Published in final edited form as:

Curr Opin Endocrinol Diabetes Obes. 2010 February ; 17(1): 33–39. doi:10.1097/MED.0b013e3283333faf8.

GASTRIN, INFLAMMATION, AND CARCINOGENESIS

Celia Chao and Mark R. Hellmich

Departments of Surgery, Neuroscience and Cell Biology, Sealy Center for Cancer Cell Biology, University of Texas Medical Branch Galveston, TX

Abstract

Purpose of review—Chronic infection of the gastric mucosa with *Helicobacter pylori* has long been recognized as a significant risk factor for gastric cancer, and indeed, this model represents the prototypical inflammation-associated cancer. In this review, we present the latest clinical and experimental evidence showing that gastrin peptides and their receptors (the cholecystokinin [CCK₂] receptors) potentiate the progression of gastric cancer and other gastrointestinal malignancies in the presence of inflammation.

Recent Findings—We highlight the feed-forward mechanisms by which gastrin and CCK₂ receptor expression are upregulated during inflammation and in GI cancers, summarize gastrin's pro-inflammatory role by inducing the production cyclooxygenase-2 (COX-2) and interleukin-8 (IL-8), and relate evidence suggesting that gastrin and their receptors modulate the function of immune cells and fibroblasts following cellular stress, injury, repair, as well as during cancer progression.

Summary—We discuss trends for future studies directed toward the elucidation of gastrin peptides' role in regulating inter-cellular molecular signaling mechanisms between local and circulating immune cells, fibroblasts, epithelial cells, and other cell-types in the microenvironments of inflammation-related cancers. Elucidation of the molecular and cellular pathways that relate inflammation with cancer may provide additional opportunities to develop complementary therapies that target the inflammatory microenvironment of the cancer.

Keywords

Helicobacter pylori; gastrin; progastrin; CCK₂ receptors; gastrointestinal cancers; inflammation

Introduction

Tumor initiation, progression and metastasis rely on the reciprocal interactions and paracrine signaling that occurs between all cell types in the tumor microenvironment, including the epithelial cells, fibroblasts, immune cells and endothelial cells. There is increasing recognition that the immune cells and their elaboration of pro-inflammatory mediators, typically associated with the containment of an inflammatory process, are also involved in the pathobiology of many cancers. Clinical observations have long supported an integral link between chronic inflammation and the development of many common cancers of the gastrointestinal (GI) tract (e.g., ulcerative colitis and colon cancer), endocrine organs, and soft tissues [1]. The elucidation of the molecular and cellular pathways that relate inflammation with cancer may provide additional opportunities to develop complementary therapies [2•, 3] that target the inflammatory microenvironment of the cancer.

To whom correspondence should be addressed: Mark R. Hellmich, Ph.D., Department of Surgery, University of Texas Medical Branch, 301 University Blvd., Galveston, TX, USA 77555-0722, Telephone: (409) 772-1845; Fax: (409) 772-6368; hellmic@utmb.edu.

Under normal physiological conditions, immune cells produce pro- and anti-inflammatory mediators (e.g., chemokines, cytokines, and prostaglandins [PG]) which in turn regulate transcriptional factors related to innate immunity and inflammation, such as nuclear factor- κ B (NF- κ B) and hypoxia-inducible factor 1- α (HIF-1 α) [4••]. These same pathways become dysregulated in the pathogenesis of cancer. The transformed cells, carcinoma-associated fibroblasts (CAF), and tumor-associated macrophages, all coordinate the local production of chemokines, cytokines, lipid inflammatory mediators, which further recruit and activate immune cells. Therefore, it is not surprising that major pathways activated in inflammation, NF- κ B, for example, also emerges as one of the major promoters of inflammation-linked cancers. However, the intra- and inter-cellular mechanisms by which inflammatory processes initiate tumor development, progression and spread remain largely undefined and thus continue to be active areas of ongoing investigations.

In this review, we present the latest clinical and experimental evidence showing that gastrin peptides and their receptors potentiate the progression of GI malignancies in the presence of inflammation. We discuss recent publications demonstrating a link between gastric inflammation, hypergastrinemia, and the progression of gastric cancer. Further, we highlight the mechanisms by which gastrin expression is upregulated during inflammation and in GI cancers, summarize gastrin's role in inducing the production of pro-inflammatory mediators such as cyclooxygenase-2 (COX-2) and interleukin-8 (IL-8), and relate evidence suggesting that gastrin and gastrin receptors modulate the function of immune cells and fibroblasts following injury and repair, as well as during cancer progression. Finally, we discuss trends for future studies directed toward the elucidation of gastrin peptides' role in regulating inter-cellular molecular signaling mechanisms between local and circulating immune cells, fibroblasts, epithelial cells, and other cell-types in the microenvironments of inflammation-related cancers.

