

## ORIGINAL PAPER

# Expression of cytokeratin MNF116 and vimentin in pleural serous effusions

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### Abstract

The purpose of this study is to evaluate the value of cytokeratin (CK) MNF116 and vimentin in the differential diagnosis of malignant pleural effusions. There were evaluated smears from 30 patients with pleural effusions stained with May–Grünwald Giemsa and Papanicolaou techniques for the routine cytological diagnosis. Additional smears were immunostained with CK MNF116 and vimentin using LSAB2 technique. Two independent observers evaluated all smears. Smears were classified first by cytological examination in seven cases (23.33%) as benign, and in 23 cases (76.67%) as malignant pleural effusions. Mesothelial cells expressed CK MNF116 in 96.67% (29/30) of cases and vimentin in 33.33% (10/30) of cases. Malignant cells expressed CK MNF116 in 52.17% (12/23) of cases and vimentin in 30.43% (7/23) of cases. The pattern of immunostaining was diffuse cytoplasmic. In conclusion, CK MNF116 and vimentin may be used as a part of the panel of antibodies for differential diagnosis of malignant pleural effusions with primary unknown.

**Keywords:** immunocytochemistry, cytokeratin, vimentin, pleural effusions.

### Introduction

MNF116 antibody detects an epitope that is present in a wide range of epithelia, corresponding to keratin numbers 5, 6, 8, 17 and probably 19 [1]. In normal tissue, it reacts with epithelial cells from simple glandular to stratified squamous epithelium.

In surgical pathology, a wide range of benign and malignant epithelial tumors exhibit positive immunoreactivity for cytokeratin MNF116. Squamous cell carcinoma, small cell carcinoma, sarcomatoid carcinoma, spindle cell carcinoma, epithelioid and spindle cell component of malignant mesothelioma and adenocarcinoma demonstrate a strong pattern of immunostaining [2].

A wide range of soft tissue tumors is also positive with cytokeratin MNF116: monophasic and biphasic synovial sarcoma, vascular neoplasms including epithelioid hemangioendothelioma, epithelioid angiosarcoma [3], epithelioid sarcoma [4, 5].

Desmoplastic small round cell tumors require cytokeratin positivity for diagnosis. Smooth muscle tumors and plasmocytoma may demonstrate aberrant expression of cytokeratin MNF116 [6, 7].

Vimentin expression is specific for mesenchymal cells but can be coexpressed by a number of epithelial cells and their corresponding tumors (endometrium, thyroid, gonadal epithelial cells, renal tubules, adrenal cortex, lung, salivary gland, hepatocytes and biliary duct). It had been suggested that a variety of high-grade epithelial tumors may acquire the expression of vimentin [8].

Vimentin expression had been described in carcinoma of the skin [9], urinary bladder, breast [10], gastric mucosa [11], and prostate [12].

### Material and methods

We studied 30 patients who were admitted in County Hospital Sibiu with diagnosis of pleural effusion. Pleural effusions were centrifuged at 1000 rotations/minute; smears were stained with May–Grünwald Giemsa and Papanicolaou methods for routine cytological diagnosis.

Additional smears were immunostained with cytokeratin, clone MNF116 and vimentin, clone V9 using LSAB2 technique and DAB for visualize the reaction. Positive immunostaining were defined as cytoplasmic pattern and positivity threshold was 10% of the tumor cells. Two independent observers evaluated smears.

### Results

Cytological examination of May–Grünwald Giemsa and Papanicolaou stained smears classified the cases in 7 (23.33%) inflammatory pleural effusions and 23 (76.67%) malignant pleural effusions.

The malignant pleural effusions were four pleural metastases of small cell carcinoma, 17 pleural metastases of adenocarcinoma, one metastatic malignant melanoma and one large cell anaplastic lymphoma.

In 16 cases, the primary site was confirmed by CT, bronchoscopy, digestive endoscopy, and surgical pathology: three cases of small cell carcinoma of lung; one case of small cell carcinoma of female genital tract;

two cases of ovarian adenocarcinoma; one case of breast carcinoma; two cases of gastric adenocarcinoma; seven cases of lung adenocarcinoma; five cases were malignant pleural effusions with primary unknown.

Mesothelial cells demonstrate immunoreactivity for cytokeratin MNF116 in 96.67% (29/30) of cases with strong cytoplasmic pattern and vimentin in 33.33% (10/30) of cases with weak, focal cytoplasmic pattern.

Malignant cells expressed cytokeratin MNF116 in 52.17% (12/23) of cases with strong cytoplasmic pattern and vimentin in 30.43% (7/23) of cases, with weak, focal cytoplasmic pattern either strong diffuse cytoplasmic pattern.

