

POTENTIAL OF GLUCAGON LIKE PEPTIDE-I IN THE THERAPY OF OBESITY IN ASSOCIATION WITH METABOLIC SYNDROME

Abstract

Obesity and associated metabolic syndrome (type 2 diabetes mellitus and heart diseases) are one of the main reasons of death globally. Several drugs are currently approved and available in the market for obesity, type II diabetes mellitus and cardiovascular diseases. But, targeting a specific receptor which can potentially reduce obesity and associated metabolic syndrome will be helpful in future therapy. Current review mainly focuses on the possible mechanistic role of glucagon like peptide-1 (GLP-1), its agonists and glucagon like peptide-1 receptor (GLP1R) in the treatment of obesity and associated metabolic syndrome for future consideration.

Keywords: Glucagon like peptide-1 (GLP-1) metabolic syndrome, obesity, Type II diabetes mellitus and cardiovascular diseases

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I. INTRODUCTION

Obesity and associated metabolic syndrome are at higher risk in the individuals nowadays [1]. Enteroendocrine L- cells produce gastrointestinal incretin hormones like Glucagon like Peptide 1 (GLP-1) i.e., huge peptide hormone in intestine that increases the insulin secretion and decrease the pancreatic discharge of glucagon [2]. In addition, it is expressed in the gut, the brainstem and it acts through the GLP-1 receptor (GLP1R) [3]. In addition, hormone of huge peptide exerts its direct effects on the heart and blood vessels [4,5]. Based on these actions, GLP-1 and its active metabolites were thought to have a therapeutic role in obesity and associated metabolic disorders that includes type II diabetes and cardiovascular diseases.

II. OBESITY AND ASSOCIATED METABOLIC SYNDROME- PREVALENCE

Now a days, obesity has increased prevalence and considered as a worldwide health concern. Worldwide, more than 39% of adults were overweight in 2016 [6]. By 2030, it is estimated that the prevalence of obesity and overweight are expected to reach a level of 89% in males and 85 % in females, respectively [7]. Obesity is also associated with metabolic disturbances such as insulin resistance and dyslipidemia leading to diabetes, hypertension and atherosclerotic disease, combinedly known as metabolic syndrome [8]. Metabolic syndrome ranges approximately from 20 to 25% in adults and 0 to 19.2% in children worldwide [9]. Metabolic syndrome prevalence is estimated to increase approximately by 53% by 2035[10].

III. GLUCAGON-LIKE PEPTIDE-1 (GLP-1)

- Structure:** Peptide of glucagon is a gut derived hormone from peptide made up of 30 amino acid long chain [11]. GLP-1 was compounded and produced in two different biological active isoforms in the body concurrently. GLP-1(7-36) amide form is the amidated peptide form and GLP-1(7-37) is the glycine extended peptide form of GLP-1 [12]. However, GLP-1(7-36) amide form was observed mostly in human and rat intestines and acts as one of the most dominant isoforms in the brain [13]. Figure 1 explains the pattern of GLP-(7-36) amide form.

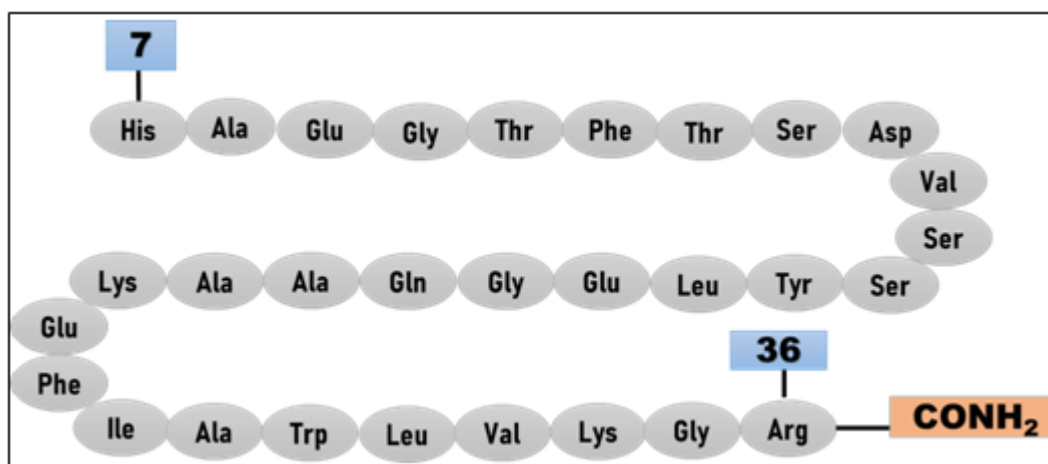


Figure 1: Glucagon like peptide-1 (7-36) amide form structure

- 2. Synthesis, distribution and secretion:** The gastrointestinal epithelium wall comprises numerous types of cells which includes enteroendocrine cells, one of the key components of the gut-brain-pancreas axis [14]. Glucagon-Like peptide-1 is synthesised peripherally from the duodenum ileum and jejunum and large bowel endocrine L- cells. Additionally, GLP-1 is also observed centrally to a lesser amount in the perikarya of neurons and also located in the olfactory bulb, tegmentum dorsal and frontal portion of medulla, and within the solitary tract nucleus [15, 16].

GLP-1 is synthesised from proglucagon, a GLP-1 precursor molecule. Proglucagon is a peptide molecule made up of 160 amino acids and cleaved in between 78-107/8 amino acids by prohormone convertase 1/3 in L cells of intestine or the brain for formation of Glucagon-like peptide-1 [17].

