Original Research Article

Methylthioadenosine Phosphorylase and Breast Cancer 1 Protein-Associated Protein 1 as Biomarkers for the Peritoneal Mesothelioma

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Abstract

Objectives: Combination of Breast Cancer 1 protein-associated protein 1 (BAP1) and methylthioadenosine phosphorylase (MTAP) in the peritoneal mesothelioma (PeM) has yet to be explored. We aim to assess the diagnostic value of combined BAP1 and MTAP to distinguish biphasic mesothelioma (BM) from epithelioid mesothelioma (EM) with reactive stroma in peritoneum, as well as its prognostic value in PeM.

Methods: This is a retrospective study from June 2014 to December 2021. This study included 18 cases of BM and 27 cases of EM with reactive stroma, excluded sarcomatoid, and EM without reactive stroma cases, and clinicopathological information was collected. The associations between MTAP and BAP1 levels and clinicopathological features or prognosis were analyzed. Clinical follow-up data were reviewed to correlate with pathological prognostic factors using Kaplan–Meier estimator and univariate/multivariate Cox proportional hazards regression models.

Results: Loss/decrease of BAP1/MTAP was observed in 6 (33.3%) BM cases and 12 (44.4%) EM cases. In 5 (27.8%) cases, loss of or decreased BAP1/MTAP expression was observed in both EC and SC of BM. BAP1/MTAP loss/decrease was observed in 12 (44.4%) cases of only EC of EM but not in reactive stroma. Compared with histology alone, a combination of BAP1 and MTAP immunohistochemistry (IHC) in spindled PeM provides a more objective mean to distinguish BM from EM with reactive stroma. Loss/decrease of BAP1/MTAP was associated with peritoneal cancer index (PCI) score (P = 0.047) and completeness of cytoreduction (CC) score (P = 0.038). BM patients have worse overall survival (OS) than EM with reactive stroma (P = 0 .007).

Conclusions: Combination of BAP1/MTAP by IHC is helpful for differential diagnosis of peritoneal BM from EM with reactive stroma. Nevertheless, BAP1/MTAP may help to evaluate the biological behavior of PeM.

Keywords

biphasic mesothelioma, methylthioadenosine phosphorylase, immunohistochemistry, peritoneal mesothelioma, Breast Cancer 1 protein-associated protein 1

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Introduction

Malignant mesothelioma (MM) is a highly aggressive neoplasm that mainly develops from the pleura or peritoneum.[1] The peritoneal mesothelioma (PeM) is less common than pleural mesothelioma, accounting for 7%–30% of MM.[2] The PeM has a median OS of approximately 1 year due to delayed diagnosis at relatively advanced stages with nonspecific symptoms.[3] The PeM can be classified into epithelioid, sarcomatoid, or biphasic subtypes according to World Health Organization (WHO) criteria (2021).[4] The biphasic mesothelioma (BM) contains both epithelioid and sarcomatoid components, and each component accounts for at least 10% of total tumor cells. The epithelioid subtype is known to have better outcome. The prognosis of biphasic subtype was determined by the proportion of sarcomatoid component.[5] Recently, studies demonstrated that the prognosis of epithelioid mesothelioma (EM) may be influenced by morphological features.[6] Therefore, it is important to accurately differentiate MM subtypes.

When reactive stroma component is present, it is challenging to distinguish EM from BM with sarcomatoid component based on histology alone.[7] BAP1 is a tumor suppressor gene located on chromosome 3 (3p21.1).[8] The *p16/CDKN2A* gene located on chromosome 9 (9p21) can regulate cell cycle by encoding cyclin-dependent kinase inhibitors.[9] Recently, fluorescence in situ hybridization (FISH) assay of *p16/CDKN2A* and immunohistochemistry (IHC) analysis of BAP1 loss have been reported to be useful for differentiating pleural BM from EM with reactive stroma.[10] Besides, MTAP immunostaining can be taken as a highly sensitive substitute marker for *p16/CDKN2A* deletion.[11] However, to our knowledge, no report has compared loss/decrease of BAP1/MTAP in peritoneal BM and EM with reactive stroma.

The objective of our study was to investigate the differential diagnostic value of BAP1/MTAP loss/decrease in spindled PeM (BM and EM with reactive stroma). Furthermore, we examined the association between BAP1/MTAP expression and clinicopathological characteristics or prognosis of PeM.

