

Biological Actions and Therapeutic Potential of the Glucagon-like Peptides

DANIEL J. DRUCKER

The Banting and Best Diabetes Centre, Department of Medicine, Toronto General Hospital, University of Toronto, Toronto, Ontario, Canada

The glucagon-like peptides (GLP-1 and GLP-2) are proglucagon-derived peptides cosecreted from gut endocrine cells in response to nutrient ingestion. GLP-1 acts as an incretin to lower blood glucose via stimulation of insulin secretion from islet β cells. GLP-1 also exerts actions independent of insulin secretion, including inhibition of gastric emptying and acid secretion, reduction in food ingestion and glucagon secretion, and stimulation of β -cell proliferation. Administration of GLP-1 lowers blood glucose and reduces food intake in human subjects with type 2 diabetes. GLP-2 promotes nutrient absorption via expansion of the mucosal epithelium by stimulation of crypt cell proliferation and inhibition of apoptosis in the small intestine. GLP-2 also reduces epithelial permeability, and decreases meal-stimulated gastric acid secretion and gastrointestinal motility. Administration of GLP-2 in the setting of experimental intestinal injury is associated with reduced epithelial damage, decreased bacterial infection, and decreased mortality or gut injury in rodents with chemically induced enteritis, vascular-ischemia reperfusion injury, and dextran sulfate-induced colitis. GLP-2 also attenuates chemotherapy-induced mucositis via inhibition of drug-induced apoptosis in the small and large bowel. GLP-2 improves intestinal adaptation and nutrient absorption in rats after major small bowel resection, and in humans with short bowel syndrome. The actions of GLP-2 are mediated by a distinct GLP-2 receptor expressed on subsets of enteric nerves and enteroendocrine cells in the stomach and small and large intestine. The beneficial actions of GLP-1 and GLP-2 in preclinical and clinical studies of diabetes and intestinal disease, respectively, has fostered interest in the potential therapeutic use of these gut peptides. Nevertheless, the actions of the glucagon-like peptides are limited in duration by enzymatic inactivation via cleavage at the N-terminal penultimate alanine by dipeptidyl peptidase IV (DP IV). Hence, inhibitors of DP IV activity, or DP IV-resistant glucagon-like peptide analogues, may be alternative therapeutic approaches for treatment of human diseases.

Glucagon, a 29 amino acid peptide hormone, was first isolated from pancreatic extracts as a peptide hormone with hyperglycemic activity. Larger molecular

forms with glucagon-related immunoreactivity were subsequently identified in intestinal extracts; however, the relationship between gut-derived glucagon-like immunoreactivity (GLI) and pancreatic glucagon remained unclear for some time. Initial studies estimated the size of intestinal "enteroglucagon" as \sim 100 amino acids, prompting the designation glicentin. After the cloning of pancreatic, intestinal, and brain complementary DNAs encoding proglucagon, the structural relationship between the various glucagon-related peptides was clearly delineated.¹⁻⁶ Pancreatic glucagon is contained within the sequence of 69 amino acid glicentin (Figures 1 and 2). Glucagon together with an 8 amino acid carboxyterminal peptide comprises oxyntomodulin.⁷ After a short intervening peptide, 2 glucagon-like peptides, designated GLP-1 and GLP-2, are located carboxyterminally in the proglucagon molecule separated from each other by a second intervening peptide.

Fish, reptiles, and birds may contain multiple genes for proglucagon.⁸ However, only a single proglucagon gene has been identified in mammals. The nucleotide sequence of mammalian proglucagon messenger RNA (mRNA) transcripts and proglucagon gene transcription start sites appear identical in fetal and adult pancreas, intestine, and brain.^{5,6,9-11} Hence, tissue-specific post-translational processing of proglucagon accounts for the diversity in the profile of secreted proglucagon-derived peptides (PGDPs) in the pancreas and gut (Figure 1). In contrast, alternative RNA splicing generates unique proglucagon mRNA transcripts in the pancreas and intestine of fish, chicken, and reptiles.^{12,13} Although the prohormone convertase (PC) enzymes that liberate PGDPs in the central nervous system have yet to be con-

Abbreviations used in this paper: DP IV, dipeptidyl peptidase IV; DS, dextran sulfate; GIP, glucose-dependent insulinotropic polypeptide; GLP, glucagon-like peptides; ICV, intracerebroventricular; PC, prohormone convertase; PGDPs, proglucagon-derived peptides; PYY, peptide YY.

© 2002 by the American Gastroenterological Association
0016-5085/02/\$35.00
doi:10.1053/gast.2002.31068

clusively identified, PC2 is thought to play a key role in the liberation of pancreatic glucagons,^{14–16} whereas PC1/3 is important for generation of the glucagon-like peptides in enteroendocrine cells.^{17–20}

Enteroendocrine L cells producing GLP-1 and GLP-2 are detected throughout the length of the small and large bowel, with the majority of L cells localized to the distal ileum and colon. L cell subpopulations have been characterized on the basis of hormonal phenotype, with some cells producing principally PGDPs, or PGDPs in combination with other gut peptides such as peptide YY (PYY), cholecystokinin, or neurotensin. The distal location of the majority of PGDP-producing endocrine cells is somewhat paradoxical in view of the rapid increase in circulating PGDPs detectable within minutes of nutrient ingestion in both rodents and humans.^{21–24} Current concepts of L cell regulation involve the integration of humoral and neural mediators such as gastrin-releasing peptide and glucose-dependent insulinotropic polypeptide (GIP) and neural inputs in the control of intestinal PGDP secretion. Peptide administration and immunoneutralization experiments support a role for somatostatin-28 as an inhibitor of both GLP-1 and GLP-2 secretion.²⁵ The importance of neural signaling for secretion of PGDPs is illustrated by studies of vagal nerve interruption that result in marked attenuation of nutrient-stimulated PGDP secretion in the rat.²⁶ Nutrients also up-regulate proglucagon mRNA transcripts in the rodent jejunum,²⁷ and fermentable fiber feeding or fatty acid infusion is associated with increased intestinal proglucagon gene expression in the rat intestine.^{28–31}

The initial increment in levels of circulating GLP-1 and GLP-2 following meal ingestion is followed by rapid inactivation and clearance of these peptides from the circulation. Both GLP-1 and GLP-2 contain an alanine at position 2, rendering them substrates for the exopeptidase dipeptidyl peptidase IV (DP IV). Bioactive GLP-1 exists in two equipotent molecular forms (Figure 2), GLP-1^{7–37} and GLP-1^{7–36amide}.³² GLP-1 is rapidly cleaved by DP IV,³³ resulting in the generation of the largely

inactive molecules GLP-1^{9–36amide} and GLP-1^{9–37}.^{34,35} The majority of GLP-1 leaving the intestinal venous circulation has already been cleaved by DP IV expressed in capillaries surrounding gut L cells,³⁶ leading to an estimated half-life for intact GLP-1 of ~1–2 minutes *in vivo*.³⁵ GLP-2 is also inactivated by DP IV in rodents and humans^{37–39}; however, native GLP-2 exhibits a slightly longer half-life of 5–7 minutes in human subjects.⁴⁰ GLP-1 and GLP-2 are also cleared nonenzymatically, and several organs, including the kidney, have been invoked as important sites for PGDP clearance *in vivo*.^{41–43}

GLP-1 Actions in the Gastrointestinal Tract and Pancreas

The actions of GLP-1 are mediated by a single GLP-1 receptor widely expressed in the kidney, heart, central nervous system, gastrointestinal tract, and in the endocrine pancreas.^{44–46} The GLP-1 receptor has been localized to human chromosome 6p21⁴⁷; however, significant linkage to diseases such as diabetes has not yet been detected.⁴⁸ GLP-1 infusion inhibits sham feeding-induced acid secretion in normal human subjects,^{49–51} and these actions on acid secretion are dependent on both somatostatin receptor activation and intact gastric vagal innervation.^{52,53} GLP-1 receptor mRNA transcripts and GLP-1 binding have also been observed in purified rat and rabbit gastric parietal cell populations.^{54,55} In contrast, GLP-1 inhibits human gastric lipase secretion in a vagal nerve-independent manner.⁵⁶ Exogenous GLP-1 administration potentially inhibits gastric emptying in rodent and human studies.⁴⁹ Intracerebroventricular (ICV) administration of GLP-1 also inhibits gastric emptying in rodents which is abolished by vagal afferent denervation, but not by cholinergic or adrenergic blockade.⁵⁷ Adrenergic blockade eliminates the inhibitory actions of GLP-1 on antral, duodenal, and jejunal motility in the rat⁵⁸ and splanchnic nerve transection abrogates the inhibitory effects of GLP-1, mediated by the vagus nerve

The Proglucagon-Derived Peptides

RSLQDTEEKSRFSASQADPLSDPDQMNEDKRHSQGTFTSDYSKYLD	SRRQDFVQWLMNTRKRRNNIA	Glicentin
HSQGTFTSDYSKYLD	SRRQDFVQWLMNTRKRRNNIA	Oxyntomodulin
HSQGTFTSDYSKYLD	SRRAQDFVQWLMNT	Glucagon
HAEGTFTSDVSSYLEGQAAKEFIAWLVKGRG		GLP-1(7-37)
HAEGTFTSDVSSYLEGQAAKEFIAWLVKGR		GLP-1(7-36) ^{amide}
HGEGTFTSDLKQMEEEAVRLFIEWLKNGGPSSGAPPS		Exendin-4
HADGSFSDEMNTILDNLAARDFINWLIQTKITD		GLP-2
	RNRNNIA	IP-1
	DFPEEVAIVEELG	IP-2

Figure 2. Amino acid sequence alignment of the human proglucagon-derived peptides and lizard exendin-4.

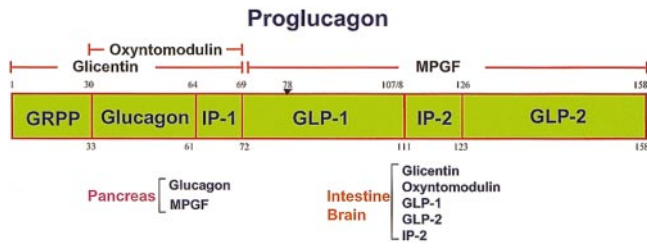


Figure 1. Structural organization of proglucagon and the proglucagon-derived peptides. MPGF, major proglucagon-derived fragment. IP, intervening peptide. The specific peptides liberated by posttranslational processing in pancreas vs. intestine are indicated below the proglucagon molecule. The numerals above and below the proglucagon structure denote the relative amino acid positions of the PGDPs within proglucagon. The *arrowhead* indicates the position of GLP-1 cleavage to generate GLP-1^{7-36amide} and GLP-1⁷⁻³⁷ from GLP-1¹⁻³⁷.

