
# ADDIS ABABA UNIVERSITY SCHOOL OF GRADUATE STUDIES

A senior paper on **‘‘The role of incretin hormone in type 2 diabetes’’** submitted to the School of Graduate Studies of Addis Ababa University in Partial Fulfillment of the Requirements for The Degree of Master of Science (MSc.) in Medical Biochemistry

**BY**

**Dereje Getachew (BSc.)**

**Department of biochemistry**

**June, 2011**

## ACKNOWLEDGMENTS

I would like to thank my advisor, Dr. Daniel Seifu, for his repeated and tireless constructive comments starting from the first draft to the final write up of my senior paper.

I would also extend my heart-felt thanks to my sister, Fantay Eshetu for her constructive advice and financial support throughout my academic life.

My acknowledgment goes to my best friend Kassa Bulcha for his close supervision and advices while I was undertaking this senior paper.

I acknowledge the department of Biochemistry, Addis Ababa University, for financial and logistic support for the study.

Last, but not least I appreciate the encouragement and support of my family and all my friends to come to this end. Above all, I would like to praise my Lord Jesus Christ, who carries my burdens day after day and saves me.

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# LIST OF ACRONYMS

T2DM –Type 2 Diabetes

GIP - Glucose Dependent Insulinotropic Peptide

GLP-1- Glucagon Like Peptide-1

I.V. -Intravenous

DPPIV- Dipeptidyl Peptidase 4

PC- Proconvertase

GRPP -Glicentin-Related Pancreatic peptide

ATP- Adenosine Tri Phosphate

$K^+$ -Potasium ion

GRP- Gastrin-Releasing Peptide

GPCR- G Protein Coupled Receptor

GPR 120,119,40- G Protein Receptor 120,119,40

MAPK- Mitogen-Activated Protein Kinase

GABA- Gama-Amino Butyric Acid

PKA,B,C- Protein Kinase A, B, C

PI-3K- Phosphatidyl Inositol-3 Kinase

PDX-1 - Pancreatic Duodenal homeobox-1

PLA2 – Phospholipase A2

GIPR – Glucose Dependent Insulinotropic Peptide Receptor

GLP-1R – Glucagon Like Peptide-1 receptor

GLUT2 –Glucose transporter 2

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## Abstract

**Background**: Type 2 diabetes mellitus is a metabolic disease associated with low quality of life and early death. It is now well established that beta-cell dysfunction and insulin resistance are two central defects in the pathophysiology of T2DM. Recently, improved understanding of the incretin effect on the pathophysiology of type 2 diabetes has led to development of new hypoglycemic agents. The incretin effect refers to the amplification of insulin secretion that occurs when glucose is ingested orally as opposed to infused intravenously in amounts that result in identical glucose excursions.

**Objective**: To review the role of incretin hormone in type 2 diabetes.

**Methode**: The review was conducted as a systemic review. Articles were searched from MEDLINE, HINARI and PUBMED in English language with the key words incretin or gut hormone, type 2 diabetes, and the role of incretin in type 2 diabetes.

**Result:** A total of 65 studies were retrieved. Of these, 12 were considered to be relevant up on initial screening. Abstract of these 12 articles were reviewed and while 5 studies excluded. 7 studies were agreed upon to meet the inclusion criteria.

**Conclusion**: The incretin effect was significantly reduced in patients with type 2 diabetes.GLP-1 concentrations and response is also reduced. But the GIP concentration is increased, reduced or normal. Incretin secretion is increased after oral glucose compared with intravenous infusion.

**Key words**; **Type 2 diabetes, Glucagon Like Peptide-1(GLP-1), Glucose Dependent Insulinotropic Polypeptide (GIP), Incretin effect.**

# 1. INTRODUCTION

Type 2 diabetes mellitus (T2DM) is the clinical manifestation of long-term metabolic abnormalities involving multiple organs and hormonal pathways that impair the body's ability to maintain glucose homeostasis (Schnabel et al. 2006). The incidence and prevalence of type 2 diabetes are rising steadily, fuelled in part by a concomitant increase in the worldwide rates of obesity (Drucker et al. 2010).T2DM comprises 90% of people with diabetes mellitus (DM) around the world. The World Health Organization estimates that more than 180 million people worldwide have DM, and, as the western lifestyle is making its entry into the developing countries, this number is likely to more than double by 2030 (Knop et al. 2009). The etiology of type 2 diabetes is unknown; however, genetic and environmental factors have been linked to its development. It is a chronic progressive disease associated with micro- and macrovascular complications such as nephropathy, neuropathy, retinopathy and cardiovascular morbidity. These complications often result in low quality of life and early death (Hansen et al. 2010).

It is now well established that beta-cell dysfunction and insulin resistance are two central defects in the pathophysiology of T2DM. Furthermore, it has been demonstrated that T2DM is a progressive disease, due to an almost linear decline in beta-cell function over time. Thus, it seems that T2DM evolves as the beta-cells lose the ability to respond adequately to the insulin need. Furthermore, evidence for inappropriate secretion of glucagon playing an important role in the pathogenesis of T2DM is accumulating; fasting and postprandial hyperglucagonemia in T2DM have been shown to result in increased glucagon-induced hepatic glucose production, which again contributes to fasting hyperglycemia and exaggerated postprandial glucose excursions (Knop et al. 2009).

The goal with diabetes treatment is to improve quality of life and prevent early death (Hansen et al. 2010 ). However, efficacy of available therapies, even when used appropriately, diminishes as the disease progresses because of a steady, relentless decline in pancreatic beta cell function (Amori et al. 2007). It is well established that tight glycemic control reduces the risk of microvascular disease while recent randomized controlled trials have failed to show a substantial benefit on macrovascular outcome (Hansen et al. 2010 ). Furthermore, current therapies for type 2 diabetes are often limited by adverse effects such as weight gain, edema, or hypoglycemia, and most do not target postprandial hyperglycemia effectively. Therefore, therapies targeting the

decline in pancreatic beta cell function without causing weight gain and with minimal adverse effects are desirable. Recently, improved understanding of the incretin effect on the pathophysiology of type 2 diabetes has led to development of new hypoglycemic agents (Amori et al. 2007).

The incretin effect refers to the amplification of insulin secretion that occurs when glucose is ingested orally as opposed to infused intravenously in amounts that result in identical glucose excursions. In healthy subjects, the incretin effect accounts 50% to 70% of the total amount of insulin released in response to an oral glucose load (Knop et al. 2009).

The concept of incretins is at least a century old (Table 1.1). In 1902, the physiologists Bayliss and Starling first proposed that food intake induces release of a chemical from the duodenal and jejunal mucosa that stimulates pancreatic secretion (Martin et al. 2011 ). In 1932, La Barre used the word “incretin” to refer to an extract from upper gut mucosa that produces hypoglycemia but does not induce exocrine secretion, although he did not prove incontrovertibly that incretins existed. Progress on the incretin concept was rapidly made once radioimmunoassays for insulin became available.

TABLE 1.1 *Time-line to use of incretin-based therapies in the treatment of diabetes*

| Year | The Development of Incretin-Based Therapies |
|---|---|
| 1869 | Islets of Langerhans (nests of cells that appeared different from the surrounding pancreatic tissue) in the pancreas were described. |
| 1901 | The role of islets of Langerhans (what ultimately became known as an endocrine function) in diabetes was described. |
| 1902 | The role of a substance (called secretin) secreted by gut cells that stimulates the digestive juices for the pancreas (what ultimately became known as exocrine function) was described. |
| 1905 | This type of substance, a presumed "chemical messenger," was now called a "hormone." |
| 1906 | The role of a gut-derived hormone to treat diabetes was first alluded to. |
| 1921–1922 | Extraction of insulin from pancreas and its potential to treat type 1 diabetes was shown. |
| 1932 | The term "incretin" was used for the first time to refer to a substance derived from the gut, presumably a hormone that regulates insulin secretion after eating. |
| 1960 | Radioimmunoassay was developed for measurement of plasma insulin levels. |
| 1964–1967 | Clinical proof that a gut-derived factor positively modulated insulin secretion. |
| 1971 | The first incretin, GIP, was isolated and sequenced. |
| 1985 | The second incretin, GLP-1, was described. |
| 1992–1994 | Studies show that exogenous GIP does not lower blood glucose in T2DM, but exogenous GLP-1 does so. |
| 2002 | Exendin-4, a GLP-1 receptor agonist extracted from Gila monster lizard saliva, was shown to powerfully stimulate insulin secretion in a glucose-dependent manner in subjects with and without T2DM. |
| 2005 | Exenatide (synthetic exendin-4) came into clinical use for T2DM. |
| 2006 | Sitagliptin, an orally active Dipeptidyl Peptidase 4 inhibitor, came into use in T2DM. |

Between 1964 and 1967, at least three groups showed independently that glucose, given orally, induced a greater insulin response (by radioimmunoassay) than i.v.( Intravenous) glucose injection even if the blood glucose levels attained were higher because of the i.v. glucose. The three groups therefore knew that the oral glucose was indeed inducing release of "incretins" into the bloodstream that subsequently increased insulin secretion, more than did glucose itself.

In 1971, John C. Brown isolated and deduced the amino acid structure of a peptide he had isolated from intestinal mucosa. Exogenous administration of the peptide inhibited gastric acid secretion in dogs, so he called it gastric inhibitory polypeptide (GIP). Brown and colleagues subsequently found that it had insulinotropic properties and suggested that it be called glucose-dependent insulinotropic peptide, retaining the acronym GIP. They not only demonstrated GIP to be insulinotropic but also demonstrated the glucose dependence of the insulinotropic activity; i.e., plasma glucose must be elevated in order for GIP to induce insulin secretion. GIP was therefore the first incretin to be isolated and its properties characterized (Kim and Egan 2008).

The discovery of a second incretin hormone, glucagon like peptide-1 (GLP-1) in 1983 followed the cloning and sequencing of mammalian proglucagon gene. Both appear to contribute relatively equally to the incretin effect (the potentiation of glucose-stimulated insulin secretion) in an additive manner in healthy subjects (Martin et al. 2011 ).

