

VISCERAL PERCEPTION

Visceral perception: sensory transduction in visceral afferents and nutrients

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The possible mechanisms that may be involved in nutrient detection in the wall of the gastrointestinal tract are reviewed. There is strong functional and electrophysiological evidence that both intrinsic and extrinsic primary afferent neurones mediate mechano- and chemosensitive responses in the gastrointestinal tract. This review focuses on the extrinsic afferent pathways as these are the ones that convey information to the central nervous system which is clearly necessary for perception to occur.

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SUMMARY

Nutrients, especially carbohydrates and lipids, in the intestinal lumen are known to inhibit gastric function. It has been postulated that enteroendocrine cells (EC) in the intestinal mucosa respond to changes in luminal contents by releasing neuroactive mediators, such as cholecystokinin (CCK) and serotonin (5-HT), which in turn activate specific receptors on primary afferent nerve terminals. Both vagal and spinal afferent pathways have been shown to be involved in intestinal feedback inhibition of gastric emptying precipitated by glucose. The response is mediated by 5-HT₃ receptors and also by sodium-glucose cotransporter1 (SGLT-1) expressed by enterochromaffin cells. Inhibition of gastric emptying and acid secretion induced by long chain fatty acids is thought to involve a vagal pathway, which is activated by CCK release. It is thought that the central nervous system (CNS) may respond either to the particular pattern of gastrointestinal motility or to impulse frequency. Research is underway to investigate how these impulses are handled by second and third order central neurones.

INTRODUCTION

It can be considered that one of the principal roles of the duodenum is to act as a sensory organ. During the intestinal phases of digestion, feedback inhibition of gastric motility and secretion, together with stimulation of pancreatic secretion, gall bladder contraction, and relaxation of the sphincter of Oddi, are regulated by signals from the duodenum. These processes are tightly regulated to match the digestive and absorptive capacity of the intestine with entry of food from the stomach and secretions from the pancreas and gall bladder. The existence of these feedback and feedforward responses implies the existence of "sensors" that can detect the presence of nutri-

THE ROLE OF NUTRIENTS: AN OVERVIEW

Our current understanding of the sensors activated by nutrients relies largely on data generated in functional experiments measuring inhibition of gastric motility or acid secretion as a measure of activation of sensors. This approach has yielded useful information about the nature of the mechanisms by which nutrients are "sensed" by the intestinal wall. It is clear that nutrients act separately from any osmotic or mechanical effects, that all macronutrient groups alter gastric secretomotor function and food intake, and that each macronutrient group acts via activation of separate and distinct pathways and mechanisms. These latter observations suggest that the responses to nutrients are not secondary to detection of caloric content.

A direct involvement of extrinsic primary afferents in mediating inhibition of gastric emptying, secretion, and food intake in response to chemical stimulation of the gastrointestinal tract wall has been demonstrated using the sensory neurotoxin capsaicin to produce functional ablation of visceral afferents.^{1,2} Intraluminal application of capsaicin has shown that mucosal afferent terminal fields play a role in mediating feedback responses of the stomach and pancreas.^{3,4} Direct application of capsaicin to vagal nerve trunks or the coeliac ganglion (to selectively denervate either vagal or spinal afferents, respectively), has shown that a vagal afferent pathway mediates inhibition of gastric emptying and gastric acid secretion in response to intestinal lipid.^{5,6} In contrast, inhibition of gastric emptying in response to monosaccharides is dependent on both a vagal and spinal capsaicin sensitive pathway.⁷

The detailed morphology of the vagal extrinsic afferent terminals has been studied using axonally transported neuronal tracers. Individual

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Abbreviations: apo, apolipoprotein; CCK, cholecystokinin; CNS, central nervous system; EC, enteroendocrine cells; GLUT, glucose transporter; SGLT, sodium-glucose cotransporter; 5-HT, serotonin.

