Chapter 1

The Value of Gastrointestinal Biopsy

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Introduction

Prior to the development of the first fibre-optic endoscopes in the 1960s, gastroenterology, in common with other traditional medical specialties, relied on biochemical and radiological techniques to investigate the gastrointestinal (GI) tract. Histological confirmation of a disease process usually required the presence of a surgeon, was invasive, and carried inherent risks. Because of a rapid improvement in technology, we are now able to sample the entire GI tract from the mouth and anus through to the ileal–jejunal junction. In addition, endoscopy can now sample beyond the mucosa into the submucosa and extra-luminal structures, because of the development of endoscopic ultrasound. Therefore, in many ways the history of GI pathology has mirrored the development of GI endoscopy. With the exponential growth in the use of endoscopy since the early 1970s, there is a continually increasing need to interpret the GI pathophysiology and immunology through the lens of a 1 to 2 mm sample of mucosa embedded in formalin. Like any investigative process, dialogue between the endoscopist framing the question and the pathologist attempting to answer it is crucial. The keys to success are clear communication, meticulous specimen labelling, sufficient clinical information, and forums for feedback and discussion such as multidisciplinary meetings, research, or simply coffee and a chat. All of these combine to embed the histopathology report within the clinical environment.

This chapter sets out to explain the value of histology to a gastroenterologist or surgeon, particularly in benign disease, and sets out the latest guidelines for endoscopic sampling in the upper, mid, and lower GI tract. Finally, it aims to explain to a histopathological audience the decision-making process that endoscopists follow when performing an endoscopic procedure and, crucially, what the endoscopist needs in return in a pathology report.

What Is the Clinical Relevance of Histopathology in Benign Gastrointestinal Disease?

Endoscopists take biopsies of the GI tract for a huge variety of reasons. The indications vary depending on the experience of the endoscopist but can be broken down into three broad categories.

1. **Macroscopic disease**. An experienced endoscopist taking a biopsy usually has a reasonable idea of the pathology. This process starts before the procedure with a clinical history, examination, and perhaps biochemical or radiological investigation. In diseases such as inflammatory bowel disease (IBD), the macroscopic appearances are often so characteristic that in combination with the pre-endoscopic history and biochemical investigations the diagnosis is clear. However, there is always a differential diagnosis and the histopathological confirmation remains crucial. Importantly, a diagnosis of IBD will probably remain a lifelong label; will underpin management decisions such as immunomodulators, biologics, and surgery for years to come; and may have a profound psychosocial impact on the patient. Other benign conditions that the endoscopist may feel confident about diagnosing macroscopically are peptic ulcer disease, colonic polyps, and GI cancers.
2. **Microscopic diseases.** There are several conditions with subtly abnormal or entirely normal macroscopic appearances but with a clinical history and supplementary investigations that may point to a high level of suspicion of abnormality. For example, in mild coeliac disease the typical features of scalloping of the second part of the duodenum may be difficult for an endoscopist to detect. Although there may be a typical history of bloating, discomfort, and diarrhoea associated with the ingestion of wheat and with serological markers that make the diagnosis highly likely, the gold standard ultimately remains histological. In other conditions such as eosinophilic oesophagitis or collagenous colitis, there is no serum or radiological marker and a diagnosis is entirely reliant on the histopathologist's skill. The value of the biopsy in these conditions is even greater than in the scenario in which there are obvious macroscopic changes. Histology can play a role in both the diagnosis and monitoring of these diseases, and repeated sampling through the lifetime of the patient may be a component of management.
3. **Reassurance.** Despite the broad range of GI conditions, the most common histopathological result is likely to state that the microscopic appearances are within the normal range. Why is this the case? Endoscopy is an invasive procedure, relatively uncomfortable, costly to the provider, and carries a small but calculable risk to the patient. Symptoms have often been ongoing for many years and at the time of endoscopy the clinician and patient are looking for confirmation that the mucosa of the GI tract is both

Practice Points 1.1 Categories of Indications for Gastrointestinal Tract Mucosal Biopsy

Macroscopic Disease
- Inflammatory bowel disease
- Unexplained ulcer
- Polyp
- Cancer

Suspected Microscopic Disease
- Eosinophilic oesophagitis/enterocolitis
- Coeliac disease
- Microscopic colitis

Reassurance
- Iron deficiency anaemia
- Chronic diarrhoea

macroscopically and microscopically normal. There are several conditions in which confirmation that the mucosa is normal at a microscopic level is useful, e.g. duodenal biopsies to exclude coeliac disease in iron deficiency anaemia (IDA), or right and left colonic biopsies to exclude microscopic colitis in chronic diarrhoea. Although in both these conditions the yield from histology is likely to be relatively low, the physician and patient are looking for reassurance. Therefore, the accompanying histopathology request form should reflect this low level of suspicion.

When and where to take biopsies in different regions of the GI tract is the subject of further discussion in the text that follows (see also Practice Points 1.1).

What Does the Gastroenterologist Want from the Histopathologist?

What the gastroenterologist wants depends on the circumstances of sampling. The endoscopist should of course submit sufficient clinical information on the accompanying request form to allow the pathologist to frame the findings within the clinical context. Ideally, a pathology request form will contain a question from the endoscopist to the pathologist – rather than '? Coeliac' or, worse, simply 'D2', the clinician should write 'Raised TTG, iron and folate deficiency, macroscopically normal duodenum; does the patient have coeliac disease?' This allows the pathologist to refute or confirm the hypothesis rather than simply list a long differential for raised intraepithelial lymphocytes. What the gastroenterologist appreciates from the pathologist is to document the changes seen microscopically and to form a differential diagnosis but then, if possible, to conclude with the most likely diagnosis. At times, this can be difficult, but a report's conclusion includes, ideally, a pathologist's best guess or an idea as to what the diagnosis is, taking into account the information from the endoscopy report and/or the request form. If possible, the histological diagnosis should also consider the interpretation of previous biopsies. For example, a patchy colitis with features of chronicity may be Crohn's disease, but if there are 10 previous reports documenting ulcerative colitis then it is likely that this is simply 'treated UC', which can often have a patchy appearance. Ideally, there is a forum for ongoing dialogue once a report has been finalised. Although a multidisciplinary team meeting (MDTM) is a common forum for the management of GI malignancy, MDTMs for non-neoplastic disease such as IBD remain hugely useful, and involvement of histopathologists enhances these meetings (Practice Points 1.2).

