Histopathological and Immunophenotypical Features of Intestinal-Type Adenocarcinoma of the Gallbladder and its Precursors
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Background: Intestinal-type adenocarcinoma of the gallbladder is an unusual malignancy associated with low- and high-grade intraepithelial neoplasms. The literature on the clinicopathologic characteristics of the precursor lesions of gallbladder cancer is limited, due in part to the variability in its definition and terminology.

Methods: Here we report one case of intestinal-type adenocarcinoma of the gallbladder with distinctive morphology and associated precursor lesions. All of the hematoxylin and eosin stained slides were reviewed. Immunostains were performed using the avidin–biotin complex method for CK20, CK7, CDX2, MUC1, MUC2, and MUC-5AC. We also reviewed the literature discussing the current terminology from the World Health Organization for these lesions.

Results: A 70-year-old man presented with epigastric abdominal pain and bloating. Computed tomography demonstrated a large heterogeneous gallbladder mass. Macroscopically, the gallbladder was 7.5 × 5.5 × 4.5 cm with smooth serosa. The lumen was occupied by a 5.0 × 4.5 × 3.0 cm irregular friable exophytic mass. The remaining mucosa had a tan brown to pink color with granular/papillary excrescences of up to 0.7 cm in thickness. Histologically, the tubulopapillary adenoma was lined by pseudostratified columnar epithelium with low and extensive high-grade dysplasia. Goblet cell and cystic dilatation were present in some glands. Immunohistochemistry showed that the intestinal type was positive for CK20, CK7, and CDX2, focally positive for MUC1/2, and negative for MUC-5AC.

Conclusion: This case showed the complete spectrum of the progression of intestinal-type intracholecystic papillary-tubular neoplasms of the gallbladder.

Introduction
Gallbladder carcinoma is a relatively uncommon neoplasm that has geographical and ethnic variations in its incidence. In the United States, it is more common in American Indians and Hispanic Americans than in Caucasians or African Americans.¹,² There is a female predominance, with the female-to-male ratio being 3–4:1.³ Most patients diagnosed with gallbladder carcinoma are in the sixth or seventh decade of life.⁴ Important risk factors for the disease include genetic backgrounds, gallstones, and abnormal choledochopancreatic junctions.⁴ The signs and symptoms are not specific, often resembling those of chronic cholecystitis. Right upper-quadrant pain is common. If the tumor is located in the gallbladder neck or its duct, then obstructive jaundice may present clinically. Gallbladder carcinoma usually forms an infiltrating, grey-white mass. Some carcinomas cause diffuse thickness of the gallbladder wall, while some present as polypoid or adenomatous in appearance. Most cases are not detectable on gross examination.⁵ Histologically, most gallbladder carcinomas exhibit pancreatobiliary-type epithelium, while others are composed of intestinal-, gastric focular-, or gastric pyloric-type epithelium. According to the most recent World Health Organization (WHO) classification, the precursor lesions of gallbladder epithelial tumors are adenoma, biliary intraepithelial neoplasia, intracystic or intraductal papillary neoplasm, and mucinous cystic neoplasm.⁶ Here we report one case of intestinal-type adenocarcinoma of the gallbladder with distinctive morphology, and we will discuss the histopathological and immunophenotypical features as well as the precursors and prognosis of the disease.
Materials and Methods
A 70-year-old man presented with epigastric abdominal pain and bloating. He was subsequently diagnosed and treated with intestinal-type adenocarcinoma of the gallbladder. His clinical, radiological, and pathological data were retrospectively reviewed following the research guidelines of the University of South Florida and the H. Lee Moffitt Cancer Center in Tampa, Florida. The tissue was processed according to the guidelines of the College of American Pathologists. The hematoxylin and eosin stain and immunohistochemical (IHC) studies were performed at the Moffitt Cancer Center, and the IHC staining was carried out with the Discovery XT System (Ventana Medical Systems, Tucson, Arizona) as per the manufacturer’s protocol.

Results
Clinical Information
Serum tumor markers, including total bilirubin, alkaline phosphatase, and carbohydrate antigen 19-9, were negative or within reference range. Computed tomography demonstrated a large, heterogeneous gallbladder mass. Cholecystectomy and lymphadenectomy were performed.

Gross Examination of the Tumor
Macroscopically, the enlarged gallbladder was 7.5 × 5.5 × 4.5 cm in size with smooth serosa. The cystic duct had a 0.1-cm luminal diameter at its opening. The lumen was occupied by a friable exophytic mass 5.0 × 4.5 × 3.0 cm in size that was irregularly pink to tan in color. The rest of the mucosa had a tan brown to pink color with granular/papillary excrescences that were up to 0.7 cm in thickness. In addition, the lumen also contained a 3.0 × 2.5 × 2.5 cm irregular frambesiform calculus that was green-brown in color.