GLP-1 secretion is dependent upon meal intake [18]. GLP-1 excitation is controlled by the neural mechanism through Gastrin Releasing peptide (GRP) and Acetylcholine [19]. Upon ingestion, digestible carbohydrates were absorbed in the form of glucose, galactose and fructose which quicken the production of GLP-1 via sodium - glucose cotransporter (SGLT-1) dependant slight inward current induction and through the glucose transporter-2 (GLUT 2) [20,21]. Whereas, ingestion of non-digestible carbohydrates undergoes fermentation that results in formation of small chain fatty acids and promotes an GLP-1 secretion via FFAR3 and FFAR2 receptors (Free fatty acid receptors 3 and 2) and G-Protein coupled receptors [22]. Ingestion of dietary lipids “majorly triglycerides” were absorbed by means of glycerol and FFA [23]. Long-chain unsaturated free fatty acids stimulate the GLP-1 secretion by binding with FFAR 1 and FFAR 4 [24]. Proteins/amino acid intake enhance the GLP-1 secretion with calcium-calmodulin dependant kinase II activation [25].

- 3. Glucagon-like Peptide -1 receptor (GLP1R):** GLP1R is one among the members of heptahelical receptor (GPCR) i.e., synthesized by a GLP1R gene located on human chromosome 6 [26]. GLP1R is composed of domains (2), one extracellular (out) domain and one transmembrane domain. Extracellular domain binds to the C- terminal helix of GLP-1 and the transmembrane domain binds to the N- terminal region of GLP-1 [27]. GLP1R is observed majorly in pancreatic beta cells and the brain; minorly in the stomach stretch-responsive vagal neurons and the intestine and also in cardiovascular systems [28,29]. Activation of pancreatic beta cells GLP1R causes insulin sensitivity and leads to synthesis and release of insulin and thereby reduces glucagon production [30]. Activation of GLP1R in the brain controls the appetite and improves memory and learning [31,32]. Changes in breathing and heart rate patterns were observed through the GLP1R activation located in the vagal neurons by communicating with other organs (Figure 2).

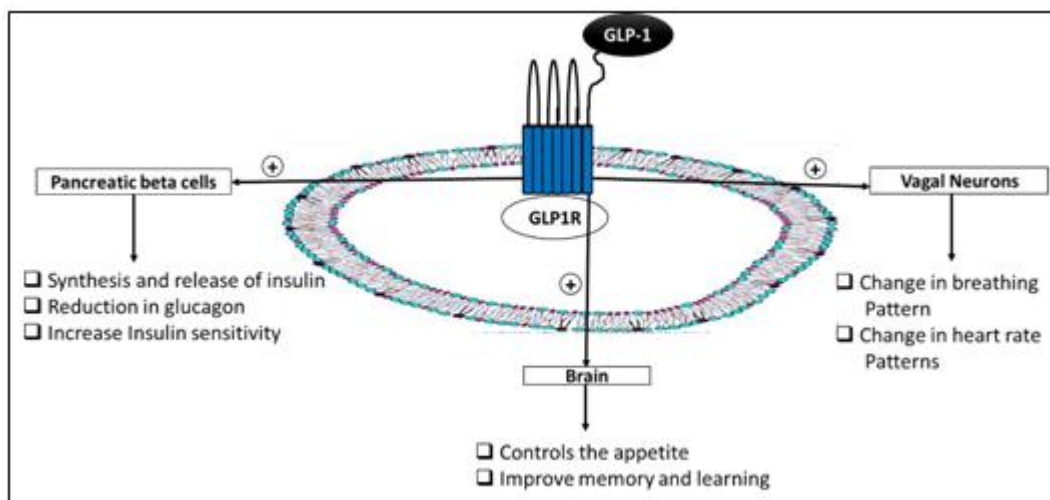


Figure 2: Various physiological functions of GLP1R upon activation by GLP-1

- 4. Purpose of GLP-1 in adult arrival diabetes:** Diabetes mellitus occurs when there is a decrease in the GLP-1 liberation succeeding oral ingestion of nutrients. Apart from this, decreased sensitivity to GLP1R in the β cells of islets of Langerhans might also contribute to an occurrence of type II diabetes mellitus (adult arrival diabetes). Additionally, in the glomerular region endothelial cells, hyperactivity of dipeptidyl peptidase-4 (DPP-IV) contributes to an increased degradation of GLP-1 and also results in stunted GLP-1 level in the plasma.

GLP-1 also encourages beta cell proliferation, apoptosis inhibition and enhances the pancreatic epithelial cells differentiation in producing insulin. However, in adult onset diabetes this regeneration. Hence, contributes as a factor to its pathogenesis [33].

- 5. Pharmacology of GLP-1 in control of Type II diabetes mellitus:** GLP-1 lowers blood glucose by stimulating the production of insulin by enhancing the action of glucose [34]. GLP-1 released in the intestine enhances the insulin secretion by means of a vagal-vagal reflex which leads in islets beta cells stimulation cholinergically. Upon binding a GLP1 agonists to the GLP1R in islets of Langerhans, adenylate cyclase signalling pathway was activated. Stimulation of this signalling pathway further promotes the conversion of ATP to cAMP. Further enhanced cAMP levels results in phosphorylation of Epac2 (cAMP-guanine nucleotide exchange factor 2 regulated) and protein kinase A (PKA) [35]. Phosphorylated PKA inhibits an ATP-dependent potassium channel (KATP) that results in the prolongation of the action potential duration, further triggers the voltage dependant L-type calcium channel and causes the calcium (Ca^{2+}) influx and action potential generation [36]. PKA also stimulates the Ryanodine receptors (RyR) and activate IP3 mediated calcium release.

Phosphorylated Epac2 activates Rap 1 and phospholipase C (PLC) and then activates IP3 and DAG pathway that results in enhanced calcium promoting calcium release (CICR) from Ryanodine receptors (RyR) and Inositol triphosphate (IP3) receptors respectively [37]. Cumulatively, phosphorylated activation of PKA and Epac2 pathways leads to increased cytoplasmic calcium levels and causes the mitochondrial synthesis of

ATP and further cause's exocytosis of granules from islets of Langerhans in pancreatic beta cells to produce insulin (Figure 3).

