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Research Article

Formulation of Insulin Bioadhesive Gel by Using Different Polymers

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Abstract
Diabetes mellitus is chronic disorder it is one of common disorder in all age groups spreaded through the world. This is treated by using many drugs in which insulin is a drug which is used to treat the type I diabetes mellitus but insulin is admistered by parental route. In this research insulin bioadhesive nasal gel is prepared by using different polymers to admistered though nasal route and increase its absorption and prolonged action.
Key words: Diabetes Mellitus, Insulin, Mucoadhesive, Nasal Gel.

INTRODUCTION

1.1 Diabetes Mellitus [Udaykumar, P, 2007; Paradkar, A.R. *et al*., 2016; medicalnewstoday.com; everydayhealth.com]

1.1.1. Definition

Diabetes mellitus is a chronic metabolic disorder characterized by hyperglycemia and altered metabolism of carbohydrates, lipids and proteins.

1.1.2 Types of Diabetes

Diabetes Mellitus is classified into 4 types
Type I- It is also called as Insulin dependent diabetes mellitus (IDDM).
Type II- It is also called as Non- Insulin dependent diabetes mellitus (NIDDM).
Type III- pancreatogenic diabetes, and
type 3c diabetes
Type IV- Gestational Diabetes

In following section we discuss about the different types of diabetes, sign and symptoms diabetes complications and about the insulin like its development, structure, its actions on the body and adverse effects.

Type I or Insulin Dependent Diabetes Mellitus:

It is an auto immune disorder where antibodies destroy the beta cells of the islet of langerhans leading to insulin deficiency. It usually occurs in the young children and adolescents. Adolescent is nothing but an irreversible onset diabetes mellitus. The incidence of this type of diabetes mellitus is fortunately low.

Sign and Symptoms:

Type I shows the following sign and symptoms

1. Polyuria,
2. Polyphagia,
3. Polydispia,
4. Weight loss
5. Polydispia
6. Decreased muscle strength and irritability
7. Blurred Vision
8. Ketoacidosis
9. Nausea and vomiting
10. Frequent infections
11. Headache
12. Fast heart rate, Etc.

Type II or Non- Insulin Dependent Diabetes Mellitus:

It is maturity onset. Most of patients are obese. There is both reduced sensitivity of tissues to utilize the insulin and converts the glucose into glycogen and also impaired regulation of insulin secretion by beta cells of langerhans in pancreas by that decrease in circulating concentration of insulin. Diabetes is discovered only when sugar appears in the urine.

Sign and Symptoms:

1. Increased Thirst
2. Frequent urination
3. Blurred vision

Type III

It is due to causes like pancreatectomy drugs and non pancreatic disease. Type III occurs when neurons in the brain become unable to respond to insulin, which is essential for basic functions including memory and learning. Some researchers believe insulin deficiency is central to the

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cognitive decline of Alzheimer's disease.

Sign and Symptoms:
1. Memory loss that interfere daily living and social interactions.
2. Difficulty in completing familiar tasks.
3. Misplacing things.
4. Decreased ability to make judgments.
5. Sudden changes in personality or demeanor.

Type IV or Gestational Diabetes:
This may be due to placental hormones that are effect insulin resistance. Yet for many pregnant women, gestational diabetes does not cause any recognizable symptoms.

That is why screening tests for the conditions are recommended for all pregnant women.

Sign and Symptoms:
1. Frequent urination
2. Increased Thirst
3. Extreme tiredness
4. Nausea and vomiting
5. Weight loss even with increased appetite
6. Blurred vision.