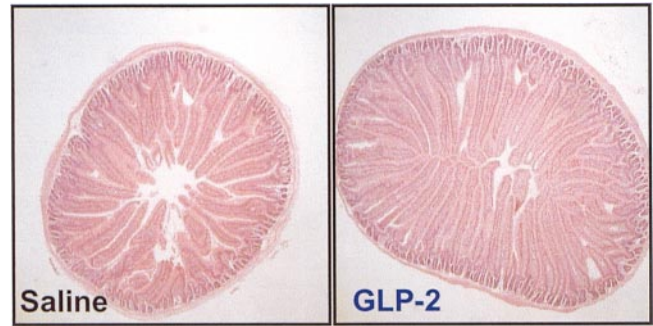


Figure 3. Histological photomicrograph of a H&E-stained cross-section of the murine jejunum following treatment with either saline or the human GLP-2 analogue h[Gly2]-GLP-2 for 6 days as described in Boushey et al.¹⁵⁴ Original magnification 40×.

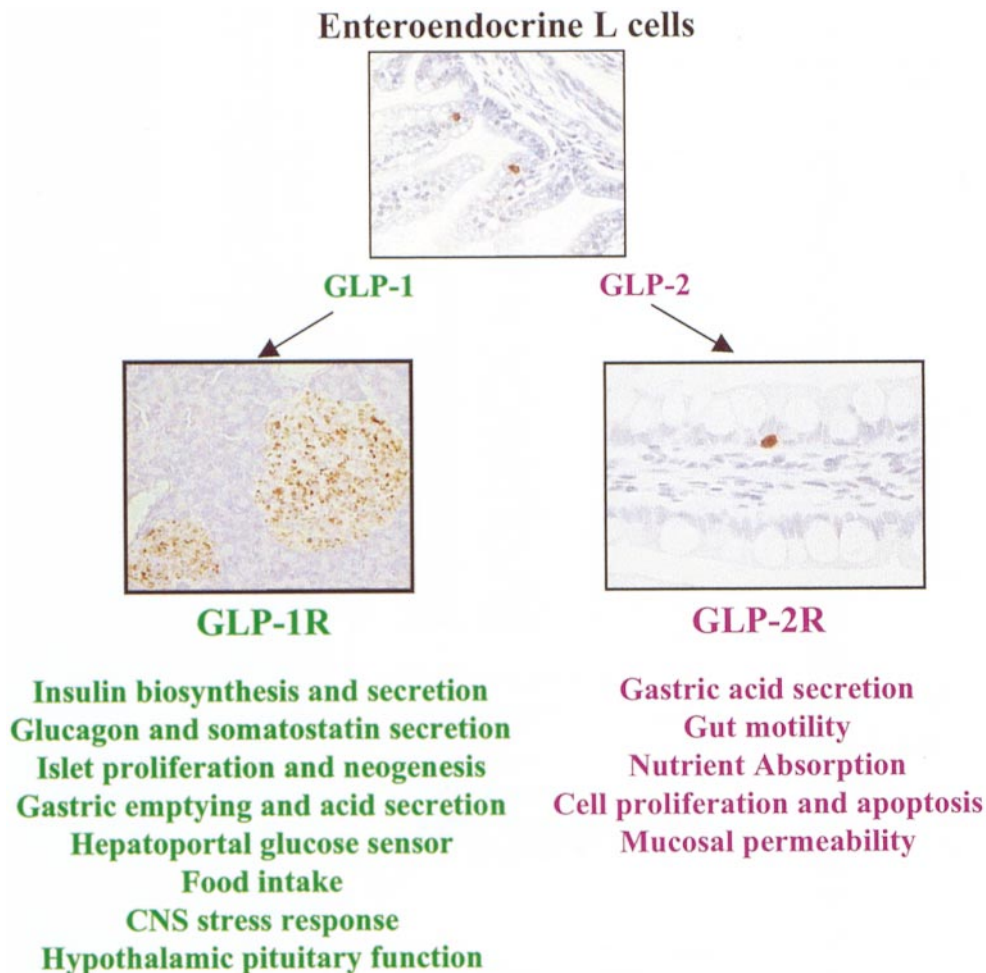


Figure 4. Representation of glucagon-like peptide action in the gastrointestinal tract. GLP-1 and GLP-2 are cosecreted from gut endocrine L cells and act in part via activation of distinct receptors expressed on distal endocrine cells (GLP-1 and GLP-2) or enteric neural populations (GLP-2) in the islets or gut.

and parasympathetic outflow, on acid secretion and gastric motility in the pig.⁵⁹ GLP-1 also inhibits small bowel motility in the rat and these actions of GLP-1 are blocked by the nitric oxide synthase inhibitor *N*^ω-nitro-L-arginine in the fasted but not fed state.⁶⁰ Taken together, the antimotility and antisecretory effects of GLP-1 suggest that GLP-1 released from the distal gut represents a candidate mediator of the ileal brake phenomenon in vivo. The actions of GLP-1 on gastric emptying result in a reduced rate of nutrient transit into the small bowel, leading to decreased glycemic excursion after meal ingestion.^{61,62} The effect of exogenous GLP-1 on gastric emptying is observed with levels of plasma GLP-1 within the physiological range of endogenous plasma levels that might be achieved after meal ingestion.⁶³

Original descriptions of GLP-1 action focused on its role as a gut-derived incretin that stimulated glucose-dependent insulin secretion.^{22,64–66} GLP-1 acts as a physiological regulator of insulin secretion as the truncated lizard peptide exendin (9-39), a functional GLP-1 receptor antagonist, reduces insulin secretion and enhances glycemic excursion following blockade of GLP-1 action in vivo.^{67–69} Similarly, mice with genetic disruption of GLP-1 receptor signaling exhibit glucose intolerance and reduced glucose-stimulated insulin secretion.⁷⁰ Although GLP-1 stimulates pancreatic somatostatin secretion and inhibits glucagon secretion,⁷¹ GLP-1R^{-/-} mice exhibit normal levels of glucagon in the fasting state and preserved suppression of plasma glucagon following oral glucose challenge.⁷²

Increasing evidence supports a role for GLP-1R signaling in the regulation of β -cell proliferation and islet neogenesis. GLP-1 promotes β -cell replication in the mouse,⁷³ and directly stimulates DNA synthesis in islet cell lines in vitro via a phosphatidylinositol 3-kinase-dependent pathway.⁷⁴ GLP-1 also promotes development of a more differentiated β -cell phenotype in islet, ductal, and exocrine cells via a pdx-1-dependent pathway.^{75–77} GLP-1 or the lizard GLP-1 agonist exendin-4 stimulate β -cell neogenesis in diabetic db/db mice in association with increased ductal pdx-1 expression, likely attributable to stimulation of pdx-1 promoter activity in both small and large pancreatic ducts in vivo.⁷⁸ Similarly, daily administration of exendin-4 to neonatal rats for 10 days after partial pancreatectomy lowered blood glucose, stimulated islet regeneration, and increased β -cell mass⁷⁹ and administration of GLP-1 or exendin-4 to neonatal rats treated with streptozotocin resulted in increased β -cell mass, which remained detectable even at 2 months

of age.⁸⁰ Conversely, elimination of GLP-1 receptor signaling is associated with reduced numbers of large islets and abnormal topographical localization of islet A cells in GLP-1R^{-/-} mice.⁸¹ Hence, these findings have fostered considerable interest in harnessing the trophic properties of GLP-1 for the treatment of type 2 diabetes characterized by progressive β cell failure.

GLP-1, Appetite, and the Central Nervous System

The finding that ICV GLP-1 administration dose-dependently inhibits food intake in rats and mice,^{70,82–84} in association with experimental data demonstrating increased food intake and weight gain following chronic ICV administration of the GLP-1 receptor antagonist exendin (9-39),⁸⁵ has engendered considerable interest in the anorexic properties of GLP-1 agonists. Subsequent studies demonstrated that peripheral administration of GLP-1 or exendin-4 to rodents also reduced short-term and long-term food intake,⁸⁶ raising the possibility that therapeutic administration of GLP-1 may prevent weight gain and perhaps induce weight loss in human subjects. Although long-term human studies of GLP-1 treatment are not yet available, GLP-1 and exendin-4 clearly reduce appetite and food consumption following peripheral administration in normal or diabetic human subjects.^{87–90}

The mechanism(s) used by GLP-1 agonists for transduction of anorectic signals remain unclear. Inhibition of gastric emptying may account for a component of the satiety experienced after GLP-1 administration. GLP-1 receptors are expressed on hypothalamic nuclei that control food intake,^{82,91,92} and peripheral GLP-1 may exert indirect effects on CNS satiety centers through neuronal relay mechanisms. Intriguingly, both GLP-1 agonists and noxious agents such as lithium chloride or lipopolysaccharide induce a similar pattern of central *c-fos* activation in the CNS and an overlapping set of aversive behavioral responses in rodents in vivo.^{93–95} Moreover, some aversive responses to noxious stimuli are blocked by the coadministration of the GLP-1 antagonist exendin (9-39),⁹⁶ suggesting the involvement of GLP-1R signaling pathways in the response to aversive stress. Whether GLP-1 signaling pathways are essential for physiological control of appetite and body weight remains unclear, as GLP-1R^{-/-} mice exhibit normal food intake and weight gain⁷⁰ and are resistant to the development of obesity, even after 6 months of high-fat feeding.⁹⁷ Taken together, the inhibitory effects of GLP-1 agonists on food intake may prove to be highly useful if sustained in

long-term treatment studies of human subjects with type 2 diabetes.

GLP-1 Agonists and Therapy of Type 2 Diabetes

The actions of GLP-1 including inhibition of gastric emptying, stimulation and inhibition of insulin and glucagon secretion, respectively, expansion of β -cell mass and reduction of food intake and weight gain, represent ideal properties for an agent designed for the treatment of type 2 diabetes.⁹⁸ Furthermore, GLP-1 actions are highly glucose-dependent, hence excess GLP-1 administration is unlikely to be associated with hypoglycemia *in vivo*. A principal obstacle to the long-term use of the native GLP-1 molecule in the clinic is the rapid inactivation of the peptide by the enzyme DP IV.^{34,35} As DP IV inhibition prolongs the duration of GLP-1 action, the development of DP IV inhibitors is being actively pursued as a potential strategy for the treatment of diabetes.⁹⁹ Although DP IV, also known as CD26, cleaves a large number of regulatory peptides and chemokines,^{100,101} mice and rats with inactivating DP IV mutations are viable and exhibit enhanced GLP-1 action and glucose clearance,^{102,103} attesting to the essential role of this enzyme for glucoregulation *in vivo*.

