

and sustentacular elements inserted among neuroendocrine elements. The intervening stroma consists of loose connective tissue, which contains neuronal axons, related to the ganglion cells and demonstrated with Bodian's silver technique. Amyloid deposition has been reported in a few cases.

### Electron microscopy

Ultrastructural analysis of the gangliocytic paragangliomas of the duodenum reveals cell types comparable to those described by light microscopy. The neuroendocrine cells contain numerous cytoplasmic secretory granules, which are membrane-bound and which have cores of variable, generally high, electron density. Some microfilaments are seen within these cells, generally arranged in bundles. The ganglion cells, in addition to containing abundant rough endoplasmic reticulum, show the presence of neurotubules and microfilaments, together with scattered vesicles. Combined neurotubules and vesicles are also found in axons emanating from the ganglion cells. The Schwann and sustentacular cells are characterized by slender cytoplasmic projections with an electron dense background containing large numbers of intermediate filaments.

### Immunocytochemistry

Numerous immunocytochemical studies have been carried out on these lesions and the results are often contradictory. The generic marker, neuron-specific enolase, is positive in the paraganglionic cells and neurons, whereas chromogranin A has been reported as positive by some and negative by others in both types of cells. Neurofilament antibodies are positive in neurons and their axons; their immunoreactivity in other cell types is, again, controversial. The interstitial cells are always positive for 5-100 protein, which is absent from the other two cell types. Numerous peptides have been investigated and detected by different authors, mostly inside paraganglionic cells and also in occasional neurons. Whereas the list of peptides reported to be contained in individual paragangliomas is very long, the most commonly found seem to be pancreatic polypeptide, somatostatin and, with lesser frequency, serotonin. The enormous variation in the immunocytochemical profiles found in the different reports may reflect in part the heterogeneity of these lesions as well as differences in technical capabilities among the authors.

### Differential diagnosis

When the complete lesion of an excision specimen is observed under the microscope, the combination of the different cellular components described above is quite typical and the diagnosis is easily made. However, when the pathologist is presented with a small endoscopic biopsy, it is quite possible that a given sample may lack the neuronal elements that characterize the gangliocytic paragangliomas, and a differential diagnosis may need to be made with a carcinoid tumor of the area. Immunocytochemical investigation with S-I 00 protein antibodies is a valuable tool, since gangliocytic paragangliomas contain immunoreactive sustentacular cells, absent from carcinoid tumors.

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## Duodenal lesions in patients with polyposis

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### Introduction

"Polyposis" refers to well-defined syndromes in which the primary feature is the presence of multiple polyps. In the commoner 'polyposis syndromes', such as familial adenomatous polyposis (FAP) and juvenile polyposis (JP), the definition is primarily associated with the number of polyps and an appropriate clinical setting with or without molecular confirmation. The major polyposis syndromes recognized to date include FAP, JP, Peutz-Jeghers syndrome, metaplastic polyposis, Cronkhite-Canada syndrome, Cowden's syndrome and benign and malignant lymphoid polyposis. The duodenum can be involved in several of these.

### **Familial adenomatous DOLyDOSiS**

FAP occurs approximately once in 8,000 births. The cardinal feature is the presence of multiple adenomatous polyps throughout the colon. The polyp-cancer sequence is accelerated in this syndrome. However, patients with FAR develop only a limited range of internal malignancies, particularly colorectal and duodenal cancer.

Polyps found in the duodenum in patients with FAP are "hyperplastic polyps" and "adenomas" (1). In a prospective study of 100 patients (mean age 34 years; range 13-73 years; 48 male) upper gastrointestinal endoscopy revealed adenomatous polyps in the duodenum in 33. The incidence appears higher in older patients (age 20: 2/14 adenomatous polyps; age 21-30: 6/33; age 31-40: 11/25; age >40: 14/28) (2). This is also true for the severity of the lesions graded according to Spigelman's criteria (3). There seems also to be a higher incidence in patients with stigmata of Gardner's syndrome.

Duodenal adenomas occur mainly in the second part but may involve the first and third part of the duodenum. They tend to have a sessile, irregular shape with a nodular or granular surface. A consistent feature is the whitish coloration in contrast to the normal darker color of the duodenal mucosa. The endoscopic appearance may be subtle. Microadenomas have indeed been reported on biopsy in the absence of visible polyps. Abnormalities of the papilla of Vater are perhaps the most difficult to recognize. The involved papilla may look normal or slightly larger, irregular or abnormally white. The number of polyps may vary from a single lesion to gross carpeting. Histologically, duodenal adenomas may be tubular or predominantly villous in character. Villous adenomas are composed of a thin fibrovascular core and crowded high columnar epithelium. Paneth cells can be present scattered all along the crypts, singly or in small clusters (and, more unusually, localized at the bottom of the crypts). Brunner's glands may be present in the endoscopic biopsies of these lesions and can appear prominent and associated with mild cystic dilatation.