# 2. LITRATURE REIVEW

## 2.1 Incretin hormones

The gastrointestinal tract is now a recognized organ critical to post-prandial glucose tolerance and perturbed in the diabetic state (Kindel 2010). Incretins are a type of gastrointestinal hormone that cause an increase in the amount of insulin released from the β-cells postprandially, and are part of the larger picture of glucose homeostasis (Funnell 2009). The difference in the insulin secretion between an amount of glucose given orally and the same amount given intravenously is called the ‘‘incretin effect’’, and the hormones responsible are called ‘‘incretins’’. The following criteria have to be fulfilled for an agent to be called an incretin: it must be released in response to oral nutrient ingestion, especially glucose; and it must reach physiological concentrations in vivo to cause insulin release (fig 2.2) (Ranganath 2008 ).

Many hormones have been suspected to contribute to the incretin effect, but today, there is ample evidence to suggest that the incretin effect mainly is conveyed by the two incretin hormones: glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP) (Knop et al. 2009). GIP and GLP-1 can account for the great majority of the incretin effect (Kindel 2010 ). Both GLP-1 and GIP is secreted by the gut in response to nutrient intake and neuronal signals (fig 2.1 ) (Funnell 2009).

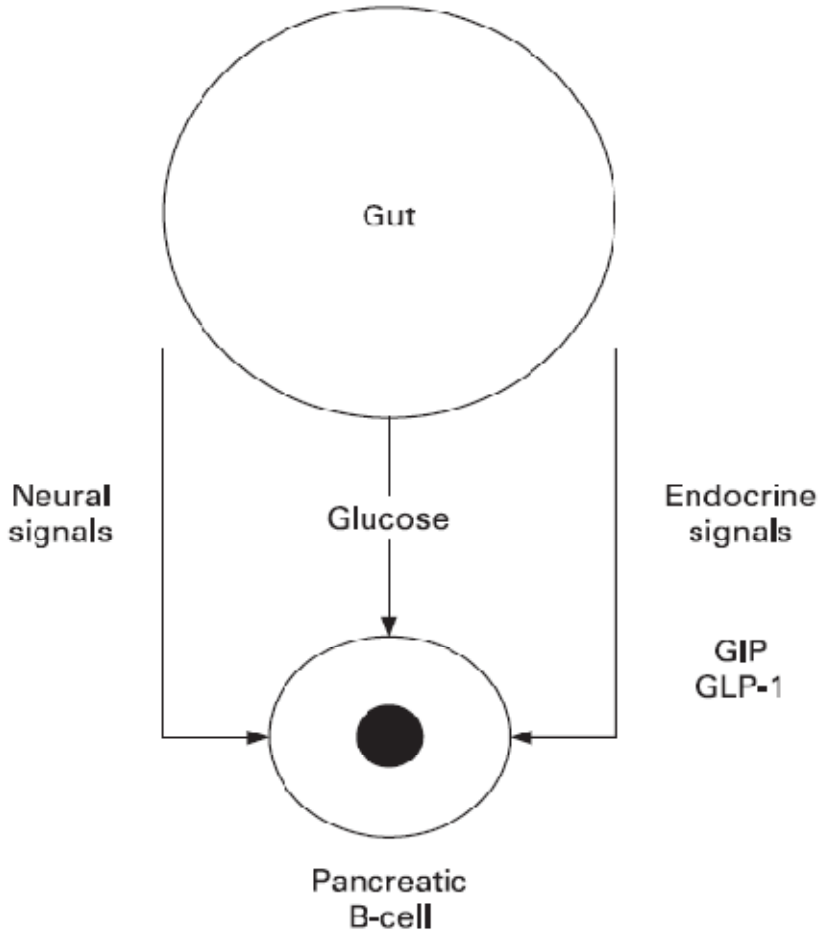


*Figure 2.1 Entero-insular axis. GIP, glucose-dependent insulinotropic polypeptide; GLP-1, glucagon-like peptide-1 (Ranganath 2008 ).*

## 2.2 Biology of incretins

### 2.2.1 Glucose-Dependent Insulinotropic Peptide (GIP)

The first incretin hormone described, GIP, is a single 42-amino acid peptide derived from the post-translational processing of a 153-amino acid precursor encoded by the *gip* gene and a member of a family of structurally related hormones that includes secretin, glucagon, and vasoactive intestinal peptide (Kim and Egan 2008). GIP is synthesized in and released in response to nutrients from enteroendocrine cells (called K cells) primarily in the proximal small intestine (duodenum and jejunum), it was named gastrointestinal inhibitory peptide or gastric

inhibitory peptide because it was thought that its purpose was to neutralize stomach acid to protect the small intestine from acid damage. However, it was later discovered that these effects are only achieved with higher than normal physiological levels, and that these results occur naturally in the body through the hormone secretin (Kindel 2010 ). It is now believed that the function of GIP is to induce insulin secretion, which is primarily stimulated by hyperosmolarity of glucose in the duodenum. After this discovery, it was given the name glucose-dependent insulinotropic peptide, but retained the acronym GIP. Postprandial GIP levels are 10 to 20 times that of fasting levels (Funnell 2009). The half life of GIP is 5-7 minutes and is inactivated by DPPIV (Kindel 2010).

GIP has no significant effect on glucagon inhibition, only modest effects on gastric emptying, and no significant effects on satiety and weight. It may, however, increase β-cell proliferation, decrease β-cell hypertrophy, and decrease β-cell death (apoptosis) (Funnell 2009). Furthermore, this action occurred at physiological plasma levels of GIP (Kim and Egan 2008).

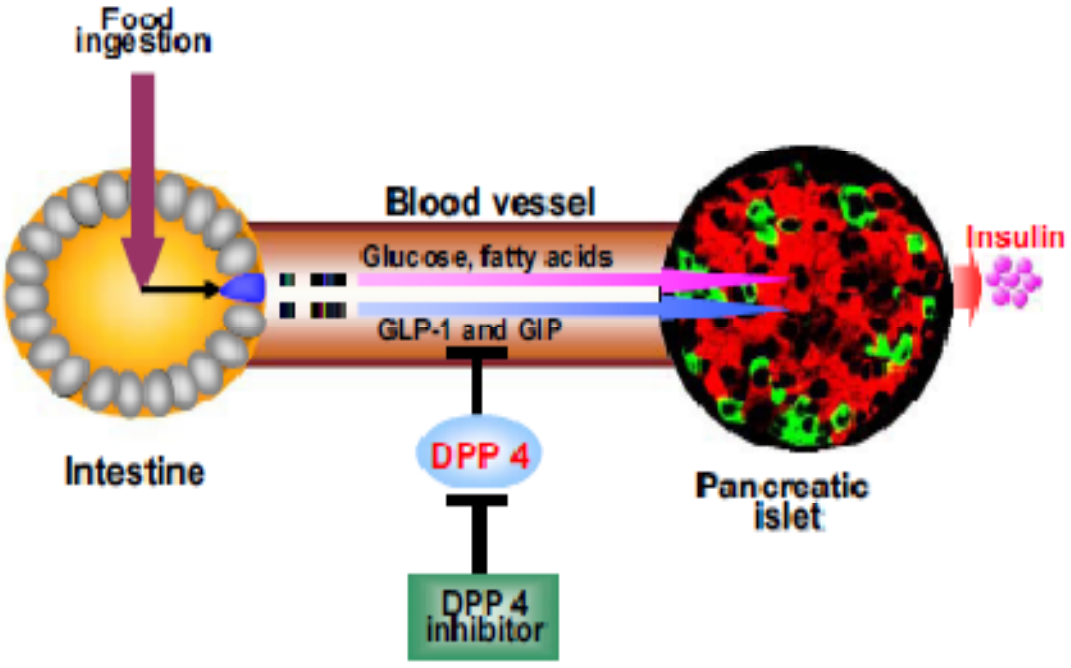


*Figure 2.2 Schematic representations of incretin secretion and action. GIP and GLP-1 are secreted after food ingestion, and they then stimulate glucose-dependent insulin secretion. Once released, GIP and GLP-1 are subject to degradation by DPP4 on lymphocytes and on endothelial cells of blood vessels. The red cells in the islets are insulin-containing (β) cells and the green cells are glucagon-containing (α) cells (Kim and Egan 2008).*

### 2.2.2 Glucagon-Like Peptide-1 (GLP-1)

GLP-1 is a 30-amino acid polypeptide produced in the endocrine L-cells of the intestinal epithelium as a product of glucagon gene expression (Knop et al. 2009). GLP-1 is derived from the proglucagon gene which is expressed in $\alpha$ cells of the pancreas, L-cells of the intestine and the central nervous system (CNS) (Kindel 2010 ).

The primary transcripts and translation products of the gene in the two types of cells, i.e. L-cell and $\alpha$ cell, are identical; but, as illustrated in Fig.2.3 the post-translational processing differs in the two tissues: In the pancreas, proglucagon is cleaved by Prohormone Convertase 2 (PC2) to glucagon, glicentin-related pancreatic peptide and the so-called major proglucagon fragment. Apart from glucagon, these fragments seem to be biologically inactive. In contrast, in the intestinal L-cells, proglucagon is processed by Prohormone Convertase 1(PC1) to GLP-1, Glucagon-Like Peptide-2 (GLP-2) and glicentin. GLP-1 is - as mentioned- secreted in response to ingestion of nutrients and is strongly insulinotropic - a true incretin hormone -and GLP-2, also secreted in response to ingestion of nutrients, is a key regulator of small intestinal growth (Knop et al. 2009) .

In humans, GLP-1 exists in multiple forms. The majority (at least 80%) of circulating biologically active GLP-1 in humans is the COOH-terminally amidated form, GLP-1 (7-36) amide, with lesser amounts of the minor glycine extended form, GLP-1 (7-37), also detectable (Shivanand 2010 ). GLP-1 has an apparent half-life of 1–2 min (Nauck 2009) .

