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REVIEW ARTICLE

GHRELIN RECEPTOR AGONISTS FOR THE TREATMENT OF GASTRIC EMPTYING DISORDERS

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ABSTRACT

Delayed gastric emptying, also known as gastroparesis or gastric stasis, is a common complication of a variety of gastrointestinal (GI) and systemic diseases. The strategies used to alleviate symptoms of gastroparesis include antiemetic and prokinetic agents, dietary adjustments and supportive treatments to correct the deficiency of nutrients and fluid. At present there is still a large unmet medical need for new drug therapies, as the currently available prokinetic agents show limited efficacy and poor side-effect profiles in the management of gastroparesis. Endogenous ghrelin, released by the stomach, is a gut hormone that stimulates GI motility and appetite via interaction with ghrelin receptors in the hypothalamus and in the periphery. Synthetic ghrelin receptor agonists that interact with peripheral or central receptors are currently under investigation for their ability to accelerate gastric emptying, induce fasting GI motility patterns, stimulate appetite and food intake, and maintain a positive energy balance. The present review summarizes the recent findings in the development of ghrelin receptor agonists and provides a critical assessment of their potential use for the treatment of gastric emptying disorders.

MOTILITY DISORDERS AFFECTING GASTRIC EMPTYING

Delayed gastric emptying occurs when the coordinated motility of the stomach is dysregulated and food is retained in the stomach. Early satiety, nausea, vomiting and abdominal pain are common symptoms caused by disturbances of gastric emptying. Delayed gastric emptying presents a significant clinical burden as it is a common feature of a number of gastrointestinal (GI) disorders with different etiologies. A long-term study in patients with gastroparesis revealed that the main etiologies were idiopathic in 36%, diabetic in 29% and postsurgical in 13% of the patients (1). A delay in gastric emptying is also caused by infections, nonobstructing malignancies, medications or eating disorders such as anorexia nervosa (2). In patients with functional dyspepsia several pathophysiological mechanisms have been suggested to cause dyspeptic symptoms; delayed gastric emptying was found in about 30% of these patients (3, 4). Impairment of gastric motor function and a delay in GI transit also develop as pathophysiological abnormalities related to anxiety disorders (5) and advanced age (6). Although the prevalence of gastroparesis in the U.S. is unknown, the most recent analysis of the trends in gastroparesis-related hospitalizations shows an increasing prevalence and a significant increase in the economic impact of this disorder. For the period 1995-2004, cases with gastroparesis as the primary or secondary diagnosis showed an increase of 158% and 136%, respectively, and the longest hospital stay with the highest or second highest total charges (7). Thus, there is clearly a need to further investigate the complex mechanisms causing impairment of gastric motility and develop better strategies for pharmacological intervention.

PHARMACOTHERAPEUTIC OPTIONS FOR THE TREATMENT OF DELAYED GASTRIC EMPTYING

Supportive therapies such as parenteral feeding and dietary adjustments are common strategies used to treat gastroparesis, as the use of antiemetic and/or prokinetic drugs alone has not been shown to provide adequate relief. Surgical procedures are used as a last resort to prevent life-threatening complications resulting from gastroparesis. The available pharmacotherapeutic options are based on the action of GI prokinetic agents, which represent a heterogeneous class of drugs known to stimulate gut contractility and promote an aboral movement of intraluminal contents. Prokinetic agents that

have been employed in the clinic to enhance gastric emptying include antidopaminergic agents (domperidone, metoclopramide), serotonergic agents (cisapride and tegaserod), motilin receptor agonists (erythromycin and other motilides) and cholinergic agents inducing GI contractility (bethanechol and acetylcholinesterase inhibitors). However, these therapies have demonstrated limited success because a significant number of patients either do not respond to the prokinetic agent or develop medication-induced side effects. Moreover, cisapride and tegaserod were withdrawn from the market owing to a high risk of adverse cardiovascular effects. Recently discovered prokinetic agents that stimulate smooth muscle contractions to enhance gastric emptying in patients with gastroparesis include itopride, a combination of a dopamine D_2 receptor antagonist and an acetylcholinesterase inhibitor (8), and the motilin receptor agonist mitemcinal (9). In addition, peripherally restricted opioid receptor antagonists (alvimopan, methylnaltrexone) have been indicated for the prevention and treatment of GI dysmotility induced by opioid drugs such as morphine administered for the management of postoperative pain. More recently, a novel class of gastroprokinetic compounds acting as agonists of the ghrelin receptor have been investigated for the treatment of gastroparesis (10).