Complication of Diabetes [Deshpande, A.D. *et al*., 2008; mayoclinic.org; Mayoclinic.org]
- Heart and blood vessel disease- complications include cardiovascular disease, stroke, and peripheral vascular disease. Peripheral vascular disease may lead to bruises or injuries that do not heal, gangrene, and, ultimately, amputation.
- Diabetic Neuropathy- caused by damage to the nerves and the small blood vessels that nourish your nerves with oxygen and nutrients.
- Diabetic Nephropathy- is defined as persistent proteinuria in patients without urinary tract infection or other diseases causing the proteinuria. In patients with type 1 diabetes, development of clinical nephropathy is a relatively late event; however, in patients with type 2 diabetes, diabetic proteinuria may be present at diagnosis. Many people with diabetes develop high blood pressure. That can also damage your kidneys.
- Diabetic Retinopathy- damage of retina. Diabetic retinopathy is the most common micro vascular complication among people with diabetes.
- Foot problems-caused by damage to the nerves and reduced blood flow to your feet.
- Gum disease and other dental problems-because a high amount of blood sugar in your saliva helps harmful bacteria grow in your mouth. The bacteria combine with food to form a soft, sticky film called plaque. Plaque also comes from eating foods that contain sugars or starches. Some types of plaque cause gum disease and bad breath. Other types cause tooth decay and cavities.
- Sexual and bladder problems-caused by damage to the nerves and reduced blood flow in the genitals and bladder
- Skin conditions- some of which are caused by changes in the small blood vessels and reduced circulation.
- People with diabetes are also more likely to have infections, including skin infections.
- Pregnancy complication
- Alzheimer disease
- Hearing impairment
- Dementia

1.2 Mucoadhesive [Puratchikody, A. *et al*., 2011]

1.2.1. Definition: Bioadhesion is a drug delivery system in which two materials are held together and in which one is biological in nature, both are held together for extended periods of time by interfacial forces. When the biological membrane is mucus or mucous membrane is called as mucoadhesion.

1.2.2. Advantage [Tangri, P. *et al*., 2011; Budharani. *et al*., 2020; Kaur, A. *et al*., 2013]
1. Increases the residence time of the dosage forms at the site absorption.
2. Drug bioavailability increases and therapeutic efficacy increase.
3. Rapid absorption because of high blood flow.
4. First pass metabolism can be avoided by that drug bioavailability is increases and dose will be reduced.
5. Stomach acidic degradation of drug can be prevented by polymer coating.
6. Improved patient compliance.
7. The unconscious and trauma patients drug can be admistered.

1.2.3. Disadvantage

1. Local ulcer effect due to long term contact of dosage form at mucous surface.
2. Patient acceptability is the big issue because of taste.
3. Eating and Drinking is prohibited.