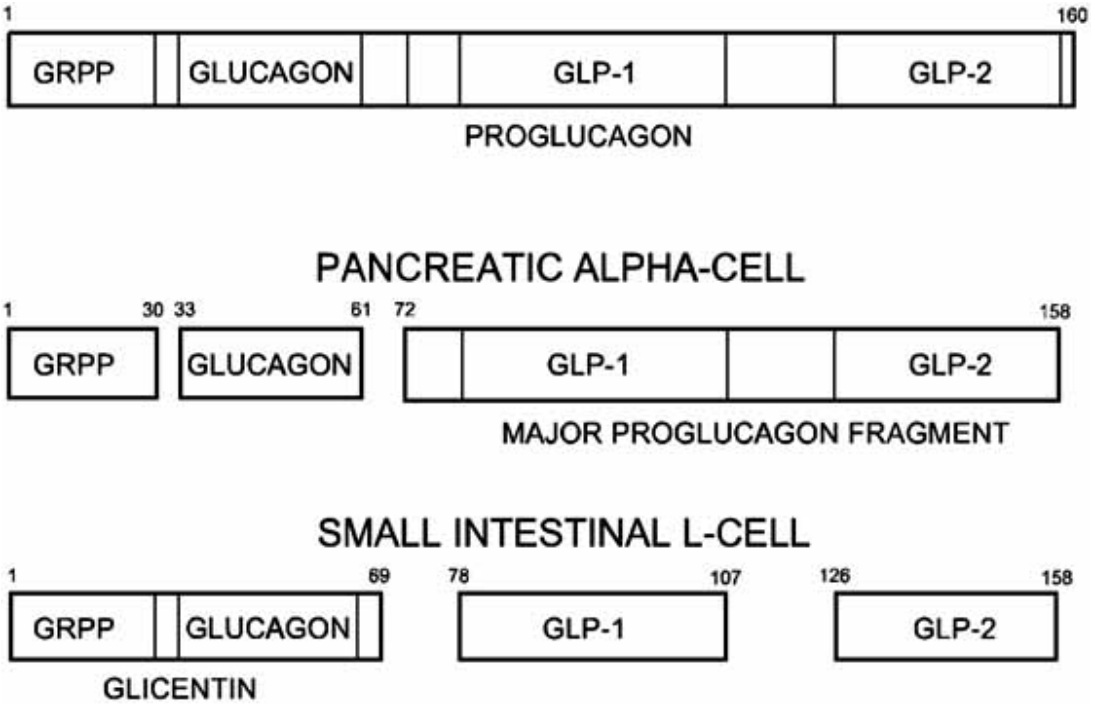


***Figure 2.3*** *Proglucagon processing in human pancreatic alpha-cells and in mucosal endocrine L-cells in the small intestine. GRPP: Glicentin-Related Pancreatic Peptide; GLP-1: glucagon-like peptide-1; GLP-2: Glucagon-Like Peptide-2(Martin et al. 2011).*

GLP-1 promotes glucose-dependent insulin secretion by acting directly on the β-cells of the pancreas. The agents that cause or stimulate secretion of this hormone include all major nutrients (e.g. carbohydrates, proteins, fats), but fats are the most potent stimulus. In addition, GLP-1 suppresses glucagon production by acting on α-cells (Funnell 2009). It also increases the biosynthesis of new insulin molecules and up regulates all the entire machinery required for insulin synthesis in the *β*-cells. Finally, and probably most important for humans, it strongly inhibits cytokine, lipid and glucose-induced apoptosis of β-cells including human *β*-cells. It also significantly inhibits appetite and food intake, and probably acts as one of the endocrine signals from the gut that terminate meal ingestion and signals inter digestive satiation (Holst 2006).

In type 2 diabetic patients, GLP-1 can improve fasting hyperglycemia caused by hyperglucagonemia and inappropriate hepatic glucose production (Kindel 2010). Most recently GLP-1 has also been suggested to exert protective effects on the heart and the blood vessels. All of these actions render GLP-1 of unusual interest in the context of diabetes treatment (Holst 2006). There is evidence that GLP-1 and GIP have 'insulin-like effects' in peripheral tissues, enhancing glucose uptake in liver, adipose and muscle tissues (Green 2007).

*Table 2.1 Action of GIP and GLP-1 that affect blood glucose level (Kim and Egan 2008).*

| | GIP | GLP-1 |
|---|---|---|
| Islets | | |
| Insulin secretion | ↑ | ↑ |
| Insulin synthesis | ↑ | ↑ |
| Insulin, glucokinase and GLUT2 expression | ↑ | ↑ |
| Glucagon secretion | ↑ | ↓ (indirect) |
| Somatostatin secretion | ↑ | ↑ (indirect) |
| β cell proliferation | ↑ | ↑ |
| β cell apoptosis | ↓ | ↓ |
| Sweet taste modulator | — | Yes |
| Gastrointestinal tract | | |
| Gastric emptying | — | ↓ |
| Gastric acid secretion | ↓ | ↓ |
| Motility | — | ↓ |
| Central nervous system | | |
| Food intake | — | ↓ |
| Satiety | — | ↑ |
| Muscle | | |
| Glucose uptake | — | ↑ |
| Liver | | |
| Glucose production | ↓ (indirect) | ↓ (indirect) |

↑, increase; ↓, decrease; —, no effect or not reported.

## 2.3 Synthesis, Secretion, and Degradation of Incretins

### 2.3.1 Synthesis and Secretion of GIP

As already stated, GIP is synthesized within and secreted from K cells in the small intestine, the highest density of K cells being in duodenum and jejunum; few, if any, K cells are present in distal ileum (Reimann 2010 ).

Human GIP is a single 42-amino acid peptide derived from the processing by PC1/3 of proGIP, a 153-amino acid precursor (Fig. 2.4) that is encoded by a 459-bp open reading frame and whose gene is localized to chromosome 17q. It is composed of six exons, and the majority of GIP-encoding sequences are in exon 3. This sequence includes a 51-amino acid N-terminal peptide containing a signal peptide with a cleavage site at glycine and a 60-amino acid C-terminal peptide flanking the 42-amino acid GIP hormone, which presently seems to be the only biologically active peptide derived from proGIP ( Baggio and Drucker 2007). GIP secretion from the gut occurs in a regulated manner (Kim and Egan 2008).

In the fasting state, the plasma concentrations of GIP is very low, although it is not immeasurable, suggesting that there is a certain basal rate of secretion. It is secreted rapidly (within 10-20 min) in response to ingestion of nutrients, with lipids and simple carbohydrates being potent stimulators of secretion (Asmar 2011 ). More specifically, it is the rate of nutrient absorption rather than the mere presence of nutrients in the intestine that stimulates GIP release. Thus, GIP secretion is reduced in individuals with intestinal malabsorption or after the administration of pharmacologic agents that reduce nutrient absorption (Baggio and Drucker 2007). Peak concentrations of GIP reached as soon as 15-30 minutes, after ingestion of e.g. glucose (Asmar 2011 ).

As GIP-secreting cells were found to express glucokinase (hexokinase-IV), which has a restricted expression profile, it was hypothesised that glucose-sensing in K-cells must involve a mechanism similar to that employed by the pancreatic β-cell, where this enzyme plays a key role. The rapid secretion following ingestion of nutrients has led to the notion of vagus-mediated stimulation of secretion. However, identification of glucokinase expression in the K-cells of action potentials, via mechanisms involving closure of adenosine 5'-triphosphate (ATP)-sensitive $K^+$ channels and trigger GIP secretion (Reimann 2010 ).

After the secretion of GIP, it is subject to degradation by the enzyme dipeptidyl peptidase 4 (DPP4). This enzyme, also known as the T-cell antigen CD26, is a serine peptidase found in numerous sites such as the intestinal and renal brush border membranes, hepatocytes and vascular endothelium, as well as in a soluble form in plasma (Knop et al. 2009).

The first two amino acids (Tyr, Ala) at the N terminus of full-length GIP (1-42) are cleaved in 1 to 2 min in rodents and 5 to 7 min in humans by DPP4 and converted to GIP (3-42), which has insignificant, if any, insulinotropic activity. It is then excreted by the kidney. The elimination rates of GIP (1-42) and GIP (3-42) are similar in subjects with T2DM and those without; hence, more rapid degradation/elimination of GIP is unlikely to be a factor in defective insulinotropic effects seen in T2DM (Asmar 2011 ).

### 2.3.2 Synthesis and Secretion of GLP-1

The human *proglucagon* gene located on the long arm of chromosome 2 has six exons, of which exons 2 to 5 encode distinct functional domains, and five introns. It spans approximately 9.4 kb. Just a single gene encodes the proglucagon sequence in mammals and proglucagon, 180 amino acids long, is very similar in all mammalian species (greater than 90% amino acid sequence homology). Glucagon is encoded in exon 3, and GLP-1 and -2 are encoded in exons 4 and 5, respectively. Recent studies have uncovered some of the intracellular signaling pathways that mediate nutrient-induced GLP-1 secretion from L cells, which is then most likely modified by the neural and endocrine factors. GLP-1 secretion from the gut occurs in a regulated manner (Kim and Egan 2008).

Peak concentration of GLP-1 is reached as soon as 30-45 minutes after ingestion of e.g. glucose (Knop et al. 2009). Several studies have shown that the autonomic nerve system, the neurotransmitter Gastrin-Releasing Peptide (GRP) and acetylcholin contribute to the early phase GLP-1 release. Studies in rodents have shown that, after a meal, GIP released by K-cells activates vagal afferents, leading to GLP-1 secretion through vagal efferents and enteric neurons that release acetylcholine and GRP. Only oral glucose intake stimulates GLP-1 release and recent experiments suggest that the action of enteral glucose could be mediated by test receptors expressed on L-cells. The Taste Receptors cells (T1Rs) of the lingual epithelium bind sweet

compounds and activate specific receptors coupled through the G protein 'gustdusin' to specific second messenger cascades: phospholipase C and a calcium activated channel. Duodenal L-cells also express the sweet T1Rs and gustducin. Gut L-cells detect glucose through the same mechanism as that used by tongue taste cells (Girard 2008).

