

Case Report

Lymphoepithelioma-like hepatocellular carcinoma: a case report with emphasis on the cytological features

Chika Miyasaka^{1*}, Mitsuaki Ishida^{1*}, Hiroko Ito¹, Masaki Kaibori², Yoshiko Uemura¹, Koji Tsuta¹

Departments of ¹Pathology and Laboratory Medicine, ²Surgery, Kansai Medical University, Osaka, Japan. *Equal contributors.

Received October 30, 2016; Accepted March 9, 2017; Epub July 1, 2017; Published July 15, 2017

Abstract: Lymphoepithelioma-like hepatocellular carcinoma (LEL-HCC) is a rare distinct histopathological subtype of HCC, which is characterized histopathologically by the presence of abundant lymphocytes around the neoplastic cells, and less than 50 cases have been reported. In this report, we describe the first cytological case of LEL-HCC. A 58-year-old Japanese male had an elevated serum alpha-fetoprotein (AFP) level, and computed tomography scan demonstrated a tumorous lesion with contrast enhancement in the liver, thus, surgical resection was performed. The Papanicolaou smear of the liver tumor demonstrated small clusters and trabeculae of large-sized polygonal cells in a background of an abundance of small lymphocytes. These tumor cells had a rich granular cytoplasm and large centrally located round to oval nuclei containing conspicuous nucleoli. Immunocytochemical analyses revealed that these cells were positive for AFP, glypican-3, and Hep-Par1. Histopathological study demonstrated LEL-HCC. The cytological features of the present case represent both lymphoepithelioma-like carcinoma and conventional HCC. The prognosis of LEL-HCC may be favorable. Therefore, the diagnosis of this type of tumor is important, and a cytological examination can provide useful information for diagnosis of LEL-HCC.

Keywords: Lymphoepithelioma-like hepatocellular carcinoma, lymphocytes, liver

Introduction

Lymphoepithelioma-like carcinoma (LEC), which was originally described in the nasopharynx, is characterized histopathologically by undifferentiated carcinoma cells intermingled with abundant tumor-infiltrating lymphocyte. This type of rare carcinoma has also been reported in various organs, including lung, stomach, and bile duct [1-3]. Albeit extremely rare, lymphoepithelioma-like hepatocellular carcinoma (LEL-HCC) has been documented [4-8].

The cytological features of LEC of the skin, urinary bladder, uterine cervix, breast, and lung have been reported [9-16]; however, those of LEL-HCC have not been documented. In this report, we describe the first cytological case of LEL-HCC.

Case report

A 58-year-old Japanese male had an elevated serum alpha-fetoprotein (AFP) level (6,852 ng/mL; range <10) in a regular medical check-up. Laboratory tests revealed that his serum HBs

antigen and HCV antibody were negative, and his PIVKA-II level was also elevated (240 AU/mL; range <40). A computed tomography scan demonstrated a well-circumscribed tumorous lesion, measuring 40 mm in diameter, with contrast enhancement in S6 of the liver. Therefore, surgical resection of the liver tumor with cholecystectomy was performed under a clinical diagnosis of HCC.

The post-operative course was uneventful, and no tumor recurrence has been observed during 2 months of medical follow-up.

The touch smear of the liver tumor was stained with Papanicolaou stain. Formalin-fixed and paraffin-embedded specimens of the resected liver tumor were processed for routine histological examination and immunohistochemical and *in situ* hybridization analyses.

In this report, immunohistochemical and *in situ* hybridization analyses were performed using an autostainer (Link48, DAKO Cytomation, Glostrup, Denmark). The primary antibodies used in this report were a rabbit polyclonal anti-

