https://doi.org/10.48047/AFJBS.6.14.2024.11929-119234

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

Role of Immunohistochemical Panel in Accurate Diagnosis of Non-Small Cell Lung Cancer Bronchoalveolar Lavage

Menatallah Hesham Mohamed Fekry[1*], Yosria Mohammed El-Gohary[1], Hesham Radwan Abd-Elaziz, Mona Mostafa Ahmed[1], Tarek hassan Abdelhamid[2]

[1] Pathology Department, Faculty of Medicine, Zagazig University

[2] Chest Diseases Department, Faculty of Medicine, Zagazig University

Corresponding author: Menatallah Hesham Mohamed Fekry

Email: fikrymenna45@gmail.com

Article History

Volume 6, **Issue** 14, 2024

Received: 28 July 2024

Accepted: 5 September 2024

Published: 6 September 2024

doi: 10.48047/AFJBS.6.15.2024.11929-11934

Abstract:

Non-small cell lung cancer shows marked genetic and clinicopathological heterogeneity, which may explain its poor prognosis despite the continuous development of target therapy. Diagnosis of lung cancer is established by examination of small tissue specimens obtained by minimally invasive techniques. It is critical to employ these tissues at maximum efficiency in order to render an accurate pathologic diagnosis and to perform studies, either genomic or by immunohistochemistry, to demonstrate genetic mutations that make patients eligible for molecularly targeted agents.

Keywords: Immunohistochemical staining, Bronchoalveolar Lavage (BAL) , Non-small cell lung cancer.

1. Introduction

Immunohistochemistry is a less challenging, widely available technique which provides clinically meaningful results quickly and allows for cellular localization of proteins in the context of tumor structure. Pathologists are required to classify lung cancer into specific subtypes and assess which one relevant to molecular targeted therapies but overlapping of microscopic features of different types of tumours sometimes makes diagnosis difficult for pathologists **[1].**

Morphology alone might not be conclusive, especially in cases due to poor differentiation, small biopsies, heterogeneity of tumors, and the artifacts. Advanced evaluation using immunohistochemistry (IHC) and molecular genetics have been useful in solving such cases. Owing to the most practical solution, usually IHC provides a suitable conclusion **[2].**

TTF1 (Thyroid transcription factor 1):

Known as NK2 homeobox 1 (NKX2–1), is a 38 kDa nuclear protein encoded by the NKX2–1 gene and physiologically expressed by surfactant-producing type 2 pneumocytes and Club cells, formerly known as Clara cells, in the lung. TTF-1 is expressed in approximately 70–80% of lung adenocarcinoma and is routinely used to distinguish lung adenocarcinoma from metastases from extrapulmonary adenocarcinomas **[3].**

So it is considered as a critical single marker for adenocarcinoma, while is p40 for squamous cell carcinoma, with napsin A also showing some diagnostic utility as a secondary marker for adenocarcinoma. When compared with corresponding surgical resection, TTF1 had slightly better performance than napsin A, whereas a combination of TTF1 and napsin A may yield greater sensitivity for adenocarcinoma **[4].**

Napsin A:

Napsin A is an aspartic proteinase involved in the maturation of the surfactant protein B in lung tissue . It is abundantly expressed in the cytoplasm of normal type II pneumocytes and Clara cells, in addition to lung ADCs, however, it is also expressed by kidney proximal and convoluted tubule cells, and renal cell carcinomas **[5]**

P40 and p63:

Both P40 and P63 are products of the P63 gene on chromosome 3q27–29, and are expressed by the basal or progenitor cell layer of bronchial epithelium but P40 had a higher specificity and lower sensitivity than that of P63 in diagnosis of lung SqCC **[6]**

At present, there are a number of reliable immunohistochemical markers that have been adopted to distinguish LUAD from LUSC as shown in the following figure (1), **"adenocarcinoma markers,"** TTF-1 and Napsin A, mainly expressed in solid adenocarcinoma. If poorly differentiated carcinoma lacking light microscopic evidence of squamous differentiation Non-keratinizing SqCC is proven by immunohistochemistry to express **"SqCC markers**," such as p40, CK5/6 and p63. Because of this classification, the proportion of large cell carcinoma has been markedly reduced **[7].**