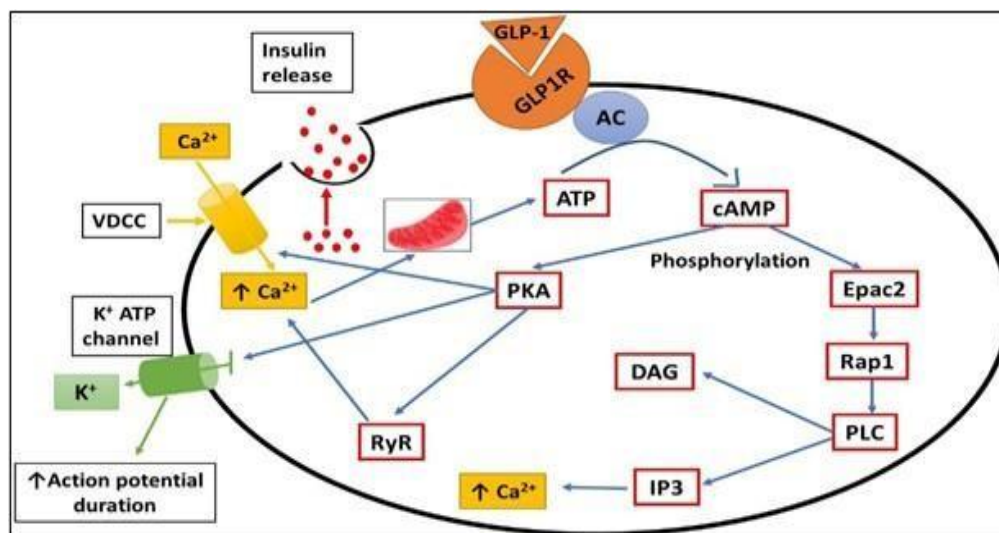


Figure 3: Mechanism of GLP-1 in insulin release for control of TDM

6. **GLP1R agonists as a target for type II diabetes therapy:** To date, several GLP1R agonists are approved therapeutically as second line therapy for the treatment of type 2 diabetes. These include liraglutide, exenatide, dulaglutide, lixisenatide, albiglutide, semaglutide and some other drugs [38].
7. **GLP-1 pharmacology in control of obesity:** Pharmacological actions of GLP1R agonists were produced by the activation of brown adipose tissue i.e., mediated by the central nervous system adrenergic & AMP-activated protein kinase (AMPK) pathways activation [39]. Preclinical studies has revealed that GLP1R agonists result in an increased energy expenditure that contributes to weight loss [40]. In addition, inhibition of food intake is one of the major approaches to the control of obesity. Post peripheral administration of GLP1R agonists, vagal afferent neurons and other brain regions contributes to the food intake suppression [41]. Some studies provide evidence that central nervous system GLP-1 signalling is essential to mediate the therapeutic effect on energy metabolism [42].

In CNS, the brain stem has important part in communication to higher brain centres for sending peripheral signals [43]. Activation of GLP1R by GLP-1 agonists in the lateral parabrachial nucleus inhibits the feed intake mechanism [44]. Additionally, the hypothalamus acts as an important centre for introspecting the therapeutic result of GLP1R agonists on energy metabolism. In specific, the arcuate nucleus (ARC) located in the hypothalamus acts as one of the main sites of action for GLP1R agonists [45]. The specific neuronal populations that mediate the anorectic action of GLP1R signalling in ARC are being investigated currently. The majority of the research that have been published point to the ARC's anorexigenic pro-opiomelanocortin (POMC) neurons being activated while orexigenic neuropeptide Y/Agouti-related peptide (NPY/AgRP) neurons are being simultaneously inhibited it leads to food intake inhibition which is one of the

critical mechanisms for pharmacological action of GLP1R agonists [46]. GLP1R is also expressed in other hypothalamic nuclei such as the dorsomedial nucleus of the hypothalamus (DMH) and the medial preoptic area (MPOA) where its function is closely related to thermogenesis control [47]. GLP-1 slows gastric emptying thereby reducing the feed intake capacity [48].

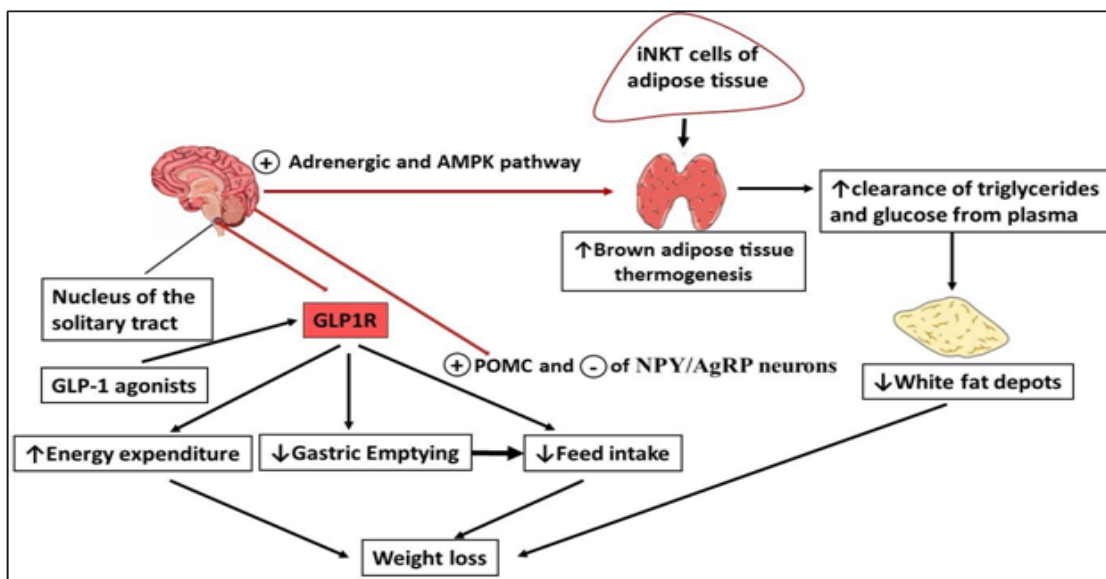


Figure 4: Possible mechanism of GLP-1 in control of obesity; iNKT- invariant natural killer T cells; POMC-proopiomelanocortin; NPY/AgRP- Neuropeptide Y/Agouti-related peptide