Lymphoepithelioma-like HCC

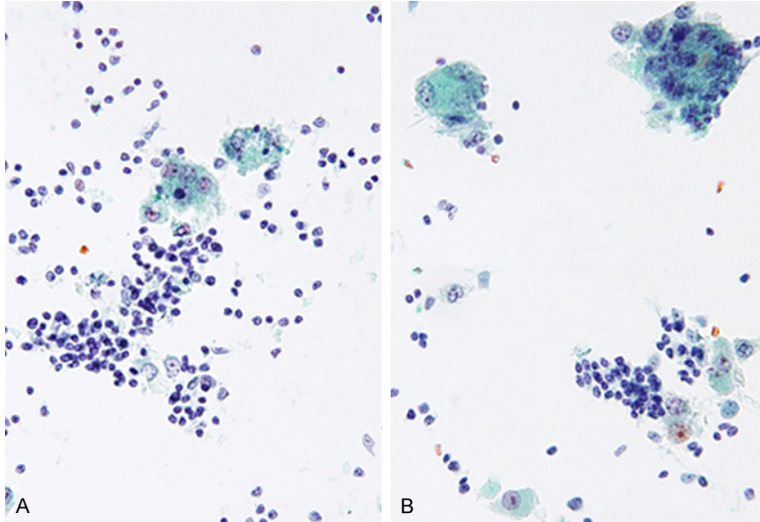


Figure 1. Cytological findings of the liver tumor. A. Small clusters of large polygonal cells in a background of many small lymphocytes, Papanicolaou stain, $\times 400$. B. These polygonal cells have a rich granular cytoplasm and centrally located large round to oval nuclei containing conspicuous nucleoli, Papanicolaou stain, $\times 400$.

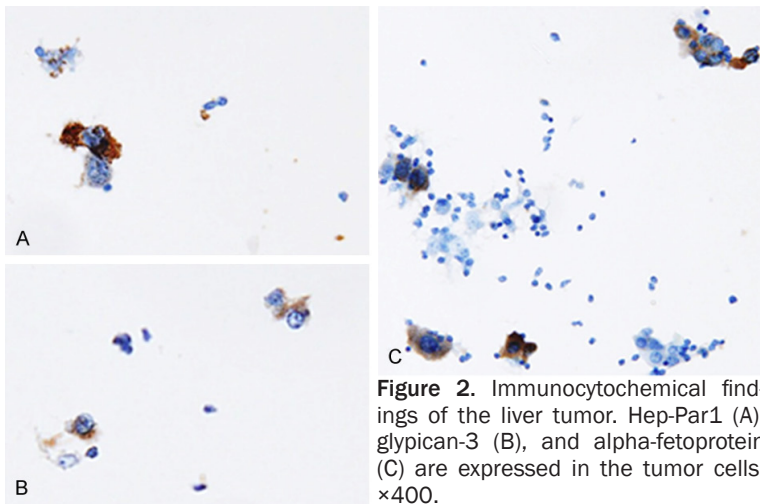


Figure 2. Immunocytochemical findings of the liver tumor. Hep-Par1 (A), glypican-3 (B), and alpha-fetoprotein (C) are expressed in the tumor cells, $\times 400$.

body against AFP (DAKO), a rabbit polyclonal antibody against CD3 (DAKO), a mouse monoclonal antibody against CD4 (4B12, DAKO), and a mouse monoclonal antibody against CD8 (C8/144B, DAKO), a mouse monoclonal antibody against CD20 (L26, DAKO), a mouse monoclonal antibody against glypican-3 (1G12, Nichirei Bioscience, Tokyo, Japan), and a mouse monoclonal antibody against Hep-Par1 (OCH1E5, DAKO).

This study was conducted in accordance with the Declaration of Helsinki, and the patient gave his informed consent.

Cytological findings

The Papanicolaou smear revealed small clusters and trabeculae of large-sized polygonal cells in a background of an abundance of small lymphocytes (**Figure 1A, 1B**). These cells had a rich granular cytoplasm and centrally located large round to oval nuclei containing vesicular chromatin and conspicuous nucleoli (**Figure 1A, 1B**).

Immunocytochemical findings

The polygonal cells were positive for AFP, glypican-3, and Hep-Par1 (**Figure 2**).