PHYSIOLOGICAL SIGNIFICANCE OF GHRELIN

Ghrelin, a 28-amino-acid peptide, was discovered as the endogenous agonist of the growth hormone secretagogue receptor (GHSR-1a) (11). According to the convention of naming receptors after their endogenous ligand, the GHSR-1a is now referred to as the ghrelin receptor (12). Ghrelin is produced mainly by endocrine cells in the gastric mucosa and serves as a neuroendocrine signal providing input from the stomach to the brain. Circulating ghrelin concentrations peak before feeding and decline immediately after food ingestion, illustrating a direct relationship between pulsatile ghrelin secretion and feeding behavior (13). Gastric vagal afferent nerves provide the major pathway conveying ghrelin signals that promote appetite and growth hormone (GH) secretion in the brain (14). At present, it is clear that the functional significance of ghrelin extends beyond stimulation of GH secretion and involves the regulation of appetite, carbohydrate and lipid metabolism, as well as stimulation of gastric motility and acid secretion. Other peripheral effects of ghrelin include regulation of cardiovascular function, exocrine and endocrine pancreatic function, and cell proliferation and survival (15). Ghrelin has also been found to stimulate bone formation (16), and recent studies have linked the expression of IL-1β and prostacyclin during states of acute inflammation induced by bacterial lipopolysaccharide to low levels of circulating ghrelin (17). The diverse functions of ghrelin imply that it has the potential to be a multitherapeutic hormone (18); however, the present review is focused specifically on the potential use of ghrelin and synthetic ghrelin receptor agonists in the pharmacotherapy of disorders associated with delayed gastric emptying.

GHRELIN AS A GUT HORMONE REGULATING GASTROINTESTINAL MOTILITY

The gastric emptying of a solid meal depends on the coordinated motor activity of the antrum, pylorus and duodenum (19). Exogenous ghrelin has been found to stimulate gastric motility (20, 21) and accelerate gastric emptying in rats (22-24), mice (25) and humans (26). Ghrelin induces fasted motor activity of the GI tract in conscious fed rats (22) and increases the coordinated antro-pyloric contractions associated with an acceleration of gastric emptying (27). In humans, the administration of ghrelin induces premature gastric phase III contractions of the migrating motor complex and accelerates gastric emptying (28). The prokinetic action of ghrelin in the upper GI tract is thought to occur via the vagal nerves (22, 29) and via direct activation of the enteric nervous system (25, 30, 31). With regard to the effect on gastric emptying, endogenous ghrelin most likely activates the ghrelin receptor expressed on vagal afferents (32, 33), as well as in the enteric nervous system, to potentiate cholinergic contractility (34, 35). Moreover, the expression of ghrelin receptors is high in the stomach and low in the colon (34, 36), a feature that is in agreement with the pronounced gastroprokinetic action of peripheral ghrelin and the lack of any direct effect on colonic motility. Overall, ghrelin acts as a gut–brain signal synchronizing growth and energy balance by regulating appetite, food intake and GI activity to meet the energy needs for GH action during growth and repair. Evidence suggests that ghrelin plays a significant role in acute, rather then chronic, adaptation, as ghrelin-null mice are not anorexic and their size, growth rate, food intake, body composition, reproduction, gross behavior and tissue pathology are not different from their wild-type littermates (37). Moreover, the absence of ghrelin in these knockout mice does not affect gastric emptying compared to wild-type littermates (38).

