International Journal of Research Publication and Reviews

Journal homepage: www.ijrpr.com ISSN 2582-7421

Gene Therapy in Diabetes Mellitus

Mr. A. Jegath Ratchagan[1], Dr.P.Udhayakumar[2]

[1]B. Pharm Final Year Student,(P.S.V College Of Pharmaceutical Science And Research, Krishnagiri)
[2]Assitant Professor,(Psv College Of Pharmaceutical Science And Research , Krishnagiri)

Abstract

Diabetes mellitus (DM) is a chronic metabolic disorder that affects millions throughout the globe, defined by defective insulin resistance or secretion. Although significant progress has been achieved in the field of insulin therapeutics and pharmacological intervention, a cure is still lacking. Over the past few years, gene therapy has been considered as a revolutionary strategy to correct the genetic and molecular abnormalities of diabetes, rather than addressing the symptoms. The present review aimed to assess the current status of gene therapy for DM, with particular attention to its applicability for restoration of endogenous insulin production, improvement of β-cell function, and long-term glycemic control. In addition, challenges associated with clinical translation such as immune responses, delivery systems, and ethics are also discussed. We did not rely on databases such as ClinicalTrials, but conducted a thorough bibliographic search using databases like PubMed, Scopus and so on. gov that are related to preclinical investigations, clinical trials, or novel gene editing methodologies (e.g., CRISPR-Cas9, viral/non-viral vectors). Here, selected studies from the last decade were reviewed looking at efficacy, safety and potential future directions. Viral vector mediated therapies (AAV, lentivirus) have also been successful in animal models in generating glucose-responsive insulin secretion. CRISPR-Cas9-mediated gene editing has shown promise in the correction of diabetes--associated mutations (e.g.: GCK, PDX1) and in prevention of destruction of the β-cells by autoimmunity. β-cells derived from stem cells along with gene therapy have normalized blood glucose levels in early human studies. Challenges include immune rejection, off-target effects and the need for targeting delivery systems. Gene therapy provides a paradigm shift in treatment of diabetes with the promise of durable insulin independence. Substantial strides have been made thus far, but additional work remains to be done to improve delivery vehicles while minimizing immune responses and long-term safety issues. Future directions include personalized gene editing, ex vivo cell engineering, and biomaterial-enhanced transplantation.

Keywords: Gene therapy, diabetes mellitus, insulin secretion, CRISPR-Cas9, viral vectors, β-cell regeneration, stem cell therapy, immunotherapy.

1. Introduction :

Diabetes mellitus, is a public health emergency currently affecting 537 million adults in 2021, and is projected to rise up to 783 million in 2045. The disease is divided in two major types: Type 1 (T1D) - where the pancreatic β-cells is destructed by the immune system - and the Type 2 (T2D), in which there is insulin resistance and β-cell dysfunction. Current treatments, including exogenous insulin injection and glucose-lowering drugs, are lifelong treatments and do not prevent progression of disease. Gene therapy represents one such transformative approach by correcting the genetic defects, stimulating insulin production in non-βcells or shielding residual β-cells against an autoimmune attack. This review summarizes most recent progress in gene therapy of diabetes with respect to delivery and therapeutic targets and clinical translation.

2. OVERVIEW OF GENE THERAPY IN DIABETES MELLITUS :

For diabetes, there is huge potential for gene therapy — not just treating the symptoms but going after the source problem. These cells are responsible for producing insulin in response to changes in blood sugar levels, and when they are damaged or lost—as is the case with type 1 diabetes—they can no longer carry out that function naturally.… Diabetes, caused by insufficient insulin production (type 1) or a resistance to insulin (type 2), requires daily management, but gene therapy could provide lasting or even permanent support. For T1D, in which the body's own immune system attacks the pancreatic beta cells to destroy the insulin-producing cells, gene transfer technologies are being developed that are applicable to replacing functional beta cells or protecting them from immune attack. And among the approaches, we have introducing genes into the pancreas that promote beta-cell regeneration, such as PDX1 or Neurogenin-3 or implanting genes that make insulin into cells that are not beta cells, the liver or muscle cells, and then they're able to release insulin in response to glucose. Another approach uses gene-editing techniques such as CRISPR-Cas9 to fix abnormal genes or bring about immune tolerance and prevent the autoimmune attack of beta cells. In Type 2 diabetes (T2D), gene therapy focuses on reversing insulin resistance and beta cell dysfunction. Such strategies could include stimulation of insulin sensitivity through the up-regulation of genes such as GLUT4 to promote glucose uptake by muscle or fat, and the regulation of genes such as those involved in inflammation or metabolism, such as PPAR-γ. In addition, gene therapy may be directed toward maintenance or regeneration of beta-cell function by the introduction of genes that promote cell survival or proliferation. Yet gene therapy for diabetes faces some obstacles such as the efficacy of delivery, immune responses of the body against viral vectors, and safety in the long-run. Accurate gene regulation needs to be maintained, to avoid hypoglycemia or off target effects. In order to tackle the

first step, that is, accurate tumour visualization, microbubbles have been developed and continue to be evaluated in clinical tests [98]; some of these preclinical studies have been promising, but they have not been applied to humans except in a few tests. If successful, the gene therapy has the potential to revolutionise the treatment of diabetes by offering a long-term cure and reducing the reliance on insulin injections or immunosuppressant medicines. However, additional studies are needed for optimization of the delivery system, demonstrate its safety and ensure consistent therapeutic efficacy.

- ❖ Restore physiological insulin production (critical in Type 1 diabetes).
- ❖ Improve insulin sensitivity (key in Type 2 diabetes).
- ❖ Protect or regenerate pancreatic β-cells from autoimmune destruction or dysfunction.

3. TYPES OF DIABETES MELLITUS :

3.1. Type 1 Diabetes (T1D) :

Type 1 diabetes mellitus (T1D) is a chronic autoimmune disease involving destruction of insulin-producing beta cells in pancreatic islets of Langerhans, leading to absolute insulin deficiency. Unlike Type 2 diabetes, which is primarily caused by insulin resistance and lifestyle, T1D is due to the immune system mistakenly attacking β-cells in the pancreas and may be caused by a combination of genetic factors and environmental factors such as viral infection (e.g. could include enteroviruses) or dietary compounds. Onset of the disease often develops in childhood or adolescence (hence it is also called "juvenile diabetes"), but the disease can occur in all age groups. The damage that the immune system does to beta cells is mediated by T-cell driven inflammation when the immune system mistakenly attacks insulin and other proteins that beta cells make, such as glutamic acid decarboxylase (GAD) and insulinoma-associated antigen-2 (IA-2). With the reduction in the number of beta cells, the pancreas slowly loses the ability to make insulin, a hormone necessary to help get glucose into cells. In the absence of insulin the blood glucose rises (hyperglycemia) leading to predictable symptoms using the 3 P's (polyuria, polydipsia, polyphagia). Sounds good, right? Well, disastrous if not handled responsibly, since severe insulin deficiency can cause diabetic ketoacidosis (DKA): a life-threatening condition marked by the accumulation of ketones, metabolic acidosis, and dehydration. T1D is treatable but the need to keep blood sugars continuously within a targeted range — a challenging task even for the most educated and diligent of patients and their careworkers — can be a struggle. Despite advances in insulin analogs and automated insulin delivery, tight glycemic control is challenging; there are potential dangers of hypoglycemia (low blood sugar) and long-term complications such as retinopathy, neuropathy, nephropathy and heart disease. Potential alternate approaches include replacing beta cells (by pancreas or islet transplantation) and strategies to modulate the immune response to halt the autoimmune attack. In addition, new technologies including beta cells derived from stem cells and gene therapy are under investigation for the restoration of insulin production. Although there is no cure for T1D at present, the potential for future therapies and preventive strategies is bright, fueled by efforts to achieve immune tolerance and to further regenerative medicine.

3.2. Type 2 Diabetes (T2D) :

Type 2 Diabetes Mellitus (T2D) is a chronic metabolic disease characterized by insulin resistance and progressive beta cell failure leading to elevated blood glucose levels (hyperglycemia). Unlike type 1 diabetes, where the onset of disease is due to autoimmune-mediated destruction of insulin-secreting beta cells, T2D results from a combination of genetic, lifestyle and environmental factors including obesity, physical inactivity and unhealthy eating (Andersen et al., 2017). While the disease predominantly strikes adults, escalating obesity levels have seen more cases in younger people. In T2D, because muscle, liver and fat cells do not respond appropriately to insulin, a condition called insulin resistance, then pancreas needs to secrete additional insulin to keep blood sugar levels within the normal range. Beta cells become worn out over time and are not able to satisfy the body's need for insulin, thus resulting in defective insulin secretion. Chronic high blood sugar leads to other metabolic issues, such as poor glucose uptake and the liver producing too much glucose. Early symptoms can include increased thirst (polydipsia) and frequent urination (polyuria), along with fatigue and blurred vision – yet some people will have no symptoms at all for a while, leaving them unaware of their diagnosis for years. Uncontrolled T2D can result in serious long-term complications like cardiovascular disease, neuropathy (nerve damage), nephropathy (kidney disease), retinopathy (vision loss), and an increased risk of infections. Treatment focuses on lifestyle changes, which all patients need, including adopting a healthy diet, increasing physical activity levels and maintaining healthy body weight, in addition to medications such as metformin (which enhances insulin sensitivity), sulfonylureas (which induce insulin secretion) and more recently introduced agents such as GLP-1 receptor agonists and SGLT2 inhibitors which improve glucose control and offer cardiovascular and kidney benefits. In more serious cases insulin might be required. Current research will finally unravel the basic mechanisms of insulin resistance and beta-cell failure, with newer therapeutic targets being inflammation, gut microbiota, and even cellular metabolism. Although T2D is preventable and manageable with lifestyle modification and pharmacotherapy, its increasing worldwide prevalence emphasizes the need for early diagnosis, public health measures, and personalized treatment strategies to reduce morbidity and improve the quality of life.