Histopathological findings

The tumor was relatively well-circumscribed and surrounded by a fibrous capsule. The neoplastic cells showed a thick trabecular growth pattern (**Figure 3A**), and had a rich eosinophilic granular cytoplasm and centrally located large round to oval nuclei and conspicuous nucleoli (**Figure 3A, 3B**). No pseudoglandular pattern was noted. The peculiar finding of this tumor was the presence of abundant infiltration of small lymphocytes around the tumor cells (>100 in ten high-power fields) (**Figure 3A, 3B**).

The liver tissue around the tumor showed no evidence of chronic hepatitis, although mild steatosis was noted.

Immunohistochemical and in situ hybridization findings

The majority of small lymphocytes were positive for CD3, and the number of CD4-positive lymphocytes was slightly larger than that of CD8-positive lymphocytes (**Figure 4A-C**). There were few CD20-positive lymphocytes. The neoplastic cells were positive for AFP, glypican-3, and Hep-Par1 (**Figure 4D**).

Lymphoepithelioma-like HCC

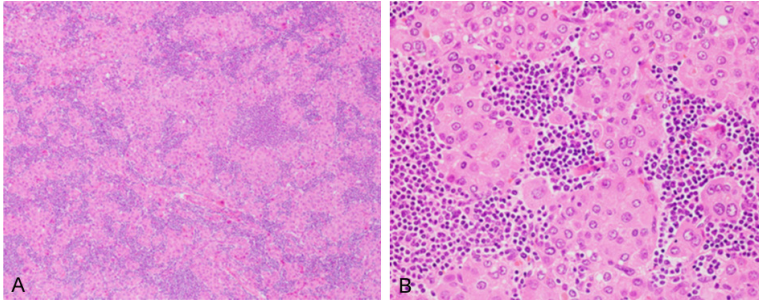


Figure 3. Histopathological features of the liver tumor. A. Thick trabecular growth of neoplastic cells intermixed with abundant lymphocytic infiltration, HE, $\times 100$. B. The tumor cells have a rich eosinophilic granular cytoplasm and centrally located large round to oval nuclei with conspicuous nucleoli, HE, $\times 400$.

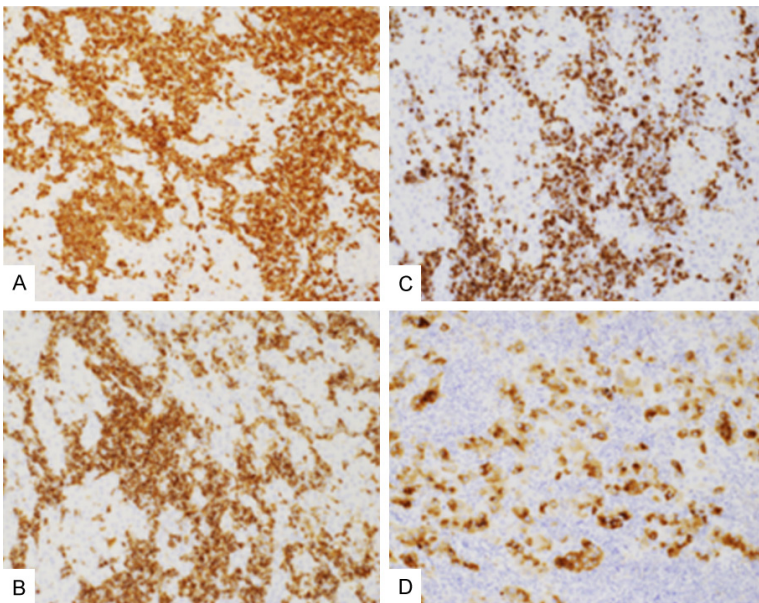


Figure 4. Immunohistochemical features of the liver tumor. (A) Abundant CD3-positive lymphocytes are present around the tumor cells ($\times 100$). CD4-positive cells (B) are slightly more predominant than CD8-positive cells (C), $\times 100$. (D) The neoplastic cells are positive for alpha-fetoprotein, $\times 100$.

No EBER-positive tumor cells were noted.

According to these results, an ultimate diagnosis of LEL-HCC was made.