Several studies show that specific GPCRs present on L cells are necessary for GLP-1 secretion. In particular, long-chain free fatty acids and lipids stimulate GLP-1 secretion through interaction with GPCRs, including GPR120, GPR119, and GPR40. GPR120 is highly expressed in the intestine and the stimulation of GPR120 by free fatty acids promotes the secretion of GLP-1 via increase of intracellular $Ca^{2+}$ levels and activation of p42/44 MAPK (Reimann 2010 ).

### *Degradation of GLP-1*

Typical basal (fasting) levels of bioactive GLP-1, measured from peripheral veins, are in the range of 5 to 10 pM and increase by 2- to 3-fold after meal ingestion, depending on the size and composition of meal. The first two N-terminal amino acids (His Ala) of native GLP-1 are rapidly cleaved by DPP4, and the resulting GLP-1 (9-36) fragment is not insulinotropic (Fig.2.4) (Seino et al. 2010).

GLP-1 is also degraded by neutral endopeptidase 24.11 (NEP-24.11), which is a membrane-bound zinc metallopeptidase, and six potential cleavage sites in GLP-1 have been identified. High levels of this enzyme are found in the kidney and GLP-1, and its metabolites are rapidly cleared through the kidneys, implying the involvement of NEP-24.11 in renal clearance of GLP-1. Therefore, many modifications have been made to synthetic GLP-1 so as to increase its biological half-life and consequently its efficacy in vivo and therapeutic strategies based on modulating GLP-1 levels and GLP-1 activity through administration of GLP-1 and its analogs or by inhibiting its degradation have been tested and/or are under development for treating T2DM. The elimination rates of GLP-1 are similar in T2DM and non diabetic subjects, and, as with GIP, more rapid degradation of GLP-1 is unlikely to be a contributory cause for glucose-responsiveness in T2DM (Kim and Egan 2008).

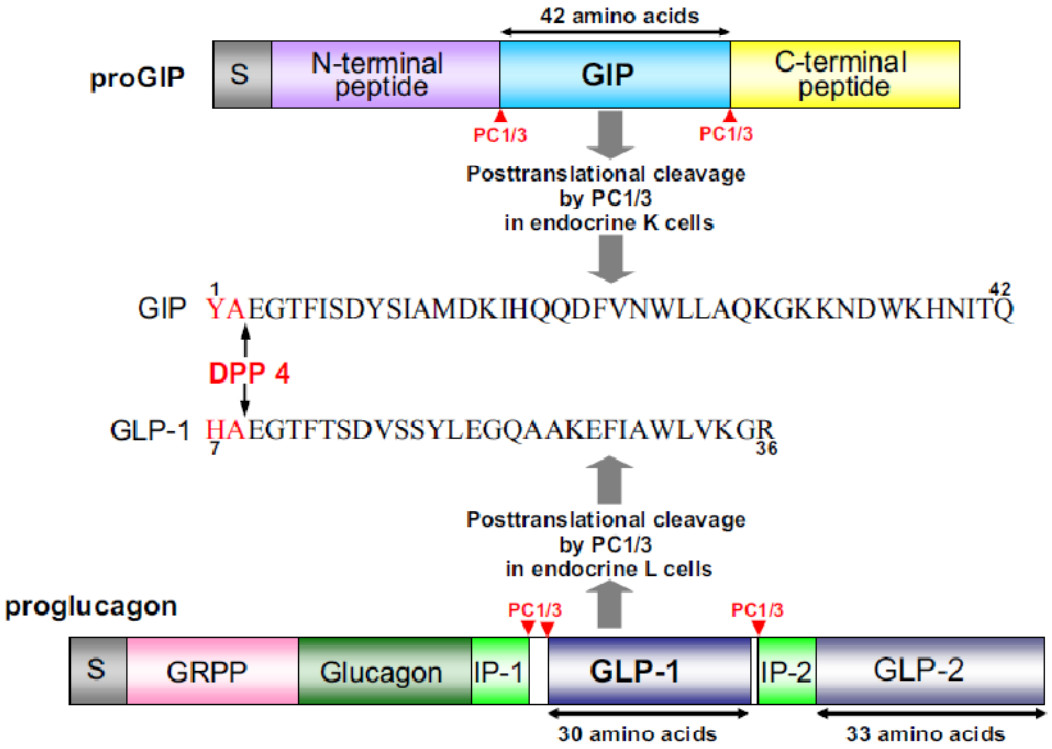


*Figure 2.4 Schematic representation of proGIP and proglucagon (Kim and Egan 2008).*

## 2.4 Regulation of incretin secretion

The L cell (as well as the K cell) is an open-type intestinal epithelial endocrine cell that is in direct contact with nutrients through its microvilli on the luminal surface, in contact with the enteric nervous system as well as the Central Nervous System (CNS) via the vagus, and in contact with the microvasculature through its basolateral membrane. Nutrient intake, composition and absorption are major factors affecting release of GLP-1(Ranganath 2008 ). This allows GLP-1 secretion from L cells, as well as GIP from K cells, to be regulated by a variety of nutrient, neural, and endocrine signals. Indeed, several studies have postulated that GLP-1 secretion from the L cells is regulated by a complex proximal-distal loop that involves both endocrine and neural factors with the vagus nerves having an essential role in this loop. In this loop also, GIP, acetylcholine, and gastrin releasing peptide act as mediators: the afferent vagus nerve is activated by GIP, which subsequently stimulates GLP-1 secretion through the efferent vagus nerve and enteric neurons that release acetylcholine and gastrin-releasing peptide. It seems

that GLP-1 secretion is also affected by other neurotransmitters and peptides, including GABA and *calcitonin* gene related peptide. Furthermore, non-nutrient factors, including leptin and insulin, have also been identified as stimulators of GLP-1 secretion. Conversely, somatostatin, which is produced from the intestinal enteroendocrine D cells (as well as from δ cells in islets of Langerhans) and whose secretion is increased by GLP-1, has been identified as the inhibitor of GLP-1 secretion, implicating the existence of a negative local feedback loop in the gut (Kim and Egan 2008).

## 2.5 Incretin Receptors

### 2.5.1 Glucose-Dependent Insulinotropic Peptide Receptor (GIPR)

The GIPR is a glycoprotein belonging to the class II G protein-coupled receptor super family that includes receptors for glucagon, GLP-1, secretin, vaso-active intestinal polypeptide, and pituitary adenylyl cyclase-activating protein. As with other GPCRs of this class, GIPR comprises an N-terminal extracellular domain that is essential for high-affinity GIP binding and receptor activation; a central transmembrane domain (the first transmembrane domain of which is important for receptor activation and cAMP (cyclic Adenosine Monophosphate) coupling); and a C-terminal cytoplasmic domain that mediates intracellular signaling by physical association with Gs protein (Seino et al. 2010).

GIP receptors are expressed in a number of cell types including pancreatic β- and α-cells, stomach, adipose tissue and brain (Ranganath 2008 ). Similar to the GLP-1R, the GIPR is a member of the 7-transmembrane–spanning, heterotrimeric G-protein–coupled receptor super family (Baggio and Drucker 2007).

### 2.5.2 Glucagon-Like Peptide-1 Receptor (GLP-1R)

GLP-1R is in the same class of receptors as GIPR. The GLP-1R is a 7-transmembrane-spanning, heterotrimeric G-protein coupled receptor (Kindel 2010 ). Under physiological conditions studied to date, however, it seems that GLP-1R activation leads to increased intracellular cAMP and $Ca^{2+}$ concentrations and activation of downstream pathways, including PKA, PKC, PI-3K, Epac2, and MAPK signaling pathway (Kim and Egan 2008).

GLP-1 receptors are found on β- and δ-cells of the pancreas, parietal cells of the stomach, pylorus, adipose tissue, lungs and the brain (Ranganath 2008 ) such as the hypothalamic centers controlling energy intake. GLP-1R is also found in kidney, liver cholangiocytes, heart, intestine and in the C-cells of the thyroid in rodents. GLP-1 binding has been detected in several neuroendocrine and lung tumours and in osteoblastic cells, GLP-1 receptor was not detected. Other groups have similarly failed to find any GLP-1R in osteoblasts or osteoclasts or any direct effects of GLP-1 on these cells in culture. In humans no changes in markers of bone turnover following acute administration of either GLP-1 or GIP have been reported (Martin et al. 2011).

