GENE EDITING- A BOON TO DIABETIC TREATMENT

INTRODUCTION

- The inheritance of transformed beta cell that is mis-expressed or coding change in gene(coding insulin secretion) generates altered insulin.
- Diabetes has become one of the major noninfectious diseases that seriously endanger public health. Thus, increases the risk of both Type 1 and Type 2 Diabetes.
- It is a metabolic disease that causes high blood sugar for prolonger time. If not treated, ultimately lapse into coma and die.
- The CRISPR/Cas9 genome editing system has been one of the greatest scientific discoveries in the last decade.
- The highly efficient and precise editing ability of this technology is of great therapeutic value and benefits the basic sciences as an advantageous research tool.
- And is powerful tool among other gene editing tools like ZFN, TALEN for understanding transformed beta cells function, growth, and survival, ultimately applies to broad range of target sequences and show accurate effectiveness towards genetically corrected cells after transplantation.

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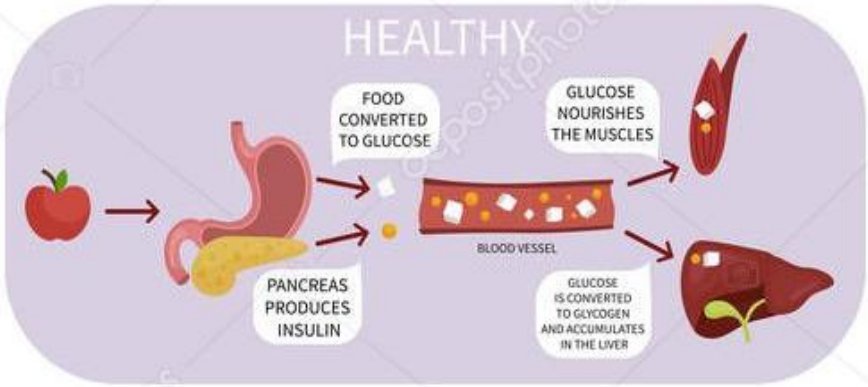
- Consequently, treating the disease
- CRISPR screens have been used primarily in cancer biology, virology, and basic cell biology, but they are been applied to diabetes in transforming the variant type of beta cell to a functional one in Type 1 and Type 2 diabetes.

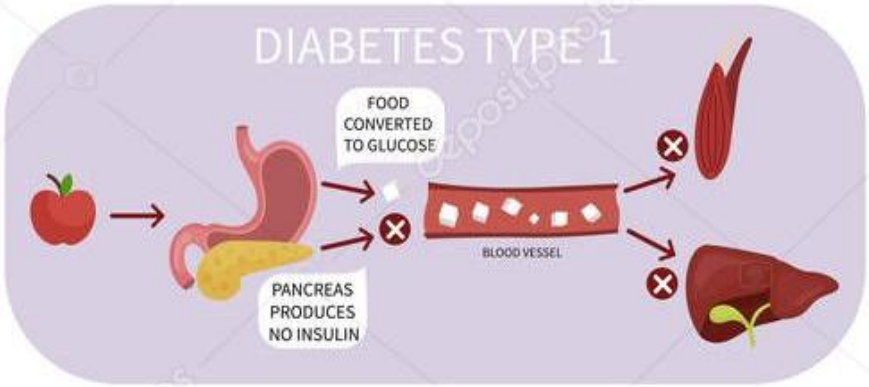
Here we review,

a) Developments in CRISPR/Cas9 technology in the process of clinical applications of diabetes.
b) The available treatments and symptoms of this chronic diabetic conditions.
c) Provide perspective on how it can be used in diabetes research.
d) Show great promise in correcting mutations inType 1 and Type 2 diabetes.