Histology and Immunohistochemical Studies
Histologically, the exophytic mass had a tubulopapillary adenomatous appearance with a large base attached to the mucosa (Fig 1A). The exophytic lesion was covered by a pseudostratified columnar epithelium.

Fig 1. — (A) Tubulopapillary adenomatous appearance of the tumor. H&E stain, ×12.5. (B) The lesion has low- to high-grade dysplasia and focal invasion. H&E stain, ×40. (C) Surrounding microadenomatous satellite. H&E stain, ×12.5. (D) CK7 IHC stain was positive in the cytoplasm of the tumor. CK7 stain, ×100. (E) CDX2 IHC stain was positive in the nuclei of the tumor. CDX2 stain, ×100. (F) MUC1 IHC stain was positive in the cytoplasm of the tumor. MUC1 stain, ×100. H&E = hematoxylin and eosin, IHC = immunohistochemical.
Adenomas of the gallbladder are an uncommon, benign epithelial neoplasm with a low incidence that ranges from 0.14% to 1.1% in different series. Most are single, small in size (< 2 cm), and are associated with cholelithiasis. Those measuring 1 cm or more are considered to be more frequently associated with cancer, and they are classified as being either tubular, papillary, or tubulopapillary according to the growth pattern and divided into pyloric, intestinal, foveolar, and biliary types.

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In 2010, the WHO introduced intracystic papillary neoplasms into the classification of neoplasms of the gallbladder, a term that included pancreatobiliary and intestinal phenotypes. These lesions were previously designated as being papillary adenoma or noninvasive papillary carcinoma. Although intracystic papillary neoplasms have more mitotic figures, greater architectural complexity, and cytological atypia than conventional adenomas, no specific and/or quantitative criteria distinguish them. Therefore, Adsay et al. proposed the term intracholecystic papillary-tubular neoplasms (ICPNs) of the gallbladder, a term that parallels the neoplasms present in the pancreas and ampullae (intraductal papillary mucinous neoplasms, intraductal tubulopapillary neoplasms, and biliary intraductal papillary neoplasms). Classifying a lesion as an ICPN requires that the lesion be an exophytic (papillary or polypoid) intramucosal gallbladder mass that measures 1.0 cm or more and is composed of preinvasive neoplastic (dysplasia) cells that form a compact lesion distinct from neighboring mucosa. This definition includes adenomas and intracystic papillary neoplasms and is recognized by the latest WHO classification. Our case showed the complete spectrum of the progression of intestinal-type ICPN with focal invasive carcinoma.

In the study by Adsay et al., invasive carcinoma arose in 55% of ICPNs at the time of diagnosis; by contrast, 6.4% of all invasive carcinomas of the gallbladder contained an ICPN component. These results indicate that ICPNs are precursors of gallbladder carcinogenesis; however, it is worth noting that they are rare lesions. In addition, ICPNs had a significantly better prognosis than pancreatobiliary-type carcinomas of the gallbladder, even when they harbored invasive carcinoma. The median survival rates were 35 months for ICPNs with invasive carcinoma versus 9 months for pancreatobiliary-type carcinomas of the gallbladder, a survival difference that was independent of tumor size and stage. These data suggest that ICPN-associated invasive carcinomas may have a distinctive biological nature.

The literature on the clinicopathological characteristics of intestinal-type tumoral intraepithelial neoplasms is fairly limited. Fewer than 50 cases have been reported. Recently, 2 large series describing adenomas or ICPNs showed that the histology of intestinal-type lesions are similar to colonic adenomas or intestinal-type intraductal papillary mucinous neo-
Immunohistochemically, these lesions are 100% CK20+, 75% to 78% CDX2+, 33% to 50% MUC2+, 25% MUC1+, and 0% MUC-5AC+. High-grade dysplasia/carcinoma in situ was recognized in 46% (13/28) of intestinal adenomas. Invasive carcinoma was seen in 3.5% (1/28) of intestinal adenomas compared with 60% (6/10) of ICPNs, which may be due to the difference of definition. No deaths were observed.\textsuperscript{20,21}

Conclusions
Invasive adenocarcinoma that arises from a tumoral intraepithelial neoplasm appears to have a better overall clinical outcome than pancreatobiliary-type adenocarcinomas unaccompanied by ICPNs and presents with unique biological properties. The unified concept of adenomatous precursor lesions will help us better understand the nature of gallbladder neoplasms using the current terminology. Further studies are warranted, including those that focus on predictive factors, pathogenesis, and molecular genetic alterations.

References