Discussion

LEL-HCC is a rare distinct histopathological subtype of HCC, and less than 50 cases have been reported in the English language literature [4-8]. Most of them were single case reports, however, recently the largest case series of this type of tumor has been published [4]. Chan *et al.* analyzed 20 surgically resected

cases of LEL-HCC. The incidence was 4.9% of all HCC cases, and patients with LEL-HCC had a relatively lower frequency of males and frequently presented as a solitary tumor compared to patients with conventional HCC [4]. However, there were no significant differences in underlying chronic viral hepatitis, cirrhotic background, and serum AFP level [4]. Most of the tumor infiltrating lymphocytes were T lymphocytes, and the majority of them were CD8⁺ [4]. The vast majority of cases were not associated with EBER, unlike LEL cholangiocarcinoma. Clinically, LEL-HCC was associated with better overall and progression-free survival compared to conventional HCC [4].

In this report, we described the first cytological case of LEL-HCC. The common cytological features of LEC are as follows; i) the neoplastic cells are present singly as well as in cohesive clusters with admixture of inflammatory cells, such as lymphocytes and histiocytes, and ii) the neoplastic cells are large with high nuclear/cytoplasmic ratios, vesicular chromatin, and prominent nucleoli [10, 13]. The typical cytological features of conventional HCC are as follows; i)

the presence of neoplastic cells in broad trabeculae or cell groups lined with endothelial cells, ii) the neoplastic cells are polygonal, and have granular or vacuolated cytoplasm and centrally located large nuclei with conspicuous nucleoli, and iii) intracytoplasmic bile and bile plugs between neoplastic cells may be found [17]. Although intracytoplasmic bile and bile plugs were not observed in the present case, the above-mentioned cytological features of i) and ii) of HCC were noted. The present case clearly demonstrated that many small lymphocytes were admixed with the neoplastic cells,

Lymphoepithelioma-like HCC

which is the characteristic feature of LEC. Thus, the present case represents the cytological features of both LEC and conventional HCC. According to these cytological features, a cytodiagnosis of LEL-HCC can be possible.

Immunocytochemical analyses may be useful for cytodiagnosis of HCC. In the present case, positive immunocytostaining for AFP and glypican-3 suggested that the neoplastic cells were neoplastic hepatocytes. Moreover, Hep-Par1 is also considered to be a useful marker of normal and neoplastic hepatocytes [17].

Regenerative nodules due to liver cirrhosis must be included in the differential diagnostic consideration of LEL-HCC because liver cirrhosis contains lymphocytes around the regenerative nodules. The most characteristic cytological features of the hyperplastic hepatocytes of the regenerative nodules are their polymorphism of the hepatocytes, which shows variation in cell size and shape, and the low nuclear/cytoplasmic ratio [18]. These features aid the differential diagnosis of LEL-HCC from regenerative nodules.

In conclusion, we report the first cytological case of LEL-HCC. The characteristic cytological feature of this rare tumor is the presence of abundant small lymphocytes around the neoplastic polygonal cells containing rich granular cytoplasm and large round to oval nuclei with conspicuous nucleoli. The prognosis of LEL-HCC may be favorable. Therefore, diagnosis of this type of tumor is important, and a cytological examination can provide useful information for diagnosis of LEL-HCC.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Mitsuaki Ishida, Department of Pathology and Laboratory Medicine, Kansai Medical University, 2-5-1, Shinmachi, Hirakata 573-1010, Osaka, Japan. Tel: +81-72-804-2690; Fax: +81-72-804-2690; E-mail: ishidamt@hirakata.kmu.ac.jp

References

- [1] Ho JC, Wong MP, Lam WK. Lymphoepithelioma-like carcinoma of the lung. *Respirology* 2006; 11: 539-545.
- [2] Herath CH, Chetty R. Epstein-Barr virus-associated lymphoepithelioma-like gastric carcinoma.