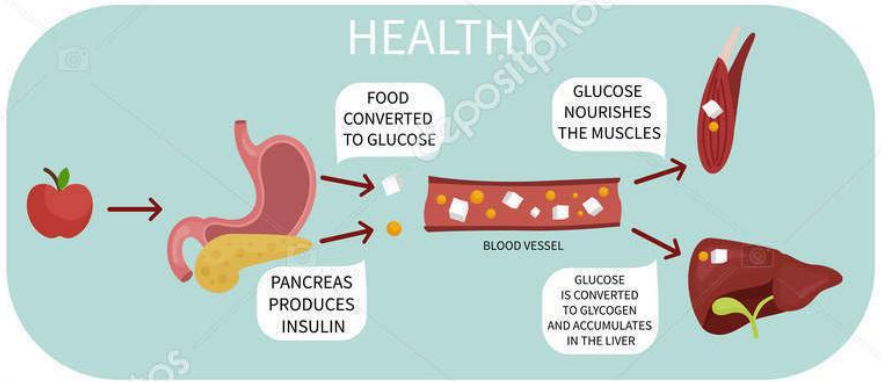
DIABETES

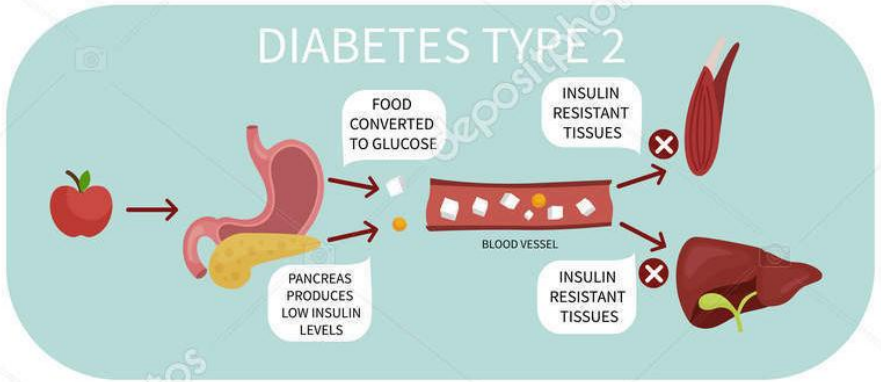
Type 1 - diabetes is an autoimmune disease. The immune system attacks and destroys own cells in the pancreas, where insulin is made. About 10 percent of people with diabetes have this type.





In contrast, Type 2 - diabetes occurs when your body becomes resistant to insulin, and sugar builds up in your blood.





GENE EDITING

The first genome editing technologies were developed in the 1900s and the method includes
1)Homologous recombination
2)Zinc finger nucleases (ZFN
3)Transcription activators like effector nucleases (TALENS)
4)Clustered regularly interspaced short palindromic repeats (CRISPR)
Genome editing also called as gene editing, is used in research fields to modify the genetic material of living organisms in order to have knowledge about the gene function and ways to develop changes in acquired and genetic diseases.

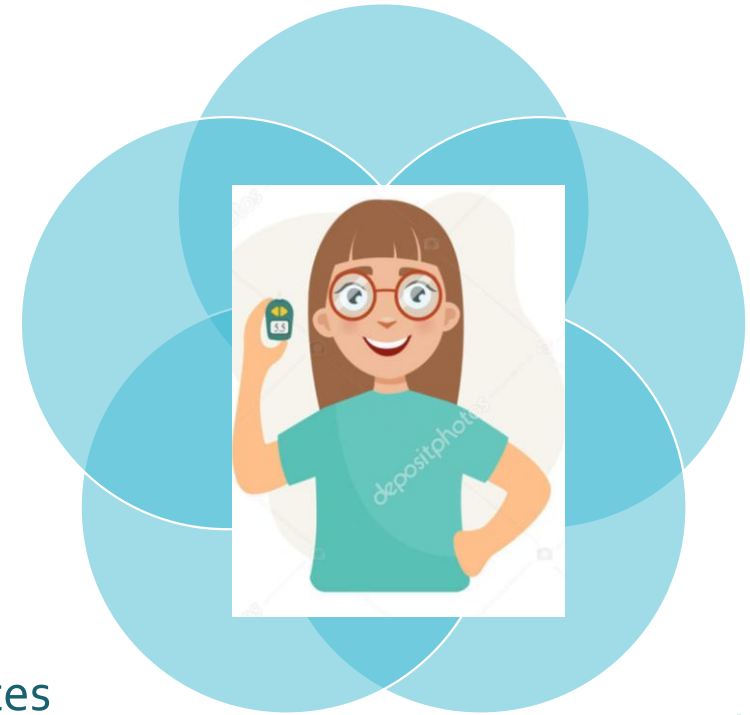
	Zinc-Finger Nuclease ZFN	Transcription Activator-Like Effector Nuclease TALEN	Clustered Regularly Interspaced Palindromic Repeats-CRISPR-Associated-9 (CRISPR–Cas9)
Construction	Protein engineering for every single target	Protein engineering for every single target	20-Nucleotide sequence of single-guide RNA (sgRNA)
Target sequence recognition	Zinc fingers protein, protein-DNA interactions	Repeat variable diresidues (RVDs) repeats, protein-DNA interactions	sgRNA, RNA-DNA interactions
Endonuclease	FokI	FokI	Cas9 and its different variants
Endonuclease construction	3–4 Zinc fingers domains	8–31 RVD repeats	sgRNA synthesis or cloning
Delivery	Two ZFNs around the target sequence	Two TALENs around the target sequence are required	sgRNA complementary to the target sequence with Cas9 protein
DNA sequence recognition size	(9 or 12 bp) × 2	(8–31 bp) × 2	17–20 bp + NGG × 1
Targeting efficiency	Low	Moderate	High
Affordability	Resource intensive and time consuming	Affordable but time consuming	Highly affordable and rapid