1.3. Insulin [Joshi, S.R. *et al*., 2007; Gualandi-Signorini, A.M. *et al*., 2001]

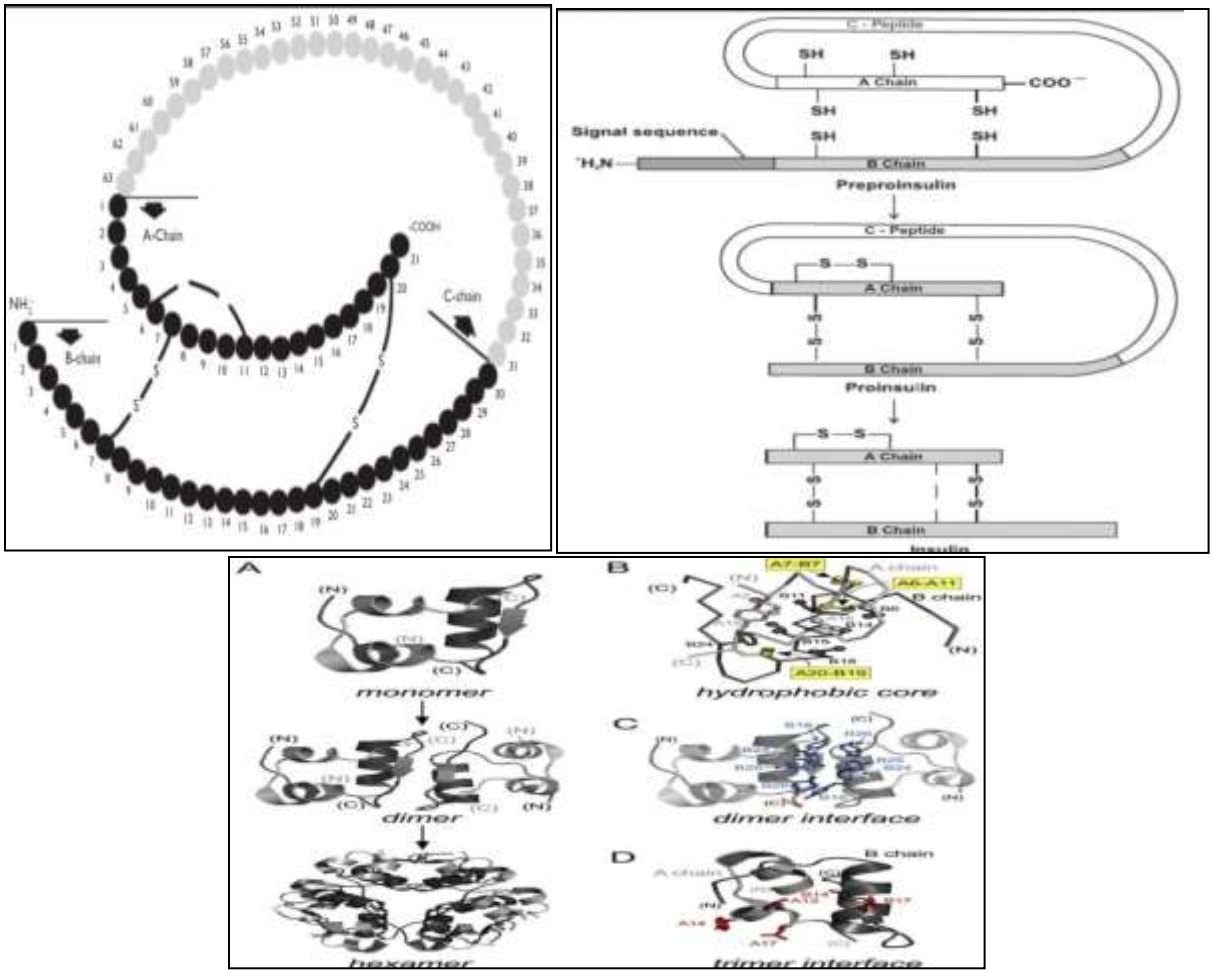
1.3.1. Insulin Development History

The discovery of insulin is a breakthrough for the treatment of diabetic and the care of diabetic patients. The development of proper purifying and modifying method for obtain insulin took 30 more years. In 1889 the scientist gives the insulin. In Germany Oskar Minkowski and Joseph von Mering notice that pancreatectomy in experimental animal causes the severe diabetes mellitus and begun the speculation that a mysterious secretion produced by the pancreas is responsible for metabolic control. In the first ten years of Twentieth Century it was broadly hypothesized that an internal solution secreted by pancreas controls carbohydrate metabolism. In 1907 Belgian Investigator J de Meyer proposed that islet cell of pancreatic secret internal secretion and it to be Insulin and in 1916 Sharpey Schafer in Britain suggest same name.

