International Journal Dental and Medical Sciences Research
Volume 4, Issue 6, Nov-Dec 2022 pp 871-874www.ijdmsrjournal.com ISSN: 2582-6018

Cytologic Diagnosis of Mesothelioma with Emphasis on Two Steps Immunohis to chemistry

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Submitted: 18-12-2022 Accepted: 31-12-2022

ABSTRACT
INTRODUCTION: Distinguishing malignant mesothelioma, adenocarcinoma and reactive mesothelial proliferation in both cytological and surgical specimens is often a diagnostic challenge. Very few studies have reported the utility and challenges in diagnosing mesothelioma on cytologic specimens. This study was done to evaluate cytological features and panel of immunohistochemical(IHC)markers useful for cytological diagnosis of mesothelioma.
AIMS AND OBJECTIVES: To study clinical, cytomorphological and IHC features of cytologically diagnosed cases of malignant mesothelioma seen over a period of five years at our hospital.
MATERIAL AND METHODS: Cases diagnosed as mesothelioma were retrieved from the medical records of Cytopathology labover a period of five years(January 2012 to December 2017). The clinical details, site, imaging, cytomorphological and IHC features were retrieved alongwith histopathological follow up wherever available.
OBSERVATION AND RESULTS: A total of eleven cytologic specimens from ten patients comprising of three ascitic fluids, one pleural fluidand sevenFNA specimens from pleural thickening(3 cases),omental thickening(1 case),mediastinal lymph nodes (1case) right hilar mass (1case) and peribronchial mass (1 case) obtained by EBUS-TBNA, CT- guidance or under ultrasound guidance were available. Mean age at diagnosis was 63.5 years including eight males and two females. On imaging, patients had omental thickening and recurrent ascitis or pleural thickening and recurrent pleural effusions. On cytology, smears were cellular with groups, fragments, ball like formations and singly dispersed polygonal cells showing moderately pleomorphic nuclei and prominent nucleoli. IHC was doneon cell blocks in two steps with first step showing positivity with calretinin and negativity with Berep4/CEA followed by second step showing positivity with WT-1, EMA, p53 and negativity with desmin.Histopathological follow up was available in fourcases which confirmed the diagnosis of mesothelioma.
CONCLUSION: Malignant mesothelioma needs to be distinguished from reactive mesothelial cells and metastatic adenocarcinoma. Preparation of cell blocks followed by use of selected panel of IHC markers can aid indiagnosis on cytology.
Keywords: Mesothelioma, cytomorphology, IHC markers.

I. INTRODUCTION:

Mesothelioma is a primary neoplasm arising from mesothelial cells of pleura, peritoneum, pericardium and tunica vaginalis testis.Malignant mesothelioma is a rare neoplasm withthe incidence being 1-2 per million.[1]Asbestos, Zeolites ,Radiation, SV-40 virus infection are known risk factors.Large number of these patients present with recurrent effusions or pleural and peritoneal masses. The cytologic examination of the fluid or FNA is one of the first diagnostic techniques attempted in these patients. Distinguishing malignant mesothelioma from metastatic adenocarcinoma and reactive mesothelialcell proliferation on morphology in both cytological and surgical specimens is often a diagnostic challenge. This is due to overlapping morphologic appearances like presence of fragments and ball like clusters of round to polygonal cells with mild to moderately pleomorphic nuclei with prominent nucleoli and moderate amount of cytoplasm. A number of techniques, including cell block preparation, immunohistochemical(IHC) and ultrastructural analysis have been used to solve these diagnostic dilemmas.[1-7]Despite the potential shown by many antibodies, it is generally agreed that no single antibody has absolute specificity or sensitivity and hence a panel of immunohistochemical markers is considered as a valuable and useful tool.[6,7,8]The current study presents a pattern based approach to the diagnosis of mesothelioma and their mimics and explores the role of limited panel of IHC markers for the diagnosis of malignant mesothelioma.