SUB TOPICS

NORMAL BLOOD GLUCOSE LEVEL
less than 140 mg/dL (7.8 mmol/L)

Symptoms of Diabetes

Ethical concerns



What are the available treatments for Type 1 and Type 2 and issues faced by patients

Reversing Diabetes

How is CRISPR used for diabetes

•The underlying cause of diabetes varies by type. But, no matter what type of diabetes you have, it can lead to excess sugar in your blood. Too much sugar in your blood can lead to serious health problems.

•Chronic diabetes conditions include type 1 diabetes and type 2 diabetes.

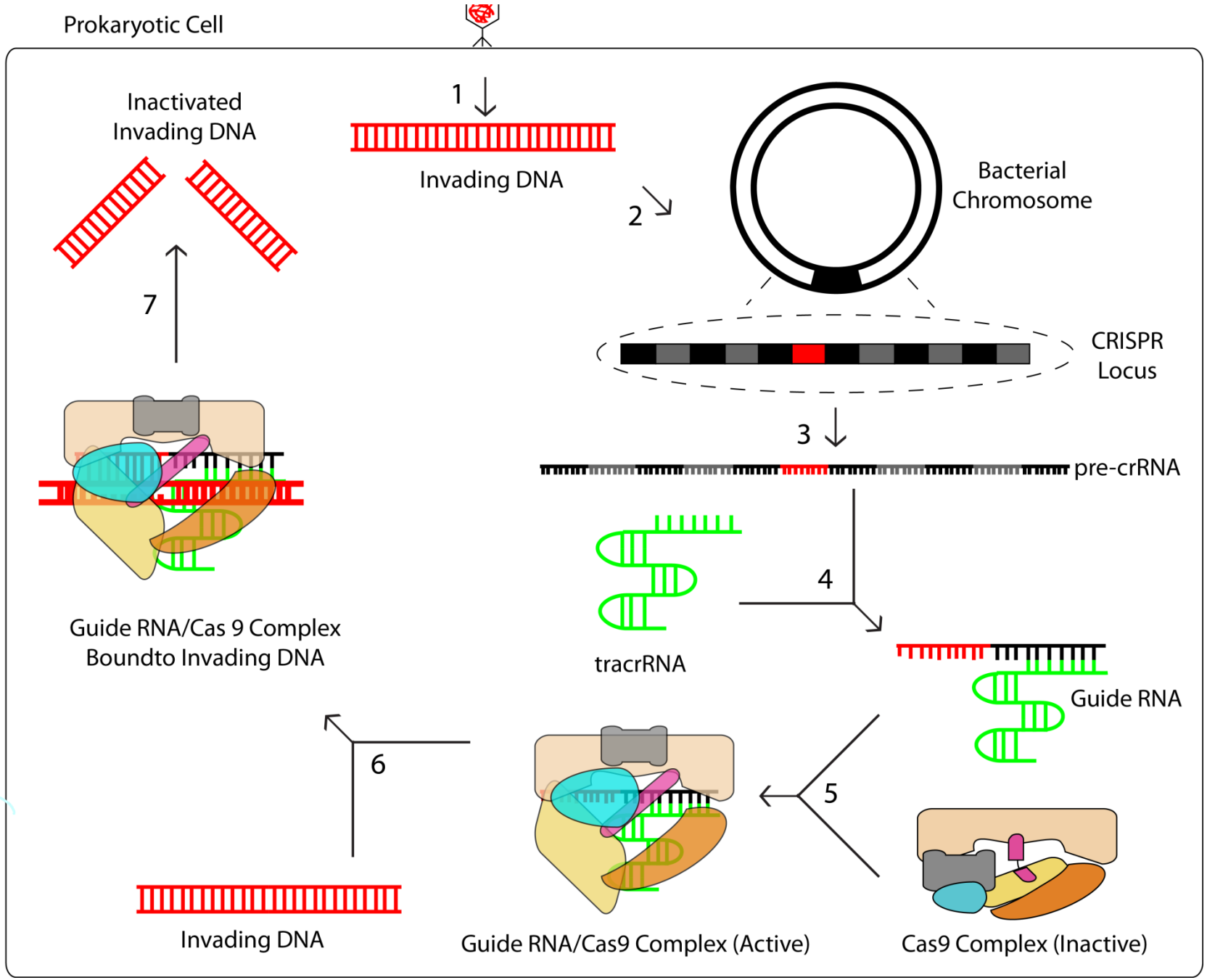
•In type 1 diabetes, symptoms tend to come on quickly and be moresevere.