Practice Points 1.2 What the Gastroenterologist Wants from the Pathologist

Document the microscopic features.
Offer a differential diagnosis.
Conclude with the most likely diagnosis, or with a best guess.
Take into account information from the endoscopy report and/or request form.
Consider previous biopsy findings and clinical picture.
Support multidisciplinary team meetings (MDTMs) for non-neoplastic disease.

Endoscopic Mucosal Tissue Sampling in Common Clinical Situations

Oesophagus

The oesophagus is easy to access in the majority of gastroscopies but is often negotiated rapidly en route to the stomach. Consequently, oesophageal lesions can be missed.[1] Routine biopsies from the oesophagus are not standard practice, unless there is a history of dysphagia. Sampling-specific lesions can be difficult because of the narrow confines of the oesophagus, the peristaltic movement of the oesophagus, and the quality of the stratified squamous epithelium. To help the pathologist, all samples should be labelled clearly with their site of origin. Typically, the label is the distance measured in centimetres from the tip of the scope to the incisors using the markings that run along the side of the scope. If relevant or required, further information can be given stating whether a lesion is seen on the anterior, posterior, left, or right wall of the oesophagus when the scope is in the neutral position (Fact Sheets 1.1 and 1.2).

Barrett's Oesophagus

The definition of Barrett's oesophagus (BE) is an oesophagus in which any portion of the normal distal squamous epithelial lining has been replaced by metaplastic columnar epithelium which is clearly visible endoscopically (≥1 cm) above the gastro-oesophageal junction and is confirmed histologically from oesophageal biopsies[2] (Figure 1.1). Although to a pathologist this may seem a fairly straightforward diagnosis, the presence or absence of a hiatal hernia, reflux oesophagitis, and difficulties defining the top of the gastric folds can all make establishing and classifying BE hazardous for an inexperienced endoscopist. The current guidelines encourage endoscopists to take their time assessing the mucosa, to wash the oesophagus

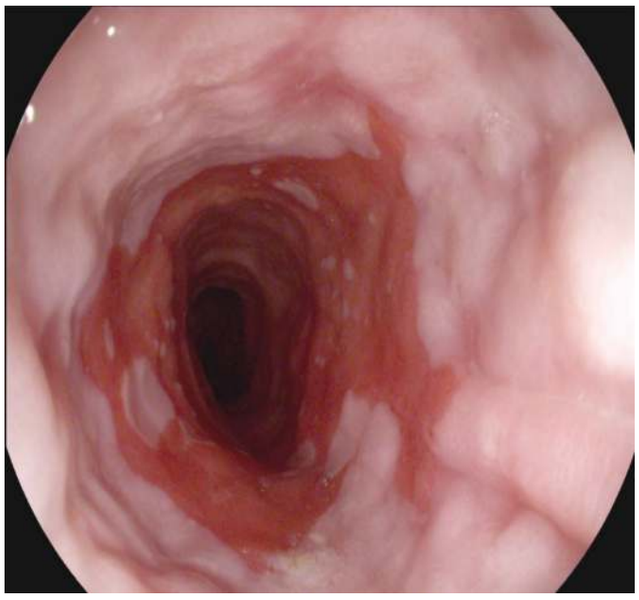

Figure 1.1 Endoscopic appearance of Barrett's oesophagus. The normal stratified squamous epithelium of the oesophagus has been partially replaced by proximally extending columnar epithelium, which is visible endoscopically.

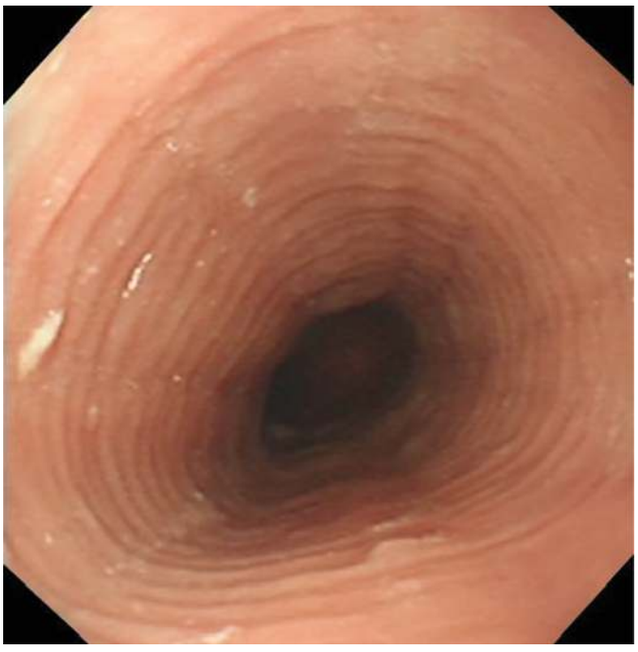

Figure 1.2 Endoscopic appearances of eosinophilic oesophagitis. There is trachealisation of the tubular oesophagus with linear furrows and white exudates, which are the hallmark endoscopic features of eosinophilic oesophagitis.

carefully, to use narrow band imaging to document the disease according to the Prague classification, and to look for obvious signs of dysplasia.[3] Suspicious areas identified as possible dysplasia should be described according to the Paris classification, sampled separately, and labelled clearly with site of origin to allow follow-up sampling. Accurate documentation guides subsequent endoscopic therapy such as endoscopic mucosal resection (EMR) and radiofrequency ablation (often documented as HALO®).[3] If no suspicious lesions are identified, then quadrantic biopsies should be taken starting from 1 cm above the top of the gastric folds and then every 2 cm, according to the Seattle protocol.[4] In patients with previous high- or low-grade dysplasia, samples at 1 cm intervals are appropriate.