Materials and Methods

Patient Selection

This is a retrospective study in Beijing Shijitan Hospital from June 2014 to December 2021. The reporting of this study conforms to STARD guidelines.[12] We collected 45 PeM patients with complete clinical data. This study included 18 cases of BM and 27 cases of EM with reactive stroma (Figure 1(A) and (B)) and excluded sarcomatoid patients and EM without reactive stroma. These patients were diagnosed independently based on morphology and conventional IHC by two pathologists. Each case was confirmed by Calretinin, Cytokeratin 5/6, D2-40, and Wilms' tumor-1 (WT-1) as positive mesothelial markers, as well as carcinoembryonic antigen (CEA) and BerEP4 as negative markers. In female patients, we excluded serous carcinomas using ER and Pax8 markers. Finally, pathological subtypes were reclassified according to BAP1 and MTAP IHC.

The clinicopathological characteristics included sex, age, asbestos exposure, peritoneal cancer index (PCI) score, carbohydrate antigen (CA) 125, completeness of cytoreduction (CC) score, ascites, treatment, pathological type, vascular tumor emboli, and tumor node metastasis (TNM) stage. The immunohistochemical characteristics included Ki-67 index and BAP1/MTAP expression status. The prognostic indices included survival status and OS.

All patients signed informed consent and agreed to use postoperative specimens and clinical data for medical research. We have de-identified all patient details.

Immunohistochemistry (IHC)

Chromogenic IHC assay was performed using an automated immunostainer (intelliPATH FLX, Beijing Zhongshan Golden Bridge Biological Technology Co., Ltd.). Briefly, formalin-fixed (10% neutral formaldehyde) paraffin-embedded 4-μm-thick sections were first deparaffinized by xylene and rehydrated in graded series of ethanol. Then, endogenous peroxidase activity was blocked by 0.3% H_2O_2. The primary antibodies and final dilutions were as follows: Ki-67 (1:100, ZM-0166, OriGene, China), BAP1 (1:75, ab255611, Abcam, UK), and MTAP (1:1000, ab126770, Abcam, UK). The sections were observed with a microscope (Nikon, Japan). KF-PRO-400 scanner (Jiangfeng, China) was used for whole slide scanning and image acquisition. Non-mesothelial cells (histiocytes, lymphocytes, fibroblasts, and endothelial cells) were used as internal positive controls for BAP1 and MTAP. BAP1 (nuclear staining) and MTAP (cytoplasmic/nuclear staining) expression was preserved in EM (Figure 1(C) and (D)). BAP1 (nuclear staining) and MTAP (cytoplasmic/nuclear staining) expression was preserved in BM (Figure 1(E) and (F)). Loss/decrease of expression of BAP1 in tumor cells was defined as completely absent nuclear staining or at an intensity lower than the internal positive control (Figure 1(G)). For MTAP, cytoplasmic/nuclear staining in tumor cells at an intensity lower than the internal positive control was defined as loss/decrease of expression (Figure 1(H)).[13] We set the cutoff value at 50% for MTAP and BAP1 IHC as described previously.[13]

Follow-Up

The PeM patients had been followed up until November 30, 2022. The frequency of follow-up was once every 3 months within 2 years after surgery, once every 6 months after 2 years, and once every 12 months after 3 years, respectively.

Statistical Analysis

Statistical analysis was performed using SPSS (version 22.0, IBM Corp, Armonk, NY) and GraphPad Prism (version 8.0.1,