There is good evidence supporting the existence of the adenoma-carcinoma sequence in the duodenum of patients with FAR; however the low incidence of duodenal cancer in FAR suggests that the adenoma-carcinoma sequence requires a longer time in this location. A lifetime risk of 3-4% of developing duodenal cancer, especially periampullary cancer by the age of 70 was found in a study of Dutch and Danish families (4). In the general population the estimated incidence of periampullary malignancy is between 0.1% and 0.4%. Biopsies are often insufficient to determine whether invasive carcinoma is present or not. They should be multiple and sample the central portion of the lesion, since it is there that malignancy first develops. Stromal changes are helpful: invasive carcinoma evokes a desmoplastic stromal response different from the loose lamina propria.

The severity of the duodenal lesions can be assessed using different methods. A staging system for duodenal polyposis has been designed whereby the lesions were subdivided according to the polyp number, size and histological type, as follows: number: 1-4 polyps = 1 point; 5-20 polyps = 2 points, >20 polyps = 3 points; size: 1-4mm = 1 point; 5-10mm = 2 points; >10mm = 3 points; histology: tubular/intlammation = 1 point; tubulovillous = 2; villous = 3; dysplasia: (mild = 1, moderate = 2, severe = 3). An overall score of 0 points is stage 0. Stage I corresponds to a score of 1-4. Stage II is between 5-6, stage III is 7-8 and stage IV is a score of 9-12. The staging system depends partly on histology and the number of biopsies is important for an accurate view. The ideal number of

biopsies that should be taken is unknown but 10 from patients with visible polyps and at least six from those without polyps has been suggested. This should include biopsies taken in the region of the papilla of Vater because villous changes in apparently normal mucosa at that site were observed.

More accurate predictive information may be yielded by studies at the molecular level. In a study of endoscopic biopsy specimens of 152 duodenal adenomatous polyps (taken from 79 patients) and from 13 surgically resected adenomatous polyps or cancers (taken from nine patients) overexpression of p53 (immunohistochemistry) was observed in 0% of normal mucosa (n=29), 25% of tubular adenomas (28/111), 72% of tubulovillous or villous adenomas (13/18) and 100% of duodenal cancers (7/7). It seems thus that overexpression of p53 increased with passage down the adenoma-carcinoma sequence and that it might be an additional marker of risk for duodenal malignancy (5). The frequency of *K-ras* mutation in duodenal adenomas in FAR patients is between 7% and 25% and is consistent with results found in colonic adenomas. Increased numbers of epidermal growth factor immunoreactive Paneth cells have been found in duodenal polyps from patients with FAP and it has been suggested that these might play a role in the growth of the polyps.

Endoscopic surveillance suggests that most patients undergo little change over a period of 40 months to 7 years. It has been suggested that upper gastrointestinal endoscopy every 3 years is safe. Treatment with Sulindac, a nonsteroidal antiinflammatory drug, seems to be of little or no benefit in the control of periampullary polyps in FAR.

### **Peutz-Jeghers syndrome**

Peutz-Jeghers syndrome (PJS) is a familial autosomal dominant disorder defined by the association of gastrointestinal polyposis with mucocutaneous pigmentation, especially in and around the mouth. In PJS, most polyps develop in the jejunum and ileum and in decreasing order of frequency in colon, stomach, duodenum and appendix. The duodenum is involved in 11% of cases. The polyps are rarely as numerous as in adenomatous polyposis coli and, even in familial cases, may be single. They may be responsible for obstructive symptoms, including biliary obstruction.

Morphologically, the lesions have a coarsely lobulated appearance. Histologically, the polyps have a central core of smooth muscle that shows conspicuous branching, each branch being covered by epithelium which usually appears normal and less commonly mildly hyperplastic. The epithelium is composed of goblet cells, absorptive cells, endocrine cells and Paneth cells. Occasionally, foci of gastric surface-type metaplasia can be found. Crypt elongation and cystically dilated glands are often present. There is normal maturation of the epithelial cells. The lamina propria appears normal. Larger polyps (>3 cm) may show features of pseudoinvasion. Cytonuclear atypia is not a feature of classical Peutz-Jeghers polyps (6). Four cases of duodenal malignancies were reported in a series of 72 patients with PJS.

### **Cowden's syndrome**

Polyps are characterized by cystically dilated glands and fibrosis of the lamina propria, with or without smooth muscle fibers, adipose tissue or hyperplastic lymphoid tissue. The duodenum is only rarely affected (8).