Small cell carcinoma of lung expressed cytokeratin MNF116 in 66.67% (2/3) of cases, and vimentin in 66.67% (2/3) of cases with diffuse cytoplasmic pattern; small cell carcinoma of female genital tract was negative for cytokeratin MNF116 and was positive for vimentin (Figure 1).

Cytokeratin MNF116 and vimentin expression was absent in ovarian adenocarcinoma; lung adenocarcinoma expressed cytokeratin MNF116 in 57.14% (4/7) of cases (Figure 2) with strong cytoplasmic, diffuse pattern, and vimentin in 28.57% (2/7) cases with diffuse cytoplasmic pattern; gastric carcinoma expressed cytokeratin MNF116 in 50% (1/2) of cases and was negative for vimentin in both cases.

Breast carcinoma react with cytokeratin MNF116 (Figure 3) and demonstrate strong, diffuse cytoplasmic pattern and with vimentin with weak, focal cytoplasmic pattern.

In malignant pleural effusions with primary unknown, malignant cells expressed cytokeratin MNF116 in 80% (4/5) of cases with strong cytoplasmic pattern (Figure 4), and vimentin in 20% (1/5) of cases with diffuse cytoplasmic pattern.

Cytokeratin MNF116 and vimentin were negative in metastatic malignant melanoma and in large cell anaplastic lymphoma. Metastatic malignant melanoma expressed S100 protein and large cell anaplastic lymphoma expressed CD30.

Co-expression of cytokeratin MNF116 and vimentin was present in three cases of reactive pleural effusion and in four cases of malignant pleural effusion: one metastatic breast carcinoma, two of small cell carcinoma of lung and one unknown primary malignant pleural effusion.

## ☒ Discussions

Cytological diagnosis of malignant pleural effusions is often difficult and requires the differential diagnosis between reactive mesothelial cells, malignant mesothelioma, adenocarcinoma, malignant melanoma, sarcoma and lymphoma. Metastases from carcinoma of unknown primary site in pleural effusion are a common clinical problem [13]. Diagnosis of malignant melanoma in serous effusion is facilitated

by the presence of cytoplasmic pigment. In the absence of obvious pigment, additional studies are required to confirm the diagnosis [14].

Sarcoma account for only 3–6% of malignant effusions and their diagnosis was frequently made in the setting of a known primary tumor; these tumors often exhibit various morphological features that differ from those of original tumors and may preclude the correct diagnosis; immunocytochemical techniques are useful in differential diagnosis between carcinoma, melanoma and sarcoma. The most sarcoma, with the exception of synovial sarcoma and epithelioid sarcoma are negative for epithelial markers [15].

Anti Human Cytokeratin, clone MNF116 stains the cytoplasm of most epithelial tumors, including tumors arising from simple epithelium and squamous cell tumors. Carcinoma of various tissue including squamous cell carcinoma, adenocarcinoma and all mesothelioma are labeled [16, 17].

Vimentin, clone V9 reacted positively with malignant melanoma, meningioma, peripheral nerve sheath tumor and sarcoma. Mesothelioma, large cell lymphoma, adenocarcinoma, small cell carcinoma, small cell undifferentiated carcinoma, carcinoid, thymoma exhibit variable positivity (10–57%). Co-expression of vimentin with keratin was often present in undifferentiated carcinoma, synovial and epithelioid sarcoma [18].

In our study, malignant cells expressed cytokeratin MNF116 in 52.17% (12/23) of cases and vimentin in 30.43% (7/23) of cases; cytokeratin MNF116 and vimentin were negative in metastatic melanoma and in anaplastic large cell lymphoma. Metastatic malignant melanoma expressed S100 protein and anaplastic large cell lymphoma expressed CD30.