These findings have spurred efforts toward development of DP IV-resistant GLP-1 analogues or related molecules such as lizard exendin-4 that exert longer durations of efficacy in human subjects.^{90,104–106} GLP-1 infusion normalizes 24-hour blood glucose profiles in subjects with type 2 diabetes,^{89,107–109} even in patients with previous sulfonylurea failure.^{110–112} The majority of these studies have been carried out for days to weeks; hence, whether treatment with GLP-1 agonists will achieve sustained control of blood glucose in long-term studies is currently under examination. Furthermore, the current lack of oral GLP-1 agonist molecules seems likely to diminish the potential for initial widespread adoption of GLP-1 therapy as a mainstream treatment for patients with type 2 diabetes well managed on oral agents alone. Nevertheless, as many patients with type 2 diabetes develop progressive β -cell failure on oral agents, the potential for GLP-1 to preserve or augment β -cell function in such patients merits careful examination.

Glucagon-like Peptide-2

GLP-2 is a 33 amino acid peptide cosecreted with GLP-1 from enteroendocrine L cells in the small and large intestine. The biological role of GLP-2 remained obscure until 1996, when GLP-2 was shown to be a

potent stimulator of mucosal epithelial proliferation in the murine small intestine.¹¹³ Studies linking proglucagon-derived peptides to intestinal growth and adaptation were fostered by the description of a patient with a glucagonoma who presented with small intestinal mucosal hyperplasia and reduced bowel motility.¹¹⁴ Tumor resection led to resolution of the small bowel villus hyperplasia. Subsequent studies in rodents correlated increased circulating levels of the PGDPs with the adaptive response to experimental intestinal injury.^{115–118} Furthermore, intestinal resection is associated with enhanced circulating levels of the PGDPs and increased levels of intestinal proglucagon mRNA transcripts in remnant intestinal segments.^{119–122} The correlation between intestinal injury, gut adaptation, and up-regulation of circulating PGDPs, demonstrated by Bloom et al.,¹¹⁸ is not restricted to rodents, as a broad variety of intestinal diseases that affect the integrity of the mucosal epithelium are associated with increased circulating levels of the PGDPs in human subjects.^{123–126} Additional reports describing glucagonoma patients with intestinal mucosal hyperplasia,^{127–129} taken together with studies linking experimental glucagonomas in mice with small bowel growth, led to definitive identification of GLP-2 as the PGDP with significant intestinotrophic properties *in vivo*.¹¹³ Although glicentin also exhibits modest intestinotrophic properties^{113,130} it is much less active as a small bowel growth factor when compared directly with GLP-2,^{113,131} and a separate receptor for glicentin has not yet been identified.

Glucagon-like Peptide-2: Synthesis and Secretion

The sequence of GLP-2 is highly conserved in vertebrates with rat and mouse GLP-2 sequences differing from human GLP-2 by a single amino acid.⁸ GLP-2, like GLP-1, contains an alanine at position 2 (Figure 2), rendering it a substrate for DP IV cleavage. Analysis of the circulating forms of GLP-2 in rats and humans demonstrates the presence of GLP-2^{1–33} and the N-terminally inactivated peptide GLP-2^{3–33}.^{37,39,40,132,133} Consistent with the importance of DP IV for degradation of bioactive GLP-2, the wild-type GLP-2^{1–33} molecule is considerably less potent than DP IV-resistant GLP-2 analogues in rat studies *in vivo*.^{38,134} In contrast to the comparatively shorter half-life of GLP-1, the disappearance half-life of exogenously administered GLP-2 is ~7 minutes in normal human subjects.⁴⁰ Furthermore, ~69% of GLP-2^{1–33} remains intact 1 hour following subcutaneous administration of 400 μ g of synthetic GLP-2

in normal human subjects.⁴⁰ The clearance of GLP-2¹⁻³³ is significantly reduced in nephrectomized rats⁴³ and levels of plasma GLP-2 are increased in human subjects with kidney failure,³⁷ in keeping with an important role for the kidney in GLP-2 clearance.⁴¹ Radioimmunoassays that only recognize mid-molecule or carboxyterminal epitopes in the GLP-2 sequence will not distinguish between intact GLP-2¹⁻³³ and partially degraded forms of the molecule and will yield overestimations of the amount of biologically active circulating GLP-2¹⁻³³.^{37,39,40}

Nutrient ingestion constitutes the principal stimulus for GLP-2 secretion.^{37,39,40,133} The levels of GLP-2 increase within minutes of nutrient ingestion.^{37,39,133} In pigs, provision of at least 40% of total nutrient intake via the enteral route is required for stimulation of significant increases in circulating GLP-2¹³⁵ whereas ingestion of a donut and coffee (~220 calories) is sufficient to significantly increase circulating GLP-2 in normal human subjects.³⁷ An intact colon in continuity with the small bowel is an important determinant of the circulating levels of GLP-2, as subjects with short bowel syndrome without a colon in continuity exhibit reduced circulating levels of meal-stimulated GLP-2.¹³⁶ In contrast, patients with less than 140 cm of remnant small bowel but with an intact continuous colon had elevated levels of GLP-2.¹³⁷ Circulating levels of GLP-2¹⁻³³ were increased in patients with active Crohn's disease or ulcerative colitis, in association with a relative increase in the ratio of intact GLP-2¹⁻³³ vs the N-terminally cleaved GLP-2³⁻³³, and a decrease in levels of plasma DP IV activity.¹³² Intriguingly, rodents with diabetes develop small bowel villous hyperplasia that may be explained in part by increased levels of circulating GLP-2 that correlate with the extent of small bowel growth.^{138,139} Treatment of diabetic rats with insulin lowers levels of circulating GLP-2 and reverses the intestinal mucosal hyperplasia.¹³⁸

GLP-2 Action in the Gastrointestinal Tract

The principal histological finding in the rodent gut after repeated GLP-2 administration is mucosal growth in the small bowel due to stimulation of cell proliferation in the crypt compartment and inhibition of enterocyte apoptosis,^{113,131,140-142} principally evident histologically as elongation of the villous epithelium (Figure 3). A modest expansion of the crypt compartment may be seen in some, but not all, studies after GLP-2 administration. The GLP-2-treated murine small bowel exhibits normal levels of digestive enzymes and normal to enhanced nutrient absorption following nutri-

ent tolerance testing.¹⁴³ Intravenous GLP-2 infusion also significantly increased the absorption of ¹⁴C-galactose and ¹⁴C-glycine from perfused intestinal segments of rats following 14 days of intravenous human GLP-2 infusion.¹⁴⁴ Ultrastructural analysis of the GLP-2-treated murine small bowel reveals longer and narrower enterocytes with longer microvilli.¹⁴⁵ Remarkably, GLP-2 enhances epithelial barrier function in the murine small bowel via reduction of tissue conductance and macromolecule flux, effects noted within 4 hours of a single GLP-2 injection *in vivo*.¹⁴⁵ Although the small bowel is significantly more sensitive to the stimulatory effects of GLP-2 compared with the colon,^{131,140,146} treatment with higher doses of GLP-2 or more potent GLP-2 agonists also produces modest increments in large bowel mucosal thickness.^{131,134} The most rapid actions of GLP-2 include enhancement of mucosal hexose transport,^{147,148} inhibition of gastric acid secretion,¹⁴⁹ and reduction of gastric motility.¹⁵⁰ In contrast to the actions of GLP-1, GLP-2 has no effect on regulation of glucose homeostasis *in vivo*.^{143,150} The minimal and optimal GLP-2 administration regimen for induction of bowel growth remains unclear; however, a single injection of GLP-2 every other day over a 10-day treatment period is sufficient to promote significant increases in small bowel growth in mice.¹⁴⁰

GLP-2 and Intestinal Injury

The mucosal atrophy observed in the intestine of parenterally fed rodents may be attributable in part to reduced circulating levels of GLP-2 in the absence of periodic nutrient stimulation of GLP-2 secretion. Intravenous infusion of GLP-2 together with parenteral nutrition prevented mucosal hypoplasia in the small bowel, but not in the large bowel of fasted rats.^{131,151} The trophic effects of exogenous GLP-2 on the rat small bowel mucosa were also preserved in parenterally fed tumor-bearing rats.¹⁵² Furthermore, GLP-2 treatment increased small bowel mass and DNA content but had no effect on the colon or on tumor growth.¹⁵² As the circulating levels of PGDPs and GLP-2 are increased in rats after small bowel resection,¹⁴¹ the effects of GLP-2 administration on small bowel adaptation have been examined in a rat model of jejunoileal resection. Rats treated for 21 days with twice daily injections of a GLP-2 analogue exhibited no differences in food intake, body weight, or small bowel length, but significant increases in mucosal weights were observed in the jejunum and ileum of GLP-2-treated rats following resection.¹⁵³ GLP-2 treatment also increased crypt plus villus height,

jejunal sucrase activity, and fractional urinary xylose excretion, the latter parameter consistent with a GLP-2-mediated improvement in intestinal sugar absorption.¹⁵³

The reparative and protective properties of GLP-2 have been assessed in the setting of experimental intestinal inflammation in the small and large bowel. Mice treated with the nonsteroidal agent indomethacin develop small bowel enteritis associated with significant mortality evident between 48–72 hours after indomethacin administration.¹⁵⁴ Treatment with a DP IV-resistant GLP-2 analogue, h[Gly²]-GLP-2, either before, concomitant with, or following indomethacin significantly reduced mortality and decreased mucosal injury.¹⁵⁴ The protective effects of h[Gly²]-GLP-2 were associated with significantly increased crypt cell proliferation and decreased crypt compartment apoptosis in the small bowel epithelium of indomethacin-treated mice. A marked reduction in myeloperoxidase activity was detected in the jejunum and ileum, in association with reduced levels of TNF- α , interleukin-2, interferon- γ , and interleukin-10 in h[Gly²]-GLP-2-treated mice.¹⁵⁴ The reduced mortality may potentially be explained by a significant reduction in the number of septic animals exhibiting bacterial culture positivity in viscera and blood following h[Gly²]-GLP-2 administration. Consistent with these findings, h[Gly²]-GLP-2 reduced intestinal permeability and the extent of bacterial translocation in rats with experimental pancreatitis.¹⁵⁵