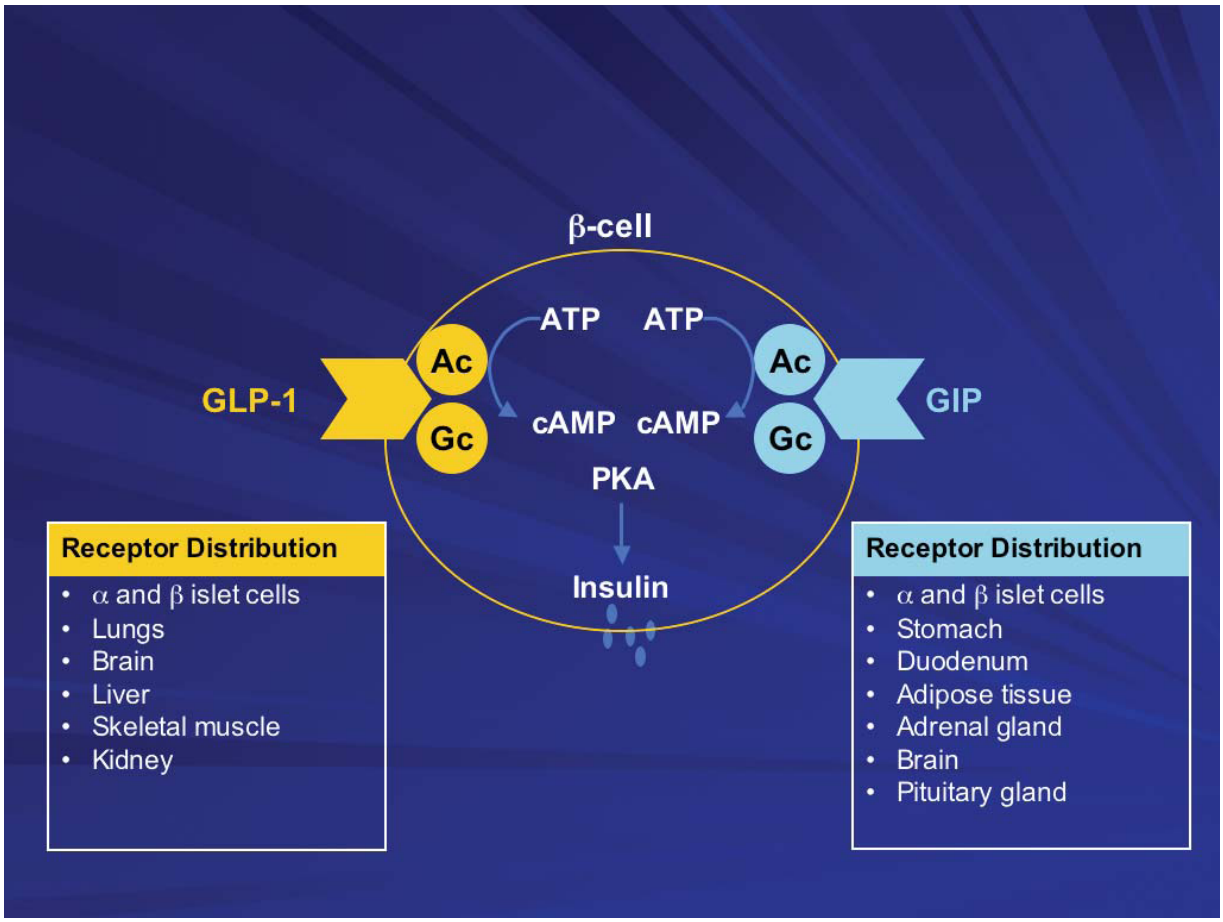


***Figure 2.5*** *Target tissues of glucagon-like peptide 1 (GLP-1) and glucose-dependent insulinotropic peptide (GIP) receptors: The progression of events involving GLP-1 and GIP leads to insulin secretion in β- cells. Also shown is the tissue-receptor distribution of GLP-1 (left) and GIP (right). Abbreviations:Ac, adenylate cyclase; ATP, adenosine triphosphate; cAMP, cyclic adenosine monophosphate; Gc, guanylate cyclase; PKA, protein kinase A (Freeman 2007).*

## 2.6 The Pleiotropic Effect of Incretin in Pancreas

### 2.6.1 The Pleiotropic Effects of GIP in Pancreas

#### *a. Effects on β-Cells*

GIP exerts glucose-dependent stimulatory effects on insulin secretion in animals and humans (Gautier et al. 2008 ). GIP binding to its specific receptor on pancreatic β-cells facilitates the secretion of insulin in the presence of Post Prandial Glucose concentration via activation of proximal signal transduction pathways, and the ability of GIP to promote glucose-stimulated insulin secretion in pancreatic β-cells is attributed to GIPR activation leading to membrane depolarization and increases in the intracellular $Ca^{2+}$ concentration as well as direct effects on insulin exocytosis (fig 2.6) (Kim and Egan 2008).

Ligand binding to GIPR activates several mechanisms, including stimulation of the adenylyl cyclase/cAMP/PKA), inhibition of $K_{ATP}$ channels, increases in intracellular $Ca^{2+}$, activation of phospholipase A2 (PLA2), specific protein kinase signaling pathways, including PKB, mitogen-activated protein kinases, and stimulation of exocytosis. The rise in cAMP significantly up-regulates activity of PKA, which leads to a cascade in which phosphorylation of regulatory proteins, including GLUT2 (a low *K*m glucose transporter), SUR1 (subunits of the pancreatic β-cell $K_{ATP}$ channel), α-SNAP (a vesicle-associated protein), and other ion channels ($Ca^{2+}$ channels, for example), induces membrane depolarization and subsequent exocytosis of insulin, as well as phosphorylation of critical transcription factors, which results in significant up-regulation of insulin gene promoter activity. PKA-independent mechanism is also involved in GIP mediated insulin secretion. The cAMP-binding protein cAMP-GEFII (Epac2) mediates cAMP-dependent, PKA independent insulin secretion, and this effect of Epac2 on insulin secretion is mediated by Rim2 and depends on intracellular $Ca^{2+}$ as well as on cAMP. (Baggio and Drucker 2007).

Metabolism of glucose has been shown to induce hydrolysis of membrane phospholipids, leading to the accumulation of arachidonic acid (AA), which amplifies insulin secretion. Incretin- and glucose-stimulated insulin secretion is also mediated via opening of voltage-dependent $Ca^{2+}$ channels as well as activation of several species of $K^+$ channels, including $K_{ATP}$ channels, $Ca^{2+}$ activated $K^+(K_{CA})$ channels, and voltage-gated $K^+$ ($K_V$) channels (Kim and Egan 2008).

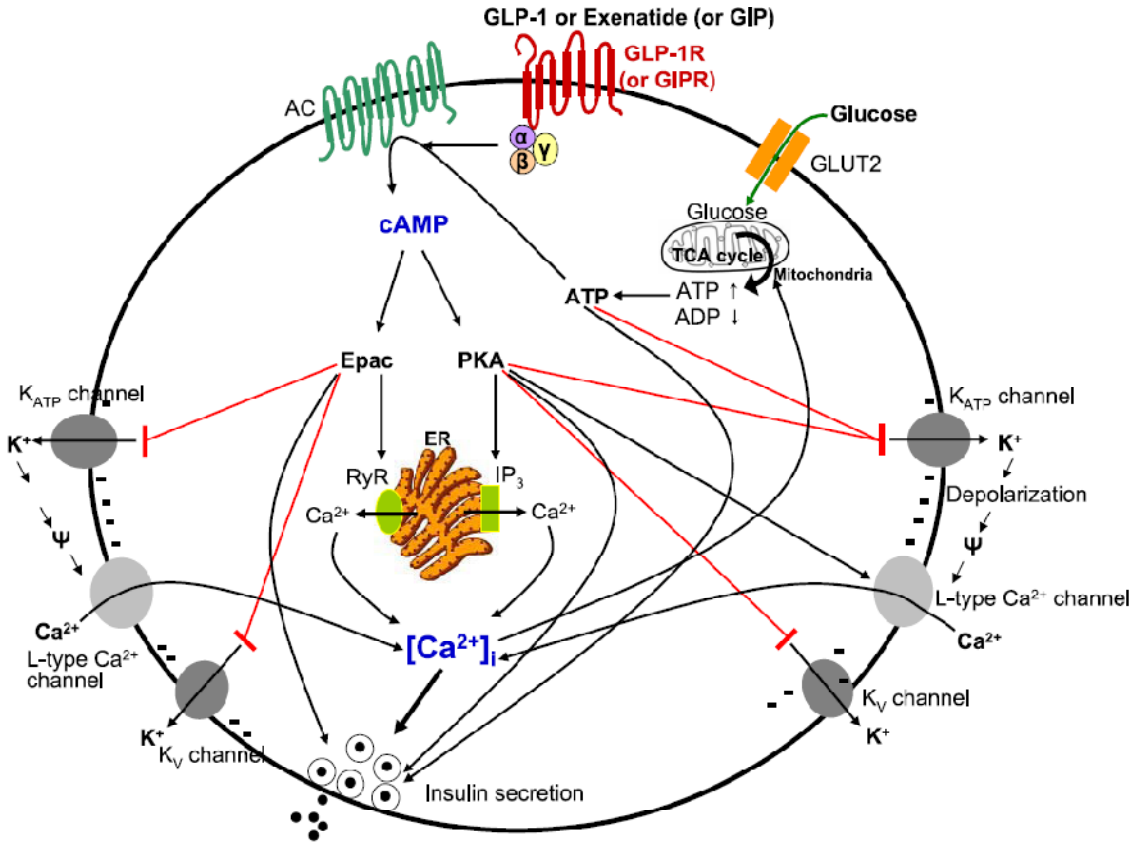


*Figure 2.6 A schematic representation of the main molecular events during incretin-induced insulin secretion from a β-cell (Kim and Egan 2008).*

In addition to potentiating the release of insulin from β-cells, GIP replenishes insulin in β-cells by increasing insulin gene transcription and biosynthesis, and enhances the glucose-sensing system by increasing the expression of components of β-cell glucose sensors (Nauck 2009). Activation of the transcription factor PDX-1, a key regulator of islet growth and insulin gene transcription may be involved (Knop et al. 2009). The molecular signaling pathways that mediate the proliferative and anti-apoptotic actions of GIP have been elucidated using heterologous cells transfected with the GIPR, rodent β-cell lines, or murine islets, and include activation of cAMP/PKA, PKA/CREB, MAPK, PI-3K–dependent activation of Akt-PKB, reductions in caspase 3 activities, and down-regulation of *bax* gene transcription. The limited information available from in vivo studies has shown that infusion of GIP into diabetic rats for 2 weeks significantly reduces β-cell apoptosis by activation of PI-3K/Akt-PKB and subsequent phosphorylation and nuclear exclusion of FoxO1, resulting in decreased expression of the pro-

apoptotic *bax* gene and up-regulation of the anti-apoptotic *bcl-2* gene (Seino et al. 2010) (fig 2.7).