Eosinophilic Oesophagitis

Eosinophilic oesophagitis (EE) is a condition diagnosed histologically and defined by the presence of >15 eosinophils per high-power field (hpf) together with other characteristic histological features in the appropriate clinical setting (see Chapter 11). Clinically it is characterised by symptoms of odynophagia, dysphagia, and, in extreme cases, food bolus obstruction.[5] Its incidence is rising through a combination of increased awareness and screening and is probably in line with overall rises in the incidence of the atopic conditions to which it is related. Endoscopically the features include trachealisation, linear furrowing, and white patches denoting immune cell aggregation, although up to 15% of patients have no endoscopic abnormalities (Figure 1.2). The sensitivity of one biopsy is 55% for detecting EE and this rises to 100% for six biopsies. Accordingly, the new British Society of Gastroenterology (BSG) guidelines suggest taking six biopsies from at least two different sites (upper, middle, or lower) when EE requires exclusion.[3]

Gastro-oesophageal Reflux Disease

Gastro-oesophageal reflux disease (GORD) is a common reason for endoscopic referral, typically to exclude Barrett's oesophagitis or cancer. The incidence of GORD varies with country, probably reflecting variations in diet, obesity, alcohol consumption levels, and smoking rates. Estimated rates are 18%–27% in North America and 8%–25% in Europe.[6] Routine biopsy is not necessary for uncomplicated GORD or for mild evidence of Los Angeles grade A–C oesophagitis. Biopsies should be performed for patients with evidence of Los Angeles grade D oesophagitis to look for the presence of dysplasia and these patients should have a repeat endoscopy 6–8 weeks after high-dose therapy with a proton pump inhibitor (PPI) to exclude Barrett's oesophagitis and to survey for dysplasia[7] (Figure 1.3).

Oesophageal Ulceration

Oesophageal ulceration is defined as a break in the mucosa of >5 mm. Biopsies should be performed for all oesophageal ulcers to look for dysplasia or malignancy and patients should have a follow-up endoscopy 6 weeks later following high-dose PPI.[8]

Benign Strictures

The finding of oesophageal strictures always raises the possibility of malignancy. If there is a suspicion of neoplasia, samples should always be taken. Other common causes of strictures are GORD, IDA, and EE. Strictures can also occur after surgery, radiofrequency ablation, radiotherapy, or endoscopic mucosal resection. There is weak evidence to suggest that even benign-looking strictures require sampling when they are first encountered (Figure 1.4). However, the sampling

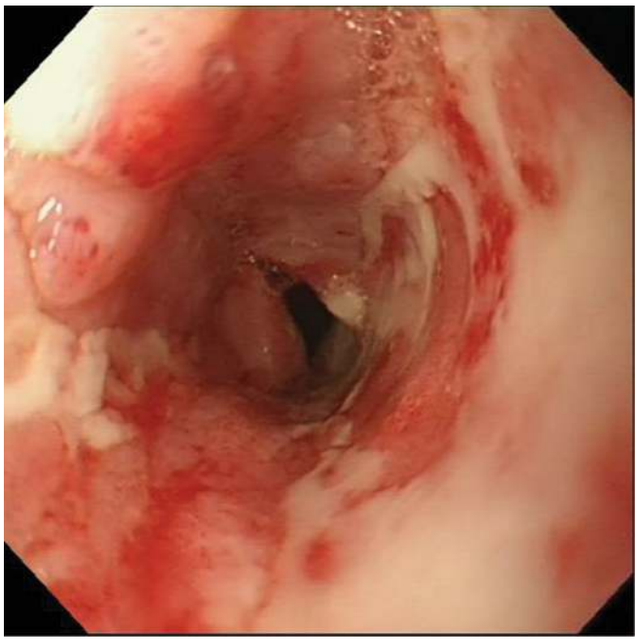

Figure 1.3 Endoscopic appearances of grade D reflux oesophagitis in the distal oesophagus. There is significant erythema involving more than 75% of the oesophageal circumference with associated superficial ulceration and friability of the oesophageal mucosa.

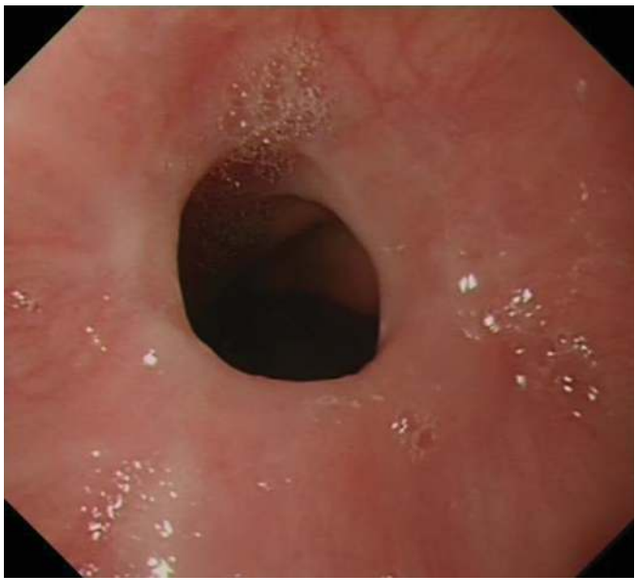

Figure 1.4 Endoscopic appearances of a benign oesophageal stricture at the gastro-oesophageal junction. The stricture has a smooth, regular contour and no depressed component. These features support a benign process.

should not precede therapeutic procedures such as dilatation because the biopsy procedure could increase the risk of perforation.[3]

Miscellaneous

Many other systemic and GI conditions can have oesophageal involvement and may require sampling. Crohn's disease, tuberculosis, human papilloma virus, candidiasis, connective tissue disorders, sarcoidosis, and Kaposi's sarcoma are all conditions that can involve the oesophagus.

Fact Sheet 1.1 Oesophageal Biopsy: General Points

Sampling-Specific Oesophageal Lesions Can Be Difficult
- Narrow confines
- Peristaltic movement
- Quality of the squamous epithelium

Routine Biopsies
- For dysphagia
- Otherwise rarely taken

Labelling
- Site of biopsy in centimetres from the incisors

Fact Sheet 1.2 Oesophageal Biopsy: Some Indications

Barrett's Oesophagus
- Narrow band imaging to document extent (Prague classification)
- Possible dysplasia is described (Paris classification) and sampled separately
- If no suspicious lesions: quadrantic biopsies from 1 cm above the gastric folds and then at 2-cm intervals
- If previous dysplasia: biopsies at 1-cm intervals

Suspected Eosinophilic Oesophagitis
- Macroscopic: trachealisation, linear furrowing, white patches
- Biopsies: at least six from at least two different sites (upper, middle, or lower oesophagus)

Gastro-oesophageal Reflux Disease
- Common reason for endoscopy; mainly to exclude Barrett's oesophagus and dysplasia
- Biopsy is unnecessary for uncomplicated GORD or mild oesophagitis
- Biopsy is usual for grade D oesophagitis to exclude dysplasia
- Repeat endoscopy 6–8 weeks after GORD therapy to exclude Barrett's oesophagus and neoplasia

Oesophageal Ulceration
- Biopsy all ulcers to exclude dysplasia or malignancy
- Follow-up endoscopy after 6 weeks following high-dose PPI.