The therapeutic effects of GLP-2 have also been examined in HLA-B27 rats after development of spontaneous small bowel inflammation. A 14-day infusion of GLP-2, 50 $\mu\text{g} \cdot \text{kg} \cdot \text{day}$, reduced mucosal damage scores in the small and large intestine, with a marked reduction in colonic expression of TNF- α and interferon- γ mRNA transcripts.¹⁵⁶ Administration of a GLP-2 analogue preserved mucosal mass and reduced mortality, with accelerated recovery of mucosal absorption of galactose and glycine after ischemia/reperfusion injury in rats.^{157,158} Reduced intestinal injury following GLP-2 treatment has also been observed in mice with dextran sulfate-induced colitis. Concomitant administration of subcutaneous h[Gly²]-GLP-2 and oral dextran sulfate (DS) for 10 days resulted in markedly reduced colonic damage and decreased weight loss in CD1 and BALB/C mice.¹⁵⁹ h[Gly²]-GLP-2 preserved large bowel length, decreased intestinal damage scores, reduced interleukin-1 expression, and stimulated colonic mucosal repair via enhanced crypt cell proliferation in mice with DS-colitis.¹⁵⁹

The observations that GLP-2 treatment reduced intestinal apoptosis¹⁵⁴ prompted assessment of the cytopro-

TECTIVE effects of GLP-2 administration in the setting of chemotherapy-induced intestinal mucositis. Rats treated with a GLP-2 analogue and 5'-fluorouracil exhibited reduced intestinal injury compared with rats treated with 5'-fluorouracil alone.¹⁶⁰ Pretreatment of mice with h[Gly²]-GLP-2 before administration of the topoisomerase inhibitor irinotecan significantly reduced bacterial infection, intestinal damage, and mortality.¹⁶¹ Histological and biochemical analyses revealed significant reductions in crypt compartment apoptosis and reduced intestinal caspase-8 activation following h[Gly²]-GLP-2-treatment, compared with mice receiving irinotecan alone.¹⁶¹ h[Gly²]-GLP-2 also reduced mortality after 5-fluorouracil treatment of BDF1 mice, and repeated cyclical administration of both h[Gly²]-GLP-2 and irinotecan resulted in significantly decreased mortality in groups of BALB/C mice implanted with subcutaneous CT-26 colon carcinomas, compared with identical groups of tumor-bearing mice treated with irinotecan alone.¹⁶¹ The antiapoptotic actions of GLP-2 have also been observed in premature parenterally fed pigs. Intravenous infusion of GLP-2 for 6 days reduced protein degradation and significantly decreased the rate of apoptosis in the jejunum of premature pigs.¹⁴² Taken together, exogenous GLP-2 administration clearly exerts protective and regenerative actions in the small and large bowel in a diverse number of animal models of experimental epithelial injury.

Identifying Mechanisms Mediating GLP-2 Action

GLP-2 exerts its actions through a recently identified G protein-coupled receptor isolated from hypothalamic and intestinal cDNA libraries. The GLP-2 receptor (GLP-2R) is comprised of 550 amino acids and was localized to human chromosome 17p13.3.¹⁶² The GLP-2R is expressed in a tissue-specific manner in the stomach, small and large intestine, central nervous system, and lung.^{162–164} GLP-2R expression in the human gut epithelium has been localized to subsets of enteroendocrine cells in the stomach, and both small and large intestine.¹⁶³ Although the majority of enteroendocrine cells do not express the GLP-2R, all GLP-2R-immunoreactive cells identified to date express one or more gut endocrine markers, including GIP, PYY, serotonin, chromogranin, and GLP-1.¹⁶³ In contrast, the GLP-2R has been localized to enteric neurons in the murine gastrointestinal tract.¹⁶⁵ These findings imply a model for GLP-2 action whereby GLP-2 released from enteroendocrine L cells acts on adjacent and distant gut endocrine cells or neurons to stimulate the release of downstream mediators of GLP-2 action (Figure 4).

GLP-2R signaling has not yet been examined in cells that express an endogenous GLP-2R. GLP-2 activates an adenosine 3',5'-cyclic monophosphate (cAMP)-dependent signaling pathway in fibroblasts transfected with the rat or human GLP-2R.^{162,166} In contrast, GLP-2 did not stimulate a significant increase in intracellular calcium accumulation and only pharmacological concentrations (100 nmol–10 μ mol) of GLP-2 increased cell proliferation in quiescent BHK-GLP-2R and Caco-2 cells.^{166,167} Activation of GLP-2R signaling in heterologous transfected cells inhibits apoptosis induced by cycloheximide or irinotecan, in association with reduced caspase activation, decreased mitochondrial cytochrome C release, and decreased cleavage of downstream effector enzymes such as poly ADP ribose polymerase.^{161,168} Hence, GLP-2 promotes cell survival via both direct and indirect actions in vitro and in vivo.

GLP-2 and Treatment of Human Intestinal Disease

Although GLP-2 exerts beneficial effects in experimental models of intestinal injury, experience with GLP-2 in the setting of human disease is limited. Treatment of 8 patients with short bowel syndrome and energy malabsorption with twice daily subcutaneous injections of native human GLP-2 for 35 days resulted in modest but statistically significant improvements in nutrient absorption as assessed by metabolic balance studies carried out before and after completion of GLP-2 therapy.¹⁶⁹ GLP-2 improved intestinal energy absorption, diminished stomal energy excretion, increased body weight and lean body mass, decreased fat mass, and enhanced urinary creatinine excretion, in association with reduced gastric emptying but normal small bowel transit times.¹⁶⁹ Evidence for increased growth of the small bowel mucosa was detected in biopsy specimens from 5 of 6 subjects. Mean fasting plasma levels of circulating GLP-2 increased from 11 ± 5 to 719 ± 281 pmol, 30 minutes after subcutaneous injection of 400 μ g of GLP-2. Compliance during the study was good, with 6 of 8 patients receiving all 70 subcutaneous injections, and 2 patients missed a single injection. There was no significant change in a broad panel of biochemical and hematological parameters assessed before and after GLP-2 treatment.¹⁶⁹ Similarly, positive results including increased wet weight absorption, decreased fecal wet weight and energy excretion, increased body weight, and enhanced fat absorption have been reported in a preliminary analysis of human short bowel patients treated with ALX-0600, a degradation-resistant GLP-2 analogue.

Glucagon-like Peptides: Future Research Directions

The original finding that GLP-1 functions as an incretin to increase insulin secretion has been followed by the delineation of multiple nonincretin actions of GLP-1 on gastric emptying, small bowel motility, glucagon secretion, and islet β -cell proliferation. The possibility that GLP-1 administration may also be associated with islet regeneration in human subjects with type 2 diabetes has engendered considerable interest in the development of GLP-1 analogues suitable for long-term diabetes treatment. Although GLP-1 decreases appetite and prevents weight gain in short term studies, its potential efficacy in the prevention of weight gain or induction of weight loss in human subjects chronically treated with GLP-1 analogues remains unknown. Intriguingly, the glucose-dependent actions of GLP-1 on the islet β cell have recently been extended by studies demonstrating that GLP-1 receptor signaling forms an essential component of the hepatoportal glucose sensor in mice.¹⁷⁰ As GLP-1 receptors have been localized to glucose-sensitive neurons in regions of the hypothalamus,⁹² future studies of the potential role of GLP-1 receptor signaling in the CNS response to hypoglycemic stress appear warranted. Most importantly, whether GLP-1 analogues will exhibit sustained efficacy and safety in the prolonged treatment of human subjects with type 2 diabetes requires additional investigation.

The results of preclinical studies have suggested that GLP-2 exerts both cytoprotective and regenerative effects in the small and large intestine. Nevertheless, defining the physiological role(s) of GLP-2 in the intestinal epithelium has proved problematic because of the lack of GLP-2 antagonists and the absence of naturally occurring or genetic models of GLP-2 deficiency. Although GLP-2 and its receptor are expressed and functional in the fetal and neonatal gut epithelium,^{142,171} the putative role of GLP-2 in gut development or in the neonatal to adult transition to a fully functional absorptive intestinal epithelium remains unclear. Similarly, although ICV administration of large amounts of GLP-2 inhibits food intake in mice and rats,^{164,172} whether GLP-2 receptor signaling is essential for physiological appetite control or whether GLP-2 subserves distinct, as yet unidentified actions in the brain unrelated to nutrient intake, requires further study. An important area requiring additional analysis is the identification of mechanisms activated by GLP-2R signaling in enteroendocrine cells and neurons and the nature of the humoral, neural, and paracrine mediators released in response to GLP-2R signaling.

Although the GLP-2R is expressed in subsets of human enteroendocrine cells and neurons, GLP-2R localization has not yet been examined in the setting of intestinal disease. Furthermore, whether GLP-2 will prove to be therapeutically useful in human subjects with inflammatory bowel disease, or in the setting of chemotherapy administration, will require careful assessment in clinical trials. Moreover, the only patients treated to date with exogenous GLP-2 have lacked a colon in continuity¹⁶⁹; hence, the therapeutic efficacy of GLP-2 in patients with an intact colon merits examination. Furthermore, the combination of enhanced proliferation and decreased apoptotic activity in the rodent small bowel observed following GLP-2 treatment suggests that intermittent surveillance of the colon in human subjects receiving chronic GLP-2 administration appears prudent. Taken together, the unique biological actions of the PGDPs in the absorption and disposal of ingested energy, taken together with the preliminary results to date from human studies of GLP-1 and GLP-2, suggests that these peptides may soon prove useful in the treatment of specific subsets of patients with diabetes and intestinal disease.