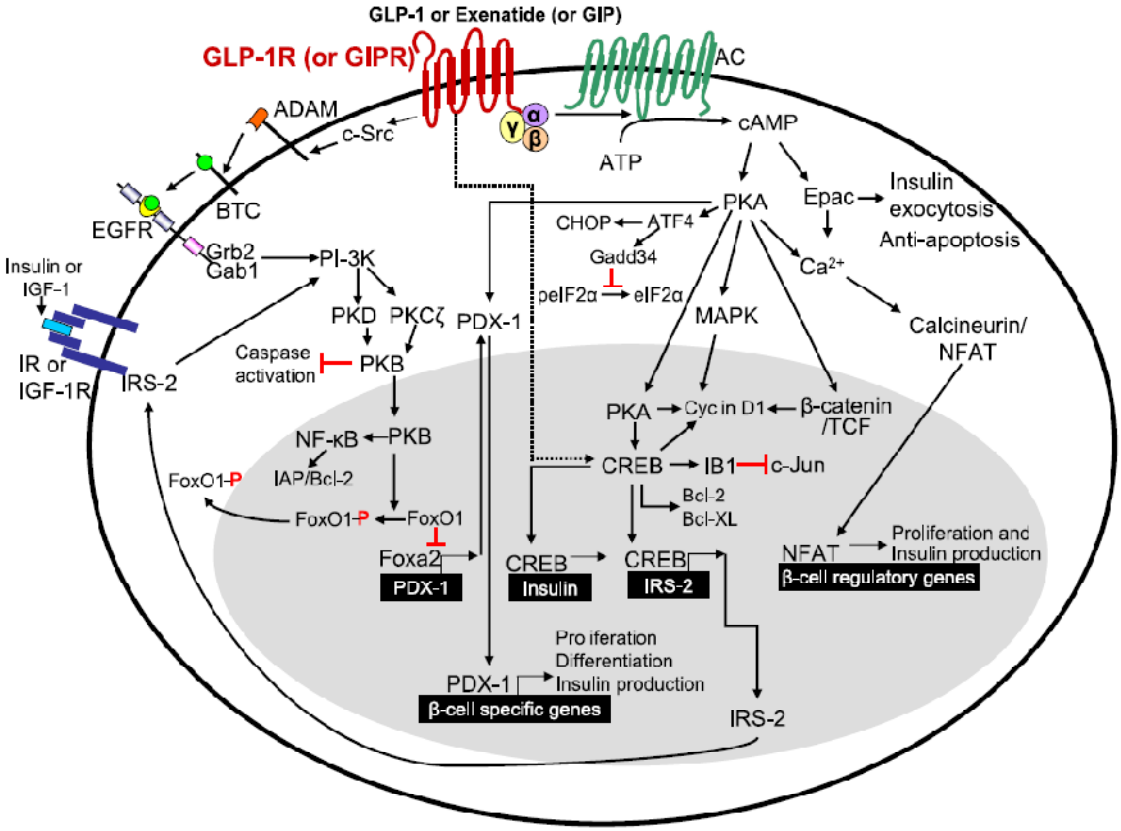


*Figure 2.7 Schema outlining the incretin downstream signal transduction pathways in a β-cell. GLP-1R activation (and GIPR activation, to some extent, but this has not been as well studied as GLP-1R activation) recruits signaling mechanisms that considerably overlap, leading to promotion of β-cell proliferation and prevention of β-cell apoptosis. Dashed line indicates mechanism that is not fully delineated(Kim and Egan 2008).*

### b. *Effects on Glucagon Secretion*

In contrast with GLP-1, GIP does not influence pancreatic α-cell secretion of glucagon in humans (Gautier et al. 2008 ).

### 2.6.2 Pleiotropic Effects of GLP-1 in Pancreas

#### *a. Effects on β-Cells*

GLP-1 is one of the most potent substances known to stimulate glucose-dependent insulin secretion from islet β-cells in animals and humans (Kim and Egan 2008). It has been demonstrated in rats, that insulin secretary responses to oral intake of nutrients and to intraduodenal glucose were suppressed when using the GLP-1 receptor antagonist exendin 9-39. In addition, mice lacking the GLP-1 receptor showed fasting hyperglycaemia and abnormal glucose excursion with significantly reduced insulin secretion after a glucose load (Gautier et al. 2008 ).

In humans, secretion of GLP-1 throughout the day is strongly correlated with the release of insulin. The effect of GLP-1 on insulin secretion is strictly glucose-dependent and there is no effect of GLP-1 on insulin secretion for glucose concentrations below a certain threshold (approximately 4.5mmol/l). At lower plasma glucose concentrations both hormones lose their insulinotropic activity completely ( Vilsbøll 2009).

Like GIP, its stimulatory activity is exerted via binding to its receptor on β-cells (fig 2.6). This binding results in activation of adenylyl cyclase with consequent production of cAMP. Subsequently, GLP-1 stimulates insulin secretion via mechanisms that include the following; (1) direct inhibition of $K_{ATP}$ channels, which leads to β-cell membrane depolarization; (2) increases in intracellular $Ca^{2+}$ levels resulting from GLP-1–dependent influx channels, activation of nonselective cation channels, and mobilization of intracellular $Ca^{2+}$ stores; (3) increases in mitochondrial ATP synthesis, which lead to further membrane depolarization; (4) closure of voltage-dependent $K^{+}$ (Kv) channels and consequent reductions in Kv currents, thereby preventing β-cell repolarization; and (5) direct effects on β-cell insulin storage granule exocytosis that occur distal to increases in ATP and intracellular $Ca^{2+}$ (Shivanand 2010 ).

The primary effector of GLP-1–induced insulin secretion is cAMP, and cAMP mediates its stimulatory effect on insulin secretion via two distinct mechanisms: (1) PKA-dependent phosphorylation of downstream targets and (2) PKA-independent activation of Epac2. Both cAMP/PKA and PI-3K/PKCζ signaling pathways mediate the antagonistic effect of GLP-1 on

Kv currents. GLP-1 can enhance $K_{ATP}$ channel–independent glucose-induced insulin secretion by PKA- and PKC-dependent signaling pathways. The glucose-dependent actions of GLP-1 are not understood completely, but may converge on the $K_{ATP}$, Kv channels, and GLUT2 glucose transporters and potentially at the level of insulin granule exocytosis (Baggio and Drucker 2007). GLP-1 has a tropic effect on β-cells, both in terms of enhancing the magnitude of insulin secretion as well as increasing their number. GLP-1 increases β-cell mass by stimulating proliferation and induction of islet neogenesis as well as by inhibiting apoptosis (Ranganath 2008 ). Thus, GLP-1 causes proliferation of already existing *β*-cells and also promotes differentiation of new *β*-cells from progenitor cells in the ducts leading to the formation of small new islets (Holst 2006). Unlike its insulinotropic effect dependent on Protein Kinase A (PKA) pathways, the trophic and survival effects of GLP-1 are mediated through different signalling pathways involving PKB (Gautier et al. 2008 ) (Fig 2.7).

**b.** ***Effects on Glucagon Secretion***

GLP-1 is able to suppress glucagon secretion in pancreatic islets, in perfused pancreas and in whole organisms. The mechanism by which GLP-1 inhibits glucagon secretion remains to be elucidated. The inhibitory effect is probably indirectly mediated *via* insulin release and *via* somatostatin secretion. A specific antagonist of the somatostatin receptors (subtype 2) completely abolished the GLP-1 effect and actually increased the secretion of glucagon, suggesting that the somatostatin-producing δ-cells of the islets transmit the effects of GLP-1 by paracrine inhibition of the alpha cells and keep them under tonic suppression. However, a direct effect of GLP-1 is not completely excluded since GLP-1 receptors are expressed on pancreatic α-cells (Holst et al. 2009). The cellular mechanisms are believed to involve an increase in cAMP / PKA, closure of K-ATP channels, membrane depolarization, inactivation of ion channels and reduction of intracellular calcium (Girard 2008).

The inhibitory effects of GLP-1 on glucagon secretion seem to represent an important mechanism for regulating elevated levels of blood glucose. In patients with type 1 diabetes (i.e. who are deprived of insulin), administration of GLP-1 decreased blood glucose levels while the secretion of glucagons was strongly inhibited, suggesting that GLP-1 suppressed the hepatic production of glucose induced by glucagon. The inhibition of glucagon secretion by GLP-1 is

glucose-dependent, meaning that GLP-1 administration is unlikely to impair the glucagon counter regulatory response to hypoglycaemia (Seino et al. 2010).

#### *Somatostatin secretion*

GLP-1 is a potent stimulator of somatostatin secretion from isolated human islets. This effect is not dependent on glucose concentrations (Gautier et al. 2008 ). Direct receptor-mediated stimulation of the δ-cell by GLP-1 has been observed, indicating a complex interplay between GLP-1 and all the major endocrine cells of the islets that may be important in the fine-tuning of homoeostatic control of glucose metabolism (Ranganath 2008 ).

## 2.7 Extrapancreatic Effects of Incretins

### 2.7.1 Extrapancreatic Effects of GIP

#### *a. Central Nervous System*

GIPRs have been detected in many areas of the brain, including the hippocampus and hippocampal progenitor cells. Thus, GIP and GIPR are indeed expressed in the CNS and seem to play a role in neural progenitor cell proliferation and behavior modification. Whether this is true for humans is unknown (Kim and Egan 2008).

#### *b. Gastrointestinal Tract*

”Gastric inhibitory polypeptide,” as already stated, was originally characterized for its inhibition of gastric acid secretion. However, although its inhibitory effect on gastric acid secretion is seen under physiologic conditions in dogs, humans require supraphysiologic plasma concentrations to bring about gastric acid inhibition. Upper and lower GI motility is inhibited by GIP at supraphysiologic plasma concentrations in healthy human subjects, but the rate of gastric emptying is unaffected by GIP, even at highly supraphysiologic plasma levels (Seino et al. 2010).

### c. *Effect on adipose tissue metabolism*

GIP receptors are present on adipocytes and there is experimental evidence indicating that GIP regulates fat metabolism in adipocytes, including enhanced insulin-stimulated incorporation of fatty acids into triglycerides, stimulation of lipoprotein lipase activity, stimulation of fatty acids synthesis. At present, the exact signalling mechanisms mediating the effects of GIP on fat cells are unknown (Gautier et al. 2008 ).