In addition to the occurrence of feed intake changes, body weight change is also regulated by the activation of central GLP1R and further causes enhanced brown adipose tissue (BAT) thermogenesis [49]. In one study it is reported that the GLP1R located in hypothalamus of ventro medial nucleus (VMH) is activated by a GLP-1 analogue and triggers the AMP-activated protein kinase (AMPK) inhibition that promotes BAT thermogenesis mediated by the sympathetic system and further leads to the browning of white adipose tissue (WAT) and reduction in the feed intake [50]. The AMPK-VMH-SNS-BAT/WAT hypothalamic pathway which is activated by several peripheral signals is a well reported pathway [51]. Some preclinical studies in mice reported that the central infusion of GLP1R agonists increases the sympathetic tone of the fibres innervating the thermogenesis of brown adipose tissue. This high brownfat activity promotes triglycerides and glucose clearance from the plasma and reduces white fat depots that lead to body weight loss [52]. Other preclinical studies suggest that GLP1R centrally mediates the metabolism of adipocytes and further inhibition of white fat depots triglyceride storage. This further signifies that GLP1R centrally elevates a catabolic state in adipose tissue by activation of the sympathetic system [53]. Some studies suggest that the immune system also plays a key role in energy homeostasis mediated by GLP1R. Among these, adipocyte resident invariant natural killer T (iNKT) cells are located in higher amounts in the fat deposits are essential for weight loss [54]. Recent data reported that iNKT cells favour BAT thermogenesis and further causes GLP-1 receptor associated weight loss effects [55]. The possible mechanism is clearly represented in figure 4.

Some of the GLP1R agonists such as liraglutide and more recently semaglutide are considered as the most efficient weight loss drugs in clinical trials [56,57].

- 8. Pharmacological actions of GLP-1 in the cardiovascular system:** GLP1R is also located in the cardiovascular system and has been observed primarily on cardiac cells and endothelium layers. GLP1R has also been located upon the autonomous nervous system peripherally that promotes active and passive action on the circulatory system [58]. The mechanistic approach regarding cardiovascular effects of GLP1R focuses mainly on lipids, microvascular function, blood pressure, pulse rate, and inflammation which are discussed briefly below.

Some studies reported that GLP1R agonists increase the heart rate slightly to minimum extent which is thought to be decreased immediately by not changing the normal heart rate [59]. Till now, a conclusive mechanism behind the increase in heart rate remains unclear. However, a recent study in mice reported that stimulation of the arterial GLP1R induces a chronotropic effect produced by the sinoatrial node [60]. Although this animal data may provide clarity on the mechanism, confirmation in human studies is needed. GLP1R agonists decrease blood pressure in subjects suffering with type II diabetes and obesity. Clear mechanisms are yet to be resolved, but some of the clinical studies demonstrated the blood pressure lowering effect may be due to GLP1R agonist induced weight loss [61]; involvement of the sympathetic nervous system as it influences both cardiac output and vascular resistance. Pre-clinical studies suggested that activation of GLP1R via serum atrial natriuretic peptide (ANP) reduces blood pressure [62]. The clinical mechanism is yet to be understood.

One of the clinical studies reported that GLP1R agonist exenatide reduces dyslipidemia by reducing the increased apo C-III, triglycerides, apoB-48 production, remnant lipoprotein cholesterol and triglycerides in type II diabetic and obese patients [63]. Further contributes to atherosclerosis and cardiovascular disease reduction [64]. Clear mechanisms are yet to be investigated. Some of the clinical studies reported that GLP1R agonists such as liraglutide and exenatide reduce some of the systemic inflammatory cytokines like tumour necrosis factor- α (TNF- α), interleukins (IL) 1 β , IL-6, c-reactive protein (CRP) and also leucocyte adhesion molecules like intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (V-CAM-1) thereby reduces inflammation for prevention of the atherosclerosis formation and cardiovascular risk [65,66,67].

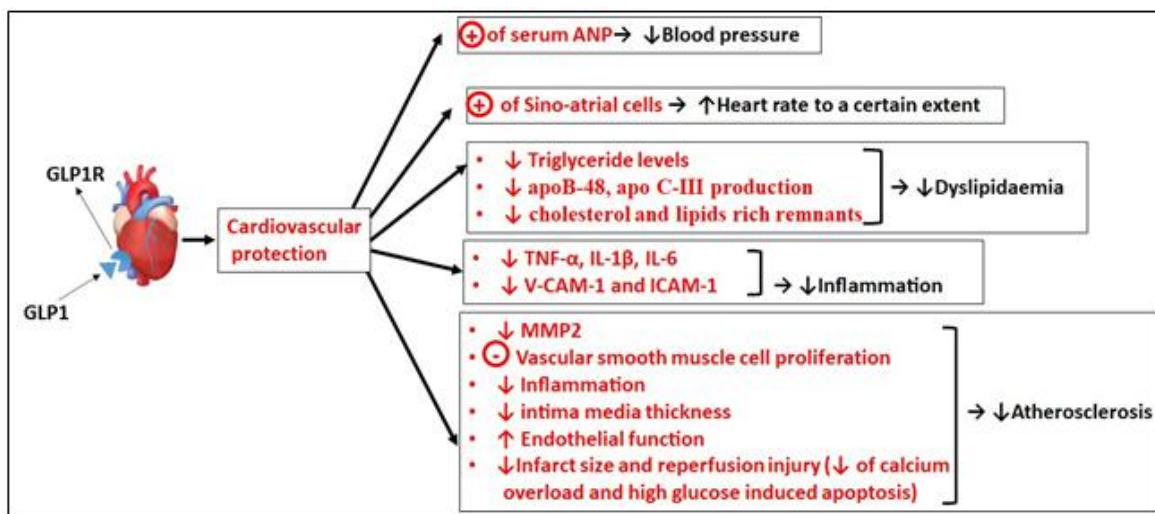


Figure 5: Possible mechanism of GLP-1 in control of cardiovascular diseases; ANP: atrial natriuretic peptide; MMP2: MatrixMetallo proteinase-2; TNF- α : Tumour necrosis factor- α ; IL-1 β : Interleukin-1 β ; IL-6: Interleukin-6; V-CAM-1: Vascular cell adhesion molecule-1; ICAM-1: intercellular adhesion molecule-1