3.3. Gestational Diabetes (GDM) :

Gestational Diabetes Mellitus (GDM) is the presence of high blood sugar if a mother had not been diabetic prior to her pregnancy and it first occurs during the second or third trimester of her pregnancy. It occurs when the hormonal changes of pregnancy, namely the rise in placental hormones such as human placental lactogen (hPL) and progesterone, cause insulin resistance, which can affect the body's ability to use insulin as it should. While most pregnant women can compensate by making more insulin, some cannot, which causes hyperglycemia. GDM affects 2-10% of pregnancies and is more common in women who are overweight, have a family history of diabetes, for those of certain ethnic backgrounds (ie Hispanic, African American, South Asian and Native American). The diagnosis is usually made by OGTT between 24 and 28 weeks of gestation. If not controlled, GDM can put mother and baby at risk; these risks include macrosomia (overly large baby during the pregnancy), increasing the rate of complications at delivery, such

as shoulder dystocia, and neonatal hypoglycemia; also, there is an increased probability of delivering via cesarean section. Women with GDM are also at an increased risk of developing preeclampsia, and have a significantly higher chance of developing Type 2 diabetes later. For the child, ongoing effects may include adult obesity and metabolic disruption. The treatment of GDM aims to control the blood glucose within the desired range with diet modification, regular exercise, and regular blood glucose measurements. When lifestyle modifications are insufficient, you will very likely require treatment, either with insulin or oral hypoglycemic agents like metformin. In the majority of cases, the condition disappears following delivery although women who have experienced GDM require regular testing for Type 2 diabetes, as they have a 50 per cent greater likelihood of having the condition within 5-10 years. These risks may be reduced through preventive measures such as weight control, healthy diet and physical activity. Early detection and proper treatment of GDM is key to a healthy pregnancy and to prevent metabolic disturbances in mother and child in long term.

3.4. Other Specific Types :

A. Monogenic Diabetes :

Monogenic Diabetes is a form of diabetes caused by a modification in one gene and constitutes approximately 1Ð2% of all diabetes cases. Unlike type 1 and type 2 diabetes, which are associated with multiple genes and environmental factors, monogenic diabetes is a result of a critical mutation(s) inherited or acquired in a gene that possesses insulin production or insulin secretion function. The two common subspecies were Maturity-Onset Diabetes of Young (MODY) and Neonatal Diabetes Mellitus (NDM) having consistent genetic aetiology, modes of inheritance and clinical profiles. Type of diabetes: MODY often develops during adolescence or early adulthood (usually before developmental age 25) and is often mistaken for either type 1 or 2 diabetes. The most common is MODY 2, caused by mutations in the GCK gene, which produces mild, persistent hyperglycemia not usually requiring treatment. MODY 3 due to HNF1A gene mutations results in progressive insulin deficiency that is highly treatable with sulfonylureas and MODY 1 (attributable to HNF4A mutations) itself can cause transient neonatal hypoglycemia and eventual diabetes. MODY unlike polygenic diabetes, follows an autosomal dominant mode of inheritance which gives a 50% probability of the mutant gene to be passed on to the offspring by the affected parent. Neonatal Diabetes Mellitus (NDM), that becomes manifest before 6 months of life, is further subdivided into transient (TNDM) and permanent (PNDM) forms. Genes such as KCNJ11 and ABCC8 (that affect the pancreatic ATP-sensitive potassium channel) seem to be the usual suspects and many times these mutations allow a patient to switch from insulin to oral sulfonylureas with much better glycemic control. Others (e.g., caused by INS or EIF2AK3 mutations) may require life-long insulin treatment. A crucial role of genetic testing is the accurate diagnosis, which not only provides guidance in developing an individualized treatment, but also allows for the detection of family members at risk. Monogenic diabetes is frequently undiagnosed as it is not common and can present with similar signs to other forms of diabetes. Diagnostic markers are the positive family history, lack of autoantibodies (Type 1 diabetes excluded) and lack of obesity or metabolic syndrome (in contrast to Type 2 diabetes). A timely genetic testing allows avoiding useless use of insulin, refining therapeutic choice and prognostication. Continued research is identifying new monogenic forms, optimizing precision medicine strategies for diabetes therapy. Detection of these cases improves the fate of individual patients and the knowledge of beta-cell biology and insulin regulation.

B. Secondary Diabetes:

Secondary diabetes refers to diabetes mellitus developed as a result of another health condition (such as chronic pancreatitis or cystic fibrosis), the use of certain medications (e.g. high-dose use of aspirin), a chemical (environmental toxin) or some other factor (e.g. drinks high in sugar content). Unlike primary diabetes, secondary diabetes is associated with identifiable aetiologies such as endocrine diseases and pancreatic diseases, genetic syndromes or drug-related metabolic perturbations. Chronic pancreatitis or pancreas damage from cystic fibrosis would also be secondary causes of the disease, as they would damage beta cells and hamper insulin production. No less important are endocrineopathies, e.g. Cushing's syndrome (increase of the cortisol level), acromegaly (increase of the growth hormone level), and hyperthyroidism, that may cause insulin resistance as well as hyperglycemia through distortion of the hormonal equilibrium. Genetic disorders, such as Down syndrome, Turner syndrome, and Prader-Willi syndrome, are also associated with higher risk of diabetes resulting from metabolic equilibrium and hormonal imbalances. Drugs constitute a further important cause for secondary diabetes. Prolonged use of glucocorticoids such as prednisone is a well-documented cause, as they promote gluconeogenesis and reduce insulin sensitivity. Other drugs like atypical antipsychotics (olanzapine), immunosuppressant (tacrolimus), and HIV protease inhibitors can also affect glucose metabolism. Furthermore, since the pancreas cannot currently be lentivirally transduced in humans, with the consequent pancreatectomy being necessary, diabetes would be induced by the loss of insulin producing cells. Secondary diabetes has similar clinical features as Type 2 diabetes including symptoms such as polyuria, polydipsia, and fatigue, although the onset may occur with the underlying disease or drug. Diagnosis involves identifying hyperglycemia on the basis of a particular risk factor, and often requires targeted investigations (e.g., hormone measurements for endocrine causes, or genetic testing for syndromic causes). Management is directed at treatment of the underlying disease and also at therapy of hyperglycemia with lifestyle modification, oral hypoglycemics, or thera- peutic insulin, depending on the severity of the presentation. Drug-induced diabetes might be reversible by discontinuing or reducing the dose of the offending drug. Secondary, but not primary diabetes, may sometimes be reversible if precipitating factors leave, which emphasizes the importance of proper diagnosis and individual treatment. Early diagnosis and treatment can prevent complications and improve the long term outcome of the sufferers.

Resulting from the conditions:

- Pancreatic diseases (chronic pancreatitis, cystic fibrosis)
- Endocrine disorders (Cushing's syndrome, acromegaly)
- Medication-induced (glucocorticoids, antipsychotics)
- Genetic syndromes (Down syndrome, Turner syndrome)

TYPE	CAUSE	WHO GETS IT?	TREATMENT
T1D	Immune attack on pancreas	Kids/Young adults	Insulin for life
T2D	Insulin resistance	Adults(Increasingly kids)	Diet, Pills, later insulin
GDM	Pregnancy hormones	Pregnant women	Diet, temporary insulin
MODY	Genetic mutation	Teens/Young adults	Gene specific meds

4. PATHOPHYSIOLOGY OF DIABETES MELLITUS :

4.1. Type 1 Diabetes Mellitus (T1D) :

Key Mechanism: Autoimmune destruction of pancreatic β-cells which leads to Absolute insulin deficiency

Detailed Process :

1. Genetic Predisposition:

- Associated with HLA class II genes (DR3, DR4)
- Other susceptibility genes (INS, PTPN22, CTLA-4)

2. Environmental Triggers:

- Viral infections (enteroviruses, rubella)
- Early diet (cow's milk exposure)
- Vitamin D deficiency

3. Autoimmune Attack:

- T-cell mediated destruction of β-cells
- Autoantibodies develop against:
- Glutamic acid decarboxylase (GAD65)
- Insulin (IAA)
- Islet cell cytoplasm (ICA)
- Tyrosine phosphatase (IA-2)

4. Progressive β-cell Loss:

- 80-90% of β-cells destroyed before symptoms appear
- Complete insulin deficiency develops

Consequences:

- ❖ Uncontrolled hepatic glucose production
- ❖ Lipolysis → ketone body formation → risk of DKA
- ❖ Inability to utilize glucose → hyperglycemia

4.2. Type 2 Diabetes Mellitus (T2D) :

Key Mechanism: Insulin resistance + Progressive β-cell dysfunction

Detailed Process:

A. Insulin Resistance:

i. Molecular Mechanisms:

a. Defects in insulin receptor signaling
b. Reduced GLUT4 translocation in muscle/adipose tissue
c. Inflammation (increased TNF-α, IL-6)
d. Lipotoxicity (FFAs impair insulin signaling)

ii. Tissue-Specific Effects:

Liver: Increased gluconeogenesis
e. Muscle: Reduced glucose uptake

f. Adipose: Increased lipolysis → elevated FFAs

B. β-cell Dysfunction:

i. Compensatory Hyperinsulinemia (early stage)

ii. Progressive β-cell Failure:

- Amyloid deposition (islet amyloid polypeptide)
- Glucolipotoxicity
- Oxidative stress
- ER stress
- Apoptosis

C. Other Contributing Factors:

- Incretin deficiency/resistance (GLP-1, GIP)
- Hyperglucagonemia
- Renal glucose reabsorption (SGLT2 overactivity)

Consequences:

- Chronic hyperglycemia
- Dyslipidemia
- Microvascular/macrovascular complications

4.3. Gestational Diabetes (GDM) :

Key Mechanism: Pregnancy-induced insulin resistance + Inadequate β-cell compensation
Detailed Process:
1. Hormonal Changes:

- Human placental lactogen
- Progesterone
- Cortisol
- Prolactin

2. Insulin Resistance:

- Develops in 2nd/3rd trimester
- Similar mechanisms to T2D

3. β-cell Adaptation Failure:

- Genetic predisposition
- Pre-existing insulin resistance
- Autoimmune factors (in some cases)

Consequences:

- Maternal hyperglycemia → fetal hyperinsulinemia
- Macrosomia
- Increased perinatal complications

4.4. Other Specific Types :

A. Monogenic Diabetes (MODY):

- Single gene mutations affecting β-cell function
- Examples:
- HNF1A (MODY3) - defective insulin secretion
- GCK (MODY2) - glucose sensing defect
- KCNJ11 (Neonatal DM) - ATP-sensitive K+ channel defect

B. Pancreatogenic Diabetes:

- Loss of β-cell mass (pancreatitis, pancreatectomy)
- Combined insulin and glucagon deficiency

C. Drug-induced Diabetes:

- Glucocorticoids: increased hepatic gluconeogenesis
- Atypical antipsychotics: weight gain/insulin resistance

5. CURRENT TREATMENT OPTIONS :

5.1. Insulin therapy for diabetes mellitus :

Insulin therapy is essential for Type 1 diabetes (T1D) and often required in Type 2 diabetes (T2D) when other treatments fail. This guide covers types of insulin, delivery methods, dosing strategies, and emerging innovations.