## 2.7.2 Extrapancreatic Effects of GLP-1

### *a. Central Nervous System*

GLP-1 has been shown to reduce caloric intake and to enhance satiety, these effects being probably related to central mechanisms (Gautier et al. 2008 ). It had been proposed that the anorectic effects of GLP-1R agonists are due to visceral illness rather than a primary effect on food intake (Kindel 2010 ). It is also likely that inhibition of gastric emptying mediated by GLP-1 increases the sensation of fullness and leads to the termination of meal ingestion, thereby participating in the regulation of food intake (Shivanand 2010 ). Alternatively, central administration of GLP-1R agonists in rodents reduced food and water intake, therefore decreasing body weight suggesting a direct effect on food intake (Kindel 2010 ).
Similar effects were observed in obese subjects, as well as in patients with type 2 diabetes. In type 2 diabetic patients treated with a subcutaneous infusion of GLP-1 for up to 6 weeks, the reduction of food intake was sustained and associated with a reduction of body weight possibly mediated by neural sensing within the lamina propria of the gastrointestinal tract as selective ablation of the nodose ganglion and vagus nerve in mice completely blocks the anorectic effects of peripheral exendin-4 (Nauck 2009).

### *b. Effects on the gastrointestinal tract*

GLP-1 exerts inhibitory effects on gastrointestinal secretion and motility, particularly on gastric emptying. Administration of GLP-1 at physiological doses in healthy volunteers results in a dose-dependent slowing of gastric emptying and of glucose absorption, which participate in a subsequent reduction of postprandial plasma glucose concentration. This suggests that GLP-1 participate in the “ileal brake” phenomenon, by which nutrients present in the distal part of the

small intestine induce a reduction in upper intestinal motility and secretary activity. The actions of GLP-1 on gastrointestinal motility and secretion probably involve neurally-mediated mechanisms, including vago-vagal pathways (Holst et al. 2009).

The physiological role of GLP-1 may be to adjust the absorptive capacity of the gut and to adjust the amount of chyme, by slowing gastrointestinal transit and decreasing secretion of digestive enzymes. Under physiological conditions, it is likely that the gastrointestinal effects of GLP-1, (i.e. reduction of gastric secretion and slowing of gastric emptying) are more important than its insulinotropic action. In pathological conditions such as diabetes, the inhibitory effects of GLP-1 on gastrointestinal motility, particularly gastric emptying, are of special interest because they potentially reduce postprandial glucose excursions (Gautier et al. 2008 ).

### *c. Effects on the bone and the heart*

It has been known for some time that there are GLP-1 receptors in the heart. GLP-1 can improve endothelial dysfunction in type 2 diabetic subjects with stable coronary heart disease. An effect on endothelium dependent vasodilation was confirmed in healthy subjects and functional receptors for GLP-1 have been identified on endothelial cells. Additionally, GLP-1 exerts a direct protective effect on the myocardium against ischemia/reperfusion injury in rat models by mechanisms independent of insulin.GLP-1 also increases left ventricular developed pressure and coronary blood flow in isolated mouse hearts, although in normal rat hearts, it reduces contractility (Holst et al. 2009).

Importantly, the GLP-1 metabolite GLP-1 amide that has no insulinotropic action, could also mediate myocardial glucose uptake, thus improving left ventricular performance in dogs with dilated cardiomyopathy. GLP-1 may control bone resorption. Mice devoid of GLP-1 receptor have cortical osteopenia, bone fragility, increased osteoclast numbers and markers of bone resorption. These effects are corrected by calcitonin treatment, indicating that GLP-1 bone-protective effects may involve a calcitonin-dependent pathway (Seino et al. 2010).

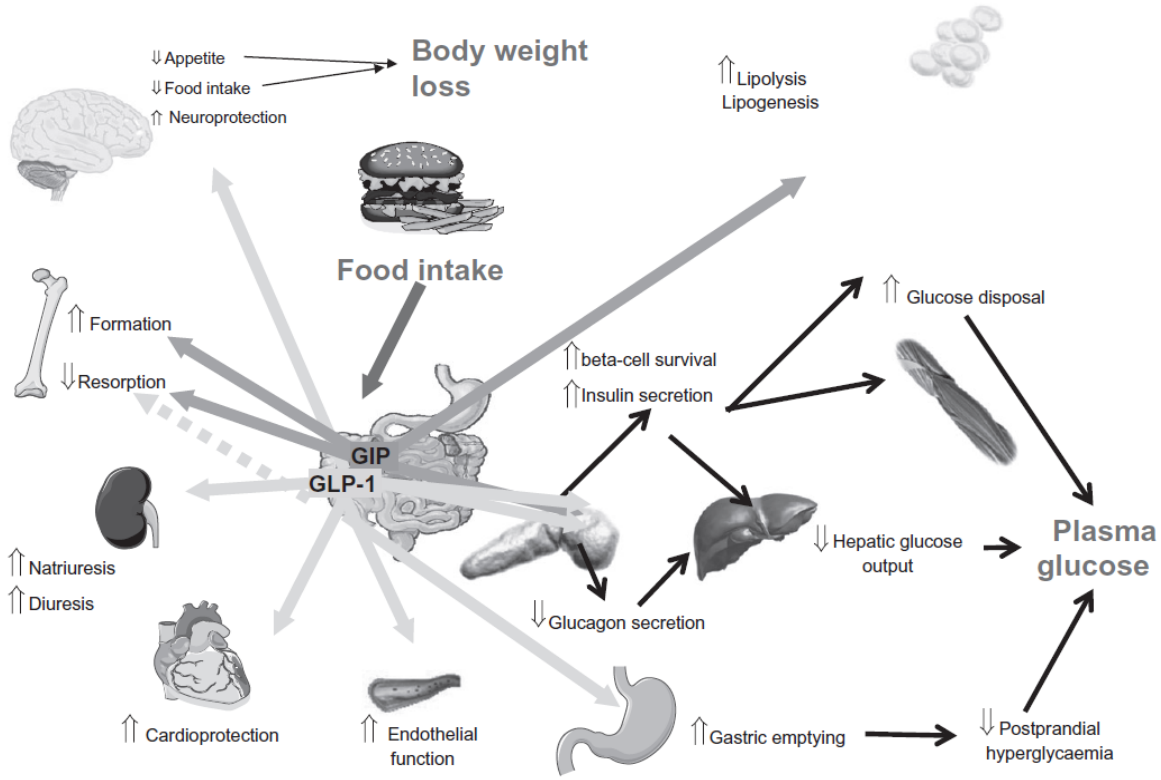


***Figure 2.8*** *Glucoregulatory effects of glucagon-like peptide (GLP) and GLP-1. GIP, gastric inhibitory peptide (Martin et al. 2011).*

# 3. OBJECTIVE

## 3.1 General objective

To review the role of incretin hormone in type 2 diabetes

## 3.2 Specific objective

To explain the incretin hormones

To explain the synthesis, secretion and degradation of the incretin hormone

To discuss the action of incretin hormones

To review incretin secretion and function in type 2 diabetes

## 4. METHODOLOGY

The review was carried out as a systemic review. Articles were searched from MEDLINE, HINARI and PUBMED in English language with the key words incretin, gut hormone, type 2 diabetes, and the role of incretin in type 2 diabetes. The search was including reviews, original or research articles. The search was also including abstracts presented at recent conference, relevant Websites, reference lists and citations.

This review includes studies which were done in randomized controlled trials and those which compared incretin effect with normal and type 2 diabetes as well as the incretin concentration. A case-control studies which were done in human subjects were included in this study.

Full texts of all studies meeting the inclusion criteria were retrieved and appropriate data extracted.

# 5. RESULT

## 5.1 Incretin secretion and function in type 2 diabetes

Many case-control studies have been conducted by exploring the role of incretin hormone in type 2 diabetes. A total of 65 studies were retrieved. Of these, 12 were considered to be relevant up on initial screening. Abstract of these 12 articles were reviewed and while 5 studies excluded. 7 studies were agreed upon to meet the inclusion criteria. The incretin secretion and effect have been investigated in seven published, randomized, controlled Trials with a total of 165 subjects. Among these, 88 were patients with type 2 diabetes and 77 Were normal subjects. A summary of the review were explained as shown below in table.

*Table 5.1 Summary of review articles and their results*

| Author | Root of adminstration | Healthy/ Type 2 DM | Parameters measured | Result |
|---|---|---|---|---|
| Skrha et.al 2010 | Mixed meal | 17/17 | Glucose<br>Insulin<br>GIP | • Impaired GIP secretion<br>• Low insulin secretion<br>• Delay GIP action<br>• High glucose |
| Virbikova et.al **2008** | OGTT | 13/21 | Glucose<br>Insulin<br>GIP and GLP-1<br>C-peptide | • High C-peptide, insulin, glucose and total GIP<br>• Low GLP-1 |
| Vollmer et.al **2008** | OGTT<br>Mixed meal | 14/17 | Glucose<br>Insulin<br>GIP and GLP-1<br>C-peptide<br>glucagon | • Plasma glucose, insulin ,glucagon and C-peptide were higher<br>• GIP level slightly higher<br>• Low GLP-1 level compared with healthy subjects |
| Knop.et.al **2007** | OGTT<br>IV | 8/8 | Glucose<br>Insulin<br>GIP and GLP-1<br>C-peptide<br>Glucagon<br>Incretin effect | • Incretin effect reduced (36±6%) compared with healthy (60±4%)<br>• Insulin, C-peptide, GIP and GLP-1 were greater in OGTT |

| | | | | |
|---|---|---|---|---|
| | | | | compared with IV<br>• High glucagon after OGTT |
| Vilsboll et.al **2003** | Mixed Meal | 8/8 | Glucose<br>Insulin<br>GIP and GLP-1<br>C-peptide<br>Glucagon | • Insulin. C-peptide, glucagon and glucose were higher<br>• Low GLP-1 and high GIP level |
| O'Donovan et.al **2004** | IV | 8/8 | Glucose<br>Insulin<br>GIP and GLP-1 | • High insulin, glucose, GLP-1 & GIP during infusion<br>• Low plasma GLP-1 compared with control<br>• But GIP was higher |
| **Nauck et.al 1993** | OGTT<br><br><br><br>IV* | 9/9 | Glucose<br>Insulin<br>C-peptide<br>Glucagon<br>GIP and GLP-1 | • Insulin. C-peptide, glucagon and glucose were higher<br>• Low GLP-1 and normal GIP level |

OGTT –Oral Glucose Tolerance Test, IV –Intravenous administration

*The investigation was designed to compare insulinotropic actions of exogenous incretin hormones (gastric inhibitory peptide (GIP) and glucagon-like peptide 1 [GLP-1] [7-36 amide]) in nine type-2 diabetic patients (fasting plasma glucose 7.8 mmol/liter; hemoglobin Alc, 6.3±0.6%) and in nine age- and weight-matched normal subjects. Synthetic human GIP (0.8 and 2.4 pmol/kg. min over 1 h each), GLP-1 [7-36 amide] (0.4 and 1.2 pmol/kg. min over 1 h each), and placebo were administered under hyperglycemic clamp conditions (8.75 mmol/liter) in separate experiments. Both GIP and GLP-1 [7-36 amide] dose-dependently augmented insulin secretion (insulin, C-peptide) in both groups ($P < 0.05$). With GIP, the maximum effect in type-2 diabetic patients was significantly lower (by 54%; $P < 0.05$) than in normal subjects. With GLP-1 [7-36 amide ] type-2 diabetic patients reached 71% of the increments in C-peptide of normal subjects (difference not significant). Glucagon was lowered during hyperglycemic clamps in normal subjects, but not in type-2 diabetic patients, and further by GLP-1 [7-36 amide ] in both groups($P<0.05$),but not by GIP (Nauck et.al 1993).