- ma. *Arch Pathol Lab Med* 2008; 132: 706-709.
- [3] Ishida M, Mori T, Shiomi H, Naka S, Tsujikawa T, Andoh A, Saito Y, Kurumi Y, Kojima F, Hotta M, Tani T, Fujiyama Y, Okabe H. Non-Epstein-Barr virus associated lymphoepithelioma-like carcinoma of the inferior common bile duct. *World J Gastrointest Oncol* 2011; 3: 111-115.
- [4] Chan AW, Tong JH, Pan Y, Chan SL, Wong GL, Wong VW, Lai PB, To KF. Lymphoepithelioma-like hepatocellular carcinoma: an uncommon variant of hepatocellular carcinoma with favorable outcome. *Am J Surg Pathol* 2015; 39: 304-312.
- [5] Cacciato Insilla A, Faviana P, Pollina LE, De Simone P, Coletti L, Filippini F, Campani D. Lymphoepithelioma-like hepatocellular carcinoma: case report and review of the literature. *World J Gastroenterol* 2015; 21: 10468-10474.
- [6] Patel KR, Liu TC, Vaccharajani N, Chapman WC, Brunt EM. Characterization of inflammatory (lymphoepithelioma-like) hepatocellular carcinoma: a study of 8 cases. *Arch Pathol Lab Med* 2014; 138: 1193-1202.
- [7] An SL, Liu LG, Zheng YL, Rong WQ, Wu F, Wang LM, Feng L, Liu FQ, Tian F, Wu JX. Primary lymphoepithelioma-like hepatocellular carcinoma: report of a locally advanced case and review of literature. *Int J Clin Exp Pathol* 2015; 8: 3282-3287.
- [8] Wada Y, Nakashima O, Kutami R, Yamamoto O, Kojiro M. Clinicopathological study on hepatocellular carcinoma with lymphocytic infiltration. *Hepatology* 1998; 27: 407-414.
- [9] Abdou AG, Asaad NY. Lymphoepithelioma-like carcinoma of the breast: cytological, histological, and immunohistochemical characteristics. *Diagn Cytopathol* 2014; 43: 210-213.
- [10] Hayashi T, Haba R, Tanizawa J, Katsuki N, Kadota K, Miyai Y, Bando K, Shibuya S, Nakano M, Kushida Y. Cytopathologic features and differential diagnostic considerations of primary lymphoepithelioma-like carcinoma of the lung. *Diagn Cytopathol* 2012; 40: 820-825.
- [11] Trihia H, Siatra H, Gklisty H, Diamantopoulos P, Arapantoni-Dadiotis P, Kalogerakos K. Lymphoepithelioma-like carcinoma of the breast: cytological and histological features and review of the literature. *Acta Cytol* 2012; 56: 85-91.
- [12] Guresci S, Simsek G, Kara C, Tezer A, Bozkurt O, Unsal A. Cytology of lymphoepithelioma-like carcinoma of the urinary bladder. *Cytopathology* 2009; 20: 268-269.
- [13] Cai G, Parwani AV. Cytomorphology of lymphoepithelioma-like carcinoma of the urinary bladder: report of two cases. *Diagn Cytopathol* 2008; 36: 600-603.

Lymphoepithelioma-like HCC

- [14] Chen KT. Cytology of lymphoepithelioma-like carcinoma of the skin. *Diagn Cytopathol* 1999; 21: 230-232.
- [15] Reich O, Pickel H, Purstner P. Exfoliative cytology of a lymphoepitheliomalike carcinoma in a cervical smear: a case report. *Acta Cytol* 1999; 43: 285-288.
- [16] Chow LT, Chow WH, Tsui WM, Chan SK, Lee JC. Fine-needle aspiration cytologic diagnosis of lymphoepithelioma-like carcinoma of the lung: report of two cases with immunohistochemical study. *Am J Clin Pathol* 1995; 103: 35-40.
- [17] Isa NM, Bong JJ, Ghani FA, Rose IM, Husain S, Azrif M. Cutaneous metastasis of hepatocellular carcinoma diagnosed by fine needle aspiration cytology and HepPar1 immunopositivity. *Diagn Cytopathol* 2012; 40: 1010-1014.
- [18] Wee A. Fine needle aspiration biopsy of hepatocellular carcinoma and hepatocellular nodular lesions: role, controversies and approach to diagnosis. *Cytopathology* 2011; 22: 287-305.