Who Needs Insulin Therapy?

Type 1 Diabetes- Lifelong requirement (autoimmune β-cell destruction)

Type 2 Diabetes - Needed when:

- Oral/injectable meds no longer control blood sugar
- Severe hyperglycemia (HbA1c >10%)
- Acute illness/surgery (temporary need)

Gestational Diabetes -If diet/exercise fails to control glucose

Pancreatic Diabetes- After pancreas removal or damage

5.2. Oral hypoglycaemic agents for diabetes mellitus :

Oral medications are first-line treatments for Type 2 Diabetes (T2D) and sometimes used in gestational diabetes (GDM). They work by:

- Stimulating insulin secretion
- Improving insulin sensitivity
- Reducing glucose absorption

Below is a detailed breakdown of major classes, mechanisms, and key drugs

Biguanides

Primary Drug: Metformin (Most widely prescribed)

Mechanism of Action

- Reduces liver glucose production (gluconeogenesis)
- Improves insulin sensitivity in muscles
- Minimal risk of hypoglycaemia

Benefits

- ❖ First-line therapy for T2D
- ❖ Weight-neutral or modest weight loss
- ❖ Cheap & effective
- ❖ Cardiovascular benefits (reduces heart disease risk)

Side Effects

- ❖ GI issues (diarrhea, nausea – usually temporary)
- ❖ Vitamin B12 deficiency (long-term use)
- ❖ Lactic acidosis (rare, avoid in severe kidney/liver disease)

Dosing

- ❖ Starting: 500 mg once daily → increase to 2000 mg/day max
- ❖ Extended-release (ER) version reduces GI side effects

6. GENE THERAPY APPROACHES:

Gene therapy offers promising strategies for treating Type 1 Diabetes (T1D) and Type 2 Diabetes (T2D) by introducing, modifying, or regulating genes involved in insulin production, β-cell function, and glucose metabolism. Below are the key approaches with detailed explanations.

6.1. Insulin Gene Therapy :

Objective: Enable non-β cells (e.g., liver, muscle) to produce and secrete insulin in a glucose-regulated manner. Methods:

A. Viral Vector Delivery (AAV, Lentivirus):

Viral vectors are modified viruses which provide "carriers" to transporting genes to specific cells. They are efficient in gene delivery and represent a viable strategy in gene therapy for diabetes. The INS gene is transfected into hepatocytes or myocytes via AAV or lentivirus vectors. Promoters sensitive to glucose(L-PK, G6Pase) insure insulin secretion after high glucose stimulation.

VECTOR	GENOME	ADVANTAGES	LIMITATIONS	DIABETES APPLICATIONS
Adeno-associated virus (AAV)	ssDNA	➢ Low immunogenicity. ➢ Long term expression. ➢ Tissue specific targeting	➢ Small cargocapacity. ➢ Pre-existing immunity in some patients.	➢ Insulin gene delivery. ➢ β-cell regeneration
Lentivirus (LV)	RNA(Integrating)	➢ Large cargocapacity. ➢ Infects dividing & non-dividing cells.	➢ Risk of insertional mutagenesis. ➢ Requires biosafety handling.	➢ Immune modulation, β-cell engineering.

Adenovirus (ADV)	DsDNA	➢ High transduction ion efficiency. ➢ Large cargocapacity.	➢ Strong immune response. ➢ Transient expression.	➢ Glucose-sensing gene circuits, immune therapy.
Retrovirus (RV)	RNA(Integrating)	➢ Stable long-term expression	➢ Only infects dividing cells. ➢ Insertional mutagenesis risk.	➢ Ex vivo β-cell engineering

B. Non-Viral Approaches (Nanoparticles, Electroporation):

Non-viral vectors are an attractive alternative to viral gene delivery due to their safety, lower immunogenicity, and flexibility in cargo size. However, they often face challenges in transfection efficiency and long-term gene expression. Plasmid DNA encoding insulin is delivered via lipid nanoparticles or electroporation for transient expression.

Objective: Generate new insulin-producing cells by reprogramming other cell types.

Vector Type	Description	Advantages	Limitations	Diabetes Applications
Naked DNA/Plasmid	Direct injection of circular DNA.	➢ Simple & low cost. ➢ No immune response.	➢ Low transfection efficiency. ➢ Rapid degradation.	➢ Localized insulin gene delivery.
Lipid Nanoparticles (LNPs)	DNA/mRNA encapsulated in lipid shells.	➢ High delivery efficiency. ➢ FDA-approved (e.g., mRNA vaccines).	➢ Transient expression. ➢ Liver-predominant uptake.	➢ mRNA-based insulin or β-cell reprogramming.
Polymer-Based (Polyplexes)	DNA condensed with cationic polymers (e.g., PEI, chitosan).	➢ Tunable properties. ➢ It Can target specific tissues.	➢ Toxicity at high doses. ➢ Variable efficiency.	➢ Oral insulin gene delivery (gut-targeted).
Inorganic Nanoparticles	Gold, silica, or magnetic NPs carrying DNA.	➢ Stability, controlled release. ➢ It Can be externally activated (e.g., light, magnets).	➢ Complex synthesis. ➢ Potential cytotoxicity.	➢ Glucose-responsive insulin release systems.
Peptide-Based	Cell-penetrating peptides (CPPs) + DNA.	➢ Low toxicity. ➢ It can cross biological barriers.	➢ Low cargo capacity. ➢ Serum instability.	➢ Targeted delivery to pancreatic islets.
Exosomes	Natural extracellular vesicles carrying nucleic acids.	➢ Biocompatible. ➢ Homing to specific tissues.	➢ Difficult to load. ➢ Low yield.	➢ Immune modulation (T1D), β-cell repair.

7. TARGET GENES IN DIABETES THERAPY:

Diabetes mellitus (DM) is a complex metabolic disorder characterized by impaired insulin secretion (Type 1 DM, T1DM), insulinresistance (Type 2 DM, T2DM), or genetic defects (e.g., MODY). Target gene therapy aims to correct these dysfunctions by modifying specific genes involved in insulin production, glucose metabolism, β-cell survival, and immune regulation.

7.1. Insulin Gene (INS):

Insulin, a hormone secreted by the pancreatic β-cells, plays a central role in the metabolism of carbohydrates, lipids, and protein. The gene INS is located on chromosome 11p15. 5, is under the control of highly controlled mechanisms by which transcription, translation, and post-translational modifications result in the activation of insulin. Dysregulation of INS expression or proteolysis also cause diabetes mellitus, a worldwide epidemic involving at least 500 million people. The gene (INS) for insulin encodes the precursor of the hormone insulin, which plays a crucial role in controlling glucose metabolism. Mutations or defects in INS are among the genetic defects causing diabetes mellitus, both type 1 (T1DM), type 2 (T2DM) and some monogenic forms (e.g., MODY 10). The insulin gene (INS) is central to the pathogenesis and therapy of diabetes. Advances in gene therapy, CRISPR technology, and in stem cell engineering are now circumventing previously existing barriers to realizing the potential for cure in at least monogenic and T1DM. The complex transcriptional control of the insulin gene relies on an elaborate interplay between tissue-specific regulators, which ensures proper expression in pancreatic β-cells. Critical regulators, such as PDX1, bind to the A3/A4 promoter elements and are required for maintenance of the identity of β-cells; MAFA is a glucose responsive transactivator, and NeuroD1/BETA2 interacts with E-box motifs. This regulatory architecture provides for a very cell-specific expression profile of the INS gene such that it is largely active only in β-cells, and selectively subject to epigenetic repression by DNA methylation and repressive histone marks in other cell types. The evolutionary conservation of the insulin gene in species reflects important functional constraints. These innovative techniques aim to replicate normal physiological insulin regulation while addressing

the challenges associated with current insulin replacement therapies, despite ongoing difficulties in achieving precise glucose-responsive control and preventing immune responses against the engineered cells.

Genomic Structure and Regulation of Insulin:

The human insulin gene (INS) resides on chromosome 11p15. 5, in an endogenous genomic region that is very conserved and also harbors: IGF2 (insulin-like growth factor 2), TH (tyrosine hydroxylase), HRAS (Harvey rat sarcoma viral oncogene homolog). INS is 1430 bp in length and is composed of 3 exons, 2 intervening introns, 5' and 3' untranslated regions (UTRs).

Region	Size (bp)	Functional Elements
Exon 1	44	5' UTR, transcriptional start site.
Intron 1	179	Contains enhancer elements.
Exon 2	204	Encodes signal peptide, B chain, part of C-peptide.
Intron 2	786	Regulatory sequences.
Exon 3	219	Encodes remainder of C-peptide, A chain, 3' UTR.

Insulin Action on Target Tissues:

Tissue	Insulin Effect
Liver	Suppresses gluconeogenesis, promotes glycogen storage
Muscle	Stimulates glucose uptake via GLUT4.
Adipose	Enhances fat storage, inhibits lipolysis.

