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## Report of a case with gallbladder carcinoma: P53 expression of the peritumor epithelium might predict biliary tract recurrence



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### ABSTRACT

**INTRODUCTION:** The over-expression of P53 protein in gallbladder carcinoma is a biomarker correlating with a poor survival. However, the significance of P53 expression in peritumor tissues is unknown. We experienced a case of gallbladder carcinoma where the operative specimen showed over-expression of P53 on the peritumor epithelium, and early recurrence developed at the biliary tract.

**PRESENTATION OF CASE:** A 74-year-old female patient was referred to our hospital due to wall thickening of the gallbladder on ultrasonography. Radiographic examinations revealed wall thickening at the fundus of gallbladder and no abnormalities of the biliary tract or surrounding lymph nodes. We performed open cholecystectomy and lymph node dissection without extrahepatic bile duct resection, as a frozen section of the surgical stump of the cystic duct was cancer-free. However, a pathological examination revealed over-expression of P53 protein in the epithelium of the peritumor to the cystic duct, which were diagnosed as normal on hematoxylin eosin staining.

The patient developed bile duct metastases, two and half years after the operation. She underwent endoscopic stenting for the obstruction of bile duct with no additional therapy, and died 6 months later. **DISCUSSION AND CONCLUSION:** The immunohistochemical staining of the GB wall or surgical stump for a surgical specimen of GBC may be crucial to predict the bile duct recurrence.

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## 1. Introduction

The incidence of gallbladder and bile duct cancer in Japan was approximately 21,000 in 2011, and the cancer mortality was 18,000 in 2014 [1]. The prognosis of gallbladder carcinoma (GBC) is poor compared with other bile duct cancers, and the 5-year survival rate of surgical resection is reported to be 40% [2].

However, the carcinogenesis of GBC has not yet been fully elucidated, and recent advancements in next-generation sequencing (NGS) analyses have revealed potentially influential mutated genes in GBC [3]. Among these genes, the significance of P53 mutation has been intensively evaluated [4,5].

TP53 is a tumor suppressor gene and its mutations are frequently observed in various cancer, such as colon cancer [6], breast cancer [7], and biliary cancer [3]. The overexpression of P53 protein

has been examined by immunohistochemical staining and found to correlate with poor survival [8,9]. However, the significance of P53 expression in peritumor tissues is unknown.

We herein report a case of GBC where the operative specimen of peritumor epithelium showed overexpression of P53, and early recurrence developed at the biliary tract. This case raised the issue of dealing with precancerous lesions of the gallbladder.

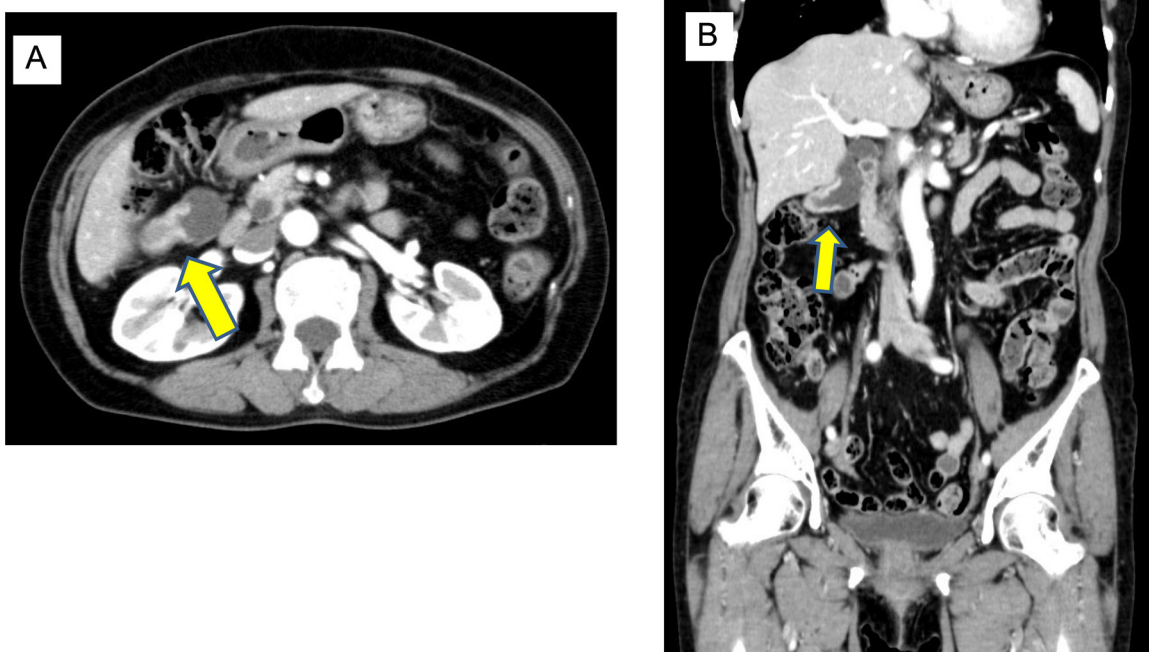
The work has been reported in line with the SCARE criteria [10].