Gastrin does not cause gastric cancer but potentiates the carcinogenic effects of *Helicobacter pylori* (*H. pylori*) infection

Epidemiological, clinical, and experimental evidence have now established that approximately 95% of gastric cancers are adenocarcinomas. They are commonly classified on the basis of histological criteria and associated risk factors: diffuse or intestinal types. Diffuse gastric adenocarcinoma is often associated with hereditary risk factors (mutations on the Ecadherin gene [5]), whereas, intestinal-type cancer often arises within an area of intestinal metaplasia and exhibits a glandular morphology that microscopically resembles colonic adenocarcinoma. Intestinal-type gastric adenocarcinoma occurs with a higher frequency in older populations, and is strongly associated with *H. pylori* infection, a gram-negative, microaerophilic bacterium that resides in the gastric pits. However, despite this clear association the molecular mechanisms mediating the transformation are only beginning to be revealed.

Gastric carcinogenesis is a multistep process that arises from superficial gastritis, chronic atrophic gastritis, progressing to intestinal metaplasia, dysplasia, and ultimately carcinoma [6]. *H. pylori* is the most common known cause of chronic gastritis in humans, and has been classified as a group I carcinogen [7]. *H. pylori* secretes urease, which converts urea to ammonia, thereby neutralizing the acid in the stomach. *H. pylori* initiates a host inflammatory response that is associated with the recruitment of mononuclear and polymorphonuclear leukocytes, and bone marrow-derived cells [8]. Specific inflammatory cytokines from immune cells are required for the initiation and promotion of carcinogenesis. Indeed, mice engineered with a deletion of the gene for cytokine interferon- γ (IFN- $\gamma^{-/-}$) do not develop atrophic gastritis [9].

In addition to local inflammation, *H. pylori* induces the systemic elevation of serum gastrin (hypergastrinemia) by several mechanisms: 1) Acutely, *H. pylori* infection suppresses gastric acid secretion by parietal cells, causing a loss of feedback inhibition by acid and a compensatory increase in gastrin production by the antral G cells. 2) Chronic infection results in parietal cell loss (atrophic gastritis), reduced acid production, which also triggers the G cells to overexpress gastrin. Additionally, a subset of patients (~15%) with chronic gastritis of the antral stomach exhibit decreased somatostatin, which removes the normal feed-back inhibition on Gcell resulting in increased gastrin secretion [10]. 3) *H. pylori*-induced inflammatory cytokines stimulate antral G cells to release gastrin [11]. Ultimately, the combination of achlorhydria and hypergastrinemia, induced by *H. pylori* infection, results in gastric bacterial overgrowth, lack of parietal cell differentiation, development of gastric metaplasia, and eventual progression to gastric carcinoma.

Although a precise assessment of the contribution of gastrin to *H. pylori*-induced gastric cancer development and progression is difficult in humans, since *H. pylori* infection and hypergastrinemia are mechanistically linked, experiments with the transgenic INS-GAS mouse model, which exhibits hypergastrinemia due to overexpression of gastrin in pancreatic beta-cells [12•, 13], indicate that gastrin can potentiate the carcinogenic effects of bacterial infection on the gastric mucosa. Compared to wild-type controls, INS-GAS mice initially develop hypertrophy/hyperplasia of parietal and ECL cells, with increased gastric acid secretion. With age, they develop progressive loss of parietal and ECL cells, metaplasia, dysplasia, and invasive cancer by 20 months. However, in the presence of *H. felis* (a bacteria similar to *H. pylori*) infection, the development of invasive gastric cancer was accelerated, occurring in mice 7–8 months old [13]. Thus, the INS-GAS mouse model demonstrated that gastrin is an important growth factor for gastric cells, potentiating the effects of a chronic inflammatory process. In humans, clinical data from hypergastrinemic patients (such as those with gastrinoma or chronic proton pump usage) clearly demonstrate that hypergastrinemia alone does not increase the risk of gastric carcinoma [14], but that chronic atrophic gastritis does predispose toward the development gastric carcinoma. The mouse data suggest that the link between gastrin and *H. pylori* infection warrants further investigation in humans.

At the molecular level *H. felis*-infected hypergastrinemic mice showed increased mRNA expression for the growth factors, regenerating islet-derived 1 (Reg1) and amphiregulin (a epidermal growth factor receptor ligand) as well as enhanced expression of message for the extracellular matrix (ECM) remodeling proteins matrix metalloproteinases MMP-9, and -13 [12•]. However, the molecular mechanisms by which hypergastrinemia potentiates the inflammation-mediated cancer progression has not been fully elucidated. For example, although both *H. pylori* and gastrin can stimulate the expression of Reg1 in primary gastric epithelial cells, Steele et al. [15] performed transfection studies using primary mouse Reg1 promoter-luciferase reporter constructs and determined that the *H. pylori* virulence factor cytotoxin-associated gene A (cagA) and gastrin are each separately controlled by distinct regulatory elements in the promoter, suggesting that Reg1 upregulation by gastrin and cagA are not mechanistically dependent on each other, but may both contribute to increase Reg1 in inflammation, injury, and cancer. Thus, further studies on the mechanistic connections between hypergastrinemia, inflammation and the various cell types in the chronic inflammatory environment will contribute to our understanding of the pathophysiology of gastric cancer.