7.2. The Glucagon-Like Peptide-1 (GLP-1) Gene:

The story of glucagon-like peptide-1 (GLP-1) begins in the 1980s when it was discovered that proglucagon, the precursor protein encoded by GCG, could be processed into several biologically active peptides besides glucagon. Landmark discoveries have included the 1983 sequence identification of GLP-1 in anglerfish proglucagon (Bell et al., 1987)! the work showing that GLP-1 could stimulate insulin release in man (Kreymann et al., and 11990 cloning of the human GCG gene and its tissue-specific processing characterization. GLP-1, an incretin hormone derived from proglucagon (GCG), plays an important role in glucose balance via stimulating insulin secretion, inhibiting glucagon release, and stimulating β-cell growth. GLP-1 RAs constitute one of the major cloud of drugs for diabetes and obesity which mimick the action of endogenous GLP-1. The drugs, including exenatide, liraglutide and semaglutide, bind to GLP-1 receptors all over the body, stimulating the release of insulin dependent on glucose and decreasing the release of glucagon. Their long half lives, obtained with fatty acid chains or through albumin binding, overcome the rapid degradation that is typical for natural GLP-1. Besides controlling blood sugar, GLP-1 RAs induce weight loss by centrally reducing appetite and by slowing gastric emptying; the newer drugs—including tirzepatide (a dual GLP-1/glucose-dependent insulinotropic polypeptide agonist)—have been found to be more effective in clinical trials. The enzyme Dipeptidyl Peptidase-4 (DPP-4) is essential in the metabolism of GLP-1, and cleaves the N-terminal amino acids thus inactivating it rapidly. The activity of endogenous GLP-1 is prolonged by blocking this degradation pathway with DPP-4 inhibitors, namely, sitagliptin and saxagliptin. Despite being less potent for reducing glucose and body weight than GLP-1 RAs, DPP-4 inhibitors are administered orally and are well tolerated. Tissue-specific distribution of DPP-4 and its effects on local GLP-1 action have been investigated in recent studies, particularly on the gut-brain axis. Preclinical trials have demonstrated amelioration of neural inflammation, synaptic plasticity and reduction of deleterious protein aggregates in Alzheimer's and Parkinson's diseases by GLP-1 RAs. Proposed mechanisms include enhanced cerebral glucose metabolism, reduced oxidative stress, and activation of neurotrophic pathways. Currently, clinical trials are investigating whether these benefits can translate to humans and, in particular, whether GLP-1 can access the brain.

7.3. Beta-Cell Regeneration genes:

Restoration of beta cells presents a promising option for therapeutic intervention in diabetes mellitus, where glucose homeostasis is impaired by the loss or dysfunction of insulin-secreting pancreatic beta cells. The ability to regenerate or repair these cells could possibly reverse not only type 1 diabetes (T1D) due to autoimmune attack and depletion of beta cells, but also type 2 diabetes (T2D) in which beta cells fail and dedifferentiate leading to progressive insulin deficiency. New discoveries from molecular biology and genetics have revealed important genes that dictate beta cell growth, neogenesis, and transdifferentiation, and these gene discoveries offer new targets for diabetes intervention. These genes control a range of biological processes, including embryonic development, cell cycle progression and cellular reprogramming and are therefore important targets for regenerative therapies. Genes encoding early developmental factors of the embryonic pancreas such as PDX1 (Pancreatic and Duodenal Homeobox 1), NGN3 (Neurogenin 3) and MAFA have been some of the most studied genes with respect to beta cell regeneration. PDX1 is essential for beta cell maturation and maintenance, and NGN3 is a major factor in the differentiation of progenitor endocrine cells. In contrast, MAFA is required for the correct GSIS. A second important research topic is transdifferentiation, i.e. differentiation of non-beta cells (eg pancreatic alpha cells, acinar cells or even liver cells) into beta cells. Crucial genes in this reprogramming are PAX4, which promotes conversion from alpha to beta cells, and ARX (Aristaless-Related Homeobox), whose suppression stimulates beta cell neogenesis. Furthermore, epigenetic regulators like DNA methyltransferases (DNMTs) and histone deacetylases (HDACs) contribute to unlocking the plasticity of beta cells, indicating that pharmacological adjustment of these pathways might enhance

regeneration. Despite these advancements, obstacles remain in translating beta cell regeneration into clinical realities. Autoimmune responses in T1D may eliminate newly created beta cells, while metabolic stress in T2D could hinder their functionality. Additionally, precise regulation of gene expression is essential to prevent unrestrained proliferation or tumor formation. Innovative technologies, including CRISPR-based gene editing, small molecule stimulators of beta cell replication, and stem cell-derived beta cell transplantation, are currently being investigated to address these challenges. By leveraging the regenerative capabilities of these important genes, scientists aspire to devise curative treatments that restore normal insulin production in individuals with diabetes, moving past lifelong insulin therapy toward genuine modification of the disease.

A. Immune Modulation Strategies for Beta Cell Regeneration in Type 1 Diabetes :

A major obstacle in beta cell regeneration for type 1 diabetes is safeguarding newly created beta cells from autoimmune damage. Current investigations focus on merging regenerative techniques with immunomodulation, such as modifying beta cells to produce immune checkpoint proteins like PD-L1 or CTLA4-Ig. These alterations lead to the formation of "immune-shielded" beta cells that can avoid destruction by T cells while retaining their normal functionality. Another promising strategy involves using CRISPR to eliminate HLA class I molecules to decrease immunogenicity, in combination with localized delivery of regulatory T cell (Treg)-stimulating cytokines such as TGF-β or IL-10. Recent research is also examining the use of encapsulation devices made from biomaterials that allow nutrient flow while preventing the entry of immune cells, which could enable the survival of regenerated beta cells in autoimmune conditions.

B. Metabolic Priming for Enhanced Beta Cell Regeneration in Type 2 Diabetes:

The metabolic microenvironment plays a crucial role in influencing the regeneration capacity of beta cells in individuals with type 2 diabetes. Persistent high blood sugar levels lead to epigenetic modifications that inhibit essential regeneration genes through DNA hypermethylation and histone deacetylation. Researchers are exploring combined strategies that integrate beta cell regeneration techniques with metabolic normalization, such as utilizing SGLT2 inhibitors to lower glucotoxicity alongside administering GLP-1 receptor agonists to encourage cell proliferation. Notably, intermittent fasting approaches have demonstrated the ability to promote beta cell regeneration by activating autophagy and diminishing oxidative stress. Compounds targeted at mitochondria, such as MitoQ, are undergoing testing to enhance the function of regenerated beta cells in insulin-resistant settings by decreasing reactive oxygen species production and improving energy metabolism.

C. Single-Cell Technologies for Decoding Beta Cell Regeneration :

Cutting-edge single-cell technologies are transforming our comprehension of beta cell regeneration by uncovering previously unknown cellular diversity. Single-cell RNA sequencing has pinpointed rare clusters of "progenitor-like" beta cells that maintain their ability to proliferate in adult pancreata. Studies using mass cytometry (CyTOF) reveal dynamic alterations in beta cell states throughout the regeneration process, highlighting distinct transitional populations that arise during reprogramming. New spatial transcriptomics methods are delineating the exact anatomical niches within pancreatic islets that support beta cell regeneration, uncovering crucial interactions with stromal cells and components of the extracellular matrix that aid this process. These advancements are paving the way for the creation of more targeted regeneration strategies focused on specific beta cell subpopulations.

D. Non-Coding RNA Networks in Beta Cell Regeneration:

Beyond genes that code for proteins, non-coding RNAs create complex regulatory frameworks that govern the regeneration of beta cells. Long non-coding RNAs like PLUTO have been demonstrated to preserve the three-dimensional chromatin structure of the insulin locus, whereas circular RNAs such as circHIPK3 influence the proliferation of beta cells by sponging miRNAs. Scientists are working on lipid nanoparticle-based systems for the targeted modulation of these RNA networks, showing preliminary success in increasing beta cell mass in animal studies. Notably intriguing is the identification of miRNA signatures associated with regeneration, including the miR-17-92 cluster, which, when overexpressed, can stimulate dormant beta cells to enter the cell cycle while retaining their ability to respond to glucose.

E. Comparative Approaches: In Vivo Reprogramming vs Cell Replacement Therapies:

The current landscape is characterized by two primary strategies for regeneration: in vivo reprogramming of native cells and ex vivo creation of beta cells for transplantation. In vivo strategies make use of viral vectors or small compounds to stimulate inactive regeneration pathways in pancreatic ductal or acinar cells, providing the benefit of preserving the original islet structure. However, these approaches struggle with issues related to the precision and extent of reprogramming. On the other hand, beta cells derived from stem cells (as seen in Vertex's VX-880 trial) present a controlled and scalable alternative but must address challenges related to implantation and long-term viability. New hybrid methods are emerging, such as employing temporary reprogramming factors to generate expandable progenitor cells that can be further differentiated in vitro prior to transplantation, potentially combining the strengths of both strategies.

8. PRECLINICAL STUDIES FOR DIABETES MELLITUS:

Preclinical diabetes research acts as a vital link between discoveries made in the lab and therapies for humans, utilizing meticulously designed models to simulate human diseases prior to clinical trials. Unlike conventional animal studies that often fail to reflect human-specific disease mechanisms, contemporary methods increasingly apply humanized systems—such as immune-reconstituted mice with human pancreatic cells and 3D organoids derived from patient stem cells. These sophisticated models tackle a significant challenge in diabetes research: the inherent metabolic and immunological differences between rodents and humans that have historically resulted in promising preclinical outcomes not translating to clinical success. For type 1 diabetes, newer preclinical investigations now employ humanized mice that are implanted with both human immune cells and pancreatic tissue, allowing for an accurate examination of autoimmune damage and the testing of immunotherapies. In studies focused on type 2

diabetes, researchers are advancing beyond basic obesity models to integrate systems that more accurately reflect the progressive β-cell dysfunction observed in humans, including organ-on-a-chip technologies that simulate the intricate interactions between liver, muscle, fat, and pancreatic cells. By merging human cell grafts with cutting-edge imaging and omics technologies, scientists can now observe the effects of therapies at the single-cell level in near-human environments. This advancement in preclinical modeling, though more intricate and expensive, provides information that translates more reliably into human trials—potentially hastening the development of genuinely transformative diabetes treatments while alleviating the ethical and financial pressures associated with unsuccessful clinical trials.