Some of the preclinical and clinical studies reported that GLP1R agonists inhibit the development of atherosclerosis by reducing matrix metalloproteinase-2 (MMP-2) levels [68], inhibiting vascular smooth muscle cell proliferation [69], regulating the inflammation [70], reducing intima-media thickening [71] and increase endothelial function via nitric oxide mediated vasodilation and decreased oxidative stress [72]; however, the specific mechanisms in clinical studies are yet to be observed. Some studies reported that GLP1R agonists prevent atherosclerotic effects by blocking the p53 protein inhibition on kruppel-like factor 2 (KLF2) located on human aortic endothelial cells [73,74]. Some of the pre-clinical and clinical studies reported that, GLP1R agonists prevents atherosclerosis via lessen myocardial ischemia and reperfusion damage mediated through intracellular calcium overload reduction and high glucose induced apoptosis [75,76]. In another study, it is reported that liraglutide can also initiates vascular smooth muscle cell cycle arrest via the AMPK pathway thereby delaying formation of atherosclerosis [77]. To date, as there are no GLP1R agonists approved for the treatment of atherosclerosis and cardiovascular diseases, these preclinical and clinical findings can provide novel approaches for the pharmacological therapy of cardiovascular diseases in association with obesity and diabetes mellitus of second type. The possible mechanism is clearly represented in figure 5.

IV. CONCLUSION AND FUTURE PERSPECTIVES

Currently, GLP1R agonists have been considered as novel antidiabetic drugs that are used in type II diabetes mellitus treatment as a second line of therapy [78]. In addition to this, GLP-1 agonists have advantages in obesity by body weight reduction, brown fat thermogenesis and elevated energy expenditure. In the United States, currently, one FDA-approved drug of GLP-1 agonist- liraglutide (saxenda) is used for the treatment of obesity. Some of the clinical data support the roles of central GLP1R and peripheral GLP1R agonistic actions in feed intake and weight reduction. Hence, it became an important target for the pharmacological therapy of obesity and overweight.

Some of the clinical trials conducted with GLP1R agonists and cardiovascular outcome trials suggested that liraglutide and semaglutide displayed beneficial effects on cardiovascular benefits in the presence of both placebo and standard treatment. Although the mechanisms through which GLP1R agonists (liraglutide and semaglutide) produce cardiovascular protection is still clearly unknown, they may be used in routine clinical practice. Still, more clinical studies should be performed to observe clear mechanisms through which GLP1R agonists act as cardioprotective agents. Since cardiovascular diseases and type II diabetes mellitus is the major cause of death in obese and overweight people, choosing GLP1R agonists as a therapeutic drug might be the best option for controlling obesity and associated metabolic syndrome (Type 2 diabetes mellitus and cardiac circulatory diseases) in the future.

REFERENCES

- [1] Zimmet P, Alberti KG, Kaufman F, Tajima N, Silink M, Arslanian S, Wong G, Bennett P, Shaw J, Caprio S, IDF Consensus Group. The metabolic syndrome in children and adolescents—an IDF consensus report. *Pediatric diabetes*. 2007;8(5):299-306.
- [2] Trujillo JM, Nuffer W, Ellis SL. GLP-1 receptor agonists: a review of head-to-head clinical studies. *Therapeutic advances in endocrinology and metabolism*. 2015;6(1):19-28.
- [3] Thorens B. Expression cloning of the pancreatic beta cell receptor for the gluco-incretin hormone glucagon-like peptide 1. *Proceedings of the National Academy of Sciences*. 1992;89(18):8641-5.
- [4] Ravassa S, Zudaire A, Díez J. GLP-1 and cardioprotection: from bench to bedside. *Cardiovascular research*. 2012;94(2):316-23.
- [5] Ussher JR, Drucker DJ. Cardiovascular biology of the incretin system. *Endocrine reviews*. 2012;33(2):187-215.
- [6] Wang YC, McPherson K, Marsh T, Gortmaker SL, Martin Brown M. Health and economic burden of the projected obesity trends in the USA and the UK. *The Lancet*. 2011;378(9793):815-25.
- [7] World Health Organization. Obesity: preventing and managing the global epidemic. Report of a WHO consultation. 2000;894:i-xii, 1-253.
- [8] Keaver L, Webber L, Dee A, Shiely F, Marsh T, Balanda K et al., Application of the UK foresight obesity model in Ireland: the health and economic consequences of projected obesity trends in Ireland. *PloS one*. 2013;8(11):e79827.
- [9] Alberti KG, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA et al., Harmonizing the metabolic syndrome: a joint interim statement of the international diabetes federation task force on epidemiology and prevention; national heart, lung, and blood institute; American heart association; world heart federation; international atherosclerosis society; and international association for the study of obesity. *Circulation*. 2009;120(16):1640-5.
- [10] do Vale Moreira NC, Hussain A, Bhowmik B, Mdala I, Siddiquee T, Fernandes VO et al., Prevalence of metabolic syndrome by different definitions, and its association with type 2 diabetes, pre-diabetes, and cardiovascular disease risk in Brazil. *Diabetes & Metabolic Syndrome: Clinical Research & Reviews*. 2020;14(5):1217-24.
- [11] Gierach M, Gierach J, Ewertowska M, Arndt A, Junik R. Correlation between body mass index and waist circumference in patients with metabolic syndrome. *International Scholarly Research Notices Endocrinology* 2014: 514589.
- [12] Ma X, Hui H, Liu Z, He G, Hu J, Meng J et al., Poly- GLP- 1, a novel long- lasting glucagon- like peptide- 1 polymer, ameliorates hyperglycaemia by improving insulin sensitivity and increasing pancreatic beta- cell proliferation. *Diabetes, Obesity and Metabolism*. 2009;11(10):953-65.
- [13] Ørskov C, Rabenhøj L, Wettergren A, Kofod H, Holst JJ. Tissue and plasma concentrations of