Local actions of gastrin promote colorectal carcinogenesis

Although hypergastrinemia has been shown to promote gastric cancer, the local upregulation of gastrin resulting in autocrine and/or paracrine signaling appears to be more important

among colorectal, pancreatic and esophageal cancers. For example, only 8% of colorectal cancers can be attributed to a hypergastrinemic state [16]. However, in the development of colorectal cancer, studies indicate that gastrin and gastrin-like peptides are upregulated locally in 78% of premalignant adenomatous polyps, before the appearance of invasive carcinoma [17], and gastrin expression has been linked to key mutations in the initiation of colorectal carcinogenesis.

The initiation of colorectal carcinogenesis involves mutations within the Wnt/adenomatous polyposis coli APC/ β -catenin signaling pathway [18]. In the inactive state, β -catenin is bound within a multiprotein inhibitory complex that includes glycogen synthase kinase-3 β (GSK-3 β) and the APC tumor suppressor protein. Normally, β -catenin is constitutively phosphorylated by GSK-3 β , and undergoes regulated degradation. Dysregulation occurs when an activating mutation in the Wnt/APC/ β -catenin signaling pathway inhibits β -catenin phosphorylation, leading to its nuclear accumulation, where it heterodimerizes with the transcription factor lymphocyte enhancer factor/T cell factor (LEF/TCF). Koh et al. [19] have elucidated a mechanistic relationship between mutations in the Wnt/APC/ β -catenin pathway, aberrant gastrin gene expression, and gastrin-mediated signal transduction using the APC^{min/+} mouse, a model of familial adenomatous polyposis. When the APC^{min/+} mouse was crossed with a gastrin gene knockout mouse, the hybrid developed fewer intestinal polyps. Gastrin transcription is linked to the Wnt/ β -catenin pathway by a binding site for the transcription factor TCF4 in the gastrin promoter. They showed that induction of the wild-type APC decreased gastrin mRNA expression, while transfection of constitutively active β -catenin increased gastrin promoter activity [19].

Aberrant signaling by the Wnt/APC/ β -catenin pathway may also appear to be involved with local upregulation of gastrin in gastric cancer cells. Infection with cagA-positive *H. pylori* strains has been associated with increased gastric mucosal inflammation and atrophic gastritis, and increased risk of gastric cancer. Carcinogenic strains of *H. pylori*, derived from Mongolian gerbils, that express cagA [20] or the outer membrane protein OipA [21] have been shown to activate β -catenin in infected gerbils that developed gastric dysplasia and carcinoma. Additionally, human gastric epithelial cells (AGS), which possess wild-type APC, when incubated with cagA or OipA, demonstrated translocation of the β -catenin from the membrane to the nucleus; however, β -catenin did not translocate to the nucleus when infected with a cagA mutant, or an OipA mutant, indicating that specific microbial factors are involved in the mechanisms of specific pathogen-associated malignancies. It is interesting to speculate that perhaps in gastric cancer, *H. pylori*-induced dysregulation of the Wnt/APC/ β -catenin signaling pathway may contribute to aberrant, cell-autonomous production of gastrin and gastrin precursor peptides by the gastric mucosal cells. Future studies will need to address if gastrin contributes to the multi-step histological and molecular changes during the progression of gastric carcinogenesis mediated by cagA or oipA.

Positive-feedback loop: Gastrin regulates the expression of pro-inflammatory mediators and cytokines induce gastrin expression

Gastrin is a transcriptional activator that targets promoter regions of cytokines, chemokines and other pro-inflammatory mediators, such as COX-2 and IL-8. COX enzymes catalyze the synthesis of PG from arachidonic acid, which then signals its pro-oncogenic effects through the E-prostanoid receptors on the target cell [22]. COX-2 expression is rapidly induced by a vast number of inflammatory cytokines (e.g., interleukins, interferon- γ , tumor necrosis factor- α), growth factors, and oncogenes [23]. The importance of downregulating the COX-2/PG system in the prevention of cancer is suggested by multiple epidemiological studies wherein subjects who regularly use aspirin or other nonsteroidal anti-inflammatory drugs have over a 30% lower incidence of mortality from colorectal cancer or lower

incidence of adenomatous polyps [24]. *In vitro*, COX-2 inhibition has been shown to mediate inhibition of cell proliferation and induce apoptosis in many GI cancer cells lines [25]. Additionally, multiple COX-2- independent effects by the inhibitors, such as inhibition of the NF- κ B pathway [26], also play a significant role in downregulating inflammation and tumor progression.