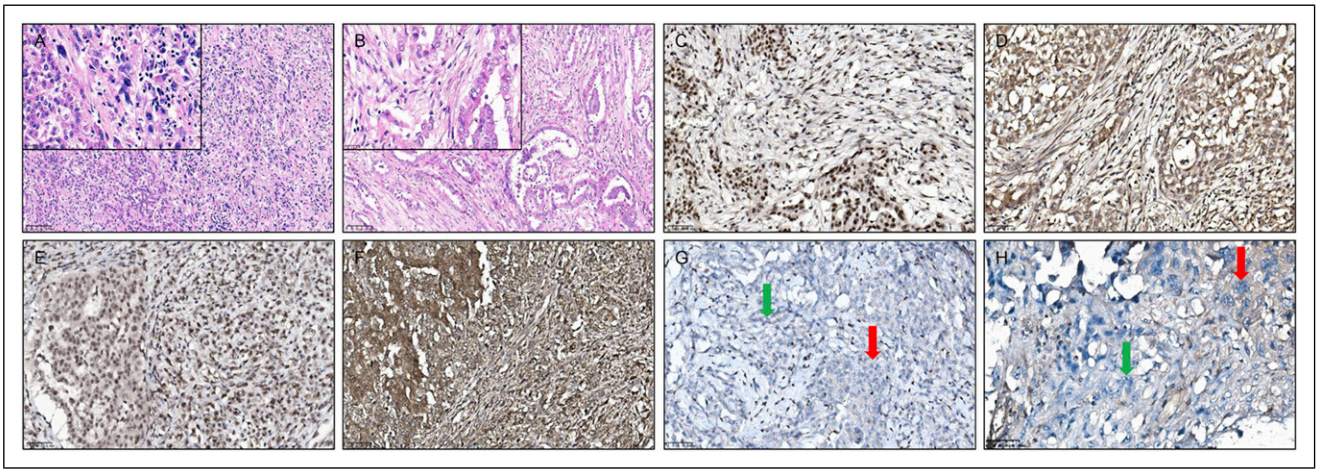

Figure 1. (A) Medium-power magnification. Inset: the biphasic mesothelioma is composed of the epithelioid component of polygonal cells with round nuclei (left) and sarcomatoid component of malignant spindle cells (right). (B) Medium-power magnification. Inset: the reactive stroma surrounding epithelioid mesothelioma of polygonal cells with round nuclei (hematoxylin and eosin [H&E] stain, ×200/800 magnification). (C) BAP1 (nuclear staining) and (D) MTAP (cytoplasmic/nuclear staining) expression in epithelioid mesothelioma. (E) BAP1 (nuclear staining) and (F) MTAP (cytoplasmic/nuclear staining) expression in epithelioid component (left) and sarcomatoid component (right) of biphasic mesothelioma. (G) Loss/decrease of BAP1 expression in epithelioid component (red arrow) and sarcomatoid component (green arrow) of biphasic mesothelioma. (H) Loss/decrease of MTAP expression in epithelioid component (red arrow) and sarcomatoid component (green arrow) of biphasic mesothelioma (immunohistochemistry staining, ×400 magnification).

GraphPad Software, San Diego, California USA). The Pearson's chi-squared test or Fisher's exact test was used for categorical variables. Univariable and multivariable Cox regression models were used to explore the potential predicative factors associated with OS, in which variables with a $P < 0.10$ in univariate analysis were selected for multivariate analysis. OS was estimated using Kaplan–Meier plot and compared with log-rank test. Statistical significance was set at a $P < 0.05$.

Results

Major Clinicopathological Characteristics

The clinicopathological characteristics of PeM patients are presented in Table 1. There were 22 males (48.9%) and 23 females (51.1%) with a median age of 56 (24–73) years. Among them, 32 (71.1%) patients were ≤60 years and 13 (28.9%) patients were >60 years. Histologically, 27 (60.0%) tumors were epithelioid, whereas 18 (40.0%) were Biphasic. Eighteen (40.0%) patients had peritoneal cancer index (PCI) < 25, and 27 (60.0%) cases had PCI ≥25. A total of 21 (46.7%) patients achieved completeness of cytoreduction (CC) 0/1, and 24 (53.3%) cases achieved CC 2/3. There were 42 (93.3%) and 3 (6.7%) patients with and without cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy (CRS + HIPEC), respectively.

BAP1/MTAP Immunohistochemistry

Three BM cases diagnosed by morphology and routine immunohistochemical staining were reclassified to EM with reactive stroma according to BAP1 and MTAP IHC (Figure 2(A) and (B)). Two of eighteen (11.1%) cases with BM were loss/decreased in both epithelioid component (EC) and sarcomatoid component (SC). Nine of twenty seven (33.3%) cases with EM with reactive stroma were loss of or decreased BAP1 expression; however, reactive stroma was positive for BAP1 in all EM. Four of eighteen (22.2%) cases with BM were loss/decreased MTAP expression, and three (16.7%) cases were loss/decreased in both EC and SC, and one case was loss/decreased in only SC (Figure 2(C) and (D)). Four of twenty seven (14.8%) cases with EM were loss/decreased staining of MTAP. However, reactive stroma was positive for MTAP in all EM. BAP1/MTAP loss/decrease was observed in 6 (33.3%) cases of BM and 12 (44.4%) cases of EM, but not in reactive stroma. In 5 (27.8%) cases, loss of or decreased BAP1 or MTAP expression was observed in both EC and SC of BM (Table 2).

Correlation Analysis Between BAP1/MTAP Expression Status and Clinicopathological Characteristics of PeM

According to expression of BAP1/MTAP, 45 PeM cases were divided into two groups: loss/decreased group (n = 18) and normal group (n = 27). There were significant differences between expression status of BAP1/MTAP with PCI score ($P = .047$) and CC score ($P = .038$), but not with gender ($P = 0.088$), age ($P = 0.893$), history of asbestos exposure ($P = 0.777$), ascites ($P = 0.220$), CA125 level ($P = 0.712$), pathological type ($P = 0.456$), history of CRS + HIPEC

Table 1. Clinicopathological Characteristics of PeM.