- amidated and glycine-extended glucagon-like peptide I in humans. *Diabetes*. 1994;43(4):535-9.
- [14] Mojsos S, Kopczyński MG, Habener JF. Both amidated and nonamidated forms of glucagon-like peptide I are synthesized in the rat intestine and the pancreas. *Journal of Biological Chemistry*. 1990;265(14):8001-8.
- [15] Gunawardene AR, Corfe BM, Staton CA. Classification and functions of enteroendocrine cells of the lower gastrointestinal tract. *International journal of experimental pathology*. 2011;92(4):219-31.
- [16] Eissele R, Göke R, Willemer S, Harthus HP, Vermeer H, Arnold RE et al., Glucagon-like peptide-1 cells in the gastrointestinal tract and pancreas of rat, pig and man. *European journal of clinical investigation*. 1992;22(4):283-91.
- [17] Merchenthaler I, Lane M, Shughrue P. Distribution of pre- pro- glucagon and glucagon-like peptide-1 receptor messenger RNAs in the rat central nervous system. *Journal of Comparative Neurology*. 1999;403(2):261-80.
- [18] Fehmman HC, Göke R, Göke B. Cell and molecular biology of the incretin hormones glucagon-like peptide-I and glucose-dependent insulin releasing polypeptide. *Endocrine reviews*. 1995;16(3):390-410.
- [19] Ørskov C, Wettergren A, Holst JJ. Secretion of the incretin hormones glucagon-like peptide-1 and gastric inhibitory polypeptide correlates with insulin secretion in normal man throughout the day. *Scandinavian journal of gastroenterology*. 1996 Jan 1;31(7):665-70.
- [20] Anini Y, Brubaker PL. Muscarinic receptors control glucagon-like peptide 1 secretion by human endocrine L cells. *Endocrinology*. 2003;144(7):3244-50.
- [21] Gribble FM, Williams L, Simpson AK, Reimann F. A novel glucose-sensing mechanism contributing to glucagon-like peptide-1 secretion from the GLUTag cell line. *Diabetes*. 2003;52(5):1147-54.
- [22] Cani PD, Holst JJ, Drucker DJ, Delzenne NM, Thorens B, Burcelin R, et al., GLUT2 and the incretin receptors are involved in glucose-induced incretin secretion. *Molecular and cellular endocrinology*. 2007;276(1-2):18-23.
- [23] Nøhr MK, Pedersen MH, Gille A, Egerod KL, Engelstoft MS, Husted AS et al., GPR41/FFAR3 and GPR43/FFAR2 as cosensors for short-chain fatty acids in enteroendocrine cells vs FFAR3 in enteric neurons and FFAR2 in enteric leukocytes. *Endocrinology*. 2013;154(10):3552-64.
- [24] Tvrzicka E, Kremmyda LS, Stankova B, Zak A. Fatty acids as biocompounds: their role in human metabolism, health and disease-a review. part 1: classification, dietary sources and biological functions. *Biomedical Papers of the Medical Faculty of Palacky University in Olomouc*. 2011;155: 117-30.
- [25] Kato M, Nakanishi T, Tani T, Tsuda T. Low-molecular fraction of wheat protein hydrolysate stimulates glucagon-like peptide-1 secretion in an enteroendocrine L cell line and improves glucose tolerance in rats. *Nutrition Research*. 2017;37:37-45.
- [26] Brubaker PL, Drucker DJ. Structure-function of the glucagon receptor family of G protein-coupled receptors: the glucagon, GIP, GLP-1, and GLP-2 receptors. *Receptors and Channels*. 2002;8(3-4):179-88.
- [27] Underwood CR, Garibay P, Knudsen LB, Hastrup S, Peters GH, Rudolph R, Reedtz-Runge S. Crystal structure of glucagon-like peptide-1 in complex with the extracellular domain of the glucagon-like peptide-1 receptor. *Journal of Biological Chemistry*. 2010;285(1):723-30.
- [28] Cork SC, Richards JE, Holt MK, Gribble FM, Reimann F, Trapp S. Distribution and characterisation of Glucagon-like peptide-1 receptor expressing cells in the mouse brain. *Molecular metabolism*. 2015;4(10):718-31.
- [29] Williams EK, Chang RB, Strohlic DE, Umans BD, Lowell BB, Liberles SD. Sensory neurons that detect stretch and nutrients in the digestive system. *Cell*. 2016;166(1):209-21.
- [30] Holst JJ. The physiology of glucagon-like peptide 1. *Physiological reviews*. 2007;87(4):1409-39.
- [31] Kinzig KP, D'Alessio DA, Seeley RJ. The diverse roles of specific GLP-1 receptors in the