Gastrin has been shown to mediate the induction of COX-2 in GI cells [27, 28], indicating that there is a direct mechanistic link between gastrin and inflammation. Sun et al. [29] demonstrated that combination treatment with a gastrin/cholecystokinin-2 receptor (CCK₂R) inhibitor AG-041R and selective COX-2 inhibitor NS-398 synergistically reduced cell viability (MTT assay) and cell proliferation (BrDU incorporation assay) in MKN-45 cells, a poorly differentiated gastric carcinoma cell line. These results indicate that gastrin activation of the CCK₂R induces cellular proliferation and inhibits apoptosis through both COX-2- dependent and COX-2-independent mechanisms. Additionally, Subramaniam and colleagues [30] recently showed that gastrin stimulated expression of COX-2 and IL-8 in the human gastric epithelial cell line AGS, transfected with the CCK₂R. Gastrin not only increased messenger RNA stability of both COX-2 and IL-8 in a p38-dependent manner, but also enhanced COX-2 and IL-8 gene transcription via the transcription factors activator protein-1 and NF- κ B, respectively.

Although gastrin can upregulate inflammatory mediators, evidence that COX-2 can induce the overexpression of gastrin also occurs, suggesting a feed-forward loop that characterizes the dysregulation of inflammatory molecules in cancer progression. Walduck et al. [31] compared gene expression profiles regulated by gastric mucosal epithelial cells of mice infected with *H. pylori*, which were treated with either COX-2 specific inhibitor NS298 (10 mg/kg) or vehicle. Gastrin expression was downregulated with NS298 treatment, indicating that gastrin gene regulation is COX-2-dependent. In times of cellular stress, feed-forward mechanisms may be adapted by cellular systems to enhance cellular survival, for example, during acute inflammation. Repeated insults or injuries, such as what occurs with chronic inflammation, renders the normal negative feedback mechanisms non-functional, in favor of the feed-forward mechanisms, and eventually contributes to the development of inflammation-related carcinomas.

Clinical evidence for a feed-forward loop

C-terminal amidated gastrin and biosynthetic precursors, including progastrin, has been shown to be highly prevalent in GI cancers [32]. A clinical study supports the hypothesis that gastrin and gastrin precursor peptides, systemic inflammatory mediators, and tumor growth may be mechanistically linked to COX-2. Konturek et al. [33] obtained tumor biopsies from 10 patients with rectal cancer before and 14-days after treatment with a COX-2 inhibitor, Celecoxib (CLX). Patients with elevated serum gastrin levels were excluded from the study. The authors reported that serum IL-8 and TNF- α were two- to three-fold higher compared to control patients. Concomitant with the increase in cytokines, nuclear extracts from tissue samples showed that the active form of NF- κ B, p65 protein, were also markedly increased, but decreased with CLX treatment. Serum progastrin levels in the cancer patients were also significantly higher, but decreased to levels not significantly different from the healthy subjects after CLX treatment. Before therapy, serum amidated gastrin levels, while not significantly different between the control patients, decreased significantly after CLX. The ratio of gastrin mRNA to GAPDH mRNA from tumor samples were double compared to that found in normal mucosa, and markedly decreased after CLX in the tumors, but remained unchanged in the normal mucosa. CCK₂ receptor mRNA ratios were the same in normal and tumor tissues but treatment with CLX decreased CCK₂R expression in only the cancerous tissues. COX-2 mRNA, and Bcl-2 and survivin protein

levels, both inhibitors of apoptosis, were significantly increased in the cancer patients, and then downregulated in the tumors post-CLX therapy. This clinical study supports the hypothesis that gastrin, progastrin, and the CCK₂ receptors, which are all aberrantly expressed in colorectal cancer, mediates pro-inflammatory effects in a feed-forward fashion, since its effects can be reversed with COX-2 inhibition.

The CCK₂ receptors mediate the oncogenic effects of gastrin and gastrin precursors

A recent report indicates that the actions of progastrin depend on CCK₂R, despite the fact that CCK₂R does not bind progastrin with high affinity. Jin and colleagues [34•] have shown that progastrin mediates colonic epithelial cell proliferation and colorectal cancer in the hGAS mouse model. The hGAS mice express a human preprogastrin transgene in the liver, and have elevated plasma concentrations of progastrin (1–100 nM) as well as normal plasma concentrations of amidated gastrin. When crossed with a *Cck2r*^{-/-} mouse, colonic crypt fission, proliferation and development of cancer are markedly attenuated. The hGas mouse significantly upregulated CCK₂R mRNA expression compared with the WT mice. Although the exact mechanism of action for progastrin and CCK₂R is not known, but involves the cell surface protease co-receptor annexin II [35], these data suggest clearly an important role for CCK₂R in a mouse model of colon carcinogenesis.