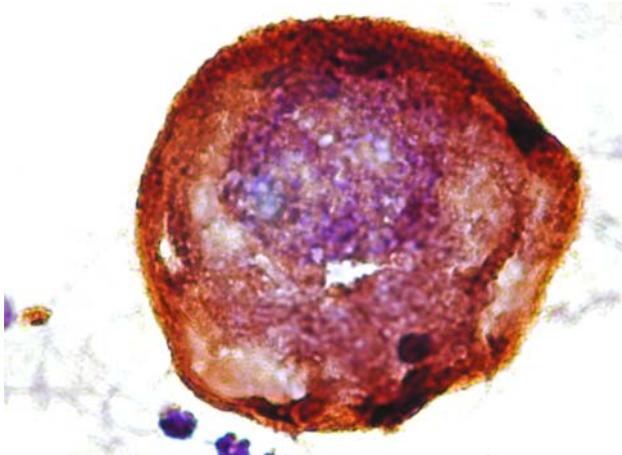
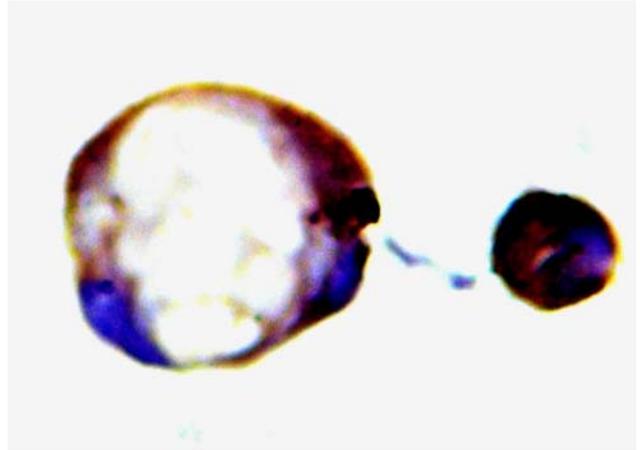
Small cell carcinoma of the lung expressed cytokeratin MNF116 in 66.67% (2/3) of cases, and vimentin in 66.67% (2/3) of cases with diffuse cytoplasmic pattern. Co-expression of cytokeratin MNF116 and vimentin was present in two cases of small cell carcinoma of the lung. Small cell carcinoma of female genital tract was negative for cytokeratin MNF116 and was positive for vimentin.

Breast carcinoma reacted with cytokeratin MNF116 and demonstrated strong, diffuse cytoplasmic pattern and with vimentin with weak, focal cytoplasmic pattern. Cytokeratin MNF116 and vimentin expression was absent in ovarian adenocarcinoma.

Lung adenocarcinoma expressed cytokeratin MNF116 in 57.14% (4/7) of cases with strong cytoplasmic, diffuse pattern and vimentin in 28.57% (2/7) of cases with diffuse cytoplasmic pattern; we did not found co-expression of cytokeratin and vimentin in lung adenocarcinoma.

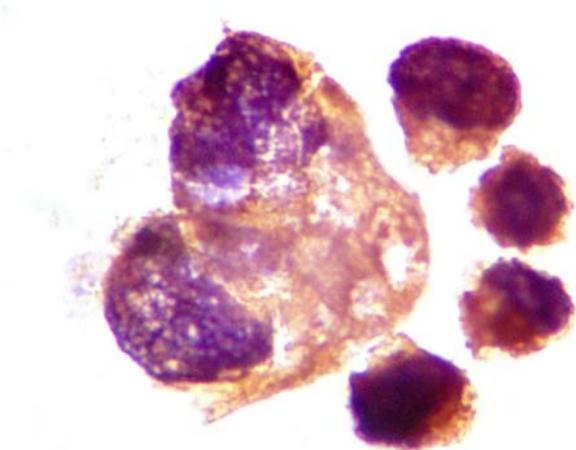
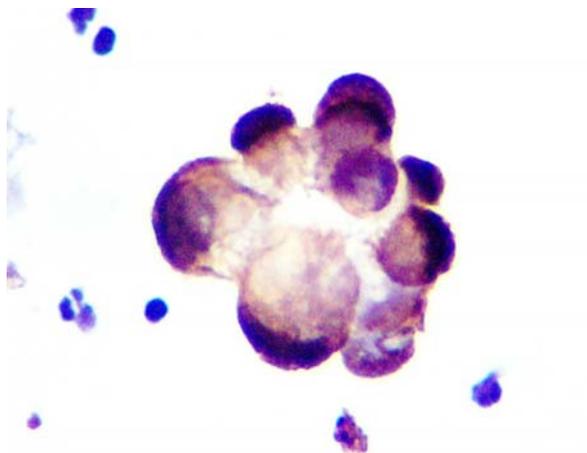
Gastric adenocarcinoma expressed cytokeratin in 50% (1/2) of cases and was negative for vimentin in both cases. In malignant pleural effusions of primary unknown, malignant cells expressed cytokeratin in 80% (4/5) of cases with diffuse, strong cytoplasmic pattern and vimentin in 20% (1/5) of cases with diffuse cytoplasmic pattern; one of cases co-expressed cytokeratin MNF116 and vimentin.

**Figure 1 – Vimentin positive malignant cell in malignant pleural effusion with primary site a lung adenocarcinoma**



**Figure 2 – Cytokeratin MNF116 positive malignant cell in a malignant pleural effusion with lung adenocarcinoma as primary site**

**Figure 3 – Cytokeratin MNF116 positive malignant cells in a malignant pleural effusion with breast adenocarcinoma as primary site**



**Figure 4 – Cytokeratin MNF116 positive malignant cells in a metastatic pleural effusion with unknown primary site**

## ☐ Conclusions

Our results strongly suggest that cytokeratin MNF116 and vimentin are useful in panel for differential diagnosis of malignant pleural effusion with primary unknown when differential diagnosis between carcinoma, melanoma, lymphoma and sarcoma is required.