In 1921 two young physician Frederick Banting and forth year medical student Charles Best along with the Prof. McLeod in the Toronto University found final link in a series of studies that guessed the pancreas secretion of substance capable of to decrease blood glucose concentrations. The isolated insulin cured the hyperglycemia in diabetic dogs. In 1922 Leonard Thompson in 14 year diabetic patient first time successfully admistered. In 1928 study proved that the hormone is nothing but protein. Banting and Best identified insulin is short acting that 6hours effective, so repeated dosing is required. To overcome this problem many scientist involve developing the better insulin. In 1936 Hans Christian Hagedorn and colleagues developed the first clinically useful protracted intermediate acting insulin in the form of neutral suspension of protamine insulin but it is chemically unstable. So further preparation is prepared by adding excess amount of protamine and small amount of zinc and it is known as protamine zinc insulin. It shows prolonged hypoglycemic effect of up to 24 hrs. But it cause hypoglycemia with reports of sudden and severe attacks and also onset of action slow that is 1-3hrs. To form the intermediate insulin by using PZI. The PZI is admixture with the soluble insulin. It act up to 16-18hrs, but it is unstable and had to be administered as separate injections, causing inconvenience to patients. Finally, in 1946, Hagedorn and colleagues introduced crystalline neutral protamine Hagedorn (NPH) insulin, which was a more stable PZI modification combining insulin and protamine in isophane proportions at neutral pH in the presence of a small amount of zinc and phenol and/or phenol derivatives to generate tetragonal oblong-shaped crystals. Unlike PZI, NPH insulin preparation could be premixed with an intermediate acting insulin and admistered, once or twice daily, used alone or in combination with soluble insulin as patient requirement.

Structure of Insulin [Bell, G.I. *et al*., 1980; Derewenda, U. *et al*., 1989].

Like most of the other hormones insulin is a made of protein comprising of 2 polypeptide chains- chain A- containing 21amino acids residue, Chain B containing the 30 amino acids residues. The A& B chains are linked by disulphide bridges. In addition A chain contains an intrachain disulphide bridge linking residue 6 and 11. The structure of insulin as follows.



C- Chain, which connects A & B, chains is liberated along with insulin after breakdown of proinsulin. Insulin monomer aggregate to form dimmers and hexamers. Although insulin is active as a monomer during its biosynthesis and storage it assembles to dimmers and in the presence of zinc to hexamers. X-ray analysis has revealed the 3-dimensional structure of the insulin molecule in its hexameric, dimeric, and monomeric states. Two main conformations of insulin which differ in the extent of helix in the B chain at B9-B20 and B1-B20 respectively have been identified. Other variations are seen in insulin when dimeric or monomeric. Reagents such as chlorides and phenol govern the conformation present in the insulin hexamers and this can influence the behavior and properties of insulin preparations.

1.3.2. Biosynthesis of Insulin [Bliss, M, 1982; Hutton, J.C, 1994]

Insulin is synthesized in the beta cells of pancreas in the form of preproinsulin which is the ultimate precursor and gene for the same is located on chromosome 11 close to that for insulin like growth factor 2(IGF-2).

Within a minute after synthesis it is discharged into cisternal space of rough endoplasmic reticulum where it is cleaved into proinsulin by proteolytic enzymes. Proinsulin with a C-chain linking A and B chains is then transported micro vesicle to the Golgi apparatus. Proinsulin is released in vesicles. Conversion of proinsulin to insulin continues to maturing granules through the action of prochrome conversate 2&3 and carboxy peptidase.

1.3.3. Insulin Pharmacology [Budhiraja, R.D]

Insulin effect on glucose uptake: Binding of insulin to the alpha-subunits leads to autophoshorylation and activation of the enzyme tyrosine kinase in the beta unit and physiological effect of insulin. Insulin stimulates glucose

transport across cell membrane by ATP dependent translocation of glucose transporters like GLTU4. GLTU has channels; through that glucose enter from extra cellular fluid to Intra cellular fluid. Then glucose metabolize. Insulin also inhibits some DNA transcription, which reduces gluconeogenesis.

Role of Insulin in Metabolism [wikipedia.org]**:** The insulin is hormone which is involved in many catabolism processes like glycogen, triglyceride, protein, DNA synthesis.

Some actions are direct and some actions are indirect. In following sections we discuss the functions of insulin in detail.