Progastrin can bind with glycosaminoglycans, negatively-charged proteoglycans that can be modified by heparin sulfate. Treatment of epithelial cells with heparinase reduces binding by progastrin [36•]. Interestingly, in a similar fashion, heparin-sulfate is a requirement for CXC chemokines, such as stromal derived factor (SDF)-1 α , to bind and signal through its receptor CXCR4 [37], and heparinase-treated cells render the actions of SDF-1 α inactive. Perhaps, in this way, similar to chemokines, the necessary modifications to progastrin are facilitated by either extracellular matrix proteoglycans or membrane-bound proteoglycans, which serve to immobilize and enhance presentation of gastrin peptides to CCK₂R. How progastrin may be processed are not known and further study of this hypothesis is warranted. Since the hGAS mouse also makes amidated gastrin, gastrin-induced CCK₂ receptor signaling most likely also contributes to tumor promotion in the CCK₂R-positive mice. Lastly, in the tumor microenvironment, many cell types may be CCK₂R-positive, and the contributions of immune blood cells or activated CAF must be considered.

Cellular stress induce gastrin and CCK₂ receptor expression

Expression of CCK₂ receptor may be induced under specific conditions of stress, injury, inflammation, and repair, for example, during Barrett's metaplasia [38], and γ -irradiated intestine of mice [39]. Recently, Ashurst et al. [40•] showed that serum starvation of the AGS and RGM1 cells caused increased transcription of CCK₂R promoter-reporter constructs as well as increased total CCK₂R mRNA via protein kinase C (PKC) and mitogen/extracellular signal-regulated kinase (MEK)-dependent pathways. In a model of cryoulcer injury to the mouse stomach, CCK₂R expression was induced in both the regenerating gastric mucosal epithelial cells, as well as the submucosal myofibroblasts (vimentin- and α -smooth muscle actin-positive, desmin-negative). Myofibroblasts are a subpopulation of fibroblasts with an "activated" phenotype; they mediate wound healing by increasing cytokine, growth factor and ECM production, and, in cancers, they contribute to cancer initiation, progression, and metastasis [41]. Previously, our laboratory reported that functional CCK₂ receptors were present not only on the epithelial cancer cells in 6/18 (33%) of primary colorectal cancer cells, but also on 7/18 (39%) of CAF [42]. Expression of CCK₂ receptors in both cell types underscore the potential importance of paracrine signaling by gastrin peptides in inflammation-initiated malignancies. One hypothesis that may explain the

expression of CCK₂ receptors by both epithelial cells and fibroblasts in the cryoinjury model may be that during injury or cancer, the epithelial cells acquired migratory and invasive properties similar to fibroblasts (epithelial to mesenchymal transition). Alternatively, CAF may become “activated” locally from normal fibroblasts, and/or may be recruited from the mesenchymal stem cell population from the bone marrow.

Gastrin recruits CCK₂R-expressing immune cells

Multiple reports indicate that CCK₂ receptors or their mRNAs are expressed by macrophages in the lamina propria [43], peripheral blood mononuclear cells [44, 45], and polymorphonuclear monocytes within the tumor stroma of human colorectal cancers [46]. Activation of the CCK₂ receptors by gastrin not only mediates leukocyte chemotaxis, adherence, and phagocytosis [47, 48], but at sites of inflammation, gastrin has a pro-inflammatory effect by recruiting CCK₂R-expressing immune cells. Using intravital microscopy, Alvarez et al. [49] studied flowing leukocytes on rat mesenteric venule endothelium and found increased leukocyte rolling and adhesion, decreased rolling velocity, and increased leukocyte extravasation into the interstitium. These pro-inflammatory features were observed when the mesentery was superfused with gastrin in a concentration- and time-dependent manner, and in hypergastrinemic states, such as pretreatment with omeprazole or *H. pylori* infection. CCK₂R antagonists (L-365,260 or proglumide), but not CCK₁R antagonist (devasepide) abrogated the results. The inflammatory response in the mesentery is mediated by the CCK₂ receptors localized in mesenteric macrophages and polymorphonuclear leukocytes, not the endothelium, and is not modulated by other neuropeptides (somatostatin or histamine).

To further study the pro-inflammatory effects of gastrin on immunocytes, we have recently shown that the CCK₂ receptor promoter contains a IFN- γ regulatory site [50]. IFN- γ is a pro-inflammatory cytokine that alters transcription of more than 30 genes which regulate a variety of physiological and cellular responses including: a) increase antigen presentation of macrophages, b) increase lysosome activity in macrophages, and c) promote adhesion and binding required for leukocyte migration. IFN- γ induces expression of genes containing the gamma-INF-activated site (GAS) within their promoter. The CCK₂ receptor promoter contains a GAS regulatory motif and preliminary data from our lab demonstrates that INF- γ induced expression of CCK₂ receptors in mouse bone marrow cells, circulating human leukocytes, and malignant monocytes cell lines HL-60, THP-1, and U937. Interestingly, a combination of INF- γ with lipopolysaccharide, the prototypical endotoxin from the outer membrane of gram-negative bacteria, further increased CCK₂R mRNA expression in the same cell lines. Furthermore, gastrin caused chemotactic migration of INF- γ -stimulated bone marrow cells *in vitro*, suggesting that gastrin is a chemoattractant that facilitates the recruitment of inflammatory cells to sites of gastrointestinal injury, inflammation and repair. Since immune cells such as bone-marrow derived cells and tumor-associated macrophages play key roles in both the destruction and progression of cancers from inflammatory backgrounds, future studies should be focused on: 1) when and how gastrin peptides and CCK₂ receptors are induced 2) how CCK₂ receptor-positive immunocytes facilitate cancer progression, invasion, angiogenesis, metastasis.