afferent fibres form an extensive system of collaterals in the submucosa, producing terminal arborisations in the mucosa that cover large areas with endings around the crypts and within the *lamina propria*. The densest innervation is found in the proximal small intestine which has been shown to be the functional site for generating many of the feedback and feed-forward responses. No fibres penetrate between epithelial cells or protrude into the lumen. Thus luminal content must signal to primary afferent nerve terminals via an interaction with the epithelial cell layer.⁸

The current hypothesis, for which there is significant experimental evidence, is that EC in the intestinal mucosa act as taste cells. A taste cell could be any cell in the mucosa that responds to nutrients by release of a neuroactive modulator or some other mechanism that activates nearby afferent nerve terminals. Gastrointestinal endocrine cells are neuroactive in that they release their content in response to nutrients in the intestinal lumen and are obvious candidates for this role. Endocrine cells have tufts of microvilli, which project to the lumen, and it is possible that these structures detect luminal nutrients via a direct effect on endocrine cells. These afferents have been shown to terminate in the proximity of EC; this has been shown for endocrine cells expressing CCK.⁹

LIPIDS AND CCK

In order to help identify the mechanisms of sensory transduction in the gastrointestinal mucosal, consideration of the pathways of absorption for each macronutrient is useful. After lipolysis of ingested triglycerides into long chain fatty acids and monoglycerides, these molecules diffuse into the enterocyte and are resynthesised to triglycerides by the endoplasmic reticulum.¹⁰ Apolipoproteins are transferred to the newly synthesised lipids, and chylomicrons are formed and released by exocytosis from the basolateral membrane of the enterocyte. Chylomicrons diffuse through the lamina propria into lymph lacteals. Fatty acids greater than 12 carbon chain (C12) are absorbed via chylomicron formation into the lymph while fatty acids less than 10 carbon chain (C10) diffuse predominantly into the portal blood and are carried bound to albumin to the liver.

Inhibition of gastric emptying or food intake by fat is induced by free fatty acids, and only by long chain fatty acids greater than C12.¹¹ Likewise, it is long chain fatty acids that are effective in releasing CCK.¹² It has recently been shown that in humans there is a good correlation between increasing plasma levels of CCK and inhibition of gastric motility by fatty acids of chain length above but not below C10.¹³

Apolipoprotein (apo) A-IV is synthesised by enterocytes in response to lipid absorption in the intestine.¹⁴ Apo A-IV synthesis and release into the mesenteric lymph depend on the transport of lipid via chylomicrons and hence secretion and release of apo A-IV are dependent on the chain length of the absorbed fatty acid.¹⁵ Formation of chylomicrons and synthesis of one of these apolipoproteins can be rapid and has been proposed to be involved in signalling intestinal lipid content to other organs.¹⁶ For example, intraperitoneal injection of exogenous apo A-IV has been shown to inhibit gastric emptying, gastric acid secretion, and food intake in rats.^{17, 18}

It is clearly of interest to determine whether lipid acts luminally, for example on the luminal aspect of EC, or whether absorption is required, and also whether it is sufficient for the lipid to enter an epithelial cell or whether a cellular event downstream is required.

Recent evidence indicates that chylomicron formation is required for lipids to inhibit gastric emptying.¹⁹ The role of chylomicron formation in this sensory transduction pathway has been investigated using a surfactant, Pluronic L-81, which inhibits chylomicron formation and apo A-IV secretion. In awake rats, lipid induced inhibition of gastric emptying is abolished when lipid is infused together with Pluronic L-81

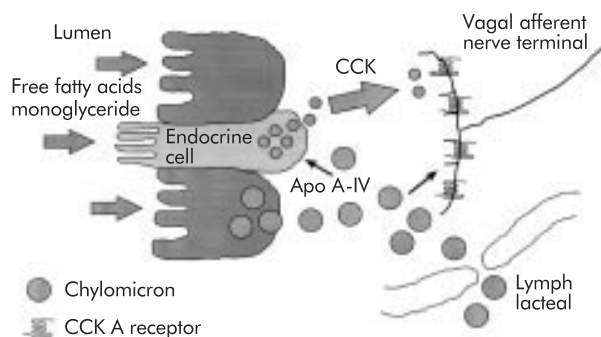


Figure 1 Diagrammatic representation of the effects of ingestion of free fatty acids on the visceral afferent nervous system. The chylomicron products are thought to signal to vagal afferent nerve terminals. Apo A-IV, apolipoprotein A-IV; CCK, cholecystokinin.