Variable	Value
Gender, n (%)	
Male	22 (48.9)
Female	23 (51.1)
Age (years), n (%)	
≤60	32 (71.1)
>60	13 (28.9)
Asbestos exposure, n (%)	
No	34 (75.6)
Yes	11 (24.4)
PCI score, n (%)	
<25	18 (40.0)
≥25	27 (60.0)
CC score, n (%)	
0/1	21 (46.7)
2/3	24 (53.3)
Ascites (mL), n (%)	
0	7 (15.6)
0–1000	20 (44.4)
>1000	18 (40.0)
CA125 level (U/mL), n (%)	
<35	19 (42.2)
≥35	26 (57.8)
CRS + HIPEC, n (%)	
No	3 (6.7)
Yes	42 (93.3)
Pathological type, n (%)	
Epithelioid	27 (60.0)
Biphasic	18 (40.0)
Vascular tumor emboli, n (%)	
No	36 (80.0)
Yes	9 (20.0)
Ki-67 index (%), n (%)	
≤10	4 (8.9)
>10	41 (91.1)
TNM stage, n (%)	
I/II	24 (53.3)
III	21 (46.7)

Abbreviations: PeM, peritoneal mesothelioma; PCI, peritoneal cancer index; CC, completeness of cytoreduction; CA125, carbohydrate antigen (CA)125; CRS + HIPEC, cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy; TNM, tumor node metastasis.

($P = 0.143$), vascular tumor emboli ($P = 0.761$), Ki-67 index ($P = 0.521$), or TNM stage ($P = 0.714$), as shown in Table 3.

Survival Analysis

As of November 30, 2022, the median follow-up time was 13.13 months (range 2.03–60.53 months), and the median OS was 16.13 months (95% CI: 10.12–22.14 months). Thirty-two patients (71.1%) were dead, and thirteen (28.9%) were alive. According to pathological type, patients with BM had worse OS than that of EM with reactive stroma ($P = .007$). There was no significant difference in OS according to BAP1, MTAP, or BAP1/MTAP expression status ($P = .375$, .235, and .859, respectively). However, when patients were stratified into two groups based on age, older patients (>60 years) had a worse prognosis ($P = .011$). In addition, patients without CRS + HIPEC had poorer OS ($P = .005$) (Figure 3).

Analysis of Risk Factors Affecting OS in PeM Patients

Univariate Cox regression analysis was applied to analyze risk factors affecting prognosis. Age (HR = 2.635, 95% *CI*: 1.217–5.706, $P = .014$), history of CRS + HIPEC (HR = 5.415, 95% *CI*: 1.443–20.323, $P = .012$), and pathological type (HR = 2.730, 95% *CI*: 1.278–5.831, $P = 0.009$) were correlated with OS of PeM patients. In order to include as many as significant risk factors, those with a *P*-value below .1 in univariate Cox regression were selected into multivariate Cox analysis model. Age (HR = 2.927, 95% *CI*: 1.311–6.536, $P = 0.009$), history of CRS + HIPEC (HR = 6.951, 95% *CI* = 1.693–28.538, $P = 0.007$), and pathological type (HR = 2.309, 95% *CI*: 1.067–5.000, $P = 0.034$) were independent risk factors for PeM prognosis. Unfortunately, the expression statuses of BAP1 or MTAP were not independent prognostic factors in PeM (Table 4).

Discussion

It is difficult to distinguish BM from EM with reactive stroma by morphology alone. In pleura, BAP1/MTAP protein determination is a diagnostic tool to differentiate biphasic mesothelioma from epithelioid mesotheliomas with reactive stroma.[14,15] To the best of our knowledge, this study is the first to evaluate BAP1/MTAP expression in peritoneal mesothelioma to differentially diagnose BM from EM with reactive stroma. The sensitivity of loss of BAP1 in EM and BM as detected by IHC had been 61%–77% and 33%–49%, respectively, which is higher than that in sarcomatoid mesothelioma (SM) (0%–22%).[16,17] Multiple studies have reported that BAP1 loss was virtually with 100% specificity for differential diagnosis of MM.[18] The sensitivity of MTAP was relatively unsatisfactory (43%–65%), but MTAP had 96%–100% specificity in pleural MM.[19] Loss of MTAP by IHC had 96%–100% specificity and ∼80% sensitivity for predicting *p16/CDKN2A* homozygous deletion (HD) by FISH.[20] Considering that FISH detection requires specialized technology and is relatively expensive, MTAP IHC offers a potential advantage, although FISH analysis is more accurate.[21] Previous studies have generally demonstrated that combination of MTAP and BAP1 by IHC in MM diagnosis had greater sensitivity (74%–90%) and specificity (96%–100%), although the sensitivity was slightly lower (approximately 10%) than that of *p16/CDKN2A* FISH combined with BAP1 IHC.[11,14,22]