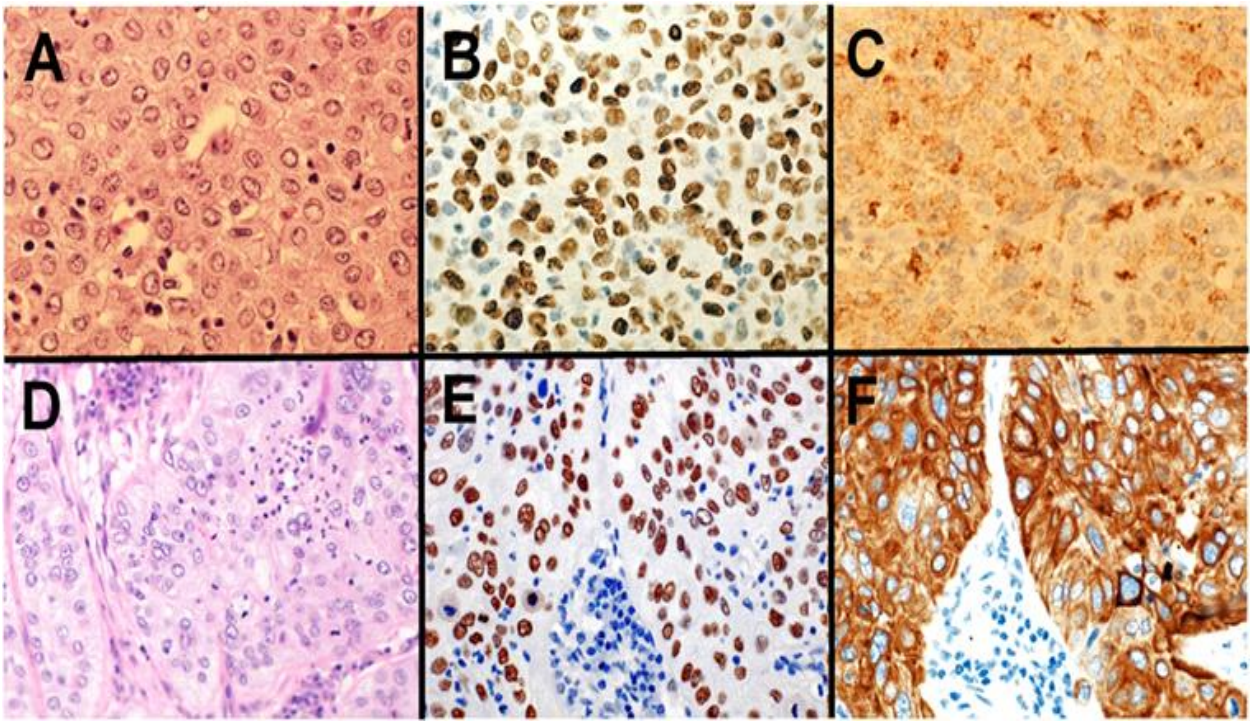

Figure (1): Solid adenocarcinoma (A–C) and non-keratinizing squamous cell carcinoma (SqCC) (D–F). Solid adenocarcinoma [(A) HE staining] is immunohistochemically positive for TTF-1 (B) and Napsin A (C). Non-keratinizing SqCC [(D) HE staining] is immunohistochemically positive for p40 (E) and CK5/6 (F) **[7].**

Panel of IHC markers used to overcome the limitation of Immunohistochemical expression especially in poorly differentiated carcinoma lacking glandular differentiation and cytokeratin formation and small biopsy specimens as shown in figure 2 **[8].**

One of limitations of individual IHC markers used in diagnosis of NSCLC is Cross-reaction with other tissue types and tumors is a limitation of current IHC markers. For example, TTF-1 is immunoreactive with normal pulmonary alveolar macrophages, whereas, both P40 and P63 are immunoreactive with normal bronchial basal epithelial cells **[6].**

In addition, individual IHC markers require multiple sections of tumor tissue for the test to be performed. It is not uncommon for tumor tissue to be exhausted by IHC studies leaving little tissue for molecular analyses, particularly when using small biopsy specimens **[8].**

Triple marker panel is used instead in practice to subclassify NSCLC. The triple marker consists of TTF-1, Napsin A and P40 and has sensitivity of 86.0% and a specificity of 100% in identifying lung ADCs, and a sensitivity of 100% and a specificity of 97.1% in identifying lung SqCCs **[9].**

That the combination panel is cost-efficient and could conserve valuable tumor tissues during the immunohistochemical study of NSCLCs and conserving tumor tissues during the immunohistochemical subclassification of difficult cases **[10].**