II. MATERIAL AND METHODS:

Cases diagnosed or suspected as mesothelioma were retrieved from the records of

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Cytopathology lab over a period of five years (January2012 toDecember2017).A total of eleven specimens from ten patients were available. These included three ascitic fluids, one pleural fluid and seven FNA specimens. FNA sites were pleural thickening(3 cases), omental thickening(1 case),peribronchial mass(1 case), right hilar mass(1 case)and mediastinal lymph node(1 case). FNA specimens were obtained by EBUS-TBNA or under CT or ultrasound guidance.The demographic details like age/sex,signs and symptoms, imaging findings and suspected clinical diagnosis were recorded. The cytomorphological features and IHC features were studied. Histopathological follow up wherever available was obtained.In all cases wet fixed smears were stained with Papanicolaou(PAP) stain while air dried smears were stained with May Grunwald giemsa(MGG) stain. Cell blocks were made in all cases and selected panel of IHC markers were applied on the cell blocks. Calretinin (mesothelial marker) and Berep4/CEA(epithelial marker) were applied as first step IHC to determine the cell of origin as mesothelial or epithelial. When mesothelial,WT-1, EMA, p53 and desmin were done in second step to characterize the mesothelial cells as reactive or malignant. Cytological diagnosis was rendered on the basis of cytomorphology and immunohistochemistry results.

III. RESULTS:

A total of eleven specimens obtained from ten patients were evaluated which included three ascitic fluids, one pleural fluid and seven FNAs. Patients ranged in age from 58 years to 70 years with majority being males (8/10). Commonest presentation was recurrent effusions(4/10) with fever(9/10), shortness of breath(8/10) and weight loss(9/10).In none of the cases history of exposure to asbestos or other predisposing factors was elicited.Imaging of three cases showed pleural thickening (case 5, 8 and 9) and one case showed omental thickening(case10). PET-CT scan was done in case no.8 and showed FDG-avid mediastinal, abdominal and supraclavicular lymph nodes.Anti-tubercular therapy had been received by three patients(case3, 5and 6).

Smears from fluids (cases 1 to 4) were cellular. Large round to polygonal cells arranged in fragments, ball like clusters as well as singly dispersed were seen. Cells hadmoderately pleomorphic nuclei, abundant cytoplasm and prominent nucleoli. Differential diagnosis considered on morphology were reactive mesothelial hyperplasia, metastatic adenocarcinoma and mesothelioma. (Fig-1)

Smears from FNA (cases 5 to 10) were cellular.Cellswere arranged in fragments, loose cell groups, papillary clusters and singly dispersed. Cells were large, round to polygonal with mild to moderately pleomorphic nuclei, prominent nucleoli and moderate amount of cytoplasm. (Fig-2)

Smears from case 8b (mediastinal lymph node) in addition showed lymphoid tissue in the background.Differential diagnosis considered in cases 5, 8a, 9 (pleural thickening) and case10 (omental thickening) were mesothelioma and reactive mesothelial hyperplasia while that in cases 6,7and 8b were metastatic adenocarcinoma,mesothelioma and reactive mesothelial cell hyperplasia.

To arrive at a definitive diagnosis, a panel of IHC markers was selected as a two step approach in all cases. Calretinin (mesothelial marker) and Berep4/CEA (epithelial markers) were done in first step to determine the cell of origin as either mesothelial or epithelial. Having recognized them as mesothelial by positivity for calretinin in all cases studied, further panel of markers were done to characterize them as malignant or reactive. For that WT1, EMA, p53, and desmin were applied. WT-1,EMA and p53 positivity along with desmin negativity confirmed the diagnosis of malignant mesothelioma(in cases 1 to 8).However, case 9 and case10 were reported as reactive mesothelial cell hyperplasia based on IHC features which showed EMA and p53 negativity and desmin positivity in the second step.

Histopathological follow up was available in four cases(case 1, 3 5and 8) which confirmed the cytologic diagnosis of mesothelioma.

IV. DISCUSSION AND CONCLUSION:

Mesotheliomas are the most frequent primary malignant tumors of serosal cavities with a poor prognosis. Common sites are pleura (75%) and peritoneum (22-23%) with rare involvementof pericardium and tunica vaginalis testis. A definitive and early diagnosis is important as newer therapies have good prognosis in patients with stage I disease[3]. Cytology is increasingly being used in the initial evaluation of effusions and pulmonary disorders.FNA specimens obtained via EBUS-TBNA, under CT or ultrasound guidance are widely accepted, safe and mininimally invasive techniques to evaluate pleural pathologies. Distinguishing malignant mesothelioma from metastatic adenocarcinoma and reactive mesothelial cell proliferation on morphology is often a diagnostic challenge due to overlapping

morphologic appearances like presence of fragments and ball like clusters of round to polygonal cells with mild to moderate pleomorphism with prominent nucleolus and moderate amount of cytoplasm. Cell block preparation, IHC markers and ultrastructural analysis have been used to solve these diagnostic dilemmas.[1-7]Since no single antibody has absolute specificity or sensitivity,a panel of immunohistochemical markers is considered as a valuable and useful tool.[6,7,8]Very few studies have been reported discussing the cytological features and panel ofIHC markers useful for the diagnosis of mesothelioma.[1-5]