## 2. Case presentation

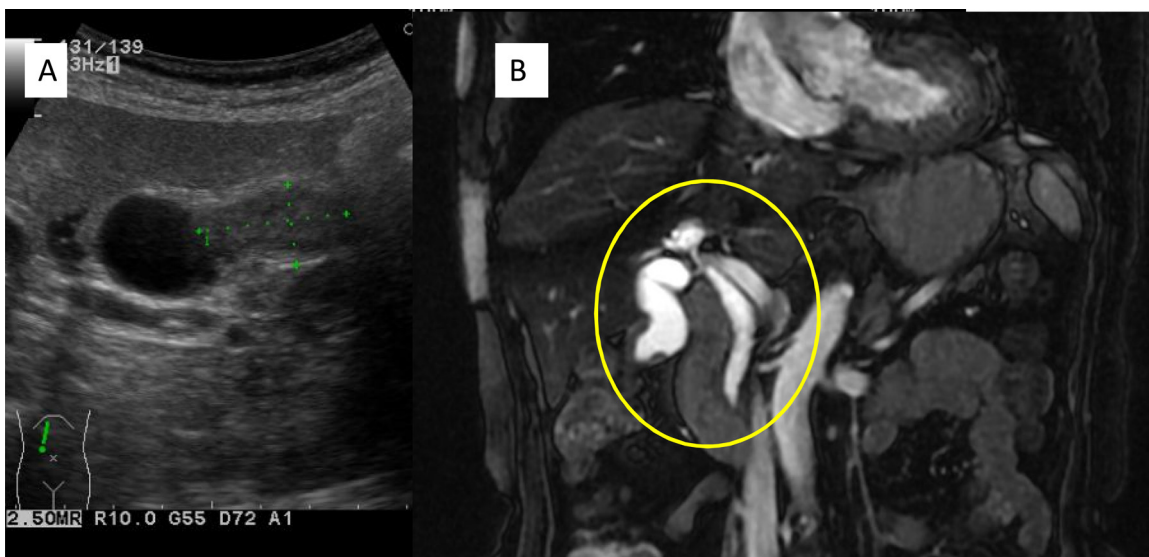
A 74-year-old female patient was referred to our hospital due to wall thickening of the gallbladder on ultrasonography (US). She underwent a health checkup program including abdominal US every year, and no diseases had been detected. The physical examination and laboratory data showed no abnormal findings at admission.

Abdominal computed tomography (CT) revealed wall thickening at the fundus of the gallbladder, which was well contrasted

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**Fig. 1.** Abdominal CT, A (axial section) and B (coronal section) indicated wall thickening at the fundus of the gallbladder, which was well contrasted, and no abnormalities of the biliary tract or surrounding lymph nodes.