which blocks the appearance of postabsorptive chylomicrons in the lamina propria and mesenteric lymph. This demonstrates that the ability of intestinal lipid to produce feedback inhibition of gastric emptying depends on chylomicron formation. However, it is not clear if it is the triglyceride content of the chylomicrons or some other component that triggers the signal to either endocrine cells or extrinsic primary afferent neurones.

There is also evidence which indicates that it is a constituent of chylous lymph that initiates feedback.²⁰ It may therefore be hypothesised that inhibition of gastric motor function is dependent on the postabsorptive components of intestinal lipid digestion and absorption (possibly chylomicrons and/or chylomicron components, for example apo A-IV) rather than the products of lipid absorption (triglyceride) itself. The effect of postabsorptive chylomicron products was studied by measuring the ability of chylous lymph given intra-arterially to inhibit gastric motility. Lymph was collected from awake lymph-fistula rats, during intestinal infusion with either a glucose-saline maintenance solution or lipid and administered close to the upper gastrointestinal tract in anaesthetised recipient rats. Injection of lymph, collected during intestinal lipid infusion, significantly inhibited gastric motility compared with injection of equivalent amounts of triglyceride. Chylous lymph was significantly more potent at inhibiting gastric motility than lymph collected during intestinal infusion of a maintenance solution. Additionally, inhibition of gastric motility was significantly reduced after injection of lymph collected from rats during lipid infusion with Pluronic L-81 compared with lymph injection from donor animals treated with Pluronic L-63 (a non-inhibitory control for Pluronic L-81). These data suggest that it is other chylomicron components rather than the lipid content of chylous lymph which cause the effect. Injection of purified recombinant apo A-IV has been shown to significantly inhibit gastric motility. Taken together, these data suggest that products of lipid digestion and absorption, other than fatty acids or triglycerides, released by the intestine during lipid digestion probably serve as signals to initiate intestinal feedback regulation of gastrointestinal function. Most likely, apo A-IV is one of the signals involved. The entire process is represented schematically in fig 1.

CARBOHYDRATES AND SEROTONIN

Dietary carbohydrates release a number of different substances from EC in the small intestine including glucagon-like peptide, glucose dependent insulinotropic polypeptide, and 5-HT which may be involved in mediating carbohydrate induced changes in gastric function and food intake.²¹

Strong evidence indicates that 5-HT has a role in mediating dietary carbohydrate induced intestinal feedback inhibition of

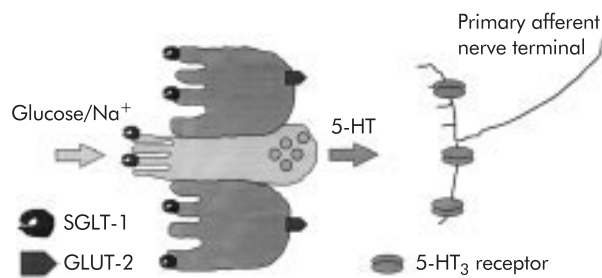


Figure 2 A working model showing how luminal hexoses signal to primary afferents. GLUT, glucose transporter; SGLT, sodium-glucose cotransporter; 5-HT, serotonin.

gastric motor function. Such evidence comes from release studies, electrophysiological recordings of visceral afferents, and functional studies using 5-HT receptor antagonists, in particular the 5-HT₃ receptor subtype.

