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Case study

Primary peritoneal epithelioid mesothelioma of clear cell type with a novel *VHL* gene mutation: a case report

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Summary Clear cell variant of epithelioid mesothelioma is an extremely rare tumor with only isolated cases reported so far in the peritoneum. Here, we report a case of peritoneal epithelioid mesothelioma, clear cell variant, in a 63-year-old female patient with a novel *VHL* gene mutation and an unusual indolent clinical course. The patient, who has no clinical history of asbestos exposure, presented with a 27.2-cm upper abdominal mass and a 5.5-cm liver lesion. Retrospective review of the patient's abdominal computed tomographic scan 4 years ago showed 2 small abdominal lesions that were felt clinically to represent hemangiomas. These were retrospectively considered to have grown in size and represented the current abdominal mass. Both masses were subsequently biopsied and showed a proliferation of monomorphic epithelioid cells with distinct cell membranes, fine chromatin, and clear to finely vacuolated pale eosinophilic cytoplasm arranged in nests and solid sheets. Immunohistochemical staining confirmed it to be malignant mesothelioma. Clear cell variant of peritoneal epithelioid mesothelioma should always be considered in patients with an abdominal or pelvic mass with clear cell features. Given the rarity of such entity, its clinical course and prognosis remains unclear.

1. Introduction

Malignant mesothelioma (MM) is a rare entity that most commonly arises from the pleura. Although less frequently seen, MM can occur in other organs lined by mesothelial cells, including the peritoneum, pericardium, and tunica vaginalis [1]. MM has a wide range of histologic subtypes that are generally grouped under 1 of 3 major patterns: epithelioid, sarcomatoid, and bipahasic [1]. The epithelioid type of mesothelioma is frequently seen with papillary or tubular components; however, a very rare entity, which has only been described in a few case reports, is the clear cell variant [2,3]. Because clear cell morphology can occur in almost any type of tumor, to diagnose a clear cell tumor as mesothelioma can be exceedingly difficult and often requires numerous immunohistochemical stains. This is especially true when it is arising in the peritoneum because of the numerous entities that are characteristically associated with clear cells in this anatomical region. In addition, because of the few reported cases of this subtype, little is known about the prognosis, molecular profile, and the disease pathogenesis [4].

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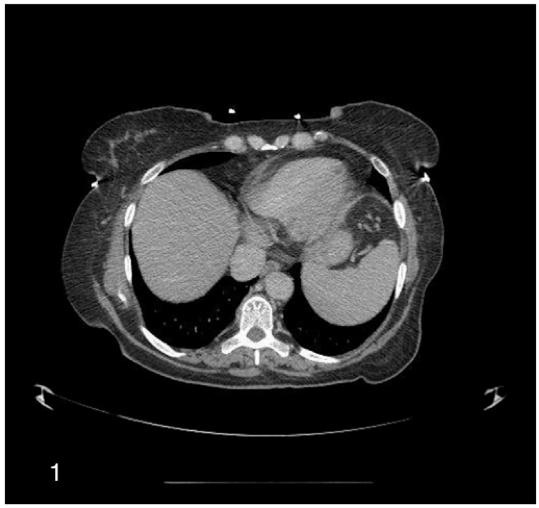

Fig. 1 Computed tomographic scans of the abdomen and pelvis show a right hepatic heterogeneous mass.

2. Case description

A 63-year-old woman presented to her primary care provider complaining of abdominal and right flank pain. A urinalysis was performed and found to be positive for hematuria. The patient was sent for imaging, and on computed tomography (CT) of the abdomen and pelvis with contrast, a complex heterogeneous mass measuring 27.2 cm was identified. The mass was extending superiorly to the liver, gallbladder, and stomach, and there was questionable anterior wall involvement of the mid to distal stomach and possible involvement of the anterior margin of the transverse colon (Fig. 1). An additional lesion in the liver measuring 5.5 cm in greatest dimension was also found. The gallbladder, pancreas, adrenal gland, kidneys, small bowel, bladder, uterus/adnexa, and remainder of colon were negative for any significant pathology. A CT of the thorax with contrast showed the chest wall to be grossly normal, with no mediastinal lymphadenopathy, and negative findings in the pericardium or pleura.