SYNTHETIC GHRELIN RECEPTOR AGONISTS

The history of the development of synthetic agonists of the ghrelin receptor goes back more then 20 years, when GH secretagogues were synthesized for their ability to stimulate GH release. A number of peptidyl and nonpeptidyl compounds were designed to induce GH release in laboratory animals and humans (Table I). These compounds tended to demonstrate low bioavailability and a short half-life, but some of them (ibutamoren [MK-0677], tabimorelin [NN-703] and SM-130686) show strong GH-releasing activity following oral administration. However, the activity of the GH secretagogues is not specific, as they also increase the release of prolactin, adrenocorticotropic hormone and cortisol, and/or induce central effects such as stimulation of food intake and influence sleep patterns (39). The GH-releasing mechanism was found to be different from that of the endogenous growth hormone-releasing hormone (GHRH) (40), as it is mediated through the G-protein-coupled GHSR-1a, now known as the ghrelin receptor. After the discovery that ghrelin was the endogenous ligand of GHSR-1a, synthetic GH-releasing peptides and peptidomimetics have attracted the attention of investigators as potential gastroprokinetic agents that may stimulate appetite and regulate homeostasis. Recently, a novel series of indolines showing potent in vitro ghrelin receptor agonist activity and acceleration of gastric emptying in rats were reported (41).

THERAPEUTIC POTENTIAL OF GHRELIN AND SYNTHETIC GHRELIN RECEPTOR AGONISTS IN DELAYED GASTRIC EMPTYING

The therapeutic potential of ghrelin and synthetic ghrelin receptor agonists for the treatment of delayed gastric emptying is based on

Table I. *Synthetic compounds developed for their action as growth hormone (GH) secretagogues.*

Name	Chemical description	Ref.
GHRP	His-D-Trp-Ala-Trp-D-Phe-Lys-NH_2	61
GHRP-6	His-D-Trp-Ala-Trp-D-Phe-Lys-NH_2	40, 62
GHRP-1	Ala-His-D-2-Nal-Ala-Trp-D-Phe-Lys-NH_2	40, 62
GHRP-2	Ala-D-Trp-Ala-Trp-D-Phe-Lys-NH_2	40, 62
Hexarelin	His-D-2Me-Trp-Ala-Trp-D-Phe-Lys-NH_2	63
Hexarelin derivatives (JMV-2874, JMV-2810)	Novel hexarelin derivatives with trisubstituted 1,2,4-triazole structure	64
Ipamorelin	Aib*-His-D-2-Nal-D-Phe-Lys-NH_2	62
L-692429, L-692585	Benzolactam nonpeptide GH secretagogues	65, 66
Ibutamoren (MK-0677, L-163191)	Spiroindanylpiperidine privileged structure	67
Tabimorelin (NN-703)	A modified tripeptide derived from ipamorelin-polyethylenimine	68
Capromorelin (CP-424391)	Pyrazolinone-piperidine dipeptide	69
SM-130686	Oxindole derivative, structurally different from ibutamoren and hexarelin	70

*Aminoisobutyryl (Aib) moiety.

their ability to stimulate GI motility. Both ghrelin and synthetic ghrelin receptor agonists were found to accelerate gastric emptying and ameliorate the symptoms of gastroparesis in a variety of animal models (Table II). Human studies in subjects with delayed gastric emptying showed similar prokinetic effects for ghrelin receptor agonists on GI motility. A clinical study in patients with idiopathic gastroparesis investigated the effect of ghrelin on the rate of gastric emptying and the associated meal-related symptom severity score evaluating epigastric pain, bloating, postprandial fullness, nausea, belching and epigastric burning (42). When the effects of an intravenous infusion of ghrelin (30 μg/30 min) or saline were assessed in a double-blind, randomized fashion, the administration of ghrelin was found to enhance gastric emptying and improve meal-related symptoms. Despite the multiple sites of ghrelin action, in humans the effect of ghrelin on gastric motility appears to be independent of the release of GH or motilin and is mediated by either the vagus nerve or direct activation of local ghrelin receptors in the stomach. Levin et al. demonstrated that ghrelin increased the rate of gastric emptying in normal-weight healthy volunteers, while there was no difference in gastric emptying before and after GH substitution in patients with GH deficiency (26). In healthy volunteers, an intravenous infusion of ghrelin (40 μg/30 min) increased the plasma levels of endogenous ghrelin, but not motilin, and induced premature phase III contractions of the migrating motor complex (42). However, the changes in motility included a prolonged increase in the tone of the proximal stomach, a finding that indicates that the use of ghrelin for the treatment of gastroparesis requires careful consideration (43).

