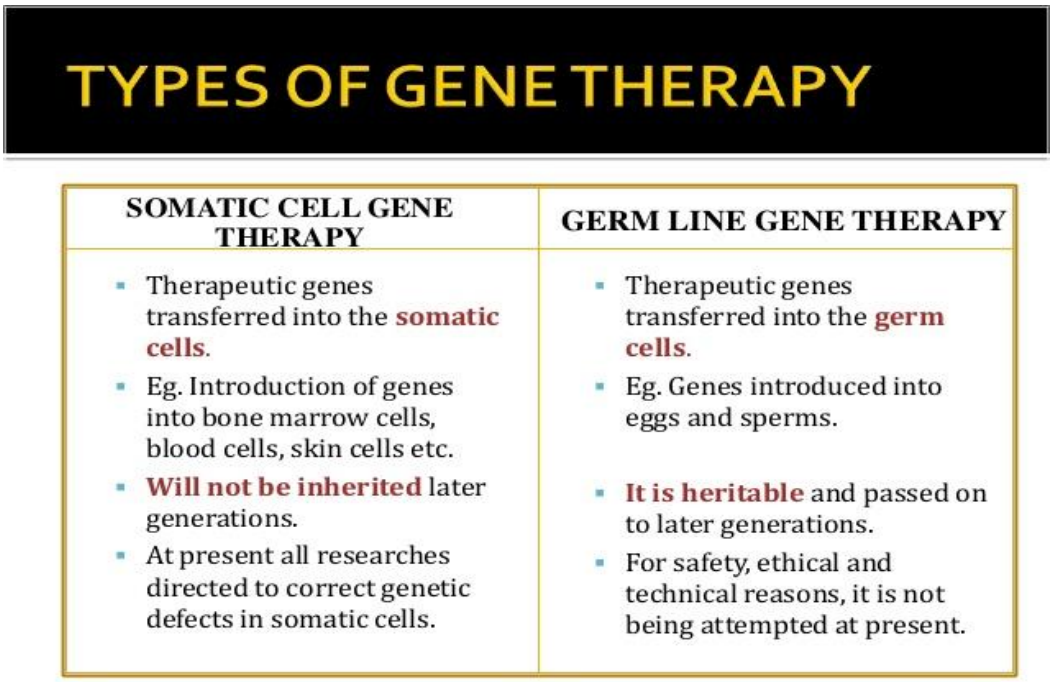
In first approach it was used for the treatment to correct the genetic disorder SCID (Severe Combined immuno Deficiency). The second approach is the direct injection of DNA into the tissue either as protein complexes or even as naked DNA into mucles/skin.

Targeted gene therapy: It uses homologous recombination to replace the endogenous gene with functional introduced gene. Two vectors are used in this case: insertion vector and replacement vector.

2. Germ line gene therapy: In germ line therapy, germ cells i.e, sperm /eggs are modified by the introduction of functional gene which is ordinarily integrated into the patient's genomes. Therefore changes due to therapy are heritable and passed on to later generation and this approach theoretically is highly effective in counteracting the genetic disorder.



Ref.By: http://fbme.utm.my/ongsimei/2016/05/10/types-of-gene-therapy/



Difference between Somatic cell and Germ line cell gene Therapy.
Ref.by:https://www.slideshare.net/damarisb/gene-therapy-27039196

4. Gene therapy and Diseases:

Human being suffers from more than 5ooo different diseases caused by a single gene mutation. Diseases resulting from single gene mutation are potentially good candidate for gene therapy. A functional copy of gene would need to be introduced in the cell together with elements to controls its expression leading to production of missing protein and restoration of function.

Gene therapy found to be effective in many diseases as – cystic fibrosis, cancer, insulin dependent diabetes, hemophilia B, cardiovascular diseases etc.

Gene therapy- deaths and problems

After almost of a decade of successful implementation of gene therapy in a 4 year child, in the year of 1999 a solitary case of death of 18 year old patient (Jesse Gelsinger) at the University of Pannsylvania's Institute of Human Gene Therapy was noticed. The patient died on 17 sep. 1999, four days after the genetically altered virus was injected into the liver. Patient was suffering from heritable disease of liver.

Problems associated with gene therapy:

1. Short lived nature of gene therapy
2. Immune response
3. Problem with viral vectors
4. Multigene disorder
5. Chances of inducing tumor.

Safety Assurance/ethics and future of gene therapy:

To calm excessive fear about gene therapy and to ensure proper societal safeguards, a multilayered review system is now in operation for scientific proposals. This system also provides opportunity for public input into scientific endeavors.

Since 1984, a multilayered review system for gene therapy experiment has been evolved. Human gene therapy proposals need approval from the food & drug administration and since 1997, comment from RAC (Recombinant DNA Advisory Committee) of NIH. The RAC has established a special working group consists of 4 lab scientists, 3 clinical scientists, 3 specialist in ethics and experts in public policy.

Human gene therapy sub-committee has prepared a document committed “points to consider” for research planning submit proposals for human gene therapy research.

It is clear that the operative recommendation for gene therapy has been proceed but with caution.

Future:

Gene therapy may be long term answer to the problem of genetic diseases. By 1999, about 1000 americans were enrolled in clinical trials in involving gene therapy, yet physicians lack an ambiguous proof that gene therapy had cured of their patient.

Nevertheless, W. French Anderson, the NIH research at the forefront of gene therapy quoted in 1995 saying...

...20 YEARS FROM NOW,GENE THERAPY WILL HAVE REVOLUTIONIZED THE PRACTISE OF MEDICINE. VIRTUALLY, EVERY DISEASE WILL HAVE GENE THERAPY H AS ONE OF ITS TREATMENT. STAY TUNED.....