**Fig. 2.** A; Abdominal US showed wall thickening of the fundus and a well-defined margin of the gallbladder. B; MRI showed no abnormal findings at the body to neck of the gallbladder and common bile duct.

(Fig. 1A and B), and no abnormalities of the biliary tract or surrounding lymph nodes.

Additionally, abdominal US showed wall thickening of the fundus and a well-defined margin of the gallbladder (Fig. 2A). Magnetic resonance imaging (MRI) showed no abnormal findings at the body to neck of the gallbladder and common bile duct (Fig. 2B). Pancreaticobiliary maljunction was not observed.

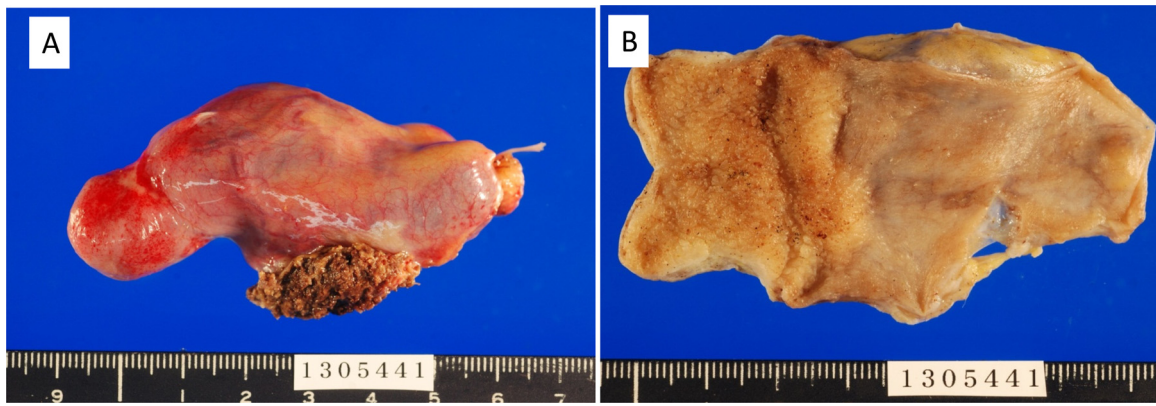
These findings suggested a diagnosis of gallbladder carcinoma rather than adenomyomatosis or other benign diseases. Therefore, we selected open laparotomy for cholecystectomy. The frozen section of the tumor confirmed the diagnosis of gallbladder carcinoma and no cancerous lesion at the stump of the cystic duct. We performed lymph node dissection of the hepato-duodenal ligament without extrahepatic bile duct resection.

Macroscopic findings showed wall thickening of the fundus of the gallbladder, and a normal mucosal layer of the body to neck (Fig. 3A and B).