8.1. Animal Models for Diabetes mellitus :

Animal models continue to be essential in diabetes research, acting as crucial platforms for investigating disease mechanisms and testing new treatments prior to human trials. Nevertheless, conventional models often fail to capture the intricacies of human diabetes, leading to the creation of more advanced, humanized systems. Current methodologies integrate genetic engineering with human cell transplantation to develop "human-like" diabetic traits in animals—such as immunocompromised mice infused with human pancreatic islets and immune cells to simulate type 1 diabetes (T1D), or obese rodents with implanted human liver cells to more accurately represent type 2 diabetes (T2D) metabolic disturbances. These hybrid models tackle significant species-specific challenges, including variations in insulin release, immune reactions, and drug metabolism, which have previously resulted in translational setbacks. For T1D investigations, non-obese diabetic (NOD) mice and their humanized counterparts (e.g., NOD-scid IL2Rγnull mice harboring human immune cells) facilitate the exploration of autoimmune β-cell destruction and the assessment of immunotherapies. Regarding T2D, models such as db/db mice or primates on high-fat diets are augmented with transplants of human gut microbiota or xenografted adipose tissue to more closely mirror human insulin resistance and inflammation. While these sophisticated models enhance predictive accuracy, challenges remain—including ethical dilemmas, financial implications, and the necessity for standardized protocols—underscoring the relevance of integrating animal studies with human cell-based systems for a thorough understanding of diabetes research.

A. Model Selection Considerations:

The choice of animal model is significantly influenced by the particular research question at hand. For studies focused on the pathogenesis of type 1 diabetes (T1D), non-obese diabetic (NOD) mice are still considered useful, even with their limitations in correlating with human disease, as they naturally develop autoimmune diabetes. In type 2 diabetes (T2D) drug testing, db/db mice and Zucker diabetic fatty (ZDF) rats are frequently utilized, although researchers need to take into consideration their severe hyperglycemia and rapid disease progression, which differs from that of human T2D. An increasing number of researchers are opting for larger species such as pigs and non-human primates, which more accurately reflect human pancreatic structure and glucose regulation, though these options come with higher costs and more significant ethical concerns.

B. Inducible and Conditional Models:

Advanced genetic engineering has created sophisticated methods that allow for precise temporal and tissue-specific regulation of genes associated with diabetes. Tet-on/off systems enable scientists to trigger β-cell destruction or induce insulin resistance at designated time points. Cre-loxP technology facilitates gene knockout specific to cell types, such as the deletion of the insulin receptor in β-cells, to investigate insulin signaling. These technologies offer unmatched accuracy in simulating various stages of diabetes advancement and evaluating targeted treatments.

C. Humanized Microbiota Models: Recognizing the gut microbiome's role in diabetes, researchers now transplant human fecal microbiota into germ-free mice to create "humanized gut" models. These help study:

- ✓ Microbial influences on insulin resistance
- ✓ Diet-microbiome interactions
- ✓ Microbial metabolite effects on β-cell function

Such models are particularly valuable for investigating the mechanisms behind observational human microbiome studies.

D. Advanced Imaging and Monitoring:

Contemporary diabetes models incorporate technologies that parallel human clinical monitoring:

- ➢ Continuous glucose monitoring systems adapted for rodents
- ➢ Miniaturized insulin pumps for closed-loop systems
- ➢ High-resolution ultrasound for pancreatic imaging
- ➢ Bioluminescent reporters for tracking β-cell mass

These tools enable longitudinal studies in individual animals that better mimic human diabetes management.

E. Comorbidity Modeling:

Newer models better replicate the multi-organ complications seen in human diabetes:

- Retinopathy models using oxygen-induced retinal damage
- Nephropathy models combining diabetes with renal injury
- Neuropathy models with detailed electrophysiological assessment

These allow testing of therapies that might affect both glycemic control and end-organ damage.

F. Limitations and Mitigation Strategies:

While invaluable, animal models have inherent limitations:

- Species differences in immune system function (addressed through humanized models)
- Varied drug metabolism pathways (addressed by human liver chimeric mice)
- Different islet architecture (addressed by human islet transplantation)

Researchers increasingly use multiple complementary models to overcome these limitations.

G. Emerging Alternatives:

The field is developing alternatives to traditional animal models:

- Organoid-humanized systems combining animal hosts with human organoids
- "Diabetes-on-a-chip" microphysiological systems
- Computational models integrating animal and human data

These approaches aim to reduce animal use while improving translational relevance. These developments reflect an evolving understanding that while animal models remain essential for diabetes research, their design and application must continually adapt to better serve human therapeutic development. The most impactful studies now carefully match model strengths to specific research questions while acknowledging each system's limitations.

8.2. In Vitro Studies in Diabetes Mellitus:

In vitro research plays a crucial role in diabetes studies, allowing for in-depth mechanistic exploration of β-cell functionality, insulin signaling, and the development of the disease in well-controlled environments. These cell-based models include a variety of options, such as immortalized β-cell lines (like INS-1, MIN6), primary human islets, and β-like cells derived from stem cells, providing adaptable platforms for large-scale drug testing and toxicity evaluations. Recent developments in 3D culture techniques, such as pancreatic organoids and microphysiological "islet-on-a-chip" systems, are now more effectively simulating the natural islet microenvironment by integrating components of the extracellular matrix, vascular endothelial cells, and immune cells to model type 1 diabetes autoimmunity or type 2 diabetes metabolic stress. In vitro systems are especially useful for examining disease mechanisms specific to humans through patient-derived induced pluripotent stem cells (iPSCs), enabling researchers to study genetic variations, β-cell dedifferentiation, and individualized drug responses in the context of personalized medicine. Although these reductionist methods lack systemic complexity, they offer unmatched detail for unraveling molecular pathways, screening potential therapies, and refining cell replacement techniques prior to transitioning to animal models or clinical trials. Current advancements aim to improve physiological relevance via dynamic glucose stimulation setups, co-cultures with liver and adipose tissue, and the incorporation of omics technologies to bridge the divide between isolated cellular investigations and whole-organism physiology.

A. Advanced 3D Culture Models:

i. Islet Organoids: Self-organizing 3D structures derived from stem cells or primary islets that better preserve β-cell polarity, gap junctions, and glucose-responsive insulin secretion compared to 2D cultures.

ii. Microfluidic "Islet-on-Chip" Systems: Mimic pancreatic vasculature with:

- Perfusion channels for nutrient/oxygen gradient control
- Integrated sensors for real-time insulin/glucose monitoring
- Immune cell recruitment chambers to model T1D insulitis

B. Patient-Specific Disease Modeling:

i. iPSC-Derived β-Cells: Reprogrammed from T1D/T2D patients to:

- Study genetic predispositions (e.g., TCF7L2 variants in T2D)
- Test personalized drug responses
- Model β-cell dedifferentiation/regeneration

ii. KMCRISPR-Edited Disease Lines: Introduce specific mutations (e.g., INS, HNF1A) to dissect mechanistic contributions.

C. Multi-Tissue Interaction Platforms:

i. Liver-Islet Co-Cultures: Study hepatic insulin resistance effects on β-cell function.
ii. Neuro-insular Systems: Model autonomic nervous system regulation of insulin secretion.

Gut-Pancreas Axis Models: Incorporate enteroendocrine cells (L-cells) to probe incretin effects.

D. Dynamic Stimulation Systems:

a. Pulsatile Glucose Exposure: Simulate physiological postprandial conditions
b. Cyclic Mechanical Stress: Replicate pancreas stiffness changes in fibrosis
c. Hypoxia Chambers: Study islet transplantation stress responses.

E. High-Content Screening (HCS) Applications:

i. Automated Imaging: Quantify β-cell mass/function via:

- ❖ Calcium flux assays
- ❖ Insulin granule trafficking
- ❖ Apoptosis/regeneration markers

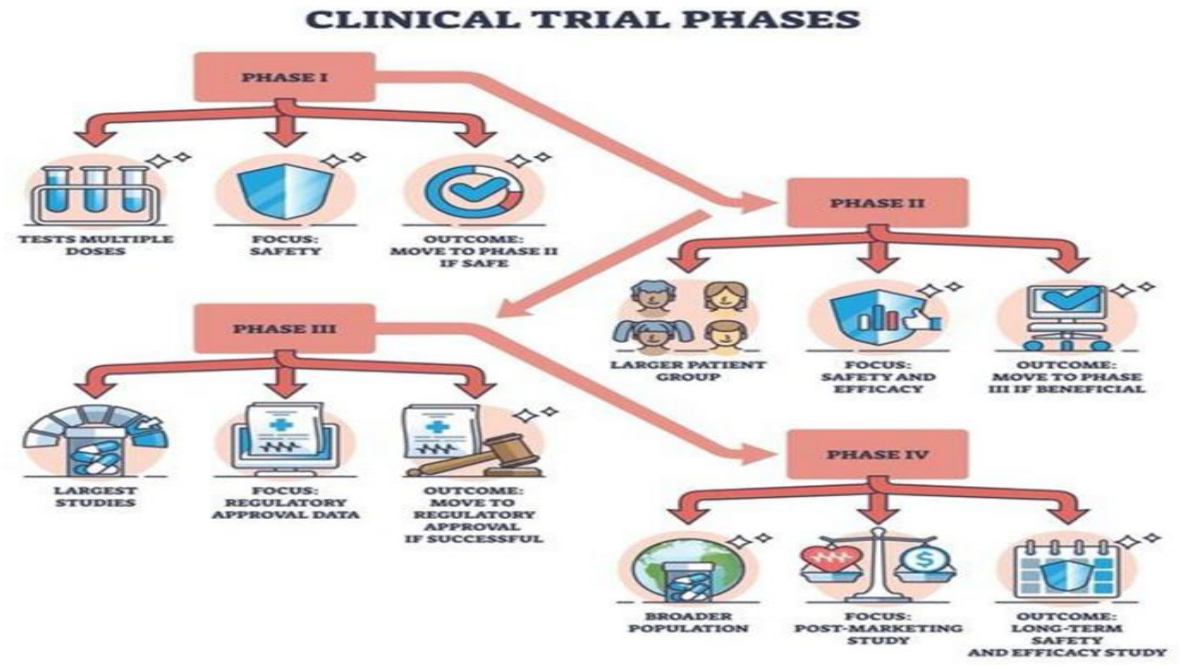
ii. Multi-Parametric Drug Testing: Simultaneously assess:

- ❖ Insulin secretion
- ❖ Cell viability
- ❖ Oxidative stress

F. Limitations & Mitigation Strategies:

Challenge	Innovative Solution
Lack of systemic metabolism	Multi-organ microphysiological systems.
Short-term viability	Perfusion bioreactors with ECM support.
Donor variability	Gene-edited isogenic iPSC controls.
Immune component absence	Co-cultures with autologous immune cells.