- control of food intake and the response to visceral illness. *Journal of Neuroscience*. 2002;22(23):10470-6.
- [32] During MJ, Cao L, Zuzga DS, Francis JS, Fitzsimons HL, Jiao X et al., Glucagon-like peptide-1 receptor is involved in learning and neuroprotection. *Nature medicine*. 2003;9(9):1173-9.
- [33] Das A, Geetha KM, Hazarika I. Contemporary updates on the physiology of glucagon like peptide-1 and its agonist to treat type 2 Diabetes mellitus. *International Journal of Peptide Research and Therapeutics*. 2020;26(3):1211-21.
- [34] Davis EM, Sandoval DA. Glucagon-like peptide-1: actions and influence on pancreatic hormone function. *Comprehensive Physiology*. 2020;10(2):577.
- [35] Leech CA, Chepurny OG, Holz GG. Epac2-dependent rap1 activation and the control of islet insulin secretion by glucagon-like peptide-1. *Vitamins & hormones*. 2010; 84:279-302.
- [36] Doyle ME, Egan JM. Mechanisms of action of glucagon-like peptide 1 in the pancreas. *Pharmacology & therapeutics*. 2007;113(3):546-93.
- [37] Gromada J, Holst JJ, Rorsman P. Cellular regulation of islet hormone secretion by the incretin hormone glucagon-like peptide 1. *Pflügers Archiv*. 1998 Mar;435(5):583-94.
- [38] Collins L, Costello RA. Glucagon-like peptide-1 receptor agonists. In *StatPearls* [internet] 2021. StatPearls Publishing.
- [39] Beiroa D, Imbernon M, Gallego R, Senra A, Herranz D, Villarroya F et al., GLP-1 agonism stimulates brown adipose tissue thermogenesis and browning through hypothalamic AMPK. *Diabetes*. 2014;63(10):3346-58.
- [40] Lockie SH, Heppner KM, Chaudhary N, Chabenne JR, Morgan DA, Veyrat-Durebex C et al., Direct control of brown adipose tissue thermogenesis by central nervous system glucagon-like peptide-1 receptor signaling. *Diabetes*. 2012;61(11):2753-62.
- [41] Kanoski SE, Fortin SM, Arnold M, Grill HJ, Hayes MR. Peripheral and central GLP-1 receptor populations mediate the anorectic effects of peripherally administered GLP-1 receptor agonists, liraglutide and exendin-4. *Endocrinology*. 2011;152(8):3103-12.
- [42] Sisley S, Gutierrez-Aguilar R, Scott M, D'Alessio DA, Sandoval DA, Seeley RJ. Neuronal GLP1R mediates liraglutide's anorectic but not glucose-lowering effect. *The Journal of clinical investigation*. 2014;124(6):2456-63.
- [43] Abbott CR, Monteiro M, Small CJ, Sajedi A, Smith KL, Parkinson JR et al., The inhibitory effects of peripheral administration of peptide YY3–36 and glucagon-like peptide-1 on food intake are attenuated by ablation of the vagal–brainstem–hypothalamic pathway. *Brain research*. 2005 May 17;1044(1):127-31.
- [44] Alhadeff AL, Baird JP, Swick JC, Hayes MR, Grill HJ. Glucagon-like peptide-1 receptor signaling in the lateral parabrachial nucleus contributes to the control of food intake and motivation to feed. *Neuropsychopharmacology*. 2014;39(9):2233-43.
- [45] Tang-Christensen M, Vrang N, Larsen PJ. Glucagon-like peptide 1 (7-36) amide's central inhibition of feeding and peripheral inhibition of drinking are abolished by neonatal monosodium glutamate treatment. *Diabetes*. 1998;47(4):530-7.
- [46] Seo S, Ju S, Chung H, Lee D, Park S. Acute effects of glucagon-like peptide-1 on hypothalamic neuropeptide and AMP activated kinase expression in fasted rats. *Endocrine journal*. 2008;0805020133.
- [47] Merchenthaler I, Lane M, Shughrue P. Distribution of pre- pro- glucagon and glucagon- like peptide- 1 receptor messenger RNAs in the rat central nervous system. *Journal of Comparative Neurology*. 1999;403(2):261-80.
- [48] Willms BE, Werner JE, Holst JJ, Orskov C, Creutzfeldt WE, Nauck MA. Gastric emptying, glucose responses, and insulin secretion after a liquid test meal: effects of exogenous glucagon-like peptide-1 (GLP-1)-(7-36) amide in type 2 (noninsulin-dependent) diabetic patients. *The Journal of Clinical Endocrinology & Metabolism*. 1996;81(1):327-32.
- [49] Lockie SH, Stefanidis A, Oldfield BJ, Perez-Tilve D. Brown adipose tissue thermogenesis in the resistance to and reversal of obesity: a potential new mechanism contributing to the metabolic benefits of proglucagon-derived peptides. *Adipocyte*. 2013;2(4):196-200.