The role of the vagus nerve in the prokinetic effect of ghrelin was evaluated in patients with gastroparesis attributed to neuronal deficiencies caused by diabetes or surgical vagotomy (44). When synthetic ghrelin (1-4 mg/kg) or saline was injected i.v. at the end of a test meal, ghrelin was found to accelerate gastric emptying regardless of the deficiency in gastric innervation, suggesting that ghrelin receptor agonists are capable of exerting gastroprokinetic effects in patients with neurogenic gastroparesis. These findings are of clinical significance, as gastroparesis is a common complication in patients with long-standing diabetes for which there is no consistently effective treatment. A double-blind, placebo-controlled study in diabetic patients with gastroparesis requiring insulin treatment and showing vagal neuropathy showed that a 2-h i.v. infusion of ghrelin (5 pmol/kg/min) increased gastric emptying (45).

To determine whether alterations in plasma ghrelin are part of the etiology of gastroparesis, Gaddipati et al. performed an elegant study in patients with diabetic, postsurgical or idiopathic gastroparesis and healthy subjects (46). The increase in systemic pancreatic polypeptide levels induced by a standard sham feeding protocol was studied as a correlate of vagal nerve integrity. The results showed that sham feeding induces an increase in plasma ghrelin and pancreatic polypeptide in healthy subjects and patients with idiopathic gastroparesis, but has no effect in patients with diabetic or postsurgical gastroparesis, suggesting impaired vagal function and regulation of systemic ghrelin levels in diabetic and postsurgical gastroparesis.

Although the levels of circulating ghrelin and glucagon-like peptide 1 (GLP-1) appear to be inversely related during glucose ingestion (47), the role of ghrelin in glucose homeostasis and diabetes remains to be clarified. A study by Pöykkö et al. showed an independent association between low plasma ghrelin levels, hypertension and the prevalence of type 2 diabetes (48). However, there is a considerable controversy regarding the results of studies analyzing ghrelin–insulin interactions: Shiiya et al. found an association between low fasting plasma ghrelin and insulin resistance in obese patients with type 2 diabetes (49), while English et al. reported elevated ghrelin levels in obese human subjects with insulin resistance (50). Studies in patients with type 1 diabetes examining the postprandial regulation of plasma ghrelin in the absence of insulin showed that ghrelin failed to decrease after a meal, and that the replacement of insulin restored meal-related ghrelin suppression (51, 52). On the other hand, ghrelin administration induces an increase in plasma glucose

Table II. *Effects of ghrelin and ghrelin receptor agonists in animal models of delayed gastric emptying.*

Animal model	Ghrelin receptor agonist (dose)	Therapeutic effect	Ref.
Postoperative ileus in rats	Ghrelin/MTLRP (5, 20 μg/kg i.v.)	Accelerates gastric emptying and GI transit	71
Postoperative ileus in dogs	Ghrelin/MTLRP (100 μg/kg i.v. on day 2; 4 μg/kg i.v. on day 4)	Normalizes gastric emptying	72
Septic ileus in mice	Ghrelin/GHRP-6 (20 μg/kg i.p.)	Accelerates gastric emptying; no effect on intestinal transit	73
Postoperative and morphine-induced ileus in rats	RC-1139 (1, 2.5, 10 mg/kg i.v.)	Dose-dependent acceleration of gastric emptying	74
Chemotherapy-associated delayed gastric emptying and decreased food intake in rats and mice	Ghrelin (0.5 mg/kg i.p. in rats; 1 mg/kg i.p. b.i.d. in mice)	Increase in food intake and gastric emptying	75
Burn-induced delayed GI transit in rats	Ghrelin (2 nmol i.p.)	Normalizes gastric emptying and small intestinal transit	76
Surgery- and morphine-induced ileus in rats	TZP-101 (0.3-1 mg/kg i.v.)	Accelerates gastric emptying and small intestinal transit	58, 77
Alloxan-induced diabetes in mice	GHRP-6 (20-200 μg/kg i.p.)	Increases gastric emptying and small intestinal transit; no effect on colonic transit	78
Streptozotocin-induced diabetes in guinea pigs	Ghrelin/GHRP-6 (20-100 μg/kg i.p.)	Accelerates gastric emptying	79