Strictures
- Causes: GORD, IDA, EE, neoplasia, post-procedure
- Biopsy if neoplasia is suspected

Miscellaneous
- Crohn's disease, tuberculosis, human papilloma virus, candidiasis, connective tissue disorders, sarcoidosis, and Kaposi's sarcoma may require biopsy

Stomach

The stomach is a large organ, easily accessed with the endoscope. Accurate labelling of samples aids the pathologist and informs future endoscopy. Briefly stated, the stomach is divided into the cardia, fundus, body, incisura, antrum, and pylorus. Further labelling may denote anterior or posterior and lesser or greater curve (Fact Sheet 1.3).

Helicobacter pylori

Typically, initial diagnosis of *Helicobacter pylori* (*H. pylori*) infection relies on a point of care urease-based test in the endoscopy department. To avoid a false-negative result, PPIs should be discontinued 2 weeks prior to the gastroscopy.[3] If patients are taking a PPI at the time of endoscopy and there is a suspicion of *H. pylori* infection, confirmatory biopsies should be taken to look for the presence of the bacteria. Whenever possible, a urease test should be performed, as it is fast (result within a few minutes), inexpensive, sensitive, and specific.[2] For a urease test, one or two biopsies should be taken 5 cm proximal to the pylorus, in the lesser curvature near the incisura angularis or the greater curvature opposite the incisura angularis. In the presence of a PPI (that should ideally be stopped 1 week prior to endoscopy), bismuth, or antibiotics, the sensitivity of a urease test decreases dramatically.[9] In this circumstance, adherence to a biopsy protocol such as the three-biopsy protocol or the Sydney protocol may be appropriate. The three-biopsy protocol comprises one biopsy from the greater curvature of the body, one from the greater curvature of the antrum, and one from the incisura angularis. The Sydney protocol comprises five biopsies encompassing the lesser and greater curvature of the antrum (2–3 cm from the pylorus), lesser and greater curvature of the body (8 cm distal to the cardia), and incisura angularis. These protocols appear equal as regards *H. pylori* detection rates (100% from both protocols in a retrospective study of 46 patients),[10] so the three-biopsy protocol is often preferred. Typical endoscopic findings that would raise the suspicion of *H. pylori* infection include gastritis, duodenitis, and gastric and duodenal ulceration (Figure 1.5). The latest recommendations from the BSG suggest that *H. pylori* testing should also be performed in the presence of IDA.[3]

Gastric Ulcers

Peptic ulcer disease can present with central epigastric pain that improves after meals, and occasionally with anaemia and GI bleeding (haematemesis, melaena from a bleeding ulcer). The characteristics of gastric ulcers, including site, size, morphological appearance, and suspicion of malignancy, should be documented carefully (Figure 1.6). Biopsies from the edge of gastric ulcers are necessary to exclude dysplasia. Testing for *H. pylori* should be performed.[11]

Gastric Polyps

Gastric polyps, particularly fundal gland polyps, are common, the latter due in part to the rise in PPI use. Current guidelines advise a representative biopsy when polyps are first encountered to exclude malignancy and to confirm the nature of the polyp.[12] Further routine surveillance is not necessary.

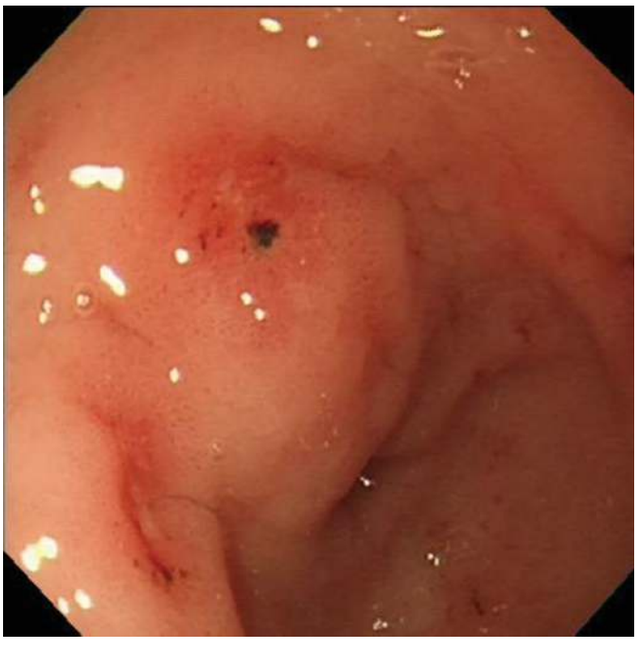

Figure 1.5 Endoscopic appearances of gastritis. There are multiple superficial erosions with relative hypertrophy of the pre-pyloric folds in the distal stomach.

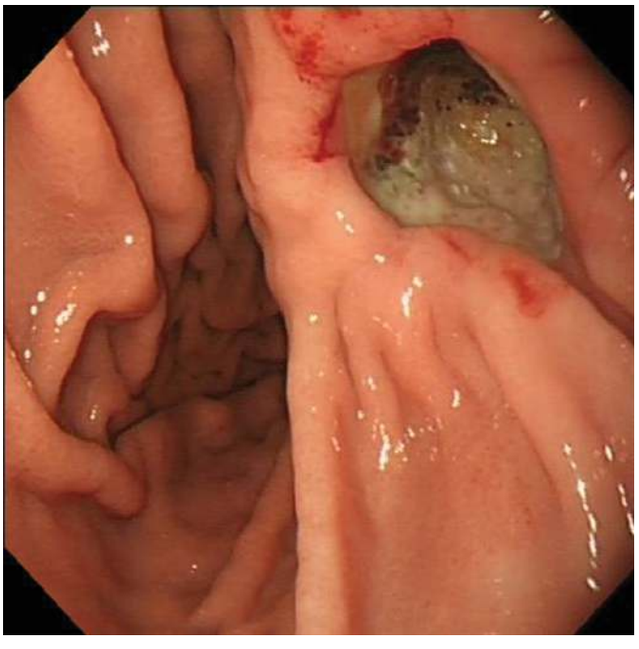

Figure 1.6 Endoscopic appearances of a cratered gastric ulcer at the incisura seen in retroflexion. The ulcer base contains a fibrin clot indicative of recent haemorrhage.