9. CLINICAL TRIALS:



Clinical trials focused on diabetes mellitus are organized research studies aimed at assessing the safety, effectiveness, and efficiency of new treatments, medications, devices, or lifestyle changes for managing or preventing diabetes. These studies follow a structured multi-phase methodology to ensure scientific rigor and patient safety. Phase I trials involve a small cohort of healthy individuals or patients to evaluate the safety, tolerance, and pharmacokinetics of a new medication. Phase II trials include a larger group of diabetic patients to establish the optimal dosage and initial efficacy, while observing side effects. Phase III trials are extensive, randomized, controlled studies that compare the new treatment with standard therapies to validate effectiveness, track adverse reactions, and adjust dosing recommendations. If these trials are successful, the treatment may receive approval from regulatory authorities. Phase IV trials, also known as post-marketing surveillance, further investigate long-term safety and practical effectiveness following approval. Important research areas in diabetes trials consist of innovative insulin formulations (e.g., ultra-rapid or ultra-long-acting analogs), non-insulin treatments (such as GLP-1 receptor agonists, SGLT2 inhibitors, and DPP-4 inhibitors), artificial pancreas technology, stem cell and gene therapies, as well as digital health tools like continuous glucose monitoring (CGM) systems. Additionally, trials examine preventive measures for prediabetes, precision medicine strategies, and the effects of dietary and exercise interventions. Regulatory organizations like the FDA (U.S.), EMA (Europe), and CDSCO (India) supervise these trials to ensure ethical practices, data reliability, and participant safety. Involvement in clinical trials grants patients access to groundbreaking therapies while aiding in the progress of diabetes management. Thorough trial designs, which include randomization, blinding, and placebo controls, help reduce bias and create trustworthy evidence for enhancing diabetes management around the globe.

9.1. Phase I Trials:

Phase 1 clinical trials for diabetes mellitus represent the initial phase of evaluating new medications, devices, or treatments in humans following laboratory and animal studies conducted in preclinical stages. These trials mainly concentrate on determining safety, tolerability, pharmacokinetics (how the drug is absorbed, circulated, metabolized, and eliminated by the body), and pharmacodynamics (the drug's impact on the body). Usually carried out with a small group of 20 to 100 participants, Phase 1 trials may involve healthy volunteers or individuals with type 1 or type 2 diabetes, depending on the associated risks of the treatment. The study design often includes dose escalation, where participants are administered progressively higher doses to establish the maximum tolerated dose (MTD) without experiencing severe side effects. Researchers carefully monitor for adverse effects, vital signs, blood glucose levels, and metabolic responses to ensure the safety of participants. In diabetes research, Phase 1 trials may investigate new insulin analogs, oral antidiabetic medications, incretin-based treatments (such as GLP-1 agonists), or innovative technologies like glucose-responsive insulin or artificial pancreas systems. Given that diabetes necessitates long-term management, early-phase trials also evaluate short-term metabolic effects and potential risks such as hypoglycemia. Regulatory authorities, including the FDA and EMA, enforce stringent compliance with ethical standards, which encompass obtaining informed consent and approval from institutional review boards (IRBs). Although Phase 1 trials do

not assess the efficacy of treatments, they yield crucial information that propels promising therapies into Phase 2 trials, where larger populations of diabetic patients are examined for preliminary effectiveness. Positive outcomes from Phase 1 are vital for the development of safer and more effective treatment options for diabetes.

A. Study Design and Objectives:

The primary objective of Phase 1 diabetes trials is to establish initial safety parameters rather than demonstrate therapeutic efficacy. These studies typically enroll a small number of participants (usually 20-100), who may include healthy volunteers or carefully selected patients with diabetes, depending on the mechanism of action and potential risks of the investigational product. The trial design often incorporates single ascending dose (SAD) and multiple ascending dose (MAD) protocols, where successive cohorts receive progressively higher doses under close medical supervision.

B. Pharmacokinetic and Pharmacodynamic Assessments:

Comprehensive pharmacokinetic (PK) analyses measure how the body processes the investigational drug, including absorption rates, peak concentration times, distribution patterns, metabolism, and elimination characteristics. Pharmacodynamic (PD) assessments evaluate the drug's biological effects, which for diabetes therapies might include glucose-lowering activity, insulin secretion patterns, or effects on counter-regulatory hormones. Frequent blood sampling, continuous glucose monitoring, and specialized metabolic tests provide detailed PK/PD profiles.

C. Safety Monitoring Protocols:

Phase 1 diabetes trials implement intensive safety monitoring that typically includes:

- Continuous clinical observation during dosing periods
- Frequent vital sign measurements
- Electrocardiogram monitoring for cardiac safety
- Comprehensive laboratory testing (hematology, chemistry, urinalysis)
- Specialized diabetes monitoring (continuous glucose monitoring, frequent point-of-care glucose testing)
- Regular assessment for adverse events and hypoglycemic episodes.

9.2. Phase II Trials :

Phase 2 clinical trials mark an important transitional phase in the development of diabetes treatments, where investigational therapies that have shown acceptable safety profiles in Phase 1 are tested for initial effectiveness and optimal dosage in patient populations. These medium-sized studies serve as a vital link between initial human trials and larger Phase 3 confirmatory studies, playing a crucial role in deciding whether a potential treatment deserves further investment and development. Structured with rigorous scientific methods, Phase 2 trials for diabetes mellitus generally utilize randomized, controlled designs to evaluate both short-term therapeutic impacts and ongoing safety monitoring in carefully chosen patient groups. The primary goals of these trials aim to establish proof of biological concept, ascertain dose-response relationships, and determine the most effective and tolerable dosing regimen for future studies. In contrast to Phase 1 trials that focus on pharmacokinetics and safety in small populations, Phase 2 studies incorporate clinically relevant endpoints such as changes in hemoglobin A1c (HbA1c), fasting plasma glucose levels, and time-in-range metrics from continuous glucose monitoring. The increasing appreciation of diabetes heterogeneity has inspired more targeted Phase 2 trials that explore personalized medicine approaches and specific diabetes subtypes. These studies also provide valuable chances to investigate mechanistic biomarkers, examine combination therapies, and identify potential predictors of treatment response. As diabetes management progresses beyond solely glycemic metrics, numerous contemporary Phase 2 trials include additional endpoints related to weight management, cardiovascular risk factors, and quality of life assessments. The data produced from well-executed Phase 2 trials not only inform decisions regarding progression to Phase 3 development but also greatly enhance our understanding of disease pathophysiology and therapeutic mechanisms for both type 1 and type 2 diabetes.

A. The primary objectives of Phase 2 diabetes trials include:

I. Establishing Proof of Concept – Determining whether the investigational treatment has measurable biological effects relevant to diabetes management (e.g., glucose lowering, improved insulin sensitivity, or beta-cell preservation).
II. Dose Optimization – Identifying the most effective and tolerable dose range for further testing in Phase 3.
III. Evaluating Short-Term Efficacy – Assessing improvements in key diabetes metrics such as HbA1c, fasting plasma glucose, and postprandial glucose control.
IV. Expanding Safety Profiling – Detecting less common or longer-term adverse effects in a larger patient population.

Phase 2 trials typically employ randomized, controlled designs, often comparing multiple doses of the investigational drug against a placebo or standard therapy. Many incorporate dose-ranging methodologies to determine the optimal therapeutic window. Some Phase 2 trials may also explore biomarkers (e.g., C-peptide levels in type 1 diabetes or insulin sensitivity indices in type 2 diabetes) to provide mechanistic insights.

B. Common Investigational Therapies in Phase 2 Diabetes Trials:

Recent Phase 2 trials in diabetes have explored several innovative approaches:

I. Novel Insulin Formulations – Ultra-long-acting insulins, glucose-responsive "smart" insulins, and alternative delivery methods (oral or inhaled insulin).
II. Incretin-Based Therapies – Next-generation GLP-1/GIP dual agonists (e.g., tirzepatide in early trials) and oral GLP-1 formulations.
III. SGLT2 Inhibitors and Beyond – Newer agents with improved renal or cardiovascular safety profiles.
IV. Immunomodulatory Therapies (for T1D) – Drugs targeting autoimmunity to preserve residual beta-cell function.
V. Gene and Stem Cell Therapies – Experimental approaches for beta-cell regeneration or replacement.
VI. Digital Therapeutics – AI-driven insulin dosing algorithms and closed-loop artificial pancreas systems.

Phase 2 clinical trials play a pivotal role in diabetes drug development, bridging early safety assessments with definitive efficacy testing. These studies refine dosing, confirm biological activity, and identify promising candidates for Phase 3 evaluation. As diabetes management evolves, Phase 2 trials continue to incorporate innovative designs, biomarkers, and therapeutic strategies to address unmet needs in this global epidemic. The insights gained from these trials not only advance new treatments but also deepen our understanding of diabetes pathophysiology, paving the way for more personalized and effective therapies.