Conclusions

These recent studies have expanded the role for gastrin, gastrin-related peptides, and CCK₂ receptors in inflammation and inflammation-associated cancers beyond simply the epithelial-derived carcinoma cell type, and suggest that their expression in CAFs and immune cells contribute to the initiation and progression of GI cancers. Contrary to earlier interpretations that their expression in non-malignant cells were “contaminants” and not

likely to contribute to the malignant phenotype [51], the cells in the surrounding stroma are now recognized as important. Current trends warrant that future studies investigate gastrin peptides and their receptors in the context of the complex interactions that characterize the microenvironment of inflammation-associated cancers.

Acknowledgments

This work was supported by grants from the National Institutes of Health: P01 DK35608, 5 R01 DK048345 (to M Hellmich), 1K08CA125209 (to C. Chao) and the Society of Surgical Oncology Clinical Investigator Award (to C. Chao).

References and Recommended Reading

Papers of particular interest, published within the annual period of review, have been highlighted as: • of special interest, •• of outstanding interest

1. Kundu JK, Surh YJ. Inflammation: gearing the journey to cancer. *Mutat Res.* 2008; 659:15–30. [PubMed: 18485806]
2. Black JW. Reflections on some pilot trials of gastrin receptor blockade in pancreatic cancer. *Eur J Cancer.* 2009; 45:360–364. [PubMed: 19131241] A recent review on gastrin receptor antagonist in pancreatic cancer clinical trials.
3. Laverman P, Roosenburg S, Gotthardt M, et al. Targeting of a CCK(2) receptor splice variant with (111)In-labelled cholecystokinin-8 (CCK8) and (111)In-labelled minigastrin. *Eur J Nucl Med Mol Imaging.* 2008; 35:386–392. [PubMed: 17934729]
4. Mantovani A, Allavena P, Sica A, et al. Cancer-related inflammation. *Nature.* 2008; 454:436–444. [PubMed: 18650914] A recent, comprehensive review on the molecular mechanisms that link inflammation and cancer.
5. Guilford P, Hopkins J, Harraway J, et al. E-cadherin germline mutations in familial gastric cancer. *Nature.* 1998; 392:402–405. [PubMed: 9537325]
6. Correa P. Human gastric carcinogenesis: a multistep and multifactorial process--First American Cancer Society Award Lecture on Cancer Epidemiology and Prevention. *Cancer Res.* 1992; 52:6735–6740. [PubMed: 1458460]
7. Chiba T, Marusawa H, Seno H, et al. Mechanism for gastric cancer development by *Helicobacter pylori* infection. *J Gastroenterol Hepatol.* 2008; 23:1175–1181. [PubMed: 18637055]
8. Houghton J, Stoicov C, Nomura S, et al. Gastric cancer originating from bone marrow-derived cells. *Science.* 2004; 306:1568–1571. [PubMed: 15567866]
9. Smythies LE, Waites KB, Lindsey JR, et al. *Helicobacter pylori*-induced mucosal inflammation is Th1 mediated and exacerbated in IL-4, but not IFN-gamma, gene-deficient mice. *J Immunol.* 2000; 165:1022–1029. [PubMed: 10878379]
10. Schubert ML, Peura DA. Control of gastric acid secretion in health and disease. *Gastroenterology.* 2008; 134:1842–1860. [PubMed: 18474247]
11. Lehmann FS, Golodner EH, Wang J, et al. Mononuclear cells and cytokines stimulate gastrin release from canine antral cells in primary culture. *Am J Physiol.* 1996; 270:G783–G788. [PubMed: 8967489]
12. Takaishi S, Tu S, Dubeykovskaya ZA, et al. Gastrin is an essential cofactor for *helicobacter*-associated gastric corpus carcinogenesis in C57BL/6 mice. *Am J Pathol.* 2009; 175:365–375. [PubMed: 19556515] A follow-up study to reference #10. The effects of both gastrin overexpression (INS-GAS) or deficiency (GAS-KO) in *H. felis* infected or uninfected mice are examined in C57BL/6 mice. Many findings confirm the original study, which was in FVB/N mice. Differences between the two studies are discussed.
13. Wang TC, Dangler CA, Chen D, et al. Synergistic interaction between hypergastrinemia and *Helicobacter* infection in a mouse model of gastric cancer. *Gastroenterology.* 2000; 118:36–47. [PubMed: 10611152]
14. Orlando LA, Lenard L, Orlando RC. Chronic hypergastrinemia: causes and consequences. *Dig Dis Sci.* 2007; 52:2482–2489. [PubMed: 17415644]