Gastric Atrophy

Gastric atrophy and intestinal metaplasia may increase the risk of gastric adenocarcinoma through the inflammation–metaplasia–dysplasia pathway (Figure 1.7). These changes can be difficult to pick up, and surveillance with both white light endoscopy and chromoendoscopy is recommended (Figure 1.8). The current Sydney protocol[13] suggests carefully labelled

targeted biopsies of suspicious lesions and two non-targeted biopsies from the antrum, body, and incisura.

Iron Deficiency Anaemia

IDA is a common reason for endoscopic referral. Current guidelines advise biopsies from the antrum and body of the stomach to exclude gastric atrophy.[3]

Miscellaneous

Numerous conditions can cause a diffuse gastric inflammation or gastritis. The most common are *H. pylori*, use of non-steroidal anti-inflammatory drugs (NSAIDs), and excess alcohol consumption. Table 1.1 lists additional, rarer causes.

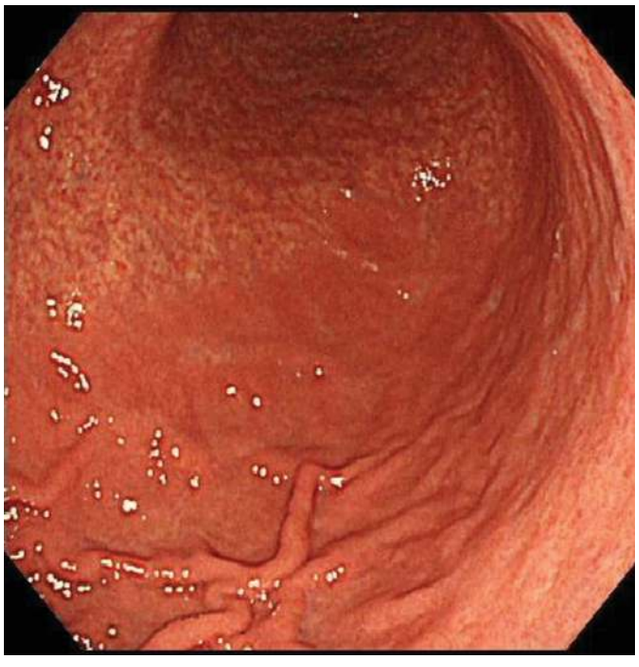

Figure 1.7 Endoscopic appearances of atrophic gastritis in the gastric body. There is a relative paucity of gastric folds with generalised pallor. Consequently, more prominent underlying vasculature is visible.

Duodenum and Proximal Small Bowel

The majority of proximal small intestinal sampling is at gastroscopy, which in most patients can easily reach the first and second parts of the duodenum and in skilled hands can extend through to the third part and potentially the fourth part. For pathology beyond the duodenum, an enteroscope is required. Enteroscopes take several forms, including a simple push enteroscope and single or double balloon scopes that can reach deep into the jejunum and potentially as far as the proximal ileum. Usually, targeted small bowel radiology or video capsule endoscopy precedes these investigations (Fact Sheet 1.4).

Duodenal Ulceration

Duodenal ulceration is commonly (approx. 70%) associated with *H. pylori* infection; other causes include NSAIDs and excessive alcohol intake (Figure 1.9). Duodenal ulcers tend to heal with PPIs and *H. pylori* eradication (if infection is confirmed).[15] If there are no malignant features in simple duodenal ulcers, follow-up gastroscopy to ensure healing is not routinely necessary. This is in contrast to the management of gastric ulcers, for which a 6-week follow-up endoscopy to ensure resolution is required.

Table 1.1 Causes of gastritis

Infectious	*H. pylori*, actinomycosis, *Mycobacterium paratuberculosis*, cytomegalovirus, syphilis
Autoimmune disease	Crohn's disease, eosinophilic gastritis, sarcoidosis, collagenous gastritis, graft-versus-host disease
Drugs	NSAIDs, alcohol, cocaine
Other	Portal hypertensive gastropathy, radiation, biliary reflux, ischaemia

Adapted from Sleisenger et al.[14]

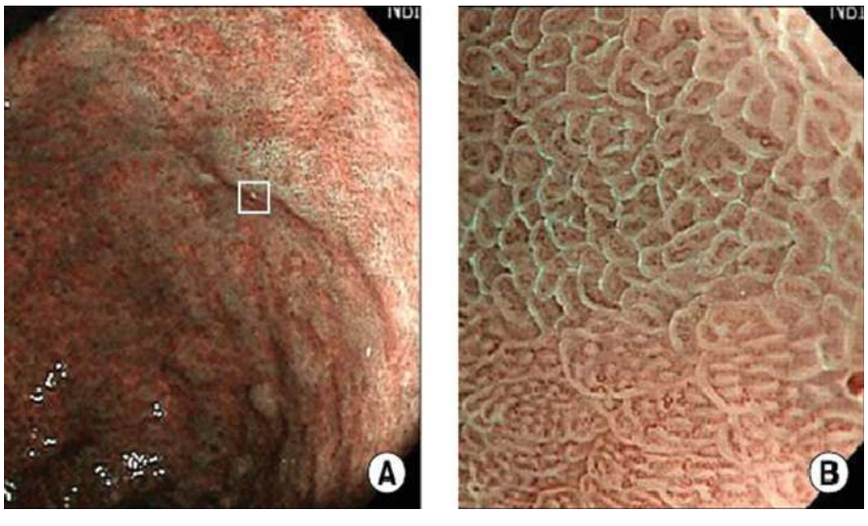

Figure 1.8 Endoscopic appearances of atrophic gastritis with intestinal metaplasia on narrow-band imaging (NBI) and zoom magnification. (A) The 'light blue crest' sign is detectable on conventional focus NBI. (B) Zoom magnification highlights the tubulovillous pits, which are the hallmark feature of intestinal metaplasia.