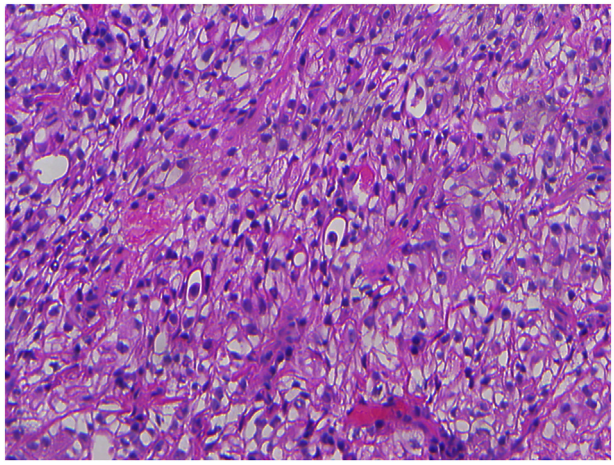

Fig. 3 Higher-power magnification showing the fine nuclear chromatin and cytoplasmic clearing (hematoxylin-eosin, original magnifications ×20).

Retrospective review of a CT performed in January of 2014 for a hematuria workup found 2 abdominal masses, one of which had been previously described in 2010 when it was incidentally found on a lumbar spine CT. At the time, it was felt that these were likely hemangiomas, and no further investigation was conducted. In light of these new findings, it is felt that these abdominal lesions represent the current abdominal mass, which had grown in size over several years. Both the abdominal and liver masses were subsequently biopsied and showed a proliferation of monomorphic epithelioid cells with distinct cell membranes, fine chromatin, and clear to finely vacuolated pale eosinophilic cytoplasm arranged in nests and solid sheets (Figs. 2 and 3). The tumor cells were immunoreactive for calretinin (Fig. 4), D2-40 (Fig. 5), HBME-1, CK5/6 (Fig. 6), CK19, CK7, and WT-1 (Fig. 7). Periodic acid–Schiff

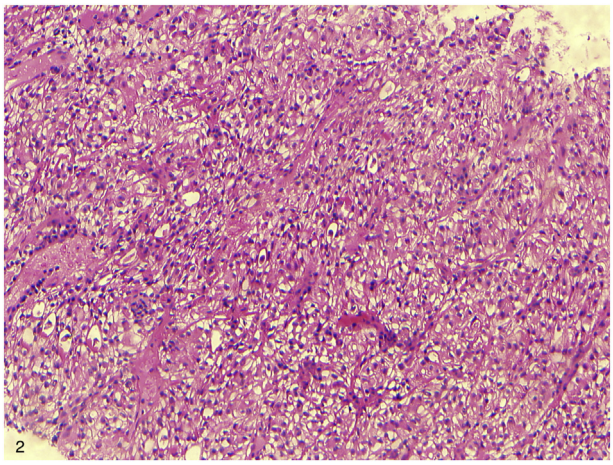

Fig. 2 Low-power magnification showing nested proliferation of monomorphic epithelioid cells (hematoxylin-eosin, original magnifications ×4).

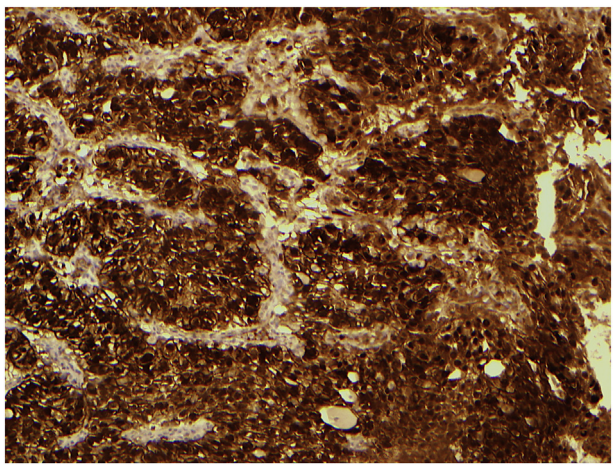

Fig. 4 Strong calretinin positivity seen in tumor cells (original magnifications ×20).