•Some of the signs and symptoms of type 1 diabetes and type 2 diabetes are:

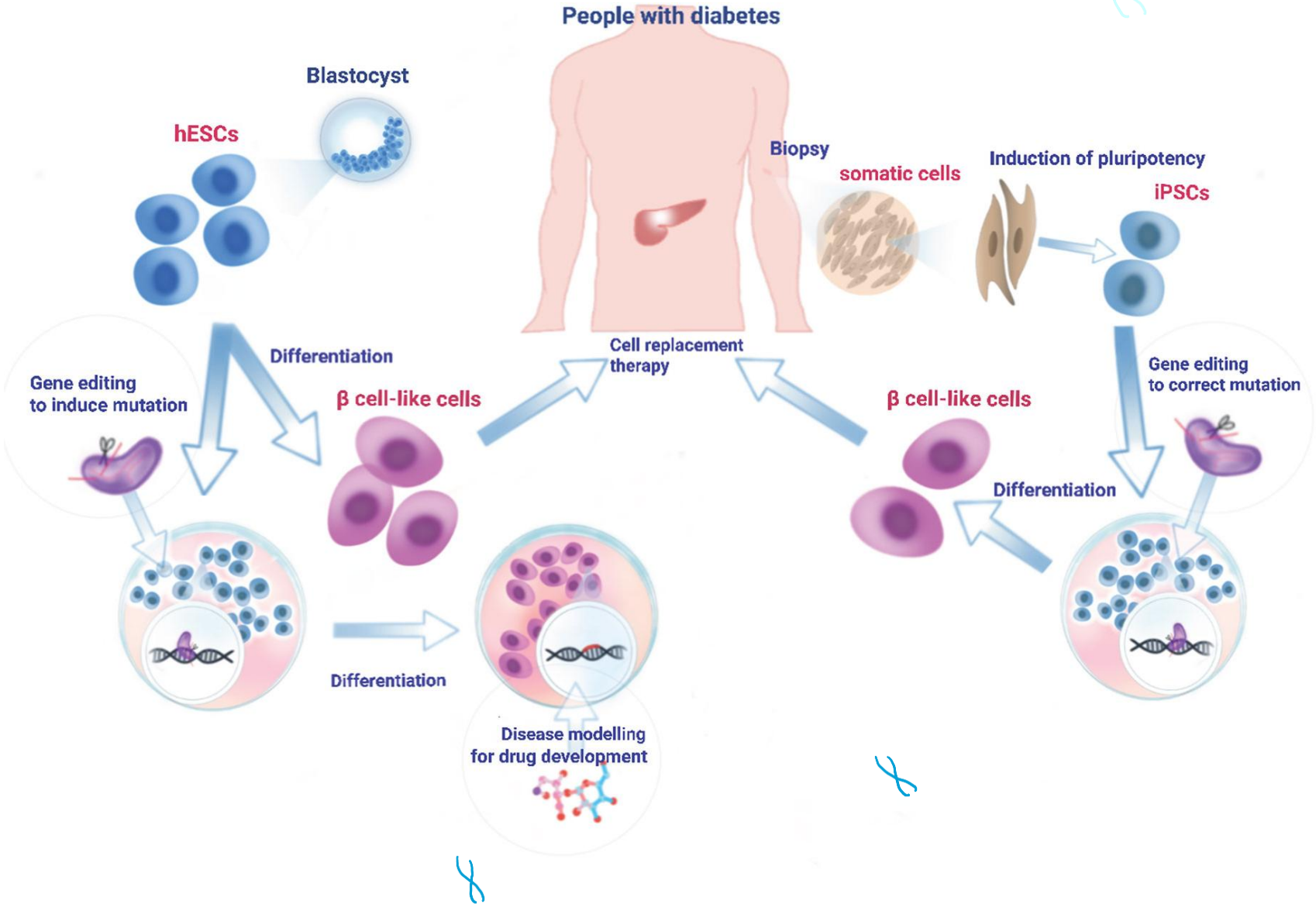
Increased thirst, Frequent urination, Extreme hunger, Unexplained weight loss, Presence of ketones in the urine, Fatigue,Irritability, Blurred vision, Slow-healing sores, Frequent infections,such as gums or skin infections and vaginal infections.