Fact Sheet 1.3 Indications for Gastric Biopsy

Helicobacter pylori

- Features that raise the possibility of *H. pylori* infection include gastritis, duodenitis, and gastric and duodenal ulceration
- Initial diagnosis relies on the urease test
- If there is a suspicion of *H. pylori* infection in a patient receiving a PPI, biopsies from the antrum and from above the incisura are taken

Gastric Ulcers

- Biopsies from the edge of gastric ulcers to exclude dysplasia

Gastric Polyps

- Biopsy at presentation to exclude malignancy and to confirm polyp type; further routine surveillance unnecessary

Gastric Atrophy

- Targeted biopsies of suspicious lesions plus two non-targeted biopsies from the antrum, body, and incisura (Sydney)

Iron Deficiency Anaemia

- Common reason for endoscopy
- Biopsies of antrum and body to exclude gastric atrophy

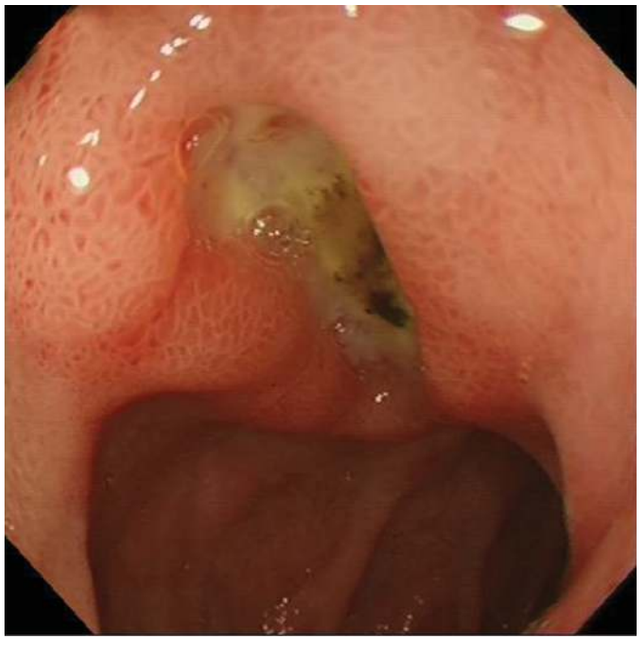

Figure 1.9 Endoscopic appearances of an ulcer on the anterior wall of the duodenal bulb. The ulcer base contains a haematin spot suggestive of recent haemorrhage.

Coeliac Disease

Coeliac disease (CD) may present with a variety of non-specific symptoms including fatigue, weight loss, bloating, and diarrhoea. Biochemical abnormalities include anaemia and micronutrient deficiencies. Diagnosis of coeliac disease is by serology and duodenal biopsies. Biopsies, while the patient is on a gluten-containing diet, remain essential for the diagnosis. Serology cannot replace biopsy.

To make a definite diagnosis of CD, villous atrophy is required. However, lesser degrees of damage (e.g. >25 intraepithelial lymphocytes [IELs] but no villous atrophy) combined with positive serology (e.g. raised tissue transglutaminase) may also represent CD ('probable CD') and in these circumstances a trial with a gluten-free diet may help support the diagnosis of CD. HLA status may also aid diagnosis (a useful test to rule out CD but not confirm it).[16]

Multiple biopsy samples taken from multiple sites help to avoid inadequate sampling of patchy disease.[17–19] In patients with suspected CD, four to six biopsy samples should be obtained from the duodenal bulb (where disease may be localised)[20–22] and more distal duodenum. Four biopsies are associated with a doubling of the diagnostic rate when compared to fewer than four biopsies.[23] Abnormal mucosa should be targeted preferentially for sampling, although microscopic disease may underlie macroscopically normal mucosa. Recent data suggest that increased IELs are the most useful finding in CD. Furthermore, assessment of this feature does not rely on good specimen orientation.[24]

There is little evidence about the relationship between re-biopsy in typical cases of CD and alterations in clinical outcome. One study (7,648 individuals) failed to show that overall mortality was increased in patients with CD with ongoing biopsy-proven villous atrophy after a median follow-up of >11 years.[25] Biopsies may be worth considering but are not mandatory if the patient with CD is asymptomatic on a gluten-free diet. However, follow-up biopsies are appropriate in patients with CD whose condition does not respond to a gluten-free diet to ensure there is not an alternative diagnosis (Fact Sheet 1.5).[17]

Miscellaneous

There are numerous histological mimics of CD in seronegative patients, and these conditions may merit further investigation in an appropriate clinical context (Table 1.2). Concurrent CD, particularly in patients with autoimmune disease, may be further investigated with serology and HLA typing.

Suspected Small Bowel Malignancy or Stricturing Disease

If a there is a suspicion that a duodenal ulcer is malignant, or there is stricturing disease, at least six to eight biopsies from the edges of the possible lesion should be taken. A stricture can be the result of a number of pathologies including coeliac disease, inflammatory bowel disease, tuberculosis, haematological malignancies (lymphoma), and GI malignancies (adenocarcinoma, neuroendocrine tumours). If diagnostic uncertainty exists, and the lesion is distal to the duodenum, a single or double balloon enteroscopy may be necessary for histological sampling. A lesion distal to the duodenum may have been visualised on previous axial imaging or capsule endoscopy (Fact Sheets 1.6 and 1.7).

Table 1.2 Conditions that mimic coeliac disease histologically

Immune disorders	Common variable immunodeficiency syndrome, glomerulonephritis, hypogammaglobulinaemia, IgA deficiency
Autoimmune disease	Autoimmune enteropathy, Grave's disease, haemolytic anaemia, Hashimoto's thyroiditis, multiple sclerosis, psoriasis, non-coeliac gluten sensitivity, protein intolerance (cow's milk, soy, eggs, peanuts, cereals), rheumatoid arthritis, Sjögren's syndrome, systemic lupus erythematosus, thymoma-associated autoimmune enteropathy, type 1 diabetes mellitus
Infection	AIDS, *Cryptosporidium* giardiasis, *Helicobacter pylori* gastritis, post-infectious diarrhoea, small intestinal bacterial overgrowth, tropical sprue, tuberculosis, viral, Whipple's disease
Drugs	Chemotherapy, non-steroidal anti-inflammatory drugs, olmesartan, mycophenolate mofetil
Neoplasia	Enteropathy-associated T-cell lymphoma, immunoproliferative small intestinal disease, refractory coeliac disease type 2, CD4 T-cell proliferation
Other	Abetalipoproteinaemia, collagenous colitis, eosinophilic gastroenteritis, glycogen storage disease, collagenous duodenitis, radiation enteritis, Crohn's disease, small bowel ischaemia

Adapted from Goddard et al.[12]

Fact Sheet 1.4 Duodenal and Proximal Small Bowel Biopsy: General Points

Gastroscopy
- Most biopsies are taken using a gastroscope
- Can reach the first and second parts of the duodenum easily
- Sometimes reaches third and fourth parts