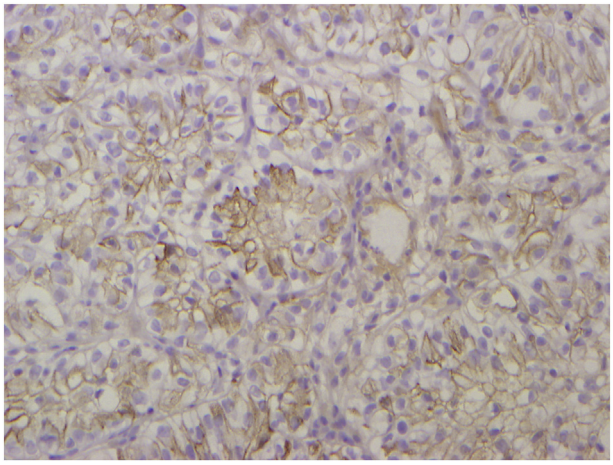

Fig. 5 D2-40–positive staining in tumor cells (original magnifications ×50).

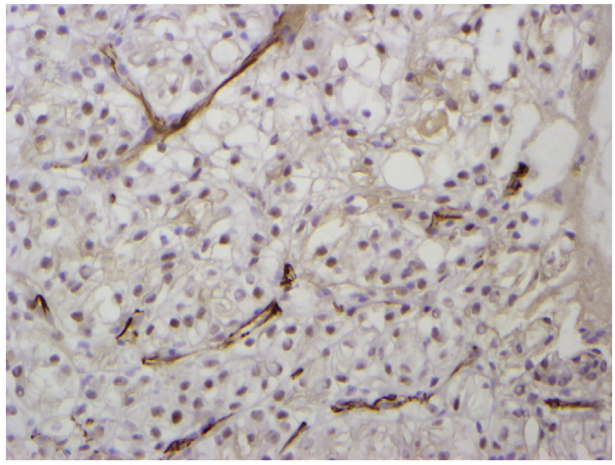

Fig. 7 Nuclear positivity is seen with WT1 (original magnifications ×50).

(Fig. 8) highlighted the intracytoplasmic glycogen deposits, which disappeared when treated with diastase. The tumor cells were negative for GATA-3, CK20, inhibin, arginase, HMB45, p63, glypican-3, CD117, DOG1, CDX2, Pax8, TTF-1, CEA and BER-EP4, chromogranin, synaptophysin, and albumin in situ. Based on the immunophenotype and morphology, the tumor was diagnosed as epithelioid mesothelioma, clear cell variant. Further molecular testing using an investigational polymerase chain reaction panel showed a *VHL* gene alteration involving exon 2 (exon2 p.F136Nfs 25 [c.401_405dupAATTA]), which is considered a novel genetic finding in MM. None of the common mesothelioma-associated molecular alterations (*BAP1*, *p16/CDKN2A*, *NF2*, *LATS2*, *SETD2*, and *DDX3X*) were positive in the clinical polymerase chain reaction panel.

The patient underwent wide excision of the mesenteric mass and partial liver resection at the end of February 2018. She was also placed on a chemotherapy regiment. On clinical follow-up, 2 months later, the patient was found to be doing well without evidence of any new lesions.

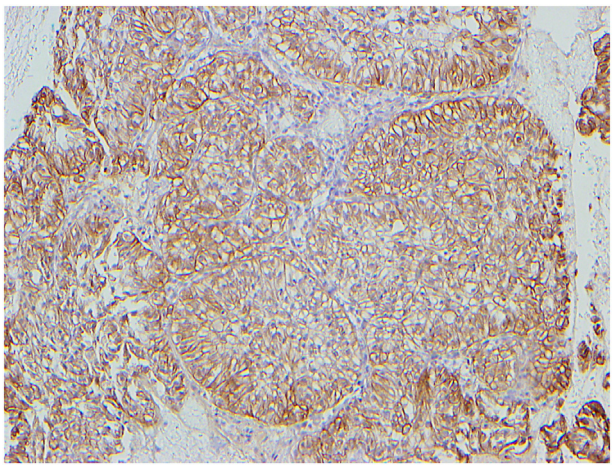

Fig. 6 CK5/6-positive membranous staining in tumor cells (original magnifications ×10).

3. Discussion

Clear cell mesothelioma, also called glycogen-rich or foamy mesothelioma, is considered a variant of epithelioid MM. It is a rare entity that was first described by Ordoñez et al [2,5] in 1996, with only isolated cases reported so far in the pleura [2,3,5-7], peritoneum [2,8,9], and testis [10].