- Glycogen Synthesis: Insulin increases the glucose uptake by activating the GLUT4 transport system in the liver. The absorb glucose converted into glycogen.
- Glycogenolysis and Gluconeogenesis: It prevents the conversion of glycogen into glucose in the liver by inhibiting the phosphorylase and cyclic AMP enzyme. By inhibiting gluconeogenesis enzyme it prevent the formation of glucose from non carbohydrate sources (protein and lipids).
- Protein and DNA Synthesis: Insulin increase the glucose uptake in the skeletal muscles and also amino acids uptake by that it increases the DNA replication and protein synthesis and it is anabolic reaction.
- Proteolysis: Insulin decreases the protein breakdown.
- **Fatty acid Synthesis and Esterification:** Increase the absorption of blood glucose in the adipose tissue and converted into glycerol, alpha glycerophosphate and fatty acid and its synthesis into triglyceride (fat).
- **Lipolysis:** Reduce the conversion of triglycerides into glycerol and fatty acids in adipose tissues by that Gluconeogenesis is reduced.
- **Autophagy:** Postprandial level inhibits the autophagy completely and decreases the level of degradation of damaged organelles.
- Sodium renal excretion decrease.
- Entry of insulin in the brain increases the verbal memory, learning and memory.
- Insulin stimulates the gonadotropin releasing hormone in the hypothalamus reason in the brain thus it favors the fertility.

Adverse Effect [Tripathi, K.D, 2019; Murgesh, N; Dandiya, P.C. *et al*., 2008; Kale, R.S. *et al*., 2016; statpearls.com**].**

Hypoglycemia: It is most frequent and most serious reaction. Hypoglycemic episodes are more common. It is common in all type of diabetic patients. Common reason for hypoglycemia is inadvertent injection of large dose or by missing of meal after injection or performing vigorous exercise.

Allergy Reaction: Some allergic reaction causes at the site of injection like itching, redness and swelling.

Weight Gain: Increase in body weight.

Lipodystrophy: Atrophy of subcutaneous fat at the site of injection takes place.

Presbyopia: It is a loss visual accommodation due to alterations in the physical properties of the lens occurring due to rapidly controlled diabetes with insulin.

Somogyi Effect: Some patients who take insulin before bed wake up with high blood sugar levels. This effect occurs when the insulin causes a hypoglycemic condition in the body, by that activates the antihyperglycemic hormones like cortisol and adrenaline, resulting in rebound hyperglycemia. This can be corrected by reducing the dose of bedtime insulin or changing the time of insulin dosing.

Dawn Phenomenon: It is the presence of high blood glucose levels in the body in the early hours of the day due to inadequate insulin concentration in the body. To correct this phenomenon, the dose of bedtime insulin needs to increase to be able to keep the blood glucose levels under normal condition throughout the night and the early hours of the morning.

Table-1: Materials of the study

Serial no	Chemical name	Source
1	INSULIN	Gift Sample
2	Gelatin	Molychem Chemicals Mumbai
3	Carbopol 934	Charco, Hoshiarpur, Punjab
4	Lecithin	Molychem Chemicals Mumbai
5	Sodium Alginate	Molychem Chemicals Mumbai
6	Starch	Molychem Chemicals Mumbai
7	Chitosan	Molychem Chemicals Mumbai
8	HPMC	BRM Chemicals Pvt. Ltd. New Delhi
9	B-Cyclodextrin	Molychem Chemicals Mumbai
10	Benzylalkonium Chloride	Molychem Chemicals Mumbai
11	Glycerol	Molychem Chemicals Mumbai
12	Sodium Metabisulphite	Molychem Chemicals Mumbai
13	Sodium Bicarbonate	Molychem Chemicals Mumbai
14	Sodium Chloride	Molychem Chemicals Mumbai
15	Distilled Water	Aqualine Distillation

2. METHODS

2.1. Analytical Methods

2.1.1 Determination of λ max [cdn.juniata.edu]
In quartz cuvetts take a blank solution and correct the base line in the range of 450-700nm. Take the sample solution in the cuvett and scan in the range of 450-700nm.