- [50] Beiroa D, Imbernon M, Gallego R, Senra A, Herranz D, Villarroya F et al., GLP-1 agonism stimulates brown adipose tissue thermogenesis and browning through hypothalamic AMPK. *Diabetes*. 2014;63(10):3346-58.
- [51] Lopez M, Nogueiras R, Tena-Sempere M, Dieguez C. Hypothalamic AMPK: a canonical regulator of whole-body energy balance. *Nature Reviews Endocrinology*. 2016 Jul;12(7):421-32.
- [52] Kooijman S, Wang Y, Parlevliet ET, Boon MR, Edelschaap D, Snaterse G et al., Central GLP-1 receptor signalling accelerates plasma clearance of triacylglycerol and glucose by activating brown adipose tissue in mice. *Diabetologia*. 2015;58(11):2637-46.
- [53] Nogueiras R, Diaz-Arteaga A, Lockie SH, Velásquez DA, Tschop J, López M et al., The endocannabinoid system: role in glucose and energy metabolism. *Pharmacological research*. 2009;60(2):93-8.
- [54] Lynch L, O'Shea D, Winter DC, Geoghegan J, Doherty DG, O'Farrelly C. Invariant NKT cells and CD1d+ cells amass in human omentum and are depleted in patients with cancer and obesity. *European journal of immunology*. 2009;39(7):1893-901.
- [55] Lynch L, Hogan AE, Duquette D, Lester C, Banks A, LeClair K et al., iNKT cells induce FGF21 for thermogenesis and are required for maximal weight loss in GLP1 therapy. *Cell metabolism*. 2016;24(3):510-9.
- [56] O'Neil PM, Birkenfeld AL, McGowan B, Mosenzon O, Pedersen SD, Wharton S et al., Efficacy and safety of semaglutide compared with liraglutide and placebo for weight loss in patients with obesity: a randomised, double-blind, placebo and active controlled, dose-ranging, phase 2 trial. *The Lancet*. 2018;392(10148):637-49.
- [57] Wilding JP, Batterham RL, Calanna S, Davies M, Van Gaal LF, Lingvay I et al., Once-weekly semaglutide in adults with overweight or obesity. *New England Journal of Medicine*. 2021 Feb 10.
- [58] Nauck MA, Meier JJ, Cavender MA, Abd El Aziz M, Drucker DJ. Cardiovascular actions and clinical outcomes with glucagon-like peptide-1 receptor agonists and dipeptidyl peptidase-4 inhibitors. *Circulation*. 2017;136(9):849-70.
- [59] Heuvelman VD, Van Raalte DH, Smits MM. Cardiovascular effects of glucagon-like peptide 1 receptor agonists: from mechanistic studies in humans to clinical outcomes. *Cardiovascular research*. 2020;116(5):916-30.
- [60] Baggio LL, Ussher JR, McLean BA, Cao X, Kabir MG, Mulvihill EE et al., The autonomic nervous system and cardiac GLP-1 receptors control heart rate in mice. *Molecular metabolism*. 2017;6(11):1339-49.
- [61] Meier JJ. GLP-1 receptor agonists for individualized treatment of type 2 diabetes mellitus. *Nature Reviews Endocrinology*. 2012;8(12):728-42.
- [62] Kim M, Platt MJ, Shibasaki T, Quaggin SE, Backx PH, Seino S et al., GLP-1 receptor activation and Epac2 link atrial natriuretic peptide secretion to control of blood pressure. *Nature medicine*. 2013;19(5):567-75.
- [63] Schwartz EA, Koska J, Mullin MP, Syoufi I, Schwenke DC, Reaven PD. Exenatide suppresses postprandial elevations in lipids and lipoproteins in individuals with impaired glucose tolerance and recent onset type 2 diabetes mellitus. *Atherosclerosis*. 2010;212(1):217-22.
- [64] Hamal S, Cherukuri L, Shaikh K, Kinninger A, Doshi J, Birudaraju D et al., Effect of semaglutide on coronary atherosclerosis progression in patients with type II diabetes: rationale and design of the semaglutide treatment on coronary progression trial. *Coronary artery disease*. 2020;31(3):306-14.
- [65] Nauck MA, Meier JJ, Cavender MA, Abd El Aziz M, Drucker DJ. Cardiovascular actions and clinical outcomes with glucagon-like peptide-1 receptor agonists and dipeptidyl peptidase-4 inhibitors. *Circulation*. 2017;136(9):849-70.
- [66] Daousi C, Pinkney JH, Cleator J, Wilding JP, Ranganath LR. Acute peripheral administration of synthetic human GLP-1 (7–36 amide) decreases circulating IL-6 in obese patients with type 2 diabetes mellitus: a potential role for GLP-1 in modulation of the diabetic pro-inflammatory

- state?. *Regulatory peptides*. 2013;183:54-61.
- [67] Lee YS, Jun HS. Anti-inflammatory effects of GLP-1-based therapies beyond glucose control. *Mediators of inflammation*. 2016.
- [68] Wang M, Kim SH, Monticone RE, Lakatta EG. Matrix metalloproteinases promote arterial remodeling in aging, hypertension, and atherosclerosis. *Hypertension*. 2015;65(4):698-703.
- [69] Nagayama K, Kyotani Y, Zhao J, Ito S, Ozawa K, Bolstad FA et al., Exendin-4 prevents vascular smooth muscle cell proliferation and migration by angiotensin II via the inhibition of ERK1/2 and JNK signaling pathways. *PLoS one*. 2015;10(9):e0137960.
- [70] Sharma A, Verma S. Mechanisms by which glucagon-like-peptide-1 receptor agonists and sodium-glucose cotransporter-2 inhibitors reduce cardiovascular risk in adults with type 2 diabetes mellitus. *Canadian Journal of Diabetes*. 2020;44(1):93-102.
- [71] Goto H, Nomiyama T, Mita T, Yasunari E, Azuma K, Komiya K et al., Exendin-4, a glucagon-like peptide-1 receptor agonist, reduces intimal thickening after vascular injury. *Biochemical and biophysical research communications*. 2011;405(1):79-84.
- [72] Gaspari T, Liu H, Welungoda I, Hu Y, Widdop RE, Knudsen LB et al., A GLP-1 receptor agonist liraglutide inhibits endothelial cell dysfunction and vascular adhesion molecule expression in an ApoE^{-/-} mouse model. *Diabetes and Vascular Disease Research*. 2011;8(2):117-24.
- [73] Chang W, Zhu F, Zheng H, Zhou Z, Miao P, Zhao L et al., Glucagon-like peptide-1 receptor agonist dulaglutide prevents ox-LDL-induced adhesion of monocytes to human endothelial cells: an implication in the treatment of atherosclerosis. *Molecular immunology*. 2019;116:73-9.
- [74] Yue W, Li Y, Ou D, Yang Q. The GLP-1 receptor agonist liraglutide protects against oxidized LDL-induced endothelial inflammation and dysfunction via KLF2. *IUBMB life*. 2019;71(9):1347-54.
- [75] Nikolaidis LA, Mankad S, Sokos GG, Miske G, Shah A, Elahi D et al., Effects of glucagon-like peptide-1 in patients with acute myocardial infarction and left ventricular dysfunction after successful reperfusion. *Circulation*. 2004;109(8):962-5.
- [76] Younce CW, Burmeister MA, Ayala JE. Exendin-4 attenuates high glucose-induced cardiomyocyte apoptosis via inhibition of endoplasmic reticulum stress and activation of SERCA2a. *American Journal of Physiology-Cell Physiology*. 2013;304(6):C508-18.
- [77] Jojima T, Uchida K, Akimoto K, Tomotsune T, Yanagi K, Iijima T et al., Liraglutide, a GLP-1 receptor agonist, inhibits vascular smooth muscle cell proliferation by enhancing AMP-activated protein kinase and cell cycle regulation, and delays atherosclerosis in ApoE deficient mice. *Atherosclerosis*. 2017;261:44-51.
- [78] R Drab S. Glucagon-like peptide-1 receptor agonists for type 2 diabetes: a clinical update of safety and efficacy. *Current diabetes reviews*. 2016;12(4):403-13.