Given the rarity of such entity, the final diagnosis of clear cell mesothelioma, in our case, was reached after excluding more common intra-abdominal tumors with clear cell morphology that entered the differential diagnosis, which included carcinomas arising from upper or lower gastrointestinal tract, pancreas, adrenal, kidney, liver and Müllerian system/primary peritoneum, or metastatic carcinomas from breast or lung origins. Other intra-abdominal tumors with clear cell morphology, within the differential diagnosis, included myoepithelial carcinoma, epithelioid leiomyosarcoma, melanoma, perivascular epithelioid cell tumor, paraganglioma, gastrointestinal

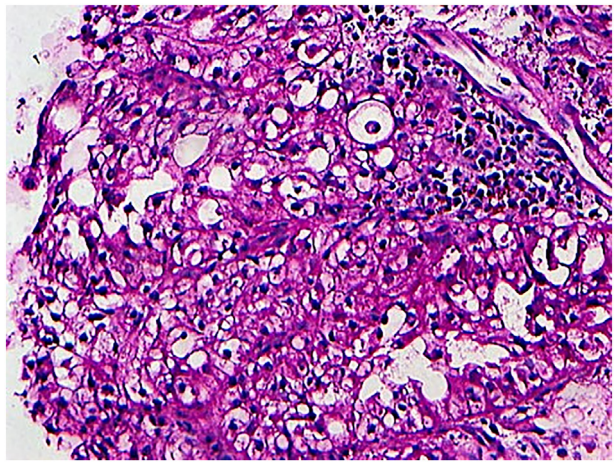

Fig. 8 Periodic acid–Schiff demonstrating intracytoplasmic glycogen deposits, confirmed with periodic acid–Schiff–diastase (original magnifications ×20).

Table List of intra-abdominal tumors with clear cell morphology and their corresponding immunostains

Intra-abdominal tumor(s) with clear cell morphology	Corresponding immunostain(s)	Immunostain(s) results	Tumor(s) ruled in/out
Clear cell mesothelioma	Calretinin, D2-40, HBME-1, CK5/6, CK7, WT-1	Positive	Ruled in
Upper and lower GI tract carcinoma/pancreatic carcinoma	CK7, CK20, CDX-2	Negative (except for CK7)	Ruled out
Urothelial/metastatic breast carcinoma/paraganglioma	GATA3	Negative	Ruled out
Renal carcinoma	Pax-8	Negative	Ruled out
Müllerian system/primary peritoneal carcinoma	Pax-8, B72.3, Ber-EP4, poly-CEA	Negative	Ruled out
Adrenal carcinoma	Inhibin, synaptophysin	Negative	Ruled out
Hepatocellular carcinoma	Glypican 3, HepPar-1, arginase 1, albumin in situ	Negative	Ruled out
Metastatic lung adenocarcinoma	TTF-1, B72.3, Ber-EP4, poly-CEA	Negative	Ruled out
GIST	CD117, DOG-1	Negative	Ruled out
Melanoma	S100, HMB-45	Negative	Ruled out
Epithelioid angiosarcoma	ERG/FLI-1	Negative	Ruled out
Epithelioid sarcoma	INI-1	Positive (retained)	Ruled out
Epithelioid leiomyosarcoma	SMA, Desmin	Negative	Ruled out
Myoepithelial carcinoma	SMA, p63	Negative	Ruled out
PEComa	SMA, HMB-45	Negative	Ruled out

Abbreviations: GI, gastrointestinal; GIST, gastrointestinal stromal tumor; PEComa, perivascular epithelioid cell tumor.

stromal tumor, epithelioid angiosarcoma, and epithelioid sarcoma. Carcinomas in general were excluded by absent immunostaining for B72.3, Ber-EP4, and poly-CEA immunomarkers [1,11]. Further immunostains were used to eliminate specific intra-abdominal tumors with clear cell morphology (see Table). Ultimately, positivity of the tumor cells for the mesothelioma markers calretinin, D2-40, HBME-1, CK5/6, CK19, CK7, and WT-1 and negativity for all of the listed immunomarkers favored the diagnosis of clear cell mesothelioma [1,4,11].

Although the risk factors for malignant peritoneal mesothelioma are not very well defined, patients with the disease are more commonly younger women and have a better prognosis compared with patients with pleural mesothelioma [12]. In addition, although most pleural mesotheliomas occur in patients with a history of asbestos exposure, there has been some recent evidence that suggests that peritoneal mesothelioma in women is rarely associated with asbestos exposure [13,14]. Having localized disease, as seen in our case, has been suggested to add an additional layer of prognostic benefit, as there has been evidence that localized MMs are biologically different in their behavior and have a markedly better prognosis than their diffuse counterpart. This may partially explain the seemingly indolent course that our patient has had [15].