Loss/decrease of BAP1/MTAP in the spindle cell component supports the diagnosis of BM and that if BAP1/MTAP loss/decrease is confined to the EC a diagnosis of BM should be made only if the spindled component shows unequivocal

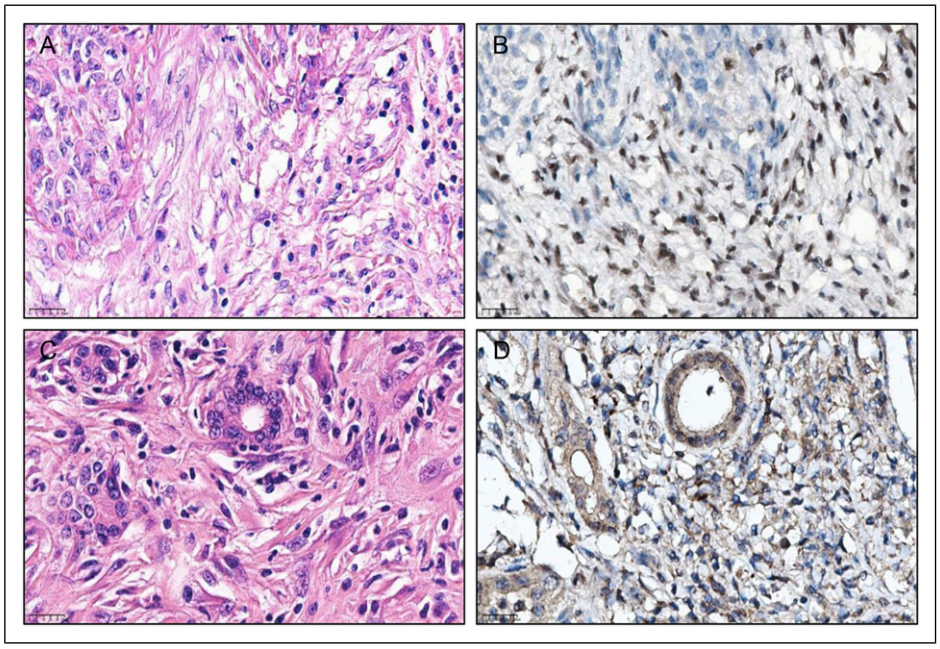

Figure 2. (A, B) Biphasic mesothelioma (BM) was reclassified to epithelioid mesothelioma (EM) with reactive stroma according to BAP1 expression [loss/decreased was observed in only epithelioid component (EC)]. (C, D) 1 case of BM was loss/decreased MTAP expression only in sarcomatoid component (SC). (hematoxylin and eosin [H&E] stain, immunohistochemistry stain, ×800 magnification).

Table 2. Summary of the Results of BAP1 and MTAP Expression in PeM.

		Biphasic Mesothelioma (n = 18)			Epithelioid Mesothelioma (n = 27)	
		EC/SC	EC	SC	EC	Stroma
BAP1	Normal	16 (88.9%)	16 (88.9%)	16 (88.9%)	18 (66.7%)	27 (100.0%)
	Loss/decreased	2 (11.1%)	2 (11.1%)	2 (11.1%)	9 (33.3%)	0 (0.0%)
MTAP	Normal	14 (77.8%)	15 (83.3%)	14 (77.8%)	23 (85.2%)	27 (100.0%)
	Loss/decreased	4 (22.2%)	3 (16.7%)	4 (22.2%)	4 (14.8%)	0 (0.0%)
BAP1/MTAP	Normal	12 (66.7%)	13 (72.2%)	12 (66.7%)	15 (55.6%)	27 (100.0%)
	Loss/decreased	6 (33.3%)	5 (27.8%)	6 (33.3%)	12 (44.4%)	0 (0.0%)

Abbreviations: PeM, peritoneal mesothelioma; EC, epithelioid component; SC, sarcomatoid component; BAP1, Breast Cancer 1 protein-associated protein 1; MTAP, methylthioadenosine phosphorylase.