15. Steele IA, Dimaline R, Pritchard DM, et al. Helicobacter and gastrin stimulate Reg1 expression in gastric epithelial cells through distinct promoter elements. *Am J Physiol Gastrointest Liver Physiol.* 2007; 293:G347–G354. [PubMed: 17463184]
16. Thorburn CM, Friedman GD, Dickinson CJ, et al. Gastrin and colorectal cancer: a prospective study. *Gastroenterology.* 1998; 115:275–280. [PubMed: 9679032]
17. Smith AM, Watson SA. Gastrin and gastrin receptor activation: an early event in the adenoma-carcinoma sequence. *Gut.* 2000; 47:820–824. [PubMed: 11076881]
18. Fodde R, Smits R, Clevers H. APC, signal transduction and genetic instability in colorectal cancer. *Nat Rev Cancer.* 2001; 1:55–67. [PubMed: 11900252]
19. Koh TJ, Bulitta CJ, Fleming JV, et al. Gastrin is a target of the beta-catenin/TCF-4 growth-signaling pathway in a model of intestinal polyposis. *J Clin Invest.* 2000; 106:533–539. [PubMed: 10953028]
20. Franco AT, Israel DA, Washington MK, et al. Activation of beta-catenin by carcinogenic *Helicobacter pylori*. *Proc Natl Acad Sci U S A.* 2005; 102:10646–10651. [PubMed: 16027366]
21. Franco AT, Johnston E, Krishna U, et al. Regulation of gastric carcinogenesis by *Helicobacter pylori* virulence factors. *Cancer Res.* 2008; 68:379–387. [PubMed: 18199531]
22. Chell S, Kaidi A, Williams AC, et al. Mediators of PGE2 synthesis and signalling downstream of COX-2 represent potential targets for the prevention/treatment of colorectal cancer. *Biochim Biophys Acta.* 2006; 1766:104–119. [PubMed: 16859832]
23. Hla T, Neilson K. Human cyclooxygenase-2 cDNA. *Proc Natl Acad Sci U S A.* 1992; 89:7384–7388. [PubMed: 1380156]
24. Rostom A, Dube C, Lewin G, et al. Nonsteroidal anti-inflammatory drugs and cyclooxygenase-2 inhibitors for primary prevention of colorectal cancer: a systematic review prepared for the U.S. Preventive Services Task Force. *Ann Intern Med.* 2007; 146:376–389. [PubMed: 17339623]
25. Grosch S, Maier TJ, Schiffmann S, et al. Cyclooxygenase-2 (COX-2)-independent anticarcinogenic effects of selective COX-2 inhibitors. *J Natl Cancer Inst.* 2006; 98:736–747. [PubMed: 16757698]
26. Yamamoto Y, Yin MJ, Lin KM, et al. Sulindac inhibits activation of the NF-kappaB pathway. *J Biol Chem.* 1999; 274:27307–27314. [PubMed: 10480951]
27. Abdalla SI, Lao-Sirieix P, Novelli MR, et al. Gastrin-induced cyclooxygenase-2 expression in Barrett's carcinogenesis. *Clin Cancer Res.* 2004; 10:4784–4792. [PubMed: 15269153]
28. Guo YS, Cheng JZ, Jin GF, et al. Gastrin stimulates cyclooxygenase-2 expression in intestinal epithelial cells through multiple signaling pathways. Evidence for involvement of ERK5 kinase and transactivation of the epidermal growth factor receptor. *J Biol Chem.* 2002; 277:48755–48763. [PubMed: 12239223]
29. Sun WH, Zhu F, Chen GS, et al. Blockade of cholecystokinin-2 receptor and cyclooxygenase-2 synergistically induces cell apoptosis, and inhibits the proliferation of human gastric cancer cells in vitro. *Cancer Lett.* 2008; 263:302–311. [PubMed: 18258354]
30. Subramaniam D, Ramalingam S, May R, et al. Gastrin-mediated interleukin-8 and cyclooxygenase-2 gene expression: differential transcriptional and posttranscriptional mechanisms. *Gastroenterology.* 2008; 134:1070–1082. [PubMed: 18395088]
31. Walduck AK, Weber M, Wunder C, et al. Identification of novel cyclooxygenase-2-dependent genes in *Helicobacter pylori* infection in vivo. *Mol Cancer.* 2009; 8:22. [PubMed: 19317916]
32. Aly A, Shulkes A, Baldwin GS. Gastrins, cholecystokinins and gastrointestinal cancer. *Biochim Biophys Acta.* 2004; 1704:1–10. [PubMed: 15238241]
33. Konturek PC, Rembiesz K, Burnat G, et al. Effects of cyclooxygenase-2 inhibition on serum and tumor gastrins and expression of apoptosis-related proteins in colorectal cancer. *Dig Dis Sci.* 2006; 51:779–787. [PubMed: 16615003]
34. Jin G, Ramanathan V, Quante M, et al. Inactivating cholecystokinin-2 receptor inhibits progastrin-dependent colonic crypt fission, proliferation, and colorectal cancer in mice. *J Clin Invest.* 2009; 119:2691–2701. [PubMed: 19652364] A provocative study demonstrating that the proliferative effects of progastrin are dependent on CCK₂R expression. The authors postulate that deletion of the cck2r gene decreases the colon "progenitor" cells, indicated by "stem cell" markers such as