References

- Lund PK, Goodman RH, Dee PC, Habener JF. Pancreatic preproglucagon cDNA contains two glucagon-related coding sequences arranged in tandem. *Proc Natl Acad Sci U S A* 1982;79:345–349.
- Bell GI, Santerre RF, Mullenbach GT. Hamster preproglucagon contains the sequence of glucagon and two related peptides. *Nature* 1983;302:716–718.
- Lopez LC, Frazier ML, Su CJ, Kumar A, Saunders GF. Mammalian pancreatic preproglucagon contains three glucagon-related peptides. *Proc Natl Acad Sci U S A* 1983;80:5485–5489.
- Heinrich G, Gros P, Lund PK, Bentley RC, Habener JF. Preproglucagon messenger ribonucleic acid: Nucleotide and encoded amino acid sequences of the rat pancreatic complementary deoxyribonucleic acid. *Endocrinology* 1984;115:2176–2181.
- Drucker DJ, Asa S. Glucagon gene expression in vertebrate brain. *J Biol Chem* 1988;263:13475–13478.
- Drucker DJ, Brubaker PL. Proglucagon gene expression is regulated by a cyclic AMP-dependent pathway in rat intestine. *Proc Natl Acad Sci U S A* 1989;86:3953–3957.
- Bataille D, Gerspach C, Tatamoto K, Marie JC, Coudray AM, Rosselin G, Mutt V. Bioactive enteroglucagon (oxyntomodulin): present knowledge on its chemical structure and its biological activities. *Peptides* 1981;2:41–44.
- Irwin DM. Molecular evolution of proglucagon. *Regul Pept* 2001; 98:1–12.
- Mojsov S, Heinrich G, Wilson IB, Ravazzola M, Orci L, Habener JF. Preproglucagon gene expression in pancreas and intestine diversifies at the level of post-translational processing. *J Biol Chem* 1986;261:11880–11889.
- Novak U, Wilks A, Buell G, McEwen S. Identical mRNA for preproglucagon in pancreas and gut. *Eur J Biochem* 1987;164: 553–558.
- Lee YC, Brubaker PL, Drucker DJ. Developmental and tissue-specific regulation of proglucagon gene expression. *Endocrinology* 1990;127:2217–2222.
- Irwin DM, Wong J. Trout and chicken proglucagon: alternative splicing generates mRNA transcripts encoding glucagon-like peptide 2. *Mol Endocrinol* 1995;9:267–277.
- Chen YE, Drucker DJ. Tissue-specific expression of unique mRNAs that encode proglucagon-derived peptides or exendin 4 in the lizard. *J Biol Chem* 1997;272:4108–4115.
- Rouillé Y, Martin S, Steiner DF. Differential processing of proglucagon by the subtilisin-like prohormone convertases PC2 and PC3 to generate either glucagon or glucagon-like peptide. *J Biol Chem* 1995;270:26488–26496.
- Rouillé Y, Westermark G, Martin SK, Steiner DF. Proglucagon is processed to glucagon by prohormone convertase PC2 in aTC1-6 cells. *Proc Natl Acad Sci U S A* 1994;91:3242–3246.
- Furuta M, Yano H, Zhou A, Rouille Y, Holst JJ, Carroll R, Ravazzola M, Orci L, Furuta H, Steiner DF. Defective prohormone processing and altered pancreatic islet morphology in mice lacking active SPC2. *Proc Natl Acad Sci U S A* 1999;94:6646–6651.
- Dhanvantari S, Seidah NG, Brubaker PL. Role of prohormone convertases in the tissue-specific processing of proglucagon. *Mol Endocrinol* 1996;10:342–355.
- Dhanvantari S, Brubaker PL. Proglucagon processing in an islet cell line: effects of PC1 overexpression and PC2 depletion. *Endocrinology* 1998;139:1630–1637.
- Rothenberg ME, Eilertson CD, Klein K, Zhou Y, Lindberg I, McDonald JK, Mackin RB, Noe BD. Processing of mouse proglucagon by recombinant prohormone convertase 1 and immunopurified prohormone convertase 2 in vitro. *J Biol Chem* 1995; 270:10136–10146.
- Rothenberg ME, Eilertson CD, Klein K, Mackin RB, Noe BD. Evidence for redundancy in propeptide/prohormone convertase activities in processing proglucagon: an antisense study. *Mol Endocrinol* 1996;10:331–341.
- Ghatei MA, Uttenthal LO, Christofides ND, Bryant MG, Bloom SR. Molecular forms of human enteroglucagon in tissue and plasma: Plasma responses to nutrient stimuli in health and in disorders of the upper gastrointestinal tract. *J Clin Endocrinol Metab* 1983;57:488–495.
- Kreymann B, Ghatei MA, Williams G, Bloom SR. Glucagon-like peptide-1 7-36: A physiological incretin in man. *Lancet* 1987;2: 1300–1304.
- Orskov C, Holst JJ. Radio-immunoassays for glucagon-like peptides 1 and 2 (GLP-1 and GLP-2). *Scand J Clin Lab Invest* 1987;47:165–174.
- Roberge JN, Brubaker PL. Secretion of proglucagon-derived peptides in response to intestinal luminal nutrients. *Endocrinology* 1991;128:3169–3174.
- Hansen L, Hartmann B, Bisgaard T, Mineo H, Jorgensen PN, Holst JJ. Somatostatin restrains the secretion of glucagon-like peptide-1 and -2 from isolated perfused porcine ileum. *Am J Physiol Endocrinol Metab* 2000;278:E1010–E1018.
- Rocca AS, Brubaker PL. Role of the vagus nerve in mediating proximal nutrient-induced glucagon-like peptide-1 secretion. *Endocrinology* 1999;140:1687–1694.
- Hoyt EC, Lund PK, Winesett DE, Fuller CR, Ghatei MA, Bloom SR, Ulshen MR. Effects of fasting, refeeding and intraluminal triglyceride on proglucagon expression in jejunum and ileum. *Diabetes* 1996;45:434–439.
- Reimer RA, McBurney MI. Dietary fiber modulates intestinal proglucagon messenger ribonucleic acid and postprandial secretion of glucagon-like peptide-1 and insulin in rats. *Endocrinology* 1996;137:3948–3956.
- Tappenden KA, Thomson AB, Wild GE, McBurney MI. Short-chain fatty acids increase proglucagon and ornithine decarboxylase

- messenger RNAs after intestinal resection in rats. *J Parenter Enteral Nutr* 1996;20:357–362.
30. Tappenden KA, McBurney MI. Systemic short-chain fatty acids rapidly alter gastrointestinal structure, function, and expression of early response genes. *Dig Dis Sci* 1998;43:1526–1536.
 31. Tappenden KA, Drozdowski LA, Thomson AB, McBurney MI. Short-chain fatty acid-supplemented total parenteral nutrition alters intestinal structure, glucose transporter 2 (GLUT2) mRNA and protein, and proglucagon mRNA abundance in normal rats. *Am J Clin Nutr* 1998;68:118–125.
 32. Orskov C, Wettergren A, Holst JJ. Biological effects and metabolic rates of glucagon-like peptide-1 7-36 amide and glucagon-like peptide-1 7-37 in healthy subjects are indistinguishable. *Diabetes* 1993;42:658–661.
 33. Mentlein R, Gallwitz B, Schmidt WE. Dipeptidyl-peptidase IV hydrolyses gastric inhibitory polypeptide, glucagon-like peptide-1(7-36)amide, peptide histidine methionine and is responsible for their degradation in human serum. *Eur J Biochem* 1993;214:829–835.
 34. Kieffer TJ, McIntosh CHS, Pederson RA. Degradation of glucose-dependent insulinotropic polypeptide and truncated glucagon-like peptide 1 in vitro and in vivo by dipeptidyl peptidase IV. *Endocrinology* 1995;136:3585–3596.
 35. Deacon CF, Nauck MA, Toft-Nielsen M, Pridal L, Willms B, Holst JJ. Both subcutaneously and intravenously administered glucagon-like peptide 1 are rapidly degraded from the NH₂-terminus in type II diabetic patients and in healthy subjects. *Diabetes* 1995;44:1126–1131.
 36. Hansen L, Deacon CF, Orskov C, Holst JJ. Glucagon-like peptide-1-(7-36)amide is transformed to glucagon-like peptide-1-(9-36)amide by dipeptidyl peptidase IV in the capillaries supplying the L cells of the porcine intestine. *Endocrinology* 1999;140:5356–5363.
 37. Brubaker PL, Crivici A, Izzo A, Ehrlich P, Tsai C-H, Drucker DJ. Circulating and tissue forms of the intestinal growth factor, glucagon-like peptide 2. *Endocrinology* 1997;138:4837–4843.
 38. Drucker DJ, Shi Q, Crivici A, Sumner-Smith M, Tavares W, Hill M, Deforest L, Cooper S, Brubaker PL. Regulation of the biological activity of glucagon-like peptide 2 by dipeptidyl peptidase IV. *Nat Biotechnol* 1997;15:673–677.
 39. Xiao Q, Boushey RP, Drucker DJ, Brubaker PL. Secretion of the intestinotropic hormone glucagon-like peptide 2 is differentially regulated by nutrients in humans. *Gastroenterology* 1999;117:99–105.
 40. Hartmann B, Harr MB, Jeppesen PB, Wojdemann M, Deacon CF, Mortensen PB, Holst JJ. In vivo and in vitro degradation of glucagon-like peptide-2 in humans. *J Clin Endocrinol Metab* 2000;85:2884–2888.
 41. Ruiz-Grande C, Pintado J, Alarcon C, Castilla C, Valverde I, Lopez-Novoa JM. Renal catabolism of human glucagon-like peptides 1 and 2. *Can J Physiol Pharmacol* 1990;68:1568–1573.
 42. Deacon CF, Pridal L, Klarskov L, Olesen M, Holst JJ. Glucagon-like peptide 1 undergoes differential tissue-specific metabolism in the anesthetized pig. *Am J Physiol* 1996;271:E458–E464.
 43. Tavares W, Drucker DJ, Brubaker PL. Enzymatic and renal-dependent catabolism of the intestinotropic hormone glucagon-like peptide-2 in the rat. *Am J Physiol* 1999;278:E134–E139.
 44. Thorens B. Expression cloning of the pancreatic β cell receptor for the gluco-incretin hormone glucagon-like peptide 1. *Proc Natl Acad Sci U S A* 1992;89:8641–8645.
 45. Bullock BP, Heller RS, Habener JF. Tissue distribution of messenger ribonucleic acid encoding the rat glucagon-like peptide 1 receptor. *Endocrinology* 1996;137:2968–2978.
 46. Campos RV, Lee YC, Drucker DJ. Divergent tissue-specific and developmental expression of receptors for glucagon and glucagon-like peptide-1 in the mouse. *Endocrinology* 1994;134:2156–2164.
 47. Stoffel M, Espinosa R III, Le Beau MM, Bell GI. Human glucagon-like peptide-1 receptor gene: Localization to chromosome band 6p21 by fluorescence in situ hybridization and linkage of a highly polymorphic simple tandem repeat DNA polymorphism to other markers on chromosome 6. *Diabetes* 1993;42:1215–1218.
 48. Zhang Y, Cook JT, Hattersley AT, Firth R, Saker PJ, Warren-Perry M, Stoffel M, Turner RC. Non-linkage of the glucagon-like peptide 1 receptor gene with maturity onset diabetes of the young. *Diabetologia* 1994;37:721–724.
 49. Wettergren A, Schjoldager B, Mortensen PE, Myhre J, Christiansen J, Holst JJ. Truncated GLP-1 (proglucagon 78-107-amide) inhibits gastric and pancreatic functions in man. *Dig Dis Sci* 1993;38:665–673.
 50. Wettergren A, Petersen H, Orskov C, Christensen J, Sheikh SP, Holst JJ. Glucagon-like peptide-1 7-36 amide and peptide YY from the L-cell of the ileal mucosa are potent inhibitors of vagally induced gastric acid secretion in man. *Scand J Gastroenterol* 1994;29:501–505.
 51. Wettergren A, Pridal L, Wojdemann M, Holst JJ. Amidated and non-amidated glucagon-like peptide-1 (GLP-1): non-pancreatic effects (cephalic phase acid secretion) and stability in plasma in humans. *Regul Pept* 1998;77:83–87.
 52. Rossowski WJ, Cheng B-L, Jiang N-Y, Coy DH. Examination of somatostatin involvement in the inhibitory action of GIP, GLP-1, amylin, and adrenomedullin on gastric acid release using a new SRIF antagonist analogue. *Br J Pharmacol* 1998;125:1081–1087.
 53. Wettergren A, Wojdemann M, Meisner S, Stadil F, Holst JJ. The inhibitory effect of glucagon-like peptide-1 (GLP-1) 7-36 amide on gastric acid secretion in humans depends on an intact vagal innervation. *Gut* 1997;40:597–601.
 54. Schmidler J, Dehne K, Allescher H-D, Schusdziarra V, Classen M, Holst JJ, Polack A, Schepp W. Rat parietal cell receptors for GLP-1-(7–36) amide: Northern blot, cross-linking, and radioligand binding. *Am J Physiol* 1994;267:G423–G432.
 55. Gros L, Hollande F, Thorens B, Kervran A, Bataille D. Comparative effects of GLP-1 (7-36)amide, oxyntomodulin and glucagon on rabbit gastric parietal cell function. *Eur J Pharmacol* 1995;288:319–327.
 56. Wojdemann M, Wettergren A, Sternby B, Holst JJ, Larsen S, Rehfeld JF, Olsen O. Inhibition of human gastric lipase secretion by glucagon-like peptide-1. *Dig Dis Sci* 1998;43:799–805.
 57. Imeryuz N, Yegen BC, Bozkurt A, Coskun T, Villanueva-Pennacarrillo ML, Ulusoy NB. Glucagon-like peptide-1 inhibits gastric emptying via vagal afferent-mediated central mechanisms. *Am J Physiol* 1997;273:G920–G927.
 58. Giralt M, Vergara P. Sympathetic pathways mediate GLP-1 actions in the gastrointestinal tract of the rat. *Regul Pept* 1998;74:19–25.
 59. Wettergren A, Wojdemann M, Holst JJ. Glucagon-like peptide-1 inhibits gastropancreatic function by inhibiting central parasympathetic outflow. *Am J Physiol* 1998;275:G984–G992.
 60. Tolessa T, Gutniak M, Holst JJ, Efendic S, Hellstrom PM. Inhibitory effect of glucagon-like peptide-1 on small bowel motility. *J Clin Invest* 1998;102:764–774.
 61. Wishart JM, Horowitz M, Morris HA, Jones KL, Nauck MA. Relation between gastric emptying of glucose and plasma concentrations of glucagon-like peptide-1. *Peptides* 1998;19:1049–1053.
 62. Willms B, Werner J, Holst JJ, Orskov C, Creutzfeldt W, 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 (non-insulin dependent) diabetic patients. *J Clin Endocrinol Metab* 1996;81:327–332.