# 6. DISCUSSION

Postprandial glucose concentration is controlled by insulin secretion regulated both directly by the absorbed nutrients and through the secretion of incretin hormones, namely glucagon-like peptide 1 (GLP-1) and glucose-dependent insulinotropic peptide (GIP). The incretin effect contributes to the postprandial insulin release by about 50-70 % in healthy persons, whereas in type 2 diabetic patients this effect is markedly reduced. To understand the mechanism of this impairment, many studies have evaluated the secretion of GLP-1 and GIP or the response to exogenous administration of GLP-1 and GIP in type 2 diabetic patients (Gautier et al. 2008 ).
According to the review plasma glucose, insulin and C-peptide concentrations were higher in the subjects with type 2 diabetes after both the OGTT and the mixed meal, (Vrbikova et al. 2008, Volmer et al. 2008, Knop et al. 2007, Vilsbøll et al. 2003, Nauck et.al 1993), indicating insulin resistance.

According to Volmer et al. 2008 and his colleagues with 14 normal and 17 type 2 diabetes subjectes found that, plasma concentrations of GIP and GLP-1 increased after both the OGTT and the mixed meal. There was no difference in GIP levels between the groups after the OGTT and only minor differences after the mixed meal (there were slightly higher peak responses in type 2 diabetic patients compared with controls).

The six case-control studies showed that plasma concentration of GLP-1 was decreased in patients with type 2 diabetes compared with Healthy subjects (Volmer et al. 2008, Vilsbøll et al. 2003, Vrbikova et al. 2008, Nauck et.al 1993, O'donova et al. 2004, Knop et al. 2007), which may be caused either by defects in GLP-1 secretion or by increased inactivation through dipeptidyl peptidase IV (DPP IV). This mechanism is supported by the clinical evidence that the administration of GLP-1 analogues as well as DPP IV inhibitors improves insulin secretion and contributes to better glucose control in Type 2 diabetic patients.

The mechanism of the impaired secretion of GLP-1 is unknown, but several factors such as the severity of diabetes or the body mass index have been suggested to influence GLP-1 secretion. But there were a controversy on the level of GIP. In this review most of the authors showed that

there were an increase in GIP concentration in type 2 diabetic patients (Vrbikova et al. 2008, Vilsbøll et al. 2003, O'donova et al. 2004).

Skrha et al. 2010 and his colleagues which were done in a total of 17 Type 2 diabetic patients, 10 obese non-diabetic persons and 17 non-obese healthy controls which were confirmed by oral glucose tolerance test suggested that GIP level was decreased, the same result also found in Knop et al. 2007 and his colleagues. In addition, Nauck et al. 1993 and his colleagues also found that GIP responses after oral glucose tended to be lower in the type-2 diabetic patients (by 31%, P = 0.10; difference not significant). Mechanism of the increase and decrease in GIP level is still unclear.

Furthermore, Knop et al. 2007 and his colleagues, which were done in eight patients with chronic pancreatitis and normal glucose tolerance eight patients with type 2 diabetes and eight healthy subjects have shown that, in the type 2 diabetes group, the incretin effect amounted to 36 ± 6%, significantly ($P < 0.05$) lower than in chronic pancreatitis patients with NGT and in healthy subjects, respectively. These results suggest that the reduced incretin effect is not a primary event in the development of type 2 diabetes, but rather a consequence of the diabetic state.
Similarly, Nauck et al. 1993 and his colleagues, Estimated that the incretin effect in first-degree relatives of patients with type 2 diabetes and found it to be similar to that of matched healthy subjects, suggesting the deficiency to be a consequence of the diabetic state.

To evaluate the action of incretin in type 2 diabetes, the response to exogenous infusions has been studied, with significant differential results between GIP and GLP-1. According to Nauck et al. 1993 and his colleagues, investigated the effects of intravenous infusions of GLP-1 and GIP compared to placebo in type 2 diabetic patients versus healthy controls. The insulinotropic response to GIP administration was 54% lower in diabetic patients compared to normal subjects. The precise cellular mechanisms contributing to impaired GIP function in diabetes remain to be elucidated. In contrast to GIP, the insulinotropic response to GLP-1 was similar to that in controls, and its glucagonostatic activity was also preserved. This impairment is due to a defect at the receptor level induced by the diabetic state, particularly hyperglycemia. The different

responsiveness is somewhat surprising since both hormones GIP and GLP-1 are very similar (close structural homology, same family of receptors, same signal transduction mechanisms). Another four case- control studies confirmed the preservation of the effects of exogenous GLP-1 on insulin secretion, glucagon suppression and ability to decelerate gastric emptying (Volmer et al. 2008, Knop et al. 2007, Vilsbøll et al. 2003, Nauck et al. 1993).

Vilsbøll et al. *2003* and his colleagues which were done in eight patients with type 2 diabetes and eight healthy subjects after ingestion of mixed meal, Confirmed that there were an increase in glucagon concentration in patients with type 2 diabetes compared with healthy subjects. Similarly, Knop et al. 2007 and his colleagues also found that, patients with type 2 diabetes exhibited significantly higher basal levels compared with the control group (9.5 ± 0.9 vs. 7.6 ± 0.6 pmol/l, $P = 0.01$). Otherwise no significant differences between the groups were observed. In the control group equal suppression of plasma glucagon concentrations was observed on both experimental days with similar nadirs of 5.4 ± 0.4 and 5.3 ± 0.5 pmol/l. Volmer et al. 2008 and his colleagues also agreed that Glucagon levels were also higher. This is may be due to hyperglucagonemia has been associated with lower GLP-1 concentrations. It is possible that there is a direct or indirect feedback relationship between glucagon and GLP-1 secretion, as GLP-1 indirectly inhibits glucagon secretion.

In addition, Knop et al. 2007 and his colleagues recently shown that the suppression of glucagon secretion is impaired during oral glucose tolerance tests (OGTTs) as opposed to isoglycemic intravenous glucose infusion in patients with type 2 diabetes. This suggests that the inflammatory condition in the pancreas blunts the glucagon suppression during the OGTT independently of normal insulin secretion and normal glucose homeostasis. Responses and concentration of GLP-1 and GIP were significantly greater during oral glucose compared with isoglycemic intravenous glucose in type 2 diabetes.

It was therefore possible that ingestion of a small meal with a relatively higher proximal absorption than after a large meal would result in a relatively lower secretion of the distal incretin hormone, GLP-1 (Vilsbøll et al. 2003). The increased incretin response to the larger

meal is probably best explained by the increased exposure of the incretin hormone producing endocrine K and L-cells of the intestinal mucosa to nutrients (Vilsbøll 2004).

The underlying cause for the diverging properties of GIP and GLP-1 regarding the altered incretin effect in type 2 diabetes is not completely understood. The promising therapeutic potential of GLP-1 as a pharmacological tool for treating type 2 diabetes was proposed in the 1990s, along with the further characterization of the incretin effect. In contrast to other insulinotropic agents, e.g. the sulfonylureas, the insulinotropic effect of GLP-1 depends even more closely on the actual glucose concentration providing the possibility of glucose normalization without the risk of hypoglycemias. As well as the glucose lowering effect via the stimulation of insulin secretion, GLP-1 has a variety of additional physiological effects that may be advantageous in type 2 diabetes therapy (Gallwitz 2005).

## 7. CONCLUSION

The incretin hormones are released during meals from gut endocrine cells. They potentiate glucose induced insulin secretion and may be responsible for up to 70% of postprandial insulin secretion. The incretin hormones include glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP), both of which may also promote proliferation/neogenesis of beta cells and prevent their decay (apoptosis). Both hormones contribute to insulin secretion from the beginning of a meal and their effects are progressively amplified as plasma glucose concentrations rise.

This review suggest that in patients with type 2 diabetes: 1) the secretion of GLP-1 is impaired while the secretion of GIP is unaffected; 2) GIP administration has very little insulinotropic activity whereas GLP-1 maintains its ability to stimulate glucose-dependent insulin secretion and to inhibit glucagon secretion as well. Interestingly, glucagon secretion was differentially regulated during oral glucose and isoglycemic intravenous glucose infusion.
Reduced incretin effect in type 2 diabetes is a consequence of the diabetic state rather than a primary event leading to type 2 diabetes.

## 8. RECOMMENDATION

The current therapeutic treatments for type 2 diabetes have had adverse effect such as weight gain and sever hypoglycemia. Such effects are now solved by synthesizing incretin based drug. But it was unfinished so it needs further study based on this hormone.

Despite data suggesting that the decreased incretin effect in type 2 diabetes may be a consequence rather than a cause of the diabetic state , the implication of impaired incretion function in the etiology of type 2 diabetes has been very recently a subject of debate. However, understanding the precise mechanisms governing this phenomenon requires further investigations.

GIP concentrations have been found to be slightly lower, normal or even increased in type 2 diabetic patients, but the insulinotropic response is markedly decreased, though for reasons that are not clear.

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