## Juvenile DOIVDOSIS

Juvenile polyps of the duodenum are rare and nearly always occur in the context of a more widespread gastrointestinal juvenile polyposis. Clinical features and neoplastic risk are less significant, although there is a case report of a duodenal carcinoma occurring in a patient with familial juvenile polyposis. Most of the polyps resemble the more common sporadic juvenile polyp of the colon, being round and smooth-surfaced pedunculated lesions. They consist of cystically dilated, small intestinal-type glands, lined by unremarkable goblet cells, absorptive cells, endocrine cells and Paneth cells. The background is an exuberant, rather inflamed lamina propria, devoid of smooth muscle (7).

## Cronkhite-Canada syndrome

Cronkhite-Canada syndrome is characterized by multiple polyps of the entire gastrointestinal tract in association with characteristic skin and nail changes. It is an extremely rare acquired sporadic condition. Gastrointestinal involvement is often revealed by protein and electrolyte loss. Duodenal polyps are found in 75% of patients and tend to be smaller in number and size than those of the stomach. They show superficial similarities to JP polyps but are usually small and more diffuse and can be differentiated easily by the associated ectodermal changes.

Microscopy shows normal villi but also flat areas with prominent crypt openings. There is flattening of the epithelial surface cells. The crypts can be elongated, irregular and cystic. The cysts are lined by attenuated epithelium. A mixed inflammatory infiltrate can be observed in the lamina propria which is edematous and thickened (9).

## Malignant lymphomatous (lymphoid) Polyposis and benign lymphoid polyposis

Lymphomatous polyposis (Mantle cell type malignant lymphoma) is an uncommon disease which may affect any part of the gastrointestinal tract. Macroscopically the mucosa shows multiple, fleshy polyps of variable size. The smallest lesions consist of a single mucosal lymphoid nodule, diffusely replaced by lymphoma sometimes with preservation of the reactive follicle center. It must be distinguished from reactive lymphoid hyperplasia.

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## Extrahepatic biliary lesions

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### Gross anatomy and histology

The extrahepatic biliary tree includes the common hepatic duct emerging from the combined right and left hepatic ducts and continuing in the common hepatic duct after the fusion with the cystic duct. The common hepatic duct is about 5-6 cm long and drains through the papilla into the duodenum. The gallbladder is about 10 cm long and shows considerable variation in size, depending on the amount of bile present. The surface epithelium of the gallbladder is composed of a single layer of tall columnar cells with basal oriented nuclei. In addition, pencil-like and basal cells can be observed. The muscular layer is composed of loosely arranged bundles of smooth muscle fibers that do not show a well-formed layer. The muscle bundles are separated by fibrovascular connective tissue. Herniation of the mucosa into the smooth muscle or the subserosal tissue is common and named Rokitsky-Aschoff sinuses. The adventitia, composed of connective and fatty tissue, vessels and nerves, is covered by serosa on the abdominal side. On the hepatic side, bile ducts (Luschka's ducts) are present in the subserosal connective tissue. The extrahepatic biliary tracts are lined by a single layer of tall epithelia similar to the gallbladder. The epithelium invaginates into the stroma forming the sacculi of Beale, which are surrounded by mucinous glands. The dense subepithelial stroma of the common bile duct usually shows no muscle fibers except in the intrahepatic and intraduodenal portion.

### Malformations

Malformations of the gall bladder are relatively rare and include the angulation of the fundus (Phrygian cap), septations, hourglass gallbladder, cysts, congenital diverticula and hypoplasia. Agenesis, with and without concomitant extrahepatic biliary atresia, as well as duplication or triplication, can occur. Although many of these malformations are clinically irrelevant, they are more often associated with gallstones. A variety of different types of bile duct cysts have been described, which occur either as solitary extrahepatic cysts, especially in the common bile duct, or as multiple intra- and extrahepatic cysts.

### Congenital extrahepatic biliary atresia

Congenital extrahepatic biliary atresia is a serious digestive disease in infants, defined by a localized obliteration of the extrahepatic bile duct at any point from the porta hepatis to the duodenum. The obstruction of bile flow leads to chronic cholestasis with rapidly progressive fibrosis and cirrhosis associated with common complications, including portal hypertension and even hepatocellular carcinoma. The incidence of biliary atresia has been estimated as one in 10,000 live births worldwide. However, epidemiological differences have suggested that environmental as well as genetic factors may be involved in the pathogenesis of the disease, including infections with cytomegalovirus and *Reovirus* 3. None of these have, however, been unequivocally associated with biliary atresia and it has been suggested that this disease may have several causes. Two forms have been distinguished. The postnatal form