• Type 1 diabetes can develop at any age, though it often appears during childhood or adolescence. Type 2 diabetes, the more common type, can develop at any age, though it's more common in people older than 40.

•Clustered regularly interspaced palindromic repeats (CRISPR) refers to sequences in the bacterial genome.
•They afford protection against invading viruses (Adaptive immunity), when combined with a series of CRISPR-associated (Cas) proteins.
•Cas9 is an endonuclease that cuts both strands of DNA. Cas9 is directed to its target by a section of RNA. This can be synthesised as a single strand called a synthetic single guide RNA (sgRNA made up of crRNA and traRNA).
•The section of RNA which binds to the genomic DNA is 18–20 nucleotides. In order to cut, a specific sequence of DNA of between 2 and 5 nucleotides (the exact sequence depends upon the bacteria which produces the Cas9) must lie at the 3′ end of the guide RNA: this is called the protospacer adjacent motif (PAM).
•Repair after the DNA cut may occur via two pathways: non-homologous end joining, (typically leading to a random insertion/deletion of DNA)or homology directed repair (homologous piece of DNA is used as a repair template).


Prokaryotic Cell
Inactivated
Invading DNA
1
Invading DNA
2
Bacterial
Chromosome
CRISPR
Locus
3
pre-crRNA
7
Guide RNA/Cas 9 Complex
Boundto Invading DNA
tracrRNA
4
Guide RNA
6
5
Invading DNA
Guide RNA/Cas9 Complex (Active)
Cas9 Complex (Inactive)

Using induced pluripotent stem cells produced from the skin of a patient with a rare, genetic form of insulin-dependent diabetes called Wolfram syndrome, researchers transformed the human stem cells into insulin-producing cells and used the gene-editing tool CRISPR-Cas9 to correct a genetic defect that had caused the syndrome. They then implanted the cells into lab mice and cured the unrelenting diabetes in those mice



REFERENCE

1. World Health Organization. 10 facts about diabetes,2013. Available at:
2. http://www.who.int/features/factfiles/diabetes/en/.
3. https://images.app.goo.gl/PJ4XfNxBd2JYwLYA7

source:National diabetes and diabetic retinopathy survey report 2019

4.World health organisation ,what is diabetes mellitus

5.https://www.who.int/health-topics/diabetes

6.Karvonen M, Pitkaniemi M, Pitkaniemi J, Kohtamaki K, Tajima N, Tuomilehto J. Sex difference in the incidence of insulin-dependent diabetes mellitus: an analysis of the recent epidemiological data.World Health Organization DIAMOND Project Group. Diabetes Metab Rev. 1997;13(4):275–291.

7.Steck AK, Rewers MJ. Genetics of type 1 diabetes. Clin Chem. 2011;57(2):176–185.

8.Roden M., Shulman G.I. The integrative biology of type 2 diabetes. Nature. 2019;576:51–60. doi: 10.1038/s41586-019-1797-8.

9.NCD Risk Factor Collaboration Worldwide trends in diabetes since 1980: A pooled analysis of 751 population-based studies with 4.4 million participants. Lancet. 2016;387:1513–1530. doi: 10.1016/S0140-6736(16)00618-8.

10.Gaj T, Gersbach CA, Barbas CF., III ZFN, TALEN, and CRISPR/Cas-based methods for genome engineering. Trends Biotechnol. 2013;31:397–405. doi: 10.1016/j.tibtech.2013.04.004

11.https://www.synthego.com/blog/genome-editing-techniques

12.difference between zfn,talen,crispr www.researchgate.net

THANK YOU