GI, gastrointestinal.

followed by a reduction in insulin secretion (53). Finally, a recent study in healthy volunteers investigated the relationship between gastric emptying rate, postprandial ghrelin and the role of insulin and other hormones (54). The results supported the hypothesis that the postprandial ghrelin response requires a "postgastric feedback" different from insulin, and suggested that glucose-dependent insulinotropic polypeptide (GIP) plays a role in ghrelin secretion. The available studies show no evidence that gastric contractility itself modulates the release of ghrelin, implying that GI motility changes are subordinate to the metabolic effects of ghrelin. In humans, the acute effect of ghrelin infusion is associated with insulin resistance and lipolysis independent from GH signaling, supporting the notion that a major function of ghrelin is the distribution and utilization of glucose to glucose-dependent tissues during conditions of energy shortage (55).

ADVANTAGES AND LIMITATIONS OF GHRELIN RECEPTOR AGONISTS IN THE TREATMENT OF GASTROINTESTINAL DISORDERS

The potential therapeutic benefits of ghrelin and ghrelin receptor agonists in patients with delayed gastric emptying depend on the pharmacological profile of the agonists and the etiology of gastroparesis. The therapeutic limitations of ghrelin owing to its short half-life and low bioavailability were overcome by the development of synthetic small-molecule ghrelin receptor agonists with enhanced metabolic stability and high affinity for the ghrelin receptor. However, as ghrelin has multiple activities, prolonged therapeutic use of ghrelin receptor agonists poses the risk of unwanted side effects. The pulsatile nature of ghrelin release implies that ghrelin receptor agonists might show an optimal effect when applied acutely to induce GI motility and gastric emptying. The orexigenic effect could add to the benefits of this treatment, as the delay in gastric emptying is associated with early satiety and loss of appetite (56). The simultaneous release of GH induced by ghrelin receptor activation precludes long-term treatment regimens, but could be useful for recovery in patients with postoperative ileus, as well as in undernourished elderly patients with gastric stasis. In patients with diabetic gastroparesis, the risks and benefits of ghrelin receptor agonist therapy should be considered with regard to the valuable prokinetic effect in the presence of gastric neuropathy, which is a leading cause of gastroparesis in diabetic patients, and the inhibition of insulin release that may worsen the metabolic state. With respect to separating the effect on motility from the induction of GH release, progress has been made with the development of synthetic nonpeptide ghrelin receptor agonists customized for the treatment of GI disorders (57). TZP-101, a macrolytic peptidomimetic compound, has been found to stimulate gastric emptying and food intake without altering ghrelin-induced GH release in the rat (58); however, whether this is the case in humans remains to be established.

Finally, the drug–receptor interactions between synthetic agonists and the ghrelin receptor are unique, showing both allosteric binding of the agonist, as well as a large overlap with the binding site for the endogenous ligand. Holst et al. presented evidence to support a model of a dimeric ghrelin receptor, where ghrelin activates one receptor subunit, while smaller synthetic agonists bind to the other subunit and modulate ghrelin function (59). As a result, different synthetic small molecules may act both as co-agonists and as either neutral (ibatumoren), positive (L-692429), or negative (GHRP-6) modulators of ghrelin effect. These findings open a new avenue for optimizing the effect of drug candidates as agonists and positive modulators of ghrelin signaling (60).

CONCLUSIONS

Given the orexigenic and gastroprokinetic effects of endogenous ghrelin, ghrelin receptor agonists have attracted significant interest

as a potential therapy for patients with various forms of gastroparesis. This has become a dynamic area of investigation due to the current lack of an optimal pharmacological treatment relieving the symptoms and providing adequate nutrition in patients with gastric dysmotility. The review of the literature supports the feasibility and potential benefits of using ghrelin receptor agonists as a new class of prokinetic agents for accelerating gastric emptying and outlines a future direction for creating selective ghrelin analogues with specific activities relevant to the pathophysiological heterogeneity of the patient population.

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