morphologic features of malignancy.[10] In this study, 3 cases (BAP1/MTAP only loss/decrease in EC) were finally diagnosed as an EM with reactive stroma but not BM. The sensitivity of BAP1/MTAP loss/decrease was 33.3% (6/18) and 44.4% (12/27) in BM and EM in this study, respectively, as detected by IHC, with 100% specificity. In previous literature, loss of BAP1 and MTAP expression was observed in 12 (67%) PeM (11 epithelioid and 1 biphasic) and 3 (17%) PeM, respectively.[23] Comparable to this report, our results show slightly lower BAP1 or MTAP loss in MPM. It was reported that 5 of 13 (38.5%) cases of pleural BM were negative for BAP1, among which, 4 cases were negative for both EC and SC.[10] We observed that 2 of 18 (11.1%) cases were loss/decreased BAP1 for both EC and SC of peritoneal BM. These results indicate that loss of BAP1 expression in SC of peritoneal BM is similar to that in pleura. While 4 of 18 (22.2%) cases with BM were loss/decreased MTAP expression, 3 (16.7%) cases were loss/decrease in both EC and SC, and 1 case was loss/decreased in only SC. And BAP1/MTAP loss/decrease was observed in 5 (27.8%) cases in both EC and SC. Loss/decrease of BAP1/MTAP was not observed in reactive stroma of EM, which was comparable to those in reports that have been published thus far.[10,13] These results show that combination of BAP1/MTAP by IHC is helpful for differential diagnosis of peritoneal BM from EM with reactive stroma. However, due to a limited sample size, further studies should

Table 3. Correlation Between BAP1/MTAP Expression Status and Clinicopathological Characteristics of PeM.

Variable	n (%)	BAP1/MTAP Status (n, %)		*P*-Value
		Loss/Decreased (n = 18)	Normal (n = 27)	
Gender				0.088
Male	22 (48.9)	6 (33.3)	16 (59.3)	
Female	23 (51.1)	12 (66.7)	11 (40.7)	
Age (years)				0.893
≤60	32 (71.1)	13 (72.2)	19 (70.4)	
>60	13 (28.9)	5 (27.8)	8 (29.6)	
History of asbestos exposure				0.777
No	34 (75.6)	14 (77.8)	20 (74.1)	
Yes	11 (24.4)	4 (22.2)	7 (25.9)	
PCI score				**0.047**
<25	18 (40.0)	4 (22.2)	14 (51.9)	
≥25	27 (60.0)	14 (77.8)	13 (48.1)	
CC score				**0.038**
0/1	21 (46.7)	5 (27.8)	16 (59.3)	
2/3	24 (53.3)	13 (72.2)	11 (40.7)	
Ascites (mL)				0.220
0	7 (15.6)	2 (11.1)	5 (18.5)	
0–1000	20 (44.4)	6 (33.3)	14 (51.9)	
>1000	18 (40.0)	10 (55.6)	8 (29.6)	
CA125 level (U/mL)				0.712
<35	19 (42.2)	7 (38.9)	12 (44.4)	
≥35	26 (57.8)	11 (61.1)	15 (55.6)	
History of CRS + HIPEC				0.143
No	3 (6.7)	0 (.0)	3 (11.1)	
Yes	42 (93.3)	18 (100.0)	24 (88.9)	
Pathological type				0.456
Epithelioid	27 (60.0)	12 (66.7)	15 (55.6)	
Biphasic	18 (40.0)	6 (33.3)	12 (44.4)	
Vascular tumor emboli				0.761
No	36 (80.0)	14 (77.8)	22 (81.5)	
Yes	9 (20.0)	4 (22.2)	5 (18.5)	
Ki-67 index (%)				0.521
≤10	4 (8.9)	1 (5.6)	3 (11.1)	
>10	41 (91.1)	17 (94.4)	24 (88.9)	
TNM stage				0.714
I/II	24 (53.3)	9 (50.0)	15 (55.6)	
III	21 (46.7)	9 (50.0)	12 (44.4)	

Abbreviations: PeM, peritoneal mesothelioma; PCI, peritoneal cancer index; CC, completeness of cytoreduction; CA125, carbohydrate antigen (CA) 125; CRS + HIPEC, cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy; TNM, tumor node metastasis; BAP1, Breast Cancer 1 protein-associated protein 1; MTAP, methylthioadenosine phosphorylase. Bold in Table 3 means *P* -Value < 0.05.

be conducted. In addition, HD of *p16/CDKN2A* occurred in 96.6% (28/29) of pleural BM (both in EC and SC) and 77.8% (7/9) of EM. However, none of these harbored deletions of *p16/CDKN2A* in reactive stroma, which could allow for more accurate differentiation of BM from EM with reactive stroma when in combination with BAP1 expression by IHC.[10] The sensitivity of MTAP by IHC is slightly lower than that of *p16/CDKN2A* by FISH. Therefore, FISH testing of *p16/CDKN2A* will be conducted in future studies.