According to the latest report from the National Cancer Institute's Surveillance, Epidemiology, and End Results Program, epithelioid mesotheliomas have a median survival of 12 to 24 months, with epithelioid morphology being more favorable than sarcomatoid and biphasic morphologies. However, in our case, the patient's tumor had an unusually indolent clinical course and was assumed to be present for almost 4 years with gradual increase in size. In addition, unlike our case whereby the liver was the main metastatic focus, liver metastasis is thought to be uncommon in mesotheliomas, with the most common site of metastasis being to regional lymph nodes [16].

At the molecular level, mesotheliomas have been linked to several alterations, which seem to vary in frequency depending on both location and morphologic subtype, in general, the most frequent being homozygous deletions of *p16/CDKN2A* at 9p21, approximately 75% of pleural mesothelioma cases [17]. *BAP1* (BRCA-associated protein 1) mutations or deletions, however, can be seen in up to 80% of epithelioid mesothelioma cases [18]. Other alterations include inactivating mutations in *NF2* gene (20%-40% of cases), mutations in *LATS2* gene at 13q21 (<10% of cases), and inactivating mutations of SETD2 (8% of cases) and DDX3X (4% of cases) [19-21].

In a study performed at the University of California by Joseph et al [19], genomic profiling of malignant peritoneal mesotheliomas revealed recurrent alterations in epigenetic regulatory genes BAP1 (85% of cases), SETD2 (15% of cases), and DDX3X (15% of cases). Apparently, BAP1 mutation is the most frequent somatic event in peritoneal MMs [22], being more often present in the epithelioid type (80%) than the nonepithelioid type (15%) [23]. However, no study so far has reported the presence of *VHL* mutation in peritoneal mesotheliomas, more specifically primary epithelioid peritoneal mesotheliomas. Mutations involving the *VHL* gene have been reported in 2 cases of pleural mesothelioma; these mutations are, however, different from the missense mutation that was found in our case [24].

The presence of a *VHL* gene alteration involving exon 2 and the absence of the common MM molecular alteration markers including *BAP1*, *p16/CDKN2A*, *NF2*, *SETD2*, and *DDX3X* are considered a novel finding in our case.

It is unclear at this point in time whether the clear cell morphology of the epithelioid peritoneal mesothelioma in our case is related to the presence of the *VHL* gene mutation or not. Further genome-wide studies of genetic and epigenetic alterations are needed to determine this relationship.

4. Conclusions

In conclusion, we report a rare case of primary peritoneal epithelioid mesothelioma of clear cell type with a novel *VHL* gene mutation. This mesothelioma variant should always be considered in patients with an abdominal or pelvic mass with clear cell features. Given the rarity of such entity, its clinical course, prognosis, pathogenesis, and molecular profile remain unclear.

References

[1] Tandon RT, Jimenez-Cortez Y, Taub R, Borczuk AC. Immunohistochemistry in peritoneal mesothelioma: a single-center experience of 244 cases. Arch Pathol Lab Med 2018;142:236-42. https://doi.org/10.5858/arpa.2017-0092-oa.

[2] Ordóñez NG. Mesothelioma with clear cell features: an ultrastructural and immunohistochemical study of 20 cases. Hum Pathol 2005;36:465-73. https://doi.org/10.1016/j.humpath.2005.02.014.

[3] Dessy E, Falleni M, Braidotti P, Del Curto B, Panigalli T, Pietra GG. Unusual clear cell variant of epithelioid mesothelioma. Arch Pathol Lab Med 2001;125:1588-90. https://doi.org/10.1043/0003-9985(2001)125<1588:UCCVOE>2.0.CO;2.

[4] Husain AN, Colby TV, Ordóñez NG, et al. Guidelines for pathologic diagnosis of malignant mesothelioma 2017 update of the consensus statement from the international mesothelioma interest group. Arch Pathol Lab Med 2018;142:89-108. https://doi.org/10.5858/arpa.2017-0124-ra.

[5] Ordóñez NG, Myhre M, Mackay B. Clear cell mesothelioma. Ultrastruct Pathol 1996;20:331-6. https://doi.org/10.3109/01913129609016333.