63. Nauck MA, Niedereichholz U, Ettl R, Holst JJ, Orskov C, Ritzel R, Schmiegel WH. Glucagon-like peptide 1 inhibition of gastric emptying outweighs its insulinotropic effects in healthy humans. *Am J Physiol* 1997;273:E981-E988.
64. Drucker DJ, Philippe J, Mojsov S, Chick WL, Habener JF. Glucagon-like peptide I stimulates insulin gene expression and increases cyclic AMP levels in a rat islet cell line. *Proc Natl Acad Sci U S A* 1987;84:3434-3438.
65. Mojsov S, Weir GC, Habener JF. Insulinotropin: glucagon-like peptide I (7-37) co-encoded in the glucagon gene is a potent stimulator of insulin release in the perfused rat pancreas. *J Clin Invest* 1987;79:616-619.
66. Holst JJ, Orskov C, Nielsen OV, Schwartz TW. Truncated glucagon-like peptide I, an insulin-releasing hormone from the distal gut. *FEBS Lett* 1987;211:169-174.
67. Edwards CM, Todd JF, Mahmoudi M, Wang Z, Wang RM, Ghatei MA, Bloom SR. Glucagon-like peptide 1 has a physiological role in the control of postprandial glucose in humans: studies with the antagonist exendin 9-39. *Diabetes* 1999;48:86-93.
68. Kolligs F, Fehmann H-C, Goke R, Goke B. Reduction of the incretin effect in rats by the glucagon-like peptide 1 receptor antagonist exendin (9-39) amide. *Diabetes* 1995;44:16-19.
69. Schirra J, Sturm K, Leicht P, Arnold R, Goke B, Katschinski M. Exendin(9-39)amide is an antagonist of glucagon-like peptide-1(7-36)amide in humans. *J Clin Invest* 1998;101:1421-1430.
70. Scrocchi LA, Brown TJ, MacLusky N, Brubaker PL, Auerbach AB, Joyner AL, Drucker DJ. Glucose intolerance but normal satiety in mice with a null mutation in the glucagon-like peptide receptor gene. *Nature Med* 1996;2:1254-1258.
71. Creutzfeldt WO, Kleine N, Willms B, Orskov C, Holst JJ, Nauck MA. Glucagonostatic actions and reduction of fasting hyperglycemia by exogenous glucagon-like peptide I(7-36) amide in type I diabetic patients. *Diabetes Care* 1996;19:580-586.
72. Scrocchi LA, Marshall BA, Cook SM, Brubaker PL, Drucker DJ. Glucose homeostasis in mice with disruption of GLP-1 receptor signaling. *Diabetes* 1998;47:632-639.
73. Edvell A, Lindstrom P. Initiation of increased pancreatic islet growth in young normoglycemic mice (Umea +/-). *Endocrinology* 1999;140:778-783.
74. Buteau J, Roduit R, Susini S, Prentki M. Glucagon-like peptide-1 promotes DNA synthesis, activates phosphatidylinositol 3-kinase and increases transcription factor pancreatic and duodenal homeobox gene 1 (PDX-1) DNA binding activity in beta (INS-1)-cells. *Diabetologia* 1999;42:856-864.
75. Zhou J, Wang X, Pineyro MA, Egan JM. Glucagon-like peptide 1 and exendin-4 convert pancreatic AR42J cells into glucagon- and insulin-producing cells. *Diabetes* 1999;48:2358-2366.
76. Hui H, Wright C, Perfetti R. Glucagon-like peptide 1 induces differentiation of islet duodenal homeobox-1-positive pancreatic ductal cells into insulin-secreting cells. *Diabetes* 2001;50:785-796.
77. Dufayet de la Tour D, Halvorsen T, Demeterco C, Tyrberg B, Itkin-Ansari P, Loy M, Yoo S-J, Hao E, Bossie S, Levine F. β -cell differentiation from a human pancreatic cell line in vitro and in vivo. *Mol Endocrinol* 2001;15:476-483.
78. Stoffers DA, Kieffer TJ, Hussain MA, Drucker DJ, Egan JM, Bonner-Weir S, Habener JF. Insulinotropic glucagon-like peptide-1 agonists stimulate expression of homeodomain protein IDX-1 and increase β -cell mass in mouse pancreas. *Diabetes* 2000;49:741-748.
79. Xu G, Stoffers DA, Habener JF, Bonner-Weir S. Exendin-4 stimulates both beta-cell replication and neogenesis, resulting in increased beta-cell mass and improved glucose tolerance in diabetic rats. *Diabetes* 1999;48:2270-2276.
80. Tourrel C, Bailbe D, Meile M-J, Kergoat M, Portha B. Glucagon-like peptide-1 and exendin-4 stimulates β -cell neogenesis in streptozotocin-treated newborn rats resulting in persistently improved glucose homeostasis at adult age. *Diabetes* 2001;50:1562-1570.
81. Ling Z, Wu D, Zambre Y, Flamez D, Drucker DJ, Pipeleers DG, Schuit FC. Glucagon-like peptide 1 receptor signaling influences topography of islet cells in mice. *Virchows Arch* 2001;438:382-387.
82. Turton MD, O'Shea D, Gunn I, Beak SA, Edwards CMB, Meeran K, Choi SJ, Taylor GM, Heath MM, Lambert PD, Wilding JPH, Smith DM, Ghatei MA, Herbert J, Bloom SR. A role for glucagon-like peptide-1 in the central regulation of feeding. *Nature* 1996;379:69-72.
83. Tang-Christensen M, Larsen PJ, Goke R, Fink-Jensen A, Jessop DS, Moller M, Sheikh SP. Central administration of GLP-1(7-36) amide inhibits food and water intake in rats. *Am J Physiol* 1996;271:R848-R856.
84. Donahey JCK, Van Dijk G, Woods SC, Seeley RJ. Intraventricular GLP-1 reduces short- but not long-term food intake or body weight in lean and obese rats. *Brain Res* 1998;779:75-83.
85. Meeran K, O'Shea D, Edwards CM, Turton MD, Heath MM, Gunn I, Abusnana S, Rossi M, Small CJ, Goldstone AP, Taylor GM, Sunter D, Steere J, Choi SJ, Ghatei MA, Bloom SR. Repeated intracerebroventricular administration of glucagon-like peptide-1(7-36) amide or exendin(9-39) alters body weight in the rat. *Endocrinology* 1999;140:244-250.
86. Szayna M, Doyle ME, Betkey JA, Holloway HW, Spencer RG, Greig NH, Egan JM. Exendin-4 decelerates food intake, weight gain, and fat deposition in Zucker rats. *Endocrinology* 2000;141:1936-1941.
87. Flint A, Raben A, Astrup A, Holst JJ. Glucagon-like peptide 1 promotes satiety and suppresses energy intake in humans. *J Clin Invest* 1998;101:515-520.
88. Gutzwiller JP, Drewe J, Goke B, Schmidt H, Rohrer B, Lareida J, Beglinger C. Glucagon-like peptide-1 promotes satiety and reduces food intake in patients with diabetes mellitus type 2. *Am J Physiol* 1999;276:R1541-R1544.
89. Toft-Nielsen MB, Madsbad S, Holst JJ. Continuous subcutaneous infusion of glucagon-like peptide 1 lowers plasma glucose and reduces appetite in type 2 diabetic patients. *Diabetes Care* 1999;22:1137-1143.
90. Edwards CM, Stanley SA, Davis R, Brynes AE, Frost GS, Seal LJ, Ghatei MA, Bloom SR. Exendin-4 reduces fasting and postprandial glucose and decreases energy intake in healthy volunteers. *Am J Physiol Endocrinol Metab* 2001;281:E155-E161.
91. Shughrue PJ, Lane MV, Merchenthaler I. Glucagon-like peptide-1 receptor (GLP1-R) mRNA in the rat hypothalamus. *Endocrinology* 1996;137:5159-5162.
92. Navarro M, Rodriguez de Fonseca F, Alvarez E, Chowen JA, Zueco JA, Gomez R, Eng J, Blazquez E. Colocalization of glucagon-like peptide-1 (GLP-1) receptors, glucose transporter GLUT-2, and glucokinase mRNAs in rat hypothalamic cells: evidence for a role of GLP-1 receptor agonists as an inhibitory signal for food and water intake. *J Neurochem* 1996;67:1982-1991.
93. Thiele TE, Van Dijk G, Campfield LA, Smith FJ, Burn P, Woods SC, Bernstein H, Seeley RJ. Central infusion of GLP-1, but not leptin, produces conditioned taste aversion in rats. *Am J Physiol* 1997;272:R726-R730.
94. van Dijk G, Thiele TE. Glucagon-like peptide-1 (7-36) amide: a central regulator of satiety and interoceptive stress. *Neuropeptides* 1999;33:406-414.
95. Rinaman L. A functional role for central glucagon-like peptide-1 receptors in lithium chloride-induced anorexia. *Am J Physiol* 1999;277:R1537-R1540.
96. Thiele TE, Seeley RJ, D'Alessio D, Eng J, Bernstein IL, Woods SC, van Dijk G. Central infusion of glucagon-like peptide-1(7-36)