Preparation of Standard Calibration Curve [Azhar, et al., 2013]
Insulin (30mg/100mL) solution was added in 11 test tubes as 0.0, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9 and 1.0 mL, respectively in each tube. Distilled water upto 1.0 mL volume 1.0, 0.9, 0.8, 0.7, 0.6, 0.5, 0.4, 0.3, 0.2, 0.1 and 0.0 mL was added in each test tube. Then reagent I was added (0.5 mL) and the solutions were mixed well and incubated for 15 min at room temperature. The reagent II solution was added (0.5 mL) to each tube and incubated for 30 min. The spectrophotometric absorbance of each standard sample was taken at 500nm to establish the standard curve after plotting protein concentration against absorbance.

2.1.2 Determination of Melting Point
The drug was finely powdered and charged in thin glass capillary tube, one end of which was sealed. Sufficient amount of drug was filled in the glass capillary, to form a column at the bottom of the tube (2.5-3.5 mm height), when packed down closely as possible by moderate tapping on solid surface. The capillary tube was placed in a melting point apparatus and the range of temperature when the drug just starts melting and till it completely melted was noted.

Table-2. Formulation Table of Insulin Mucoadhesive Nasal Gel

Sr. No	Ingradients	F1	F2	F3	F4	F5	F6	F7
1	Insulin	0.694mg	0.694mg	0.694mg	0.694mg	0.694mg	0.694mg	0.694mg
2	Gelatin	1.7%						
3	Carbopol 934		1.7%					
4	Lecithin			1.7%				
5	Sodium Alginate				1.7%			
6	Starch					1.7%		
7	Chitosan						1.7%	
8	HPMC							1.7%
9	B-Cyclodextrin	0.3%	0.3%	0.3%	0.3%	0.3%	0.3%	0.3%
10	Benzylalkonium	0.02%	0.02%	0.02%	0.02%	0.02%	0.02%	0.02%

	Chloride							
11	Glycerol	9.5ml	9.5ml	9.5ml	9.5ml	9.5ml	9.5ml	9.5ml
12	Sodium Metabi-sulph	0.8%	0.8%	0.8%	0.8%	0.8%	0.8%	0.8%
13	Sodium Bicar-bonate	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.
14	Sodium Chloride	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.
15	Distilled Water	To 100ml	To 100ml	To 100ml	To 100ml	To 100ml	To 100ml	To 100ml

2.2.1. Preparation Method of Insulin Muco-adhesive Nasal Gel [Wang, Y. *et al*., 2017]
In this experiment the Insulin mucoadhesive gel is prepared by cold method.

As per formulation table, the insulin is weighed and stirred with sufficient quantity of double distilled water and kept overnight at 40 c in a refrigerator. The sufficient quantity of polymer is added to different beakers as per the formula and slowly add with continuous stirring and 0.02% Benzylalkonium Chloride as a preservative , 9.5ml Glycerol as a humectant and co- solvent, 0.8% Sodium Metabisulphite as a antioxidant, 0.9% NaCl is used as Osmotic agent and Sodium bicarbonate buffering agent to adjust the pH are mixed. The dispersion is then stored in a refrigerator until clear solution is formed and finally volume is adjusted with distilled water.

3. RESULT AND DISCUSSION

Nine formulations are prepared as per the formulation table and prepared formulations are to be further subject to the following evaluations.
1. Determination of Isotonicity
2. Determination of pH
3. Determination Gelation Temperature
4. Estimation of Viscosity
5. Measurement of Gel strength
6. Estimation of percentage of drug in formulations
7. Drug absorption is estimated by In-vitro Analysis.