Release of 5-HT from the intestinal mucosa is under neural, paracrine, and direct control. Importantly, there is clear evidence that 5-HT is secreted from EC in response to changes in luminal contents. Evidence from both in vivo and in vitro studies suggests that 5-HT can be released by intestinal perfusion of hyperosmotic glucose solutions or acid.²² The exact mechanism by which luminal glucose or acid stimulates 5-HT secretion and the physiological importance of these observations are not known. However, it is clear that 5-HT is capable of producing changes in activity of extrinsic and intrinsic neurones. Electrophysiological recordings of vagal afferents with terminal fields in the intestine of the ferret and rat are extremely sensitive to exogenously administered 5-HT.^{22–26} This seems to be a 5-HT₃ mediated response as it was antagonised by a specific antagonist, such as granisetron or tropisetron. Further support for this model comes from evidence that vagal afferents express 5-HT₃ receptors. In vitro preparations of nodose neurones show that 5-HT or 5-HT₃ receptor agonists depolarise neurones and that 5-HT₃ receptor antagonists block this effect.²⁷ A direct role for 5-HT acting via 5-HT₃ receptors has been demonstrated in mediating inhibition of gastric emptying in response to glucose.²⁸ Taken together, this evidence clearly supports a role for 5-HT, 5-HT₃ receptors, and extrinsic primary afferent neurones in the intestinal feedback inhibition induced by glucose in the intestinal lumen.

Only monosaccharides are absorbed across the gastrointestinal epithelium.²⁹ Ingested complex carbohydrates are digested in the lumen and by brush border enzymes to generate monosaccharides such as glucose, galactose, or fructose. Glucose and galactose enter the enterocyte via the sodium-glucose cotransporter 1 (SGLT-1) located on the apical surface of enterocytes (fig 2). They exit the cell by facilitated diffusion via the basolaterally located glucose transporter (GLUT-2). Fructose is absorbed by facilitated diffusion via GLUT-5. Preliminary evidence indicates that SGLT-1 is critical to the production of glucose induced inhibition of gastric emptying.³⁰ Perfusion of the intestine with the glucose analogue, α -methyl glucose, which is a substrate for SGLT-1, inhibits gastric emptying whereas analogues of glucose that are not substrates of SGLT-1 do not inhibit gastric emptying.

Although there is an established role for 5-HT, the question arises as to the mechanism by which glucose, possibly acting via SGLT-1, releases 5-HT from EC in the intestinal mucosa. Previous studies on 5-HT release from the intestinal mucosa have used in vitro preparations, including isolated loops of intestine, sheets of mucosa, or impure preparations of EC. Enterocytes express SGLT-1 and, therefore, it is important to discriminate between a direct effect of glucose on EC and an indirect effect mediated by enterocytes, as both mechanisms are possible. In order to help address this question, studies

have been undertaken using a human tumour cell line (BON) which releases 5-HT in response to a number of different stimuli.³¹

BON cells release glucose in response to D-glucose. Glucose evoked release of 5-HT can be mimicked by a non-metabolisable substrate of SGLT-1, α -methyl glucose, but not by mannitol or fructose. Glucose evoked release of 5-HT was also sensitive to phloridzin which is a blocker of SGLT-1. These results suggest that SGLT-1 is involved in initiating feedback responses to glucose in the intestine and that this process may involve release of 5-HT from EC by a mechanism involving SGLT-1.

CONCLUSIONS

This paper has reviewed the possible mechanisms that may be involved in nutrient detection in the wall of the gastrointestinal tract. Clearly, the majority of this information never reaches the level of consciousness but is none the less important in the regulation of normal gastrointestinal function. There is ample evidence that in a number of gastrointestinal disorders, including functional dyspepsia and irritable bowel syndrome, the gastrointestinal nutrient content may either be the source or contribute to abnormal, painful, or uncomfortable visceral perception. A better understanding of the mechanisms and pathways by which nutrients are detected in the intestinal mucosa, and of the consequent activation of extrinsic vagal and spinal primary afferents, will help in understanding the pathophysiology of these functional gastrointestinal disorders.

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