Enteroscope
- Necessary for detection of pathology distal to the duodenum
- Simple push enteroscope versus single or double balloon scopes (can reach deep into the jejunum and potentially as far as the proximal ileum)

Fact Sheet 1.5 Coeliac Disease

- Diagnosis requires both serology and duodenal biopsies (while on a gluten-containing diet)
- Four to six biopsies from the duodenal bulb (may be localised to the bulb) and from the duodenum distal to the bulb
- Targeted biopsies of abnormal mucosa
- Follow-up biopsies if no response or poor response to gluten-free diet, to look for an alternative diagnosis

Fact Sheet 1.6 Duodenal Ulceration and Strictures

- Most duodenal ulcers (70%) are associated with *H. pylori* infection
- Urease test for *H. pylori* on one or two biopsies 5 cm proximal to the pylorus
- Biopsy for *H. pylori* if patient is on PPIs, in accordance with three-biopsy protocol or Sydney protocol
- Three-biopsy protocol:
  - One biopsy from the greater curvature of the body
  - One biopsy from the greater curvature of the antrum
  - One biopsy from the incisura angularis
- Sydney protocol:
  - Five biopsies encompassing the lesser and greater curvature of the antrum (2–3 cm from the pylorus), lesser and greater curvature of the body (8 cm distal to the cardia), and incisura angularis
- Routine follow-up gastroscopy not necessary if no malignant features

Fact Sheet 1.7 Suspected Malignant Duodenal Ulcer or Stricturing Disease

- Take six to eight biopsies or more from the edges of the lesion
- Causes of stricture include coeliac disease, inflammatory bowel disease, tuberculosis, haematological malignancies, adenocarcinoma, and neuroendocrine tumours

Terminal Ileum

Biopsies from a macroscopically normal terminal ileum rarely provide clinically relevant information and are not recommended in a routine diagnostic ileocolonoscopy.[26] However, recent European guidelines still advise terminal ileum biopsies (at least two) in patients with suspected IBD.[27] An inflamed terminal ileum is frequently associated with Crohn's disease. However, a number of conditions other than Crohn's disease can cause terminal ileitis (Table 1.3), and establishing the correct diagnosis is crucial for optimising treatment. Taking multiple (at least two) targeted biopsies from the terminal ileum may help clarify the diagnosis (Fact Sheet 1.8).

Colon

Microscopic Colitis

Microscopic colitis is a category of intestinal disorder that includes two main subtypes: collagenous colitis (CC) and lymphocytic colitis (LC). It may cause intractable diarrhoea, particularly in the elderly.

Table 1.3 Differential diagnosis of Crohn's disease terminal ileitis in clinical practice

'Backwash' ileitis	Ulcerative colitis
Intestinal infection	*Actinomyces israelii*, *Anisakis simplex*, *Cryptococcus neoformans*, cytomegalovirus, *Histoplasma capsulatum*, *Mycobacterium avium* complex, *Mycobacterium tuberculosis*, neutropaenic enterocolitis, *Salmonella* spp., *Yersinia enterocolitica*, *Yersinia pseudotuberculosis*
Lymphoid nodular hyperplasia	
Medication	Non-steroidal anti-inflammatory drug enterocolopathy
Malignancy	Lymphoma, carcinoid tumour, caecal or ileal adenocarcinoma, leiomyosarcoma
Other conditions	Vasculitis, Henoch–Schönlein purpura, amyloidosis, eosinophilic gastroenteritis, systemic mastocytosis, endometriosis

Adapted from Bojic and Markovic.[28]

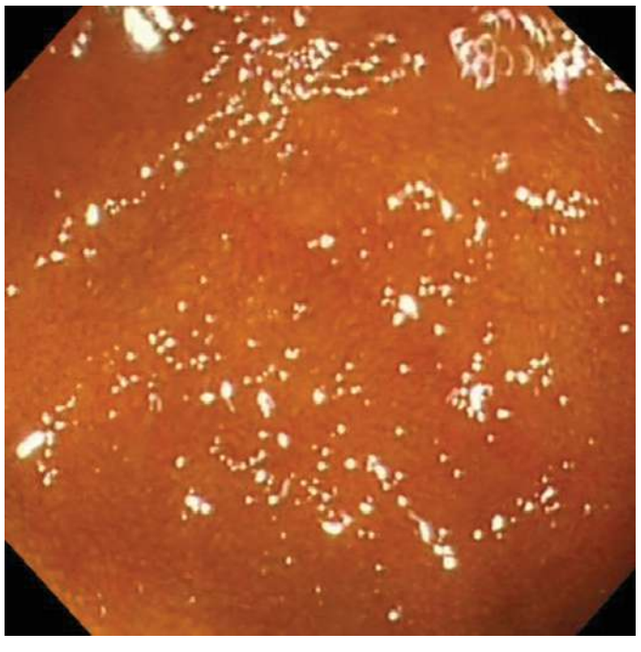

Figure 1.10 Normal macroscopic appearances of the terminal ileum visible during colonoscopy.

The potentially patchy distribution of microscopic colitis requires multiple biopsies (at least two) of the ascending and transverse colon, in addition to the descending colon, for diagnosis.[29–31] For this reason, the first-line investigation of suspected microscopic colitis should be a colonoscopy and not a flexible sigmoidoscopy (see also Chapter 20).

Inflammatory Bowel Disease: New Diagnosis

In patients undergoing an ileocolonoscopy for suspected IBD, multiple (at least two) biopsy samples should be taken from six sites (terminal ileum, ascending colon, transverse colon, descending colon, sigmoid colon, and rectum).[27] The biopsy specimens from different locations should be labelled separately to allow assessment of the extent of disease and the severity at each site.[32] Higher detection rates for granulomas can be achieved when biopsy specimens are taken from the edge of ulcers and aphthous erosions.[33]

Endoscopic evaluation with biopsies from at least one site is essential in acute severe colitis, both for diagnosis and to exclude other causes of acute colitis (e.g. cytomegalovirus colitis).[19] There remains a broad differential diagnosis for acute and chronic inflammation of the colon, which can include infection, ischaemia, vasculitis, infiltration, amyloidosis, drug reactions, and many more conditions. The endoscopist should provide sufficient macroscopic evidence such as photographs, written descriptions, and samples (see also Chapter 21).