- amide (GLP-1) receptor antagonist attenuates lithium chloride-induced c-Fos induction in rat brainstem. *Brain Res* 1998;801:164–170.
97. Scrocchi LA, Drucker DJ. Effects of aging and a high fat diet on body weight and glucose control in GLP-1R^{-/-} mice. *Endocrinology* 1998;139:3127–3132.
 98. Drucker DJ. Development of glucagon-like peptide-1-based pharmaceuticals as therapeutic agents for the treatment of diabetes. *Curr Pharm Des* 2001;7:1399–1412.
 99. Holst JJ, Deacon CF. Inhibition of the activity of dipeptidyl-peptidase IV as a treatment for type 2 diabetes. *Diabetes* 1998;47:1663–1670.
 100. Mentlein R. Dipeptidyl-peptidase IV (CD26)–role in the inactivation of regulatory peptides. *Regul Pept* 1999;85:9–24.
 101. De Meester I, Korom S, Van Damme J, Scharpe S. CD26, let it cut or cut it down. *Immunol Today* 1999;20:367–375.
 102. Marguet D, Baggio L, Kobayashi T, Bernard AM, Pierres M, Nielsen PF, Ribet U, Watanabe T, Drucker DJ, Wagtmann N. Enhanced insulin secretion and improved glucose tolerance in mice lacking CD26. *Proc Natl Acad Sci U S A* 2000;97:6874–6879.
 103. Nagakura T, Yasuda N, Yamazaki K, Ikuta H, Yoshikawa S, Asano O, Tanaka I. Improved glucose tolerance via enhanced glucose-dependent insulin secretion in dipeptidyl peptidase IV-deficient Fischer rats. *Biochem Biophys Res Commun* 2001;284:501–506.
 104. Deacon CF, Knudsen LB, Madsen K, Wiberg FC, Jacobsen O, Holst JJ. Dipeptidyl peptidase IV resistant analogues of glucagon-like peptide-1 which have extended metabolic stability and improved biological activity. *Diabetologia* 1998;41:271–278.
 105. Gallwitz B, Ropeter T, Morys-Wortmann C, Mentlein R, Siegel EG, Schmidt WE. GLP-1-analogues resistant to degradation by dipeptidyl-peptidase IV in vitro. *Regul Pept* 2000;86:103–111.
 106. Siegel EG, Gallwitz B, Scharf G, Mentlein R, Morys-Wortmann C, Folsch UR, Schrezenmeier J, Drescher K, Schmidt WE. Biological activity of GLP-1-analogues with N-terminal modifications. *Regul Pept* 1999;79:93–102.
 107. Juntti-Berggren L, Pigon J, Karpe F, Hamsten A, Gutniak M, Vignati L, Efendic S. The antidiabetogenic effect of GLP-1 is maintained during a 7-day treatment period and improves diabetic dyslipoproteinemia in NIDDM patients. *Diabetes Care* 1996;19:1200–1206.
 108. Gutniak MK, Linde B, Holst JJ, Efendic S. Subcutaneous injection of the incretin hormone glucagon-like peptide 1 abolishes postprandial glycemia in NIDDM. *Diabetes Care* 1994;17:1039–1044.
 109. Todd JF, Edwards CM, Gbatei MA, Mather HM, Bloom SR. Subcutaneous glucagon-like peptide-1 improves postprandial glycaemic control over a 3-week period in patients with early type 2 diabetes. *Clin Sci (Colch)* 1998;95:325–329.
 110. Gutniak MK, Juntt-Berggren L, Hellstrom PM, Guenifi A, Holst JJ, Efendic S. Glucagon-like peptide I enhances the insulinotropic effect of glibenclamide in NIDDM patients and in the perfused rat pancreas. *Diabetes Care* 1996;19:857–863.
 111. Nauck MA, Sauerwald A, Ritzel R, Holst JJ, Schmiegel W. Influence of glucagon-like peptide 1 on fasting glycemia in type 2 diabetic patients treated with insulin after sulfonylurea secondary failure. *Diabetes Care* 1998;21:1925–1931.
 112. Larsen J, Hylleberg B, Ng K, Damsbo P. Glucagon-Like peptide-1 infusion must be maintained for 24 h/day to obtain acceptable glycemia in type 2 diabetic patients who are poorly controlled on sulphonylurea treatment. *Diabetes Care* 2001;24:1416–1421.
 113. Drucker DJ, Ehrlich P, Asa SL, Brubaker PL. Induction of intestinal epithelial proliferation by glucagon-like peptide 2. *Proc Natl Acad Sci U S A* 1996;93:7911–7916.
 114. Gleeson MH, Bloom SR, Polak JM, Henry K, Dowling RH. Endocrine tumour in kidney affecting small bowel structure, motility, and absorptive function. *Gut* 1971;12:773–782.
 115. Holst JJ, Sorensen TI, Andersen AN, Stadil F, Andersen B, Lauritsen KB, Klein HC. Plasma enteroglucagon after jejunoileal bypass with 3:1 or 1:3 jejunoileal ratio. *Scand J Gastroenterol* 1979;14:205–207.
 116. Barry RE, Barisch J, Bray GA, Sperling MA, Morin RJ, Benfield J. Intestinal adaptation after jejunoileal bypass in man. *Am J Clin Nutr* 1977;30:32–42.
 117. Besterman HS, Mallinson CN, Modigliani R, Christofides ND, Pera A, Ponti V, Sarson DL, Bloom SR. Gut hormones in inflammatory bowel disease. *Scand J Gastroenterol* 1983;18:845–852.
 118. Bloom SR, Polak JM. The hormonal pattern of intestinal adaptation [a major role for enteroglucagon]. *Scand J Gastroenterol* 1982;17:93–103.
 119. Taylor RG, Beveridge DJ, Fuller PJ. Expression of ileal glucagon and peptide tyrosine-tyrosine genes. Response to inhibition of polyamine synthesis in the presence of massive small-bowel resection. *Biochem J* 1992;286:737–741.
 120. Rountree DB, Ulshen MH, Selub S, Fuller CR, Bloom SR, Gbatei MA, Lund PK. Nutrient-independent increases in proglucagon and ornithine decarboxylase messenger RNAs after jejunoileal resection. *Gastroenterology* 1992;103:462–468.
 121. Fuller PJ, Beveridge DJ, Taylor RG. Ileal proglucagon gene expression in the rat: characterization in intestinal adaptation using in situ hybridization. *Gastroenterology* 1993;104:459–466.
 122. Ulshen MH, Hoyt EC, Fuller CR, Gbatei MA, Bloom SR, Lund PK. Increased ileal proglucagon expression after jejunectomy is not suppressed by inhibition of bowel growth. *Dig Dis Sci* 1996;41:677–683.
 123. Sarson DL, Scopinaro N, Bloom SR. Gut hormone changes after jejunoileal (JIB) or biliopancreatic (BPP) bypass surgery for morbid obesity. *Int J Obes* 1981;5:471–480.
 124. Kilander AF, Dotevall G, Lindstedt G, Lundberg PA. Plasma enteroglucagon related to malabsorption in coeliac disease. *Gut* 1984;25:629–635.
 125. Besterman HS, Bloom SR, Sarson DL, Blackburn AM, Johnston DI, Patel HR, Stewart JS, Modigliani R, Guerin S, Mallinson CN. Gut-hormone profile in coeliac disease. *Lancet* 1978;1:785–788.
 126. Besterman HS, Adrian TE, Mallinson CN, Christofides ND, Sarson DL, Pera A, Lombardo L, Modigliani R, Bloom SR. Gut hormone release after intestinal resection. *Gut* 1982;23:854–861.
 127. Stevens FM, Flanagan RW, O’Gorman D, Buchanan KD. Glucagonoma syndrome demonstrating giant duodenal villi. *Gut* 1984;25:784–791.
 128. Jones B, Fishman EK, Bayless TM, Siegelman SS. Villous hypertrophy of the small bowel in a patient with glucagonoma. *J Comput Assist Tomogr* 1983;7:334–337.
 129. Lax E, Leibovici V, Fields SI, Gordon RL. Neglected radiologic signs of the glucagonoma syndrome. *Diagn Imaging Clin Med* 1986;55:321–326.
 130. Myojo S, Tsujikawa T, Sasaki M, Fujiyama Y, Bamba T. Trophic effects of glicentin on rat small-intestinal mucosa in vivo and in vitro. *J Gastroenterol* 1997;32:300–305.
 131. Gbatei MA, Goodlad RA, Taheri S, Mandir N, Brynes AE, Jordinson M, Bloom SR. Proglucagon-derived peptides in intestinal epithelial proliferation: glucagon-like peptide-2 is a major mediator of intestinal epithelial proliferation in rats. *Dig Dis Sci* 2001;46:1255–1263.
 132. Xiao Q, Boushey RP, Cino M, Drucker DJ, Brubaker PL. Circulating levels of glucagon-like peptide-2 in human subjects with