9.3. Phase III Trials:

Phase 3 clinical trials signify a crucial phase in the development of diabetes medications, wherein new therapies undergo thorough assessment in extensive and varied patient groups to validate their effectiveness, safety, and clinical significance. These multicenter trials build on encouraging Phase 2 findings by using randomized and controlled designs, enrolling sample sizes that range from several hundred to thousands of participants from multiple locations. Unlike earlier phases that concentrate on initial safety evaluations and dosage determination, Phase 3 trials offer conclusive evidence regarding the advantages of a treatment in comparison to standard care or placebo, establishing the essential groundwork for regulatory approval and clinical practice recommendations. In diabetes research, Phase 3 studies generally assess both immediate glycemic control and long-term effects over a duration of six months to several years. They include thorough evaluations such as reductions in HbA1c, fasting glucose levels, time-in-range data from continuous glucose monitoring, and feedback from patients. A notable aspect of contemporary diabetes trials is the required assessment of cardiovascular outcomes, reflecting regulatory demands to confirm cardiovascular safety for newly developed antidiabetic medications. These trials also investigate impacts on body weight, kidney function, lipid levels, and overall quality of life, acknowledging diabetes as a multifaceted metabolic disorder with broad implications. The design of modern Phase 3 diabetes trials increasingly focuses on real-world relevance by including a variety of patient demographics, comorbidities, and existing treatments. Adaptive trial designs paired with innovative statistical methods improve efficiency while upholding scientific integrity. As diabetes care progresses toward personalized medicine, newer studies integrate biomarker analysis and digital health tools to identify the most suitable treatment candidates. The outcomes from these extensive studies not only influence regulatory approval but also guide clinical decision-making, ultimately enhancing treatment options for millions affected by diabetes around the world.

A. Study Design and Methodological Rigor:

Phase 3 diabetes trials employ the most rigorous experimental designs, typically featuring:

- ✓ Randomized, double-blind, placebo-controlled protocols.
- ✓ Multicenter participation across diverse geographic regions
- ✓ Active comparator arms using standard therapies.
- ✓ Predefined primary and secondary endpoints
- ✓ Comprehensive statistical analysis plans

These trials often incorporate complex adaptive designs that may include:

1. Dose-response evaluations
2. Non-inferiority or superiority comparisons
3. Long-term extension studies
4. Cardiovascular outcome components

The large sample sizes (typically 500-5000 participants) enable detection of both common and rare adverse events while providing sufficient power to demonstrate clinically meaningful differences in efficacy endpoints.

Phase 3 clinical trials are regarded as the benchmark for determining the therapeutic efficacy of new diabetes treatments. These extensive studies provide the conclusive evidence necessary for gaining regulatory approval and integrating new therapies into clinical practice, while also deepening our knowledge of diabetes management. With the global rise in diabetes cases, the significance of thorough Phase 3 research is crucial—it acts as the vital link between scientific advancement and significant patient benefits. The ongoing development of Phase 3 methodologies continues to improve our capacity to create safer and more effective therapies that tackle the varied challenges of diabetes care.

10. CASE STUDIES:

Diabetes mellitus presents variably among patients, shaped by factors including its type, duration, associated diseases, and socioeconomic background. The case studies provided depict authentic scenarios of diabetes management, shedding light on diagnostic hurdles, treatment methods, and patient results. Diabetes mellitus is a multifaceted and diverse metabolic condition with various clinical manifestations, management difficulties, and long-term effects. Case studies offer essential insights into real-life situations, illustrating the subtleties of diagnosis, treatment approaches, and patient results across distinct subtypes, such as type 1 (T1D), type 2 (T2D), gestational diabetes (GDM), and uncommon forms like monogenic diabetes (MODY).

These examples demonstrate how elements such as genetics, autoimmune reactions, insulin resistance, and psychosocial factors influence disease development and treatment responses. Through comprehensive case evaluations, healthcare professionals and researchers can gain a deeper understanding of diagnostic challenges—such as differentiating T1D from T2D in adult patients or recognizing MODY in younger individuals with unusual characteristics. Case studies further highlight the real-world difficulties in managing diabetes, including challenges with medication adherence, the risk of hypoglycemia, and the emotional strain of living with a chronic illness. Moreover, they point out disparities in access to advanced treatments, like insulin pumps or GLP-1 receptor agonists, which unevenly impact marginalized communities. By exploring the unique paths of individual patients, these case studies reinforce the significance of personalized medicine, team-based care, and patient-focused strategies in managing diabetes. They function as teaching resources for healthcare professionals, providing insights on enhancing glycemic control, preventing complications, and addressing the comprehensive needs of patients. Ultimately, these real-world illustrations connect clinical guidelines with actual practice, showcasing how customized interventions can enhance the quality of life and long-term health outcomes for those managing diabetes.

Case Study 1: Late-Onset Type 1 Diabetes in an Adult:

A 32-year-old man came in with a two-week history of excessive urination, increased thirst, and an unexplained weight loss of 5 kg. Initial laboratory tests indicated a random blood glucose level of 480 mg/dL, an HbA1c of 11.5%, and the presence of ketones in urine. Although there was no family history of diabetes, autoimmune screening showed positive GAD-65 antibodies, leading to a diagnosis of latent autoimmune diabetes in adults (LADA). Unlike traditional Type 1 Diabetes, his symptoms appeared gradually, initially giving the impression of Type 2 Diabetes. However, due to his slender build and swift metabolic deterioration, it became evident that there was autoimmune destruction of the beta cells. He was initiated on a basal-bolus insulin regimen paired with diabetes education. Over the course of six months, his glycemic control improved (HbA1c of 7.2%), but he experienced issues with hypoglycemia unawareness, which necessitated the use of continuous glucose monitoring (CGM). This case highlights the diagnostic intricacies of adult-onset Type 1 Diabetes and the critical role of antibody testing in atypical cases.

Case Study 2: Type 2 Diabetes with Severe Insulin Resistance:

A 55-year-old woman with obesity (BMI 38) and a decade-long history of type 2 diabetes (T2D) came to the clinic with consistently high HbA1c levels (9.8%) despite reaching the maximum dosage of her oral medications (metformin, SGLT2 inhibitor, and sulfonylurea). She had also developed hypertension and high triglycerides, indicating the presence of metabolic syndrome. Genetic tests excluded any monogenic forms of diabetes, but her significant insulin resistance prompted an assessment for acquired lipodystrophy, which returned negative results. She was switched to a GLP-1 receptor agonist (semaglutide) along with basal insulin, leading to a 6% reduction in weight and a decrease in HbA1c to 7.5% over the course of a year. Nevertheless, financial limitations affected her ability to consistently use the GLP-1 agonist, underscoring the disparities in access to newer diabetes medications. This case demonstrates the complexities involved in managing insulin-resistant T2D and the socioeconomic challenges that impede optimal treatment.

Case Study 3: Monogenic Diabetes (MODY) Misdiagnosed as Type 1 Diabetes:

A 14-year-old girl was diagnosed with Type 1 Diabetes (T1D) after displaying hyperglycemia (fasting glucose of 200 mg/dL) and mild ketosis. There was a significant family history of diabetes spanning three generations, all exhibiting non-insulin-dependent types. Genetic analysis revealed a heterozygous HNF1A mutation, confirming the presence of MODY3. In contrast to T1D, MODY3 has a favorable response to sulfonylureas due to maintained beta-cell sensitivity. She was transitioned from insulin to glimepiride, successfully reaching an HbA1c level of 6.3% without experiencing hypoglycemia. This case highlights the importance of genetic testing in young, non-obese patients with a family history of autosomal dominant diabetes to prevent unnecessary insulin treatment.

Case Study 4: Gestational Diabetes with Postpartum Progression:

A 30-year-old pregnant woman (BMI 28) failed her 2-hour oral glucose tolerance test at 24 weeks (fasting: 95 mg/dL, 2-hour: 210 mg/dL), meeting criteria for gestational diabetes (GDM). She managed her condition with diet and metformin, delivering a healthy infant at term. However, postpartum screening revealed persistent impaired glucose tolerance, and she developed T2D within three years. Her case reflects the 50% lifetime risk of T2D after GDM and underscores the importance of long-term metabolic monitoring in this high-risk population

Case Study 5: Type 1 Diabetes with Brittle Diabetes Phenotype:

A 25-year-old female with longstanding T1D had recurrent hospitalizations for diabetic ketoacidosis (DKA) and severe hypoglycemia, meeting criteria for brittle diabetes. Psychological evaluation revealed diabetes distress and disordered eating (omitting insulin to lose weight). A multidisciplinary team, including an endocrinologist, psychologist, and dietitian, implemented a closed-loop insulin pump, cognitive behavioral therapy, and structured meal plans. Over two years, her HbA1c stabilized at 7.8%, and DKA episodes ceased. This case highlights the interplay between psychological factors and glycemic control in T1D, necessitating holistic care models.