In this study, we highlight the cytomorphological features in 11specimens comprising of 4 fluids and 7 FNA that were suspicious of mesothelioma and were later confirmed by a panel of IHC markers. Due to the overlapping features of mesothelioma with reactive mesothelial cell hyperplasia and adenocarcinoma,a diagnosis of mesothelioma on cytomorphology alone is not possible and needs ancillary tests like IHC. In the present study, a panel of IHC markers was done in a two step approach in all cases to limit the use of antibodies in each case. However review of literature shows selection of various antibodies for the diagnosis of mesothelioma like Hjerpe et al [1] suggested the panel of atleast four IHC markers, two in favour of MM(EMA and calretinin) and two excluding it(CEA and Berep4)to be done for the confirmation supported by WT-1 and podoplanin for the diagnosis of MM if the results are equivocal. To differentiate from reactive mesothelial cells desmin(positive in reactive mesothelial cells)and p53(positive in MM) have been recommended by them. Similarly study by Nishino et al [5] highlighted the use of panel of immunostains like BerEp4 , B72.3, MOC-31, and Claudin-4 as epithelial markers while WT-1,calretinin and D2-40 [podoplanin] as mesothelial markers for diagnosis of mesothelioma.

Some studies have recommended the use of special stains along with cytomorphology for diagnosis of mesothelioma. Ehya et al demonstrated in their study that positive cytological stain with PAS after diastase digestion and positivemucicarmine stain after hyaluronidase treatment are against the diagnosis of malignant mesothelioma. On the other hand,positive stain with alcian blue which becomes negative after treatment with hyaluronidase is strongly suggestive of malignant mesothelioma.[10]

One unusual case in our study was that of malignant mesothelioma metastasic to mediastinal lymph nodes (case 8). Lymph node involvement by mesothelioma is rare as biological behaviour of mesothelioma is characterized by relentless local progression to chest wall or pericardium with rare hematogenous and lymphatic spread in late stages.[8,9]The current study highlights one such rare case of pleural mesothelioma metastatic to mediastinal lymph nodes(case 8).

However,the key criteria in histopathological diagnosis of mesothelioma regarding the evidence of invasive growth in pleural soft tissues and subjacent lung parenchyma is impossible to determine on cytology specimens.[9]

Medico-legal implications constitute a significant preanalytical consideration when contemplating the cytological diagnosis of MM. History of asbestos exposure must be actively sought for. A significant exposure reinforces the suspicion of MM. The diagnosis of MM must be made based on solid grounds, including cytomorphology and ancillary techniques such as IHC.

Patients with recurrent pleural effusions and/or ascitis along with pleural nodules should raise the suspicion of mesothelioma.A practical approach to the diagnosis of malignant mesothelioma on cytology includes (1) exclusion of metastatic adenocarcinoma. (2) determing the cells as of mesothelial origin (3) Use of further panel of IHC markers to differentiate malignant meosthelioma from reactive mesothelial cells. Early and accurate diagnosis of mesothelioma on cytologyspecimens can direct appropriate treatment and improve the survival rate in these patients.

REFERENCES:

[1]. Hjerpe , Ascoli , Bedrossian , Boon , Creaney , Davidson et al.Guidelines for the Cytopathologic Diagnosis of Epithelioid and Mixed-Type Malignant Mesothelioma.ActaCytol2015;59:2-16

[2]. Sterrett , Whitaker , Shilkin , Walters .Fine needle aspiration cytology of malignant mesothelioma.ActaCyol1987;31: 185-93

[3]. Pomjanski , Grote , Sander , Heikaus , Müller , Böckinget al. Early cytological diagnosis of diffuse malignant mesothelioma of the peritoneum: a case report. DiagnCytopath 2008;36: 120-3

[4]. Husain, Colby, Ordonez , Krausz , Attanoos , Beasley et al. Guidelines for pathologic diagnosis of malignant mesothelioma: 2017 update of the consensus statement from the International Mesothelioma Interest Group.ArchPathol Lab Med 2018;142:89-108

[5]. Nishino.Is the cytodiagnosis of mesothelioma any easier these days? Cancer Cytopathol2014;122:857-8.