The diagnostic value of BAP1/MTAP expression had been explored in MM, while the association between BAP1/MTAP expression status and clinicopathological features of MM remains unclear, especially in PeM. In our study, loss/decrease of BAP1/MTAP was not related to gender, history of asbestos exposure, ascites, CA125 level, pathological type, history of CRS + HIPEC, vascular tumor emboli, Ki-67 index, or TNM stage. Notably, BAP1/MTAP loss/decrease was associated with higher PCI and CC

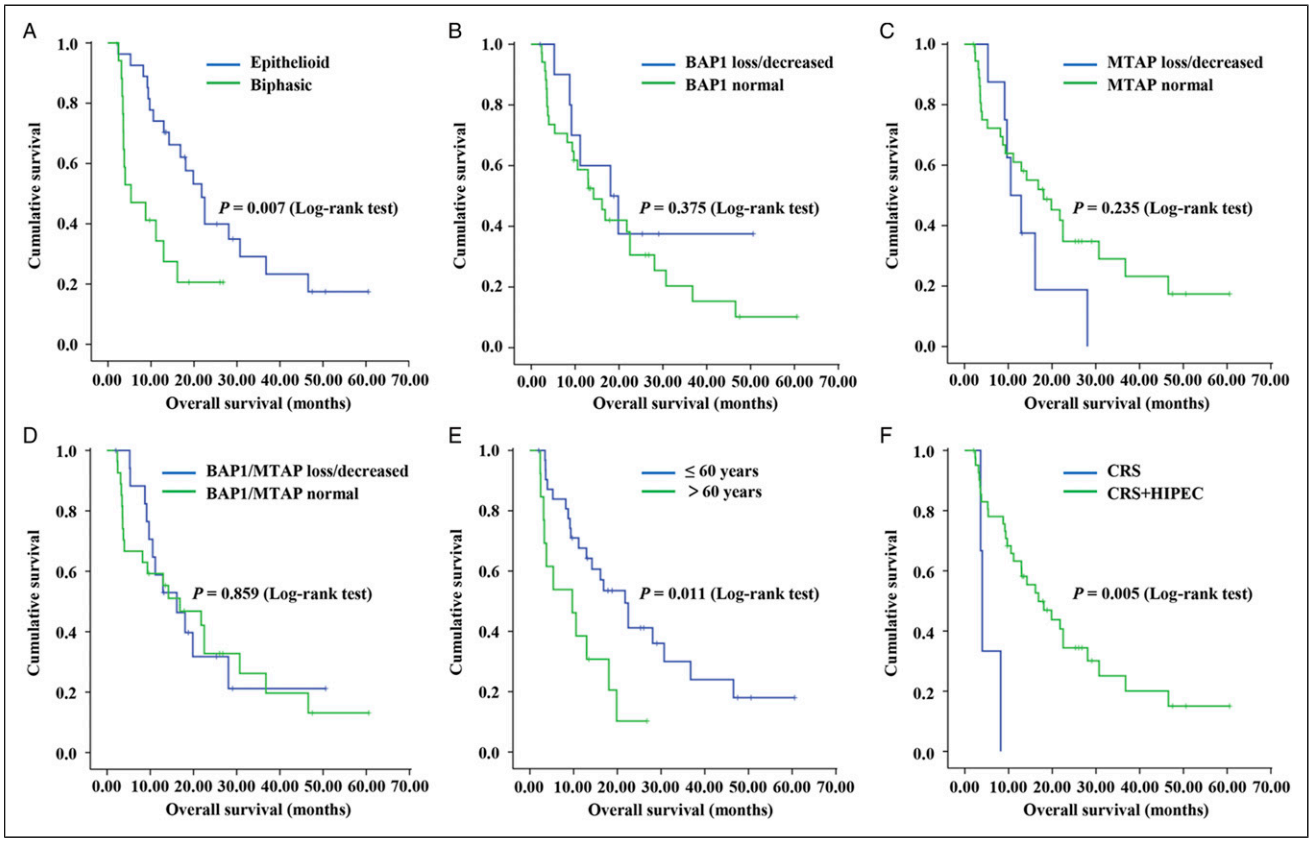

Figure 3. Survival analysis. (A) Pathological type; (B) BAP1 protein expression; (C) MTAP protein expression; (D) BAP1/MTAP protein expression; (E) age; and (F) therapy.

Table 4. Univariate and Multivariate Cox Regression Analysis of OS in PeM Patients.