11. CONCLUSION:

Diabetes mellitus continues to be one of the most significant health challenges globally in the 21st century, with its occurrence escalating among all age groups and locations. This complex metabolic condition, marked by persistent high blood sugar due to issues with insulin production, effectiveness, or both, impacts individuals, healthcare systems, and societies on a global scale. Our understanding of diabetes has shifted from perceiving it merely as a disorder of glucose metabolism to recognizing it as a multifaceted condition involving complex interactions between genetic factors, environmental influences, and physiological processes. The categorization into type 1 (caused by autoimmune destruction of β-cells), type 2 (characterized by insulin resistance alongside relative insulin deficiency), and other specific types has established a framework for diagnosis and treatment, although ongoing

research is uncovering a greater variety of cases within these categories than previously realized. The management of diabetes has experienced remarkable progress in recent years, evolving from simply extending life to allowing individuals to lead fuller and more productive lives while managing the condition. Current treatment approaches focus on personalized healthcare, acknowledging that every patient's diabetes presents uniquely and necessitates customized interventions. The range of options for achieving glycemic control has grown beyond conventional insulin therapy and oral medications to include cutting-edge drug classes like GLP-1 receptor agonists and SGLT2 inhibitors, which not only enhance glucose control but also provide cardiovascular and renal benefits. Technological advancements such as continuous glucose monitoring devices, smart insulin pens, and automated insulin delivery systems have transformed daily diabetes management, easing the self-care burden while enhancing patient outcomes. Digital health solutions, including AI-based tools for risk assessment and management support, are starting to revolutionize diabetes care delivery. Concurrently, public health efforts aimed at creating healthier food environments, urban planning that encourages physical activity, and policies designed to mitigate socioeconomic disparities in diabetes risk are gaining momentum. The financial impact of diabetes remains immense, with expenses extending well beyond direct medical costs to encompass lost productivity and diminished quality of life. This highlights the urgent need for effective prevention strategies and healthcare models that emphasize value-based care focused on patient-centered outcomes. Healthcare systems around the globe are progressively moving towards integrated, team-oriented care approaches that cater to the comprehensive needs of diabetes patients, covering everything from medical treatment to psychological support. In summary, while diabetes mellitus continues to pose significant challenges to global health, the intersection of scientific progress, technological advancements, and the evolution of care models presents real promise for enhanced prevention, management, and potential cures. The next few decades are expected to bring significant changes in our understanding, classification, and treatment of diabetes. However, achieving this potential will require ongoing investment in research, dedication to health equity, and collaboration across various disciplines and sectors. For the time being, making optimal use of existing knowledge and resources, combined with empathetic, patient-focused care, can greatly improve the lives of millions affected by diabetes worldwide. The path to overcoming diabetes continues, with each scientific advancement and clinical innovation bringing us closer to making a difference against this widespread disease.

REFERENCES

1. 1.Yechoor V, Chan L. Gene Therapy Progress and Prospects: Gene therapy for diabetes mellitus. Gene Therapy. 2004 Oct 21;12(2):101–7.https://doi.org/10.1038/sj.gt.3302412.
2. 2.Kolodka TM, Finegold M, Moss L, Woo SL. Gene therapy for diabetes mellitus in rats by hepatic expression of insulin. Proceedings of the National Academy of Sciences [Internet]. 1995 Apr 11 [cited 2021 Jun 3];92(8):3293–7. https://doi.org/10.1073/pnas.92.8.3293.
3. Yamaoka T. Gene Therapy for Diabetes Mellitus. Current Molecular Medicine. 2001 Jul 1;1(3):325–37.https://doi.org/10.2174/1566524013363717.
4. 4.Yue Z, Zhang L, Li C, Chen Y, Tai Y, Shen Y, et al. Advances and potential of gene therapy for type 2 diabetes mellitus. Biotechnology & Biotechnological Equipment. 2019 Jan 1;33(1):1150–7.https://doi.org/10.1080/13102818.2019.1643783.
5. Prud'homme GJ, Draghia-Akli R, Wang Q. Plasmid-based gene therapy of diabetes mellitus. Gene Therapy. 2007 Jan 11;14(7):553–64. https://doi.org/10.1038/sj.gt.3302907.
6. Wong MS, Hawthorne WJ, Manolios N. Gene therapy in diabetes. Self/Nonself [Internet]. 2010 Jun 9;1(3):165–75.
7. https://doi.org/10.4161/self.1.3.12643.
8. Calne RY, Gan SU, Lee KO. Stem cell and gene therapies for diabetes mellitus. Nature Reviews Endocrinology [Internet]. 2010 Mar 1;6(3):173–7.https://doi.org/10.1038/nrendo.2009.276.
9. Tan SY, Mei Wong JL, Sim YJ, Wong SS, Mohamed Elhassan SA, Tan SH, et al. Type 1 and 2 diabetes mellitus: A review on current treatment approach and gene therapy as potential intervention. Diabetes & Metabolic Syndrome: Clinical Research & Reviews [Internet]. 2019 Jan;13(1):364–72. https://doi.org/10.1016/j.dsx.2018.10.008.
10. Bornstein SR, Fraser WJ, Steenblock C. The promising potential of gene therapy for diabetes mellitus. Nature Reviews Endocrinology [Internet]. 2024 Aug 29 [cited 2024 Aug 30];1–2. https://doi.org/10.1038/s41574-024-01030-7.
11. Efrat S. Prospects for gene therapy of insulin-dependent diabetes mellitus. Diabetologia. 1998 Nov 23;41(12):1401–9. https://doi.org/10.1007/s001250051085
12. Nett PC, Sollinger HW, Alam T. Hepatic Insulin Gene Therapy in Insulin-Dependent Diabetes Mellitus. American Journal of Transplantation. 2003 Aug 11;3(10):1197–203.https://doi.org/10.1046/j.1600-6143.2003.00221.x
13. Tae Keun Oh, Mei Zi Li, Seung Taik Kim. Gene therapy for diabetes mellitus in rats by intramuscular injection of lentivirus containing insulin gene. Diabetes Research and Clinical Practice. 2006 Mar 1;71(3):233-40.https://doi.org/10.1016/j.diabres.2005.08.005
14. Docherty K. Gene Therapy for Diabetes Mellitus. Clinical Science. 1997 Apr 1;92(4):321–30.https://doi.org/10.1042/cs0920321
15. Welsh N. Prospects for gene therapy of diabetes mellitus. Gene Therapy. 2000 Feb;7(3):181–2. https://doi.org/10.1038/sj.gt.3301134.
16. Giannoukakis N, Robbins PD. Gene and Cell Therapies for Diabetes Mellitus. BioDrugs. 2002 Jan 1;16(3):149–73. https://doi.org/10.2165/00063030-200216030-00001.
17. Sena CM, Bento CF, Pereira P, Seiça R. Diabetes mellitus: new challenges and innovative therapies. EPMA Journal. 2010 Mar;1(1):138–63.https://doi.org/10.1007/s13167-010-0010-9.
18. Sasaki T, Fujimoto K, Sakai K, Nemoto M, Nakai N, Tajima N. Gene and Cell-Based Therapy for Diabetes Mellitus: Endocrine Gene Therapeutics. Endocrine Pathology. 2003 Jan 1;14(2):141–4.https://doi.org/10.1385/EP:14:2:141.

19. Barć P, Antkiewicz M, Śliwa B, Frączkowska K, Guziński M, Dawiskiba T, et al. Double VEGF/HGF Gene Therapy in Critical Limb Ischemia Complicated by Diabetes Mellitus. Journal of Cardiovascular Translational Research. 2020 Sep 1;14(3):409–15.https://doi.org/10.1007/s12265-020-10066-9.
20. Stafeev YS, Menshikov MY, Parfyonova YV. Gene therapy of type 2 diabetes mellitus: state of art. Terapevticheskii arkhiv. 2019 Feb 15;91(2):149–52. https://doi.org/10.26442/00403660.2019.02.000042.
21. Ayuso E, Jimenez V, Callejas D, Mann C, Bosch F. A1-6 AAV gene therapy for diabetes mellitus. EDP Sciences eBooks. 2012 Nov 1;62–70.https://doi.org/10.1051/978-2-84254-237-5.c011.
22. Freeman DJ, Leclerc I, Rutter GA. Present and potential future use of gene therapy for the treatment of non-insulin dependent diabetes mellitus (Review). International Journal of Molecular Medicine. 1999 Dec 1;https://doi.org/10.3892/ijmm.4.6.585.
23. Shabani A, HodaMohammad Dezashibi. A Mini-Review of Current Treatment Approaches and Gene Therapy as Potential Interventions for Diabetes Mellitus Types 1. Advanced Biomedical Research [Internet]. 2023 Jan 1;12(1):219–9.DOI: 10.4103/abr.abr_201_23.
24. Stewart C, Taylor NA, Green IC, Docherty K, Bailey CJ. Insulin-releasing pituitary cells as a model for somatic cell gene therapy in diabetes mellitus. Journal of Endocrinology. 1994 Aug;142(2):339–43.
25. Chellappan DK, Yap WS, Bt Ahmad Suhaimi NA, Gupta G, Dua K. Current therapies and targets for type 2 diabetes mellitus. Panminerva Medica. 2018 Sep;60(3).https://doi.org/10.23736/s0031-0808.18.03455-9
26. Wal P, Aziz N, Prajapati H, Soni S, Wal A. Current Landscape of Various Techniques and Methods of Gene Therapy through CRISPR Cas9 along with its Pharmacological and Interventional Therapies in the Treatment of Type 2 Diabetes Mellitus. Current Diabetes Reviews. 2024 Jul 1;20(6).https://doi.org/10.2174/0115733998263079231011073803.
27. Lee HC, Kim SJ, Kim KS, Shin HC, Yoon JW. Remission in models of type 1 diabetes by gene therapy using a single-chain insulin analogue. Nature [Internet]. 2000 Nov 1;408(6811):483–8.https://doi.org/10.1038/35044106.
28. Cheng Y, Wang H, Li M. The Promise of CRISPR/Cas9 Technology in Diabetes Mellitus therapy: How Gene Editing Is Revolutionizing Diabetes Research and Treatment. Journal of Diabetes and Its Complications. 2023 Aug 1;37(8):108524–4.https://doi.org/10.1016/j.jdiacomp.2023.108524.
29. Dieterle C, Brendel MD, J. Seissler, Eckhard M, Bretzel RG, Landgraf R. Therapie des Diabetes mellitus. Der Internist. 2006 Mar 30;47(5):489-501. https://doi.org/10.1007/s00108-006-1607-x.
30. Yamaoka T. Regeneration therapy for diabetes mellitus. Expert Opinion on Biological Therapy. 2003 Jun 1;3(3):425–33.https://doi.org/10.1517/14712598.3.3.425
31. Dhankhar S, Chauhan S, Mehta DK, Nitika, Saini K, Saini M, et al. Novel targets for potential therapeutic use in Diabetes mellitus. Diabetology & Metabolic Syndrome. 2023 Feb 13;15(1).https://doi.org/10.1186/s13098-023-00983-5