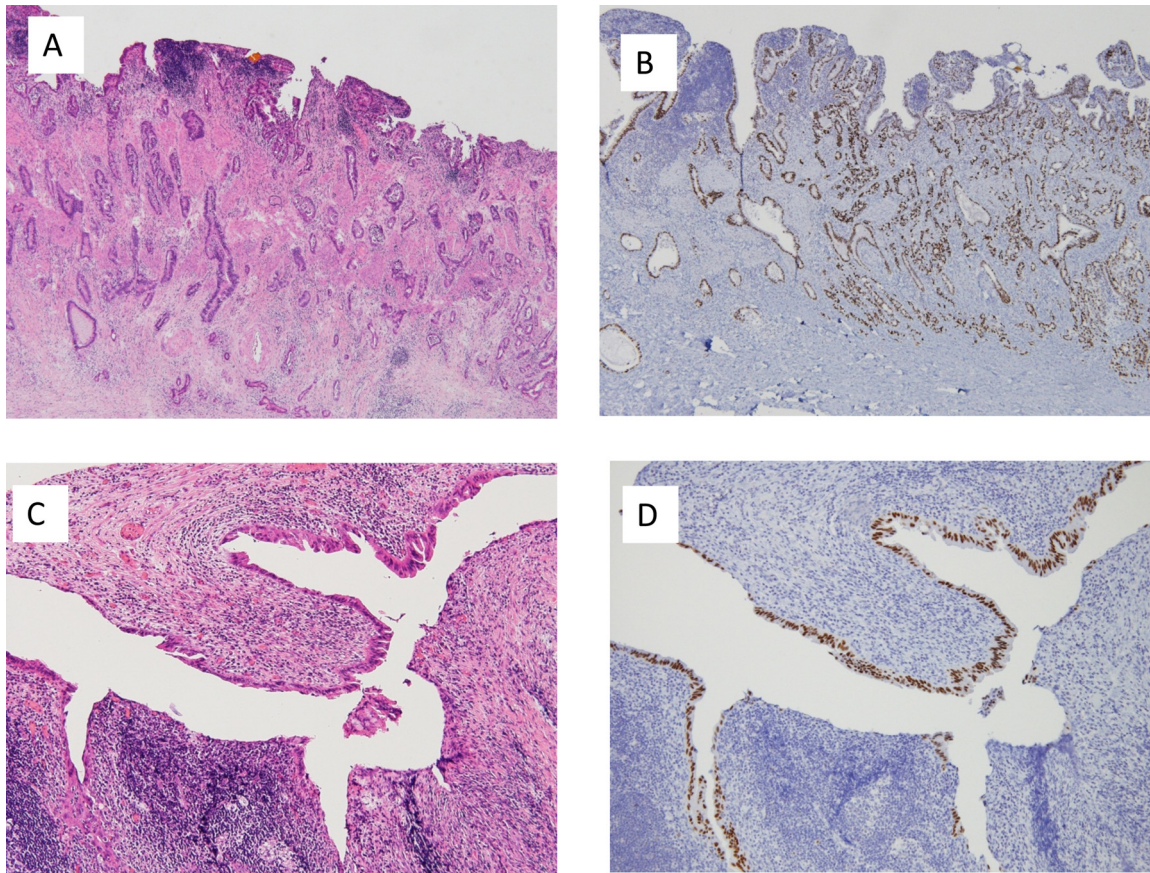
The GBC was diagnosed as well-differentiated carcinoma (Grade 1) (Fig. 4A) measuring 4 × 3 cm, with transmural invasion (T2) and no lymph node metastases (N0) indicating stage II of AJCC criteria [11]. P53 protein expression was observed in the cancer cells (Fig. 3B) and in the epithelium of the peritumor to the cystic duct (Fig. 4D), which were diagnosed as normal on HE staining (Fig. 4C).

Although, we discussed with the patient to undergo the additional surgery of hepatectomy according to the pathological diagnosis of T2 GBC, the patients denied the reoperation. We decided to conduct a careful follow-up, given the diagnosis of the surgical stump of the cystic duct as tumor-free. However,





**Fig. 3.** A and B; Macroscopic findings showed wall thickening of the fundus of the gallbladder, and a normal mucosal layer of the body to neck.



**Fig. 4.** A; GBC was diagnosed as well-differentiated carcinoma, (HE  $\times 40$ ), B P53. Protein expression was observed in the cancer cells ( $\times 40$ ), C; the epithelium of the peritumor to the cystic duct were diagnosed normal on HE staining ( $\times 100$ ) D; TP53 expression was observed at peritumor epithelium to cystic duct. ( $\times 100$ ).

the patient developed bile duct recurrence after 2.5 years after the surgery. She was admitted to our hospital due to the sudden onset of jaundice, and MRI revealed the wall thickness of bile duct indicating bile duct metastasis. And Magnetic Resonance Cholangiopancreatography (MRCP) showed dilated hepatic duct and irregular stenosis of bile duct (Fig. 5). Although, the patient underwent endoscopic stenting for the obstruction of bile duct, she denied additional therapy, and died 6 months later.