Surveillance for Colorectal Cancer in Inflammatory Bowel Disease

Studies indicate that most dysplasia is endoscopically visible.[34,35] Targeted biopsies of all visible abnormalities should be performed. To optimise detection of such abnormalities, chromoendoscopy using methylene blue or indigo carmine with targeted biopsies is recommended.[36] When chromoendoscopy is not available (extensive active disease, multiple pseudopolyps, poor preparation), random mucosal sampling (4-quadrant biopsies every 10 cm from caecum to anus for a minimum of 33 total random mucosal samples)[19] with targeted biopsies of suspicious-appearing lesions remains a reasonable alternative, although this is inferior to chromoendoscopy for the detection of neoplastic lesions.[18] All lesions that are endoscopically resectable should be removed in their entirety if possible, and biopsy specimens of the normal mucosa surrounding the resection site should be obtained to ensure that the lateral margins of the lesion are free of dysplasia.[37]

Pouchitis

In patients who have undergone subtotal colectomy with ileal pouch–anal anastomosis and have symptoms consistent with pouchitis, pouchoscopy with biopsy is indicated to confirm the diagnosis.[38] The upper pouch, lower pouch, rectal cuff, and afferent small bowel should be evaluated (see Chapter 25). Biopsies should also be performed for abnormalities in the small bowel proximal to the pouch to ensure that there is no evidence of Crohn's disease.

Colorectal Polyps and Colorectal Cancer

If a lesion is potentially amenable to endoscopic resection, biopsies should be taken with caution as there is a risk of

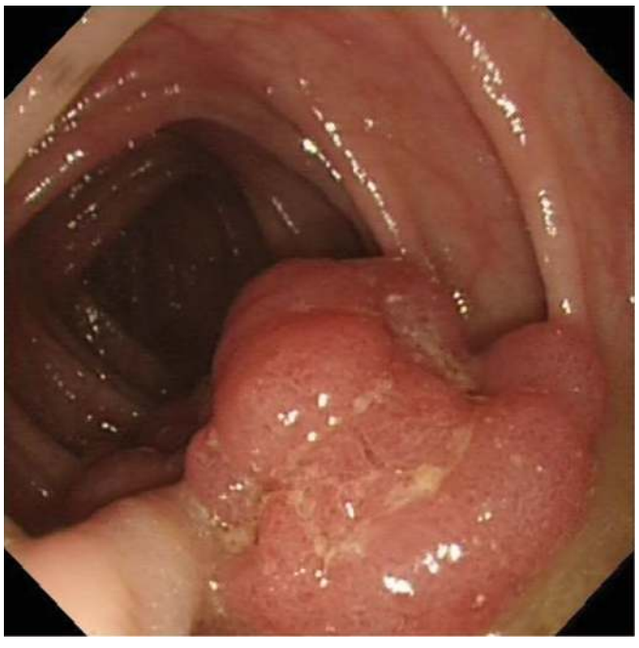

Figure 1.11 Endoscopic appearances of a pedunculated polyp in the transverse colon. Such a polyp is amenable to endoscopic mucosal resection by experienced colonoscopists.

Fact Sheet 1.8 Indications for Ileal and Colonic Biopsy

Terminal Ileum
- If macroscopically normal, biopsies are usually uninformative
- Biopsies are advisable in IBD or suspected IBD

Microscopic Colitis
- Requires at least two biopsies of ascending, transverse, and descending colon

Inflammatory Bowel Disease: New Diagnosis
- At least two biopsies from each of six sites in the ileum and large bowel
- Granuloma detection rate is higher if a biopsy is taken from the edges of ulcers and aphthous erosions

Acute Severe Colitis
- Biopsies from at least one site, for diagnosis and to exclude causes other than IBD

Suspected Pouchitis
- Biopsies from upper pouch, lower pouch, and rectal cuff to confirm and grade pouchitis
- Biopsies from afferent small bowel to exclude Crohn's disease

Gastrointestinal Acute Graft-versus-Host Disease
- Rectal or distal colonic biopsies are the most sensitive (at least four biopsies from rectum/sigmoid and at least four biopsies from left colon)
- A reasonable next investigation is gastroscopy (at least four biopsies from the body, antrum, and duodenum) or ileocolonoscopy (at least four biopsies from terminal ileum; right, transverse, and left colon; and rectum/sigmoid)

Fact Sheet 1.9 Colorectal Neoplasia

Colorectal Polyps and Cancer
- At least six to eight biopsies should be targeted to the area with features of cancer
- Avoid flat areas and lesion periphery
- Biopsies might cause submucosal tethering and consequent unresectability
- Polyps and possible cancers confined to the mucosa should be removed with margins of >1 mm

Surveillance for Colorectal Cancer in Inflammatory Bowel Disease
- Most dysplasia is endoscopically visible
- Take biopsies that target all visible abnormalities
- If chromoendoscopy is not available, random mucosal sampling (4-quadrant biopsies every 10 cm from caecum to anus with a minimum of 33 random samples) plus targeted biopsies of suspicious lesions
- Endoscopically resectable lesions require complete removal, with biopsies of the non-lesional mucosa surrounding the resection site to ensure that the margins of the lesion show no dysplasia

submucosal tethering due to scarring, rendering the lesion unresectable. Where multiple biopsies are required because of a clinical concern of cancer (at least six to eight), they should target the area exhibiting endoscopic features indicative of cancer. Flat areas and the lesion periphery should be avoided.

As a general principle, biopsies should not be taken from polyps and possible malignant growths confined to the mucosa. Instead, these lesions should be removed in their entirety by appropriately experienced colonoscopists (Figure 1.11). Resection margins should be more than 1 mm from the lesion, and some authors believe that an ideal margin should be at least 2 mm away.[39] Lesions <20 mm in maximum dimension should be removed en bloc if possible, so as to enable more accurate histopathological interpretation. Piecemeal resection should be avoided if malignancy is suspected (Fact Sheet 1.9).[40]

Miscellaneous

Gastrointestinal Acute Graft-versus-Host Disease

Two small prospective studies identify rectal or distal colon biopsies as the most sensitive test for diagnosis of GI acute graft-versus-host disease, even in patients presenting with primarily upper GI symptoms (at least four biopsies from rectosigmoid, and at least four biopsies from left colon).[41,42] If diagnostic and clinical suspicion remains high, a reasonable next investigation is a gastroscopy, with at least four biopsies from the body, antrum, and duodenum. An alternative is ileocolonoscopy, with at least four biopsies from terminal ileum; right, transverse, and left colon; and rectum/sigmoid.

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