- CD44, doublecortin and CaM kinase-like-1 (DCAMKL1), and leucine rich repeat-containing G protein-coupled receptor 5 (LgR5).
35. Singh P, Wu H, Clark C, et al. Annexin II binds progastrin and gastrin-like peptides, and mediates growth factor effects of autocrine and exogenous gastrins on colon cancer and intestinal epithelial cells. *Oncogene*. 2007; 26:425–440. [PubMed: 16832341]
 36. Dubeykovskiy A, Nguyen T, Dubeykovskaya Z, et al. Flow cytometric detection of progastrin interaction with gastrointestinal cells. *Regul Pept*. 2008; 151:106–114. [PubMed: 18674570] This paper discusses a non-radioactive, FACS-based technique that labels progastrin with biotin. A quantitative way to measure binding of tumor cells to progastrin is described.
 37. Amara A, Lorthioir O, Valenzuela A, et al. Stromal cell-derived factor-1 α associates with heparan sulfates through the first beta-strand of the chemokine. *J Biol Chem*. 1999; 274:23916–23925. [PubMed: 10446158]
 38. Haigh CR, Attwood SE, Thompson DG, et al. Gastrin induces proliferation in Barrett's metaplasia through activation of the CCK2 receptor. *Gastroenterology*. 2003; 124:615–625. [PubMed: 12612900]
 39. Ottewill PD, Duckworth CA, Varro A, et al. Gastrin increases murine intestinal crypt regeneration following injury. *Gastroenterology*. 2006; 130:1169–1180. [PubMed: 16618411]
 40. Ashurst HL, Varro A, Dimaline R. Regulation of mammalian gastrin/CCK receptor (CCK2R) expression in vitro and in vivo. *Exp Physiol*. 2008; 93:223–236. [PubMed: 17933865] CCK₂R expression is induced in both gastric epithelial cells and myofibroblasts in gastric tissue after cryoinjury to the stomach. This study demonstrates that CCK₂R may be important in mediating cellular regeneration after injury or inflammation.
 41. Kalluri R, Zeisberg M. Fibroblasts in cancer. *Nat Rev Cancer*. 2006; 6:392–401. [PubMed: 16572188]
 42. Chao C, Tallman ML, Ives KL, et al. Gastrointestinal hormone receptors in primary human colorectal carcinomas. *J Surg Res*. 2005; 129:313–321. [PubMed: 16051276]
 43. Mezey E, Palkovits M. Localization of targets for anti-ulcer drugs in cells of the immune system. *Science*. 1992; 258:1662–1665. [PubMed: 1333642]
 44. Sacerdote P, Wiedermann CJ, Wahl LM, et al. Visualization of cholecystokinin receptors on a subset of human monocytes and in rat spleen. *Peptides*. 1991; 12:167–176. [PubMed: 2052492]
 45. Schmitz F, Schrader H, Otte J, et al. Identification of CCK-B/gastrin receptor splice variants in human peripheral blood mononuclear cells. *Regul Pept*. 2001; 101:25–33. [PubMed: 11495676]
 46. Schmitz F, Otte JM, Stechele HU, et al. CCK-B/gastrin receptors in human colorectal cancer. *Eur J Clin Invest*. 2001; 31:812–820. [PubMed: 11589724]
 47. Carrasco M, Hernanz A, De La Fuente M. Effect of cholecystokinin and gastrin on human peripheral blood lymphocyte functions, implication of cyclic AMP and interleukin 2. *Regul Pept*. 1997; 70:135–142. [PubMed: 9272625]
 48. De la Fuente M, Carrasco M, Hernanz A. Modulation of human neutrophil function in vitro by gastrin. *J Endocrinol*. 1997; 153:475–483. [PubMed: 9204002]
 49. Alvarez A, Ibiza S, Hernandez C, et al. Gastrin induces leukocyte-endothelial cell interactions in vivo and contributes to the inflammation caused by *Helicobacter pylori*. *FASEB J*. 2006; 20:E1742–E1752.
 50. Kim SO, Chao C, Townsend CM Jr, et al. Interferon- γ induces CCK2 receptor expression in monocytes and bone marrow-derived cells. *Gastroenterology*. 2009; 136:A398.
 51. Reubi JC, Waser B, Schmassmann A, et al. Receptor autoradiographic evaluation of cholecystokinin, neurotensin, somatostatin and vasoactive intestinal peptide receptors in gastrointestinal adenocarcinoma samples: where are they really located? *Int J Cancer*. 1999; 81:376–386. [PubMed: 10209952]