- inflammatory bowel disease. *Am J Physiol* 2000;278:R1057–R1063.
133. Hartmann B, Johnsen AH, Orskov C, Adelhorst K, Thim L, Holst JJ. Structure, measurement, and secretion of human glucagon-like peptide-2. *Peptides* 2000;21:73–80.
 134. Drucker DJ, Deforest L, Brubaker PL. Intestinal response to growth factors administered alone or in combination with h[Gly2]-Glucagon-like peptide 2. *Am J Physiol* 1997;273:G1252–G1262.
 135. Burrin DG, Stoll B, Jiang R, Chang X, Hartmann B, Holst JJ, Greeley GH Jr, Reeds PJ. Minimal enteral nutrient requirements for intestinal growth in neonatal piglets: how much is enough? *Am J Clin Nutr* 2000;71:1603–1610.
 136. Jeppesen PB, Hartmann B, Hansen BS, Thulesen J, Holst JJ, Mortensen PB. Impaired meal-stimulated glucagon-like peptide-2 response in ileal resected short bowel patients with intestinal failure. *Gut* 1999;45:559–563.
 137. Jeppesen PB, Hartmann B, Thulesen J, Hansen BS, Holst JJ, Poulsen SS, Mortensen PB. Elevated plasma glucagon-like peptide 1 and 2 concentrations in ileum resected short bowel patients with a preserved colon. *Gut* 2000;47:370–376.
 138. Fischer KD, Dhanvantari S, Drucker DJ, Brubaker PL. Intestinal growth is associated with elevated levels of glucagon-like peptide-2 in diabetic rats. *Am J Physiol* 1997;273:E815–E820.
 139. Thulesen J, Hartmann B, Nielsen C, Holst JJ, Poulsen SS. Diabetic intestinal growth adaptation and glucagon-like peptide 2 in the rat: effects of dietary fibre. *Gut* 1999;45:672–678.
 140. Tsai C-H, Hill M, Asa SL, Brubaker PL, Drucker DJ. Intestinal growth-promoting properties of glucagon-like peptide 2 in mice. *Am J Physiol* 1997;273:E77–E84.
 141. Hartmann B, Thulesen J, Kissow H, Thulesen S, Orskov C, Ropke C, Poulsen SS, Holst JJ. Dipeptidyl peptidase IV inhibition enhances the intestinotrophic effect of glucagon-like peptide-2 in rats and mice. *Endocrinology* 2000;141:4013–4020.
 142. Burrin DG, Stoll B, Jiang R, Petersen Y, Elnif J, Buddington RK, Schmidt M, Holst JJ, Hartmann B, Sangild PT. GLP-2 stimulates intestinal growth in premature TPN-fed pigs by suppressing proteolysis and apoptosis. *Am J Physiol Gastrointest Liver Physiol* 2000;279:G1249–G1256.
 143. Brubaker PL, Izzo A, Hill M, Drucker DJ. Intestinal function in mice with small bowel growth induced by glucagon-like peptide-2. *Am J Physiol* 1997;272:E1050–E1058.
 144. Kato Y, Yu D, Schwartz MZ. Glucagonlike peptide-2 enhances small intestinal absorptive function and mucosal mass in vivo. *J Pediatr Surg* 1999;34:18–20.
 145. Benjamin MA, McKay DM, Yang PC, Cameron H, Perdue MH. Glucagon-like peptide-2 enhances intestinal epithelial barrier function of both transcellular and paracellular pathways in the mouse. *Gut* 2000;47:112–119.
 146. Kitchen PA, Fitzgerald AJ, Goodlad RA, Barley NF, Ghatei MA, Legon S, Bloom SR, Price A, Walters JR, Forbes A. Glucagon-like peptide-2 increases sucrase-isomaltase but not caudal-related homeobox protein-2 gene expression. *Am J Physiol Gastrointest Liver Physiol* 2000;278:G425–G428.
 147. Cheeseman CI, Tsang R. The effect of gastric inhibitory polypeptide and glucagon like peptides on intestinal hexose transport. *Am J Physiol Gastrointest Liver Physiol* 1996;271:G477–G482.
 148. Cheeseman CI. Upregulation of SGLT-1 transport activity in rat jejunum induced by GLP-2 infusion in vivo. *Am J Physiol* 1997;273:R1965–R1971.
 149. Wojdemann M, Wettergren A, Hartmann B, Hilsted L, Holst JJ. Inhibition of sham feeding-stimulated human gastric acid secretion by glucagon-like peptide-2. *J Clin Endocrinol Metab* 1999;84:2513–2517.
 150. Wojdemann M, Wettergren A, Hartmann B, Holst JJ. Glucagon-like peptide-2 inhibits centrally induced antral motility in pigs. *Scand J Gastroenterol* 1998;33:828–832.
 151. Chance WT, Foley-Nelson T, Thomas I, Balasubramaniam A. Prevention of parenteral nutrition-induced gut hypoplasia by coinfusion of glucagon-like peptide-2. *Am J Physiol* 1997;273:G559–G563.
 152. Chance WT, Sheriff S, Foley-Nelson T, Thomas I, Balasubramaniam A. Maintaining gut integrity during parenteral nutrition of tumor-bearing rats: effects of glucagon-like peptide 2. *Nutr Cancer* 2000;37:215–222.
 153. Scott RB, Kirk D, MacNaughton WK, Meddings JB. GLP-2 augments the adaptive response to massive intestinal resection in rat. *Am J Physiol* 1998;275:G911–G921.
 154. Boushey RP, Yusta B, Drucker DJ. Glucagon-like peptide 2 decreases mortality and reduces the severity of indomethacin-induced murine enteritis. *Am J Physiol* 1999;277:E937–E947.
 155. Kouris GJ, Liu Q, Rossi H, Djuricin G, Gattuso P, Nathan C, Weinstein RA, Prinz RA. The effect of glucagon-like peptide 2 on intestinal permeability and bacterial translocation in acute necrotizing pancreatitis. *Am J Surg* 2001;181:571–575.
 156. Alavi K, Schwartz MZ, Palazzo JP, Prasad R. Treatment of inflammatory bowel disease in a rodent model with the intestinal growth factor glucagon-like peptide-2. *J Pediatr Surg* 2000;35:847–851.
 157. Prasad R, Alavi K, Schwartz MZ. GLP-2alpha accelerates recovery of mucosal absorptive function after intestinal ischemia/reperfusion. *J Pediatr Surg* 2001;36:570–572.
 158. Prasad R, Alavi K, Schwartz MZ. Glucagonlike peptide-2 analogue enhances intestinal mucosal mass after ischemia and reperfusion. *J Pediatr Surg* 2000;35:357–359.
 159. Drucker DJ, Yusta B, Boushey RP, Deforest L, Brubaker PL. Human [Gly2]-GLP-2 reduces the severity of colonic injury in a murine model of experimental colitis. *Am J Physiol* 1999;276:G79–G91.
 160. Tavakkolizadeh A, Shen R, Abraham P, Kormi N, Seifert P, Edelman ER, Jacobs DO, Zinner MJ, Ashley SW, Whang EE. Glucagon-like peptide 2: a new treatment for chemotherapy-induced enteritis. *J Surg Res* 2000;91:77–82.
 161. Boushey RP, Yusta B, Drucker DJ. Glucagon-like peptide (GLP)-2 reduces chemotherapy-associated mortality and enhances cell survival in cells expressing a transfected GLP-2 receptor. *Cancer Res* 2001;61:687–693.
 162. Munroe DG, Gupta AK, Kooshesh P, Rizkalla G, Wang H, Demchyshyn L, Yang Z-J, Kamboj RK, Chen H, McCallum K, Sumner-Smith M, Drucker DJ, Crivici A. Prototypic G protein-coupled receptor for the intestinotrophic factor glucagon-like peptide 2. *Proc Natl Acad Sci U S A* 1999;96:1569–1573.
 163. Yusta B, Huang L, Munroe D, Wolff G, Fantaska R, Sharma S, Demchyshyn L, Asa SL, Drucker DJ. Enteroregulatory localization of GLP-2 receptor expression. *Gastroenterology* 2000;119:744–755.
 164. Lovshin J, Estall J, Yusta B, Brown TJ, Drucker DJ. Glucagon-like peptide-2 action in the murine central nervous system is enhanced by elimination of GLP-1 receptor signaling. *J Biol Chem* 2001;276:21489–21499.
 165. Bjercknes M, Cheng H. Modulation of specific intestinal epithelial progenitors by enteric neurons. *Proc Natl Acad Sci U S A* 2001;98:12497–12502.
 166. Yusta B, Somwar R, Wang F, Munroe D, Grinstein S, Klip A, Drucker DJ. Identification of glucagon-like peptide-2 (GLP-2)-activated signaling pathways in baby hamster kidney fibroblasts expressing the rat GLP-2 receptor. *J Biol Chem* 1999;274:30459–30467.
 167. Jasleen J, Shimoda N, Shen ER, Tavakkolizadeh A, Whang EE, Jacobs DO, Zinner MJ, Ashley SW. Signaling mechanisms of

- glucagon-like peptide 2-induced intestinal epithelial cell proliferation. *J Surg Res* 2000;90:13–18.
168. Yusta B, Boushey RP, Drucker DJ. The glucagon-like peptide-2 receptor mediates direct inhibition of cellular apoptosis via a cAMP-dependent protein kinase-independent pathway. *J Biol Chem* 2000;275:35345–35352.
169. Jeppesen PB, Hartmann B, Thulesen J, Graff J, Lohmann J, Hansen BS, Tofteng F, Poulsen SS, Madsen JL, Holst JJ, Mortensen PB. Glucagon-like peptide 2 improves nutrient absorption and nutritional status in short-bowel patients with no colon. *Gastroenterology* 2001;120:806–815.
170. Burcelin R, Da Costa A, Drucker D, Thorens B. Glucose competence of the hepatoportal vein sensor requires the presence of an activated glucagon-like peptide-1 receptor. *Diabetes* 2001;50:1720–1728.
171. Lovshin J, Yusta B, Iliopoulos I, Migirdicyan A, Dableh L, Brubaker PL, Drucker DJ. Ontogeny of the glucagon-like peptide-2 receptor axis in the developing rat intestine. *Endocrinology* 2000;141:4194–4201.
172. Tang-Christensen M, Larsen PJ, Thulesen J, Romer J, Vrang N. The proglucagon-derived peptide, glucagon-like peptide-2, is a neurotransmitter involved in the regulation of food intake. *Nat Med* 2000;6:802–807.

Received August 4, 2001. Accepted October 18, 2001.

Address correspondence to: Daniel J. Drucker, M.D., Banting and Best Diabetes Centre, Toronto General Hospital, 101 College Street CCRW3-845, Toronto, Ontario, Canada M5G 2C4. e-mail: d.drucker@utoronto.ca; fax: (416) 978-4108.

Supported in part by grants from the Canadian Institutes of Health Research (CIHR) and the Ontario Research and Development Challenge Foundation (ORDCF). Dr. Drucker is a Senior Scientist of the CIHR and a consultant to Merck Research Laboratories, Amylin Pharmaceuticals Inc, Conjuchem Inc, and NPS Pharmaceuticals Inc. GLP-2 is the subject of a licensing agreement between NPS Pharmaceuticals, the University of Toronto, The University Health Network, and Dr. Drucker.