	Univariate Analysis		Multivariate Analysis	
Variable	HR (95% *CI*)	*P*-Value	HR (95% *CI*)	*P*-Value
Gender (female vs male)	1.650 (.813–3.348)	0.165		
Age (;60 vs≤ 60 years)	2.635 (1.217–5.706)	**0.014**	2.927 (1.311–6.536)	**0.009**
History of asbestos exposure (yes vs no)	1.016 (.449–2.299)	0.970		
PCI score (≥25 vs < 25)	1.092 (.768–1.551)	0.624		
CC score (2/3 vs 0/1)	1.012 (.503–2.036)	0.974		
CA125 level (U/mL) (≥35 vs < 35)	1.210 (.651–2.646)	0.447		
Ascites (yes vs no)	1.695 (.853–1.715)	0.286		
History of CRS + HIPEC (no vs Yes)	5.415 (1.443–20.323)	**0.012**	6.951 (1.693–28.538)	**0.007**
Pathological type (biphasic vs epithelioid)	2.730 (1.278–5.831)	**0.009**	2.309 (1.067–5.000)	**0.034**
Vascular tumor emboli (yes vs no)	1.008 (.661–1.538)	0.969		
Ki-67 (>10% vs ≤ 10%)	4.670 (.635–34.352)	0.130		
TNM stage (III vs I/II)	1.087 (.766–1.543)	0.641		
BAP1 (loss/decreased vs normal)	.819 (.525–1.278)	0.379		
MTAP (loss/decreased vs normal)	1.296 (.840–2.000)	0.241		
BAP1/MTAP (loss/decreased vs normal)	1.068 (.517–2.204)	0.859		

Abbreviations: PeM, peritoneal mesothelioma; OS, overall survival; PCI, peritoneal cancer index; CC, completeness of cytoreduction; CA125, carbohydrate antigen (CA)125; CRS + HIPEC, cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy; TNM, tumor node metastasis; BAP1, Breast Cancer 1 protein-associated protein 1; MTAP, methylthioadenosine phosphorylase. Bold in Table 4 means *P* -Value < 0.05.

scores. PCI score (range 0–39) can evaluate the spread of peritoneal cancer during laparotomy or CT examination.[24] The residual tumors were intraoperatively classified by CC scores, including 4 groups: CC-0 (no residual tumors), CC-1 (residual tumor with a diameter of <2.5 mm), CC-2 (residual tumor with a diameter of between 2.5 mm and 2.5 cm), and CC-3 (residual tumor with a diameter of >2.5 cm).[25] Previous study had shown that PCI score and CC score were related to the prognosis of PeM.[26] We speculated that BAP1/MTAP expression status was associated with PeM prognosis. In previous study, loss of expression of BAP1 is related to a more favorable prognosis in pleural MM.[27] While in this study, although there was a trend affecting OS by BAP1 expression, it was not statistically significant. In addition, loss of MTAP and BAP1/MTAP were not associated with PeM prognosis. However, BM subtype was correlated with shorter OS than EM with reactive stroma subtype ($P = 0.007$). This can be attributed to essential difference between SC in BM and reactive stroma in EM. In addition, older patients (>60 years) and those without CRS + HIPEC also had poorer OS ($P = 0.011$ and $P = 0.005$, respectively). The above 3 factors were all independent risk factors for worse prognosis. Compared to traditional treatment, CRS + HIPEC has improved the median OS by up to 19–92 months, which is currently the preferred choice for PeM.[28]

The limitation of this study is a relatively small sample size. Thus, it is necessary to conduct multi-center studies with larger sample sizes to identify more precise molecular markers for differential diagnosis of PeM with spindle cells.

Conclusion

In our study, we found that expression of BAP1/MTAP by IHC is helpful for the differentiation between peritoneal BM and EM with reactive stroma. BAP1/MTAP expression status was correlated with PCI score and CC score, while, to a certain extent, BAP1 was associated with clinical outcome, indicating that BAP1/MTAP might help to evaluate the biological behavior of PeM.

Abbreviations

BAP1	Breast Cancer 1 protein-associated protein 1
BM	Biphasic mesothelioma
CA125	Carbohydrate antigen (CA) 125
CC	Completeness of cytoreduction
CRS + HIPEC	Cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy
EC	Epithelioid component
EM	Epithelioid mesothelioma
FISH	Fluorescence in situ hybridization
IHC	Immunohistochemistry
PeM	peritoneal mesothelioma
MTAP	Methylthioadenosine phosphorylase
OS	Overall survival
PCI	Peritoneal cancer index
SC	Sarcomatoid component
TNM	Tumor node metastasis.

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Authors' Contributions

YC was responsible for searching literature and drafting the manuscript and performed statistical analyses. XD reviewed the slides and participated in the analysis and interpretation of data. YG performed the immunohistochemistry. HW and YS collected demographic and clinical data and followed up. HZ was in charge of routine pathology.

Declaration of Conflicting Interests

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Research Ethics and Patient Consent

The study protocol was approved by the Medical Ethics Committee of Beijing Shijitan Hospital, Capital Medical University (Beijing, China; approval number: 2022-36; date of approval: August 12, 2022). All patients signed informed consent and agreed to use postoperative specimens and clinical data for medical research.

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