## References

- [1] MOLL R., FRANKE W. W., SCHILLER D. L., GEIGER B., KREPLER R., *The catalog of human cytokeratins: pattern of expression in normal epithelia, tumors and cultured cells*, Cell, 1982, 31(1):11–24.
- [2] MIETTINEN M., *Keratin immunohistochemistry: update of applications and pitfalls*, Pathol Annu, 1993, 28(Pt 2):113–143.
- [3] MENTZEL T., BEHAM A., CALONYE E., KATENKAMP D., FLETCHER C. D., *Epithelioid haemangioendothelioma of skin and soft tissue: clinicopathologic and immunohistochemical study of 30 cases*, Am J Surgical Pathol, 1997, 21(4):363–374.
- [4] EVANS H. L., BAER S. C., *Epithelioid sarcoma: a clinicopathologic and prognostic study of 26 cases*, Semin Diagn Pathol, 1993, 10(4):286–291.
- [5] GUILLOU L., WADDEN C., COINDRE J. M., KRAUSZ T., FLETCHER C. D., *“Proximal-type” epithelioid sarcoma, a distinctive aggressive neoplasm showing rhabdoid feature. Clinicopathologic, immunohistochemical, and ultrastructural study of a series*, Am J Surg Pathol, 1997, 21(2):130–146.
- [6] RAMAECKERS F. C., PRUSZCZYNSKY M., SMEDTS F., *Cytokeratins in smooth muscle cells and smooth muscle tumors*, Histopathology, 1988, 12(5):558–561.
- [7] WOTHERSPOON A. C., NORTON A. J., ISAACSON P. G., *Immunoreactive cytokeratins in plasmacytomas*, Histopathology, 1989, 14(2):141–150.
- [8] LEONG A. S. Y., *The expression of vimentin in epithelial neoplasms*. In: FENOGLIO-PREISER C. M., WOLFF M., RILKE F. (eds), *Progress in Surgical Pathology*, vol. XII, Springer Verlag, Berlin–Heidelberg–New York, 1992, 31–48.
- [9] IYER P. V., LEONG A. S.-Y., *Poorly differentiated squamous cell carcinomas of the skin can express vimentin*, J Cutan Pathol, 1992, 19(1):34–39.
- [10] RAYMOND W. A., LEONG A. S., *Vimentin a new prognostic parameter in breast carcinoma?*, J Pathol, 1989, 158(2):107–114.
- [11] TAKEMURA K., HIRAYAMA R., HIROKAWA K., INAGAKI M., TSUJIMURA K., ESAKI Y., MISHIMA Y., *Expression of vimentin in gastric carcinoma: a possible indicator for prognosis*, Pathobiology, 1994, 62:149–154.
- [12] LEONG A. S., GILHAM P., MILIOS J., *Cytokeratin and vimentin intermediate filament proteins in benign and malignant prostatic epithelium*, Histopathology, 1988, 13(4):435–442.
- [13] POMJANSKI N., GROTE H. J., DOGANAY P., SCHMIEMANN V., BUCKSTEGGE B., BÖCKING A., *Immunocytochemical identification of carcinomas of unknown primary in serous effusions*, Diagn Cytopathol, 2005, 33(5):309–315.
- [14] BEATY M. W., FETSCH P., WILDER A. M., MARINCOLA F., ABATI A., *Effusion cytology of malignant melanoma, A morphologic and immunocytochemical analysis including application of the MART-1 antibody*, Cancer Cytopathol, 1997, 81(1):57–63.
- [15] ABADI M. A., ZAKOWSKI M. F., *Cytologic features of sarcomas in fluids*, Cancer, 1998, 84(2):71–76.
- [16] PRIETO V. G., MCNUTT N. S., SHEA C. R., LUGO J., *Expression of cytokeratin 10, 17 and 18 in normal and pathological skin as detected by the monoclonal antibody MNF116*, J Invest Dermatol, 1993, 100:547.
- [17] WALSH C. B., HAMILTON V., CURRAN B., LEADER M., *Diagnostic application of the cytokeratin MNF116 in a wide range of sarcomas, carcinomas and lymphomas*, J Pathol, 1993, 167(Suppl):148A.
- [18] AZUMI N., BATTIFORA H., *The distribution of vimentin and keratin in epithelial and nonepithelial neoplasms. A comprehensive immunohistochemical study on formalin- and alcohol-fixed tumors*, Am J Clin Pathol, 1987, 88(3):286–296.

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