### 3. Discussion

The screening program for hepatobiliary morbidity using abdominal US has been widely used in Japan for several decades

[12]. Consequently, the frequency of incidental gallbladder carcinoma diagnosed during laparoscopic surgery for benign diseases is increasing [13,14]. However, the prognosis for patients with GC has not improved [15].

There are several prognostic factors associated with the recurrence of GBC: lymph node metastasis [15], depth of tumor invasion, histological grade, and perineural invasion [16–19]. However, superficial cancer spread from the main tumor has also been reported to be an independent predictive factor for a poor prognosis [20].

The clinical significance of extra-hepatic bile duct resection (EHBDR) in radical surgery has been extensively discussed, but this surgery was found to be not associated with an increased

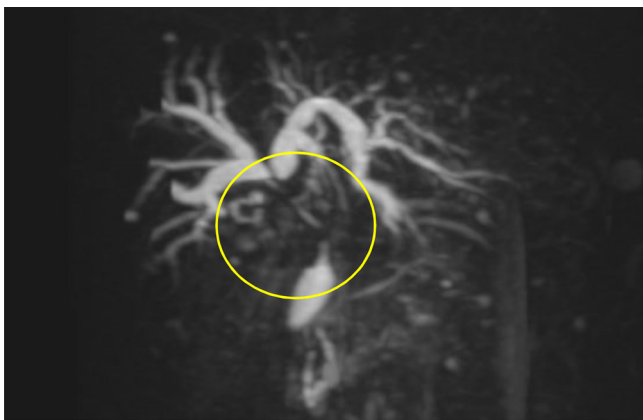


Fig. 5. MRCP showed dilated hepatic duct and irregular stenosis of bile duct.

survival rate [21,22]. EHBDR should be selectively performed only in specific cases, such as GC with extrahepatic bile duct invasion. A multi-center questionnaire survey by the Japanese Society of Biliary Surgery [23] concluded that EHBDR may be unnecessary in advanced GC without direct infiltration of the hepatoduodenal ligament and the cystic duct.

The significance of P53 immunostaining for GBC had been reported over the past several decades. Kamel et al. hypothesized that, assuming p53 expression is an early event in the carcinogenesis of a group of GCs, p53-positive dysplasias evolve to a more aggressive type of tumor, which would be indicated by association with high tumor grade and Ki67 positivity [24].

Few studies have so far examined the P53 protein expression of the normal epithelium of the gallbladder. Shu et al. reported that the p53 expression of GBC, peritumor tissue, and benign disorder (cholecystitis) was 50%, 30%, and 5%–10%, respectively [25]. In addition, Katabi et al. reported P53 immunoreactivity of 80% in carcinoma (4/5), 25% in metaplasia (2/8), and 0% in normal epithelium (0/21) [26], indicating that P53 expression occurs late in biliary carcinogenesis and highlights the sequence of tumor progression from metaplasia to carcinoma.

The present case showed that immunostaining for P53 expression is necessary to predict the carcinogenesis of the remnant bile duct. Even though the surgical stump of the cystic duct was diagnosed as tumor-free by HE staining as the present case, P53 expression was still observed. We should consider additional prophylactic bile duct resection.

The immunohistochemical staining of P53 for a surgical specimen of GBC may be crucial to predict the bile duct recurrence.

#### Conflict of interest

The authors declare no conflicts of interest (COI).

#### Sources of funding

We report no involvement of sponsors.

#### Ethical approval

The case report was approved by the institutional review board at Yamanashi Prefectural Central Hospital.

#### Consent

Written Informed Consent was obtained from the patient for publication of this case and any accompanying images. A copy of the written consent is available upon request.

#### Authors' contribution

AT HN TO and MO conceived of this case presentation and drafted the manuscript. KI, AY, HW, HN, MI, HS, MY, KF, MH and YM participated in the treatment of this case. All authors read and approved the final manuscript.

#### Guarantor

Hiroshi Nakagomi and Masao Omata have acceptable responsibility for this work and controlled the decision to publish.

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