[6]. Abutaily, Addis , Roche . Immunohistochemistry in the distinction between malignant mesothelioma and pulmonary adenocarcinoma: a critical evaluation of new antibodies. J ClinPathol2002; 55:662–8.

[7]. Soomin,In Ho Choi, Joungho, Jhingook, Myung-Ju. Pleural Mesothelioma: An Institutional Experience of 66 Cases. Korean J of Pathol 2014; 48: 91-9.

[8]. Jung. Pathological diagnosis of malignant mesothelioma. J Korean Med Assoc 2009; 52:456–64

[9]. Abdel Rahman , Gaafar, Baki , El Hosieny, Aboulkasem, Farahat. Prevalence and pattern of lymph node metastasis in malignant pleural mesothelioma.AnnThoracSurg 2008;86:391-5

[10]. Ehya. The cytologicdiagnosis of mesothelioma.SeminDiagnPathol1986;3:196-203

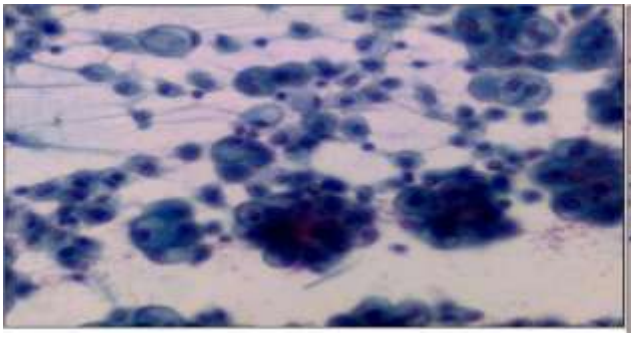

Figure 1a

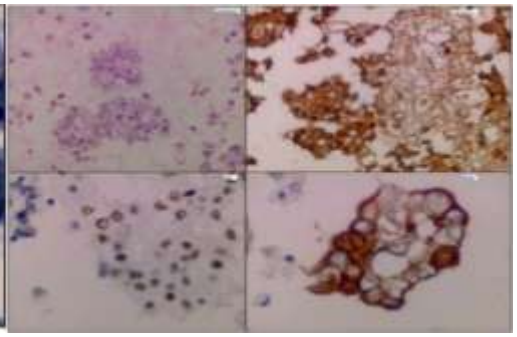

Figure 1b

Case1Ascitic fluid

Figure 1a-fragments and ball like arrangement of mesothelial cells with moderately pleomorphic nuclei and abundant vacoulatedcytoplasm,PAP(X200)

Figure1b-Cellblock showing similar morphology,H&E(X100),IHC:calretinin(nuclear positive)X100, P53-1(nuclear positive)X100, EMA(membranous positive)X400

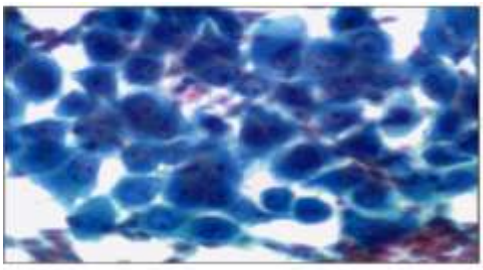

Figure 2a

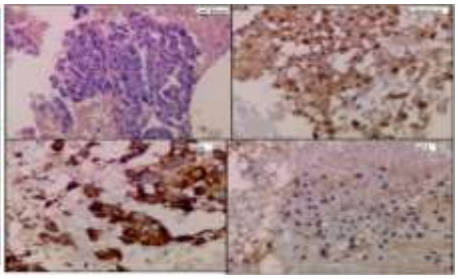

Figure 2b

CASE 5 EBUS-TBNA Peribronchial mass

Fig 2a- cells in loose groups with pleomorphic nuclei, prominent nucleoli and moderate amount of cytoplasm, PAP(X400)

Fig 2b- Cell block showing similar cells in fragments,H&E(X400).IHC:Calretinin(nuclear positive) X100,,EMA(membranous positive) X100,WT-1(nuclear positive) X100