[6] Ordonez NG, Mackay B. Glycogen-rich mesothelioma. Ultrastruct Pathol 1999;23:401-6. https://doi.org/10.1080/019131299281383.

[7] Gkogkou C, Samitas K, Foteinou M. Primary pleural epithelioid mesothelioma of clear cell type: a case report and review of current literature. Ultrastruct Pathol 2011;35:267-70. https://doi.org/10.3109/01913123.2011.606965.

[8] Ordóñez NG. Clear cell mesothelioma presenting as an incarcerated abdominal hernia. Virchows Arch 2005;447:823-7. https://doi.org/10.1007/s00428-005-0023-2.

[9] Hayashi H, Kawata T, Shimokawa I. Malignant peritoneal mesothelioma, clear cell variant, in a female and its differentiation from clear cell carcinoma. Pathol Res Pract 2017;213:580-4. https://doi.org/10.1016/j.prp.2016.12.025.

[10] Mishra A, Shet T. Clear cell mesothelioma of the testis with deciduoid areas—a case report. Indian J Pathol Microbiol 2004;47:544-6.

[11] Robinson BWS, Lake RA. Advances in malignant mesothelioma. N Engl J Med 2005;353:1591-603.

[12] Liu S, Staats P, Lee M, Alexander HR, Burke AP. Diffuse mesothelioma of the peritoneum: correlation between histological and clinical parameters and survival in 73 patients. Pathology 2014;46:604-9. https://doi.org/10.1097/pat.0000000000000181.

[13] de Ridder GG, Kraynie A, Pavlisko EN, Oury TD, Roggli VL. Asbestos content of lung tissue in patients with malignant peritoneal mesothelioma: a study of 42 cases. Ultrastruct Pathol 2016;40:134-41.

[14] Kraynie Alyssa, de Ridder GG, Sporn TA, Pavlisko EN, Roggli VL. Malignant mesothelioma not related to asbestos exposure: analytical scanning electron microscopic analysis of 83 cases and comparison with 442 asbestos-related cases. Ultrastruct Pathol 2016;40:142-6.

[15] Allen TC, et al. Localized malignant mesothelioma. Am J Surg Pathol 2005;29:866-73.

[16] Zannella S, Testi MA, Cattoretti G, Pelosi G, Zucchini N. Peritoneal malignant mesothelioma metastatic to supraclavicular lymph nodes. Int J Surg Pathol 2014;22:552-4. https://doi.org/10.1177/1066896913520037.

[17] Krasinskas AM, Bartlett DL, Cieply K, Dacic S. CDKN2A and MTAP deletions in peritoneal mesotheliomas are correlated with loss of p16 protein expression and poor survival. Mod Pathol 2010;23:531-8.

[18] Churg A, Sheffield BS, Galateau-Salle F. New markers for separating benign from malignant mesothelial proliferations: are we there yet? Arch Pathol Lab Med 2016;140:318-21. https://doi.org/10.5858/arpa.2015-0240-SA.

[19] Joseph NM, Chen Y-Y, Nasr A, et al. Genomic profiling of malignant peritoneal mesothelioma reveals recurrent alterations in epigenetic regulatory genes BAP1, SETD2 and DDX3X. Mod Pathol 2016;30:246-54. https://doi.org/10.1038/modpathol.2016.188.

[20] Sekido Y. Molecular biology of malignant mesothelioma. Environ Health Prev Med 2008;13:65-70. https://doi.org/10.1007/s12199-007-0015-8.

[21] McMillan R, Zauderer M, Bott M, Ladanyi M. Important recent insights into the genetics and biology of malignant pleural mesothelioma. Ann Cardiothorac Surg 2012 Nov;1:462-5.

[22] Harismendy O, Alakus H, Yost S, et al. BAP1 mutation is a frequent somatic event in peritoneal malignant mesothelioma. J Transl Med 2015;13:4803. https://doi.org/10.1158/1538-7445.am2015-4803.

[23] Yoshikawa Y, Sato A, Tsujimura T, et al. Frequent inactivation of the BAP1 gene in epithelioid-type malignant mesothelioma. Cancer Sci 2012;103:868-74. https://doi.org/10.1111/j.1349-7006.2012.02223.x.

[24] Shukuya T, Serizawa M, Watanabe M. Identification of actionable mutation in malignant pleural mesothelioma. Lung Cancer 2014;86:35-40.