Preparation Figures

4. REFERENCE

1. Udaykumar, P. "Textbook of Pharmacology for Dental and Allied Health Sciences." *Jaypee Brothers Medical Publishers New Delhi* 2 (2007): 364-365.
2. Paradkar, A.R. and Chunawala, S.A "Text of Hospital and Clinical Pharmacy." *Nirali Prakashan Pune* 20 (2018): 19.1-19.13.
3. https://www.medicalnewstoday.com.
4. https://www.everydayhealth.com.
5. Deshpande, A.D., Harris-Hayes, M. and Schootman, M. "Epidemiology of Diabetes and Diabetes-Related Complications." *Phys Ther*. 88.11 (2008):1254–1264.
6. https://www.mayoclinic.org/disease-conditions/type-1-diabetes.
7. https://www. Mayoclinic.org/disease-conditions/type-2-diabetes.
8. Puratchikody, A., Prasanth, V.V., Mathew, S.T. and Kumar, A. "Buccal drug delivery: past, present and future-a review." *International Journal of Drug Delivery* 3.2 (2011): 171-184.
9. Tangri, P. and Madhav, N.S. "Oral Mucoadhesive Drug Delivery System: A Review." *International Journal of Biopharmaceutics* 2.1 (2011): 36-46.
10. Budharani, et al., "Mucoadhesive Buccal Drug Delivery System: A review." *Am. J. PharmTech Res*. 10.2 (2020).
11. Kaur, A. " Mucoadhesive Drug Delivery System: A Review." *Int. J. Drug Dev. And Res*. 5.1 (2013): 11-20.
12. Joshi, S.R., Parikh, R.M. and Das, A.K. "Insulin-history, biochemistry, physiology and pharmacology." *Journal-association of physicians of India* 55.L (2007): 19.
13. Gualandi-Signorini, A.M. and Giorgi, G. "Insulin formulations-a review." *European review for medical and pharmacological sciences* 5 (2001): 73-84.
14. Bell, G.I., Pictet, R.L., Rutter, W.J., Cordell, B., Tischer, E. and Goodman, H.M. "Sequence of the human insulin gene." *Nature* 284.5751 (1980): 26-32.
15. Derewenda, U., Derewenda, Z., Dodson, G.G., Hubbard, R.E. and Korber, F. "Molecular structure of insulin: the insulin monomer and its assembly." *British Medical Bulletin* 45.1 (1989): 4-18.
16. Bliss, M. "The discovery of Insulin." *Chicago Press* (1982).
17. Hutton, J.C. "Insulin Secretary Granules biogenesis and the proinsulin- processing

endopeptidases." *Dialectological* 37.2 (1994): 548-56.
18. Budhiraja, R.D. and Krishan, P. "Text of Elementary Pharmacology and Toxicology." *Vallabh Prakashan Dehli* 5: 363.
19. https://en.wikipedia.org/wiki/Insulin
20. Tripathi, K.D. "Text of Essential of Medical Pharmacology." *Jaypee Brothers Medical Publishers New Dehli* 8 (2019): 288.
21. Dandiya, P.C. and Kulkarni, S.K. "Text of Introduction to Pharmacology." *Vallabh Prakashan Dehli* 7 (2008): 383.
22. Kale, R. S., Kale, R. R. "Text of Pharmacology and Toxicology." *Nirali Prakashan Pune* 18 (2016): 10.5.
23. https://www.statpearls.com/ArticleLibrary/viewarticle/23563
24. https://cdn.juniata.edu/documents/departments/sim/_Labs-and-Equipment/Chemistry/Visible-Spectroscopy/ProceduralLabs/Determination%20of%20Wavelength%20of%20Maximum%20Absorbance.doc
25. Azhar, et al. "Isolation and characterization of *Pseudomonas stutzari* strain K-1 protein." *International Journal of Chemical and Biochemical Sciences* 3 (2013): 69-73.
26. Wang, Y., Jiang, S., Wang, H. and Bie, H. "A mucoadhesive, thermoreversible in situ nasal gel of geniposide for neurodegenerative diseases." *PloS one* 12.12 (2017): e0189478.

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Conflict of Interest- Nil

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