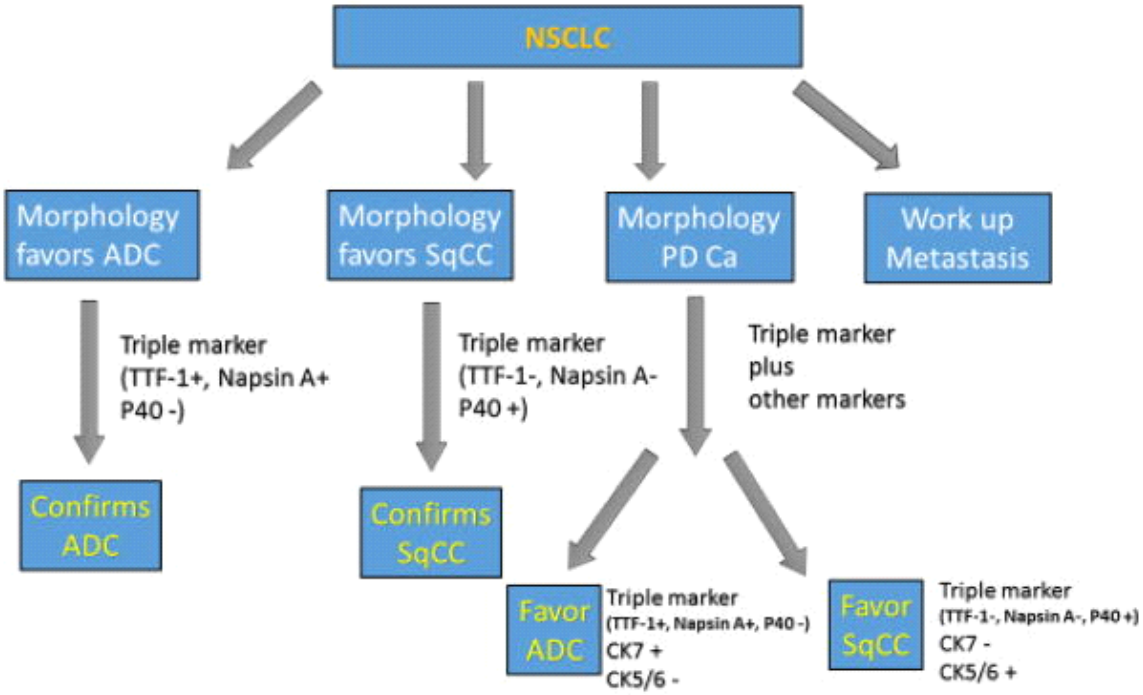

Figure (2): The work flow for the differential diagnosis of NSCLC using the triple marker. Based on the histomorphological assessment of the specimen, different immunomarkers are performed **(**PD Ca: poorly differentiated carcinoma**) [8].**

Finally, IHC may not be needed if samples are large enough for routine H&E; however, when used, IHC can provide a considerable amount of diagnostic information. In small specimens, IHC with one lung adenocarcinoma (LUAD) marker (e.g., TTF1 or napsin A) and one lung squamous cell carcinoma (LUSC) marker (e.g., p40 or p63) will suffice. TTF1 is expressed in 77% of patients with LUAD and is not expressed in LUSC, while p40 is highly expressed in LUSC and minimally expressed in LUAD **[11].**

Lung cytopathology Bronchoalveolar lavage (BAL):

Biopsies are time-consuming and there is increased pressure on pathologist to report cases as early as possible. Cytologic techniques are safer, economical, and provide quick results **[12].**

BAL provided the highest diagnostic yield when cytopathological correlation was done as compared with other cytological techniques like brush cytology in diagnosis of primary and secondary malignancies especially those involving lung parenchyma close to the smaller airways **[13].**

For Normal range composition of BAL comprises of macrophages (83%-94%), neutrophils (1%-4%) (increased to 6% in smokers), lymphocytes (5%-16%), eosinophils (<1%) and columnar respiratory epithelial cells (>2%). In smokers the yield increases 4 times. Alteration in normal composition of BAL and increase in specific cell line cut offs may indicate certain specific diagnoses as infection , immune diseases and cancers **[14].**

Although paraffin blocks are routinely processed in pathology laboratories, and these blocks provide multiple sections for various analyses. However, as many as 40%–70% of all advanced

NSCLC are diagnosed by cytological evaluation alone, with no concurrent histological examination of tissue material **[15].**

References:

1. N. Pandya, V. Popat. (2019). THE ROLE OF IMMUNOHISTOCHEMISTRY IN DIAGNOSIS OF LUNG CANCER WITH CORRELATION OF SERUM TUMOUR MARKER CARCINOEMBRYONIC ANTIGEN. Journal of Evolution of Medical and Dental Sciences, 8(10), 701-706.
2. S. Umakanthan, A. C. Rao, W. Mohammed. (2021). Role of immunohistochemistry markers in neoplastic lung lesions. Journal of Cancer Research and Therapeutics, 17(6), 1382-1388.
3. S. Schallenberg, G. Dernbach, M. P. Dragomir, G. Schlachtenberger, K. Boschung, C. Friedrich, N. Frost. (2024). TTF-1 status in early-stage lung adenocarcinoma is an independent predictor of relapse and survival superior to tumor grading. European Journal of Cancer, 197, 113474.
4. Y. Yatabe, S. Dacic, A. C. Borczuk, A. Warth, P. A. Russell, S. Lantuejoul, A. L. Moreira. (2019). Best practices recommendations for diagnostic immunohistochemistry in lung cancer. Journal of thoracic oncology, 14(3), 377-407.
5. S. Nageshwar Rao, K. Anuradha Vutukuru, K. Durga, K. B. Mithila V. Sailaja, Viaya Sreedhar. (2024). Study of p40 is superior to p63 for the diagnosis of pulmonary squamous cell carcinoma on cytological smears International Journal of Community Medicine and Public Health Rao SN et al. Int J Community Med Public Health. Apr;11(4):1507-1512
6. M. T. Lilo, D. Allison, Y. Wang, M. Ao, E. Gabrielson, S. Geddes, Q. K. Li. (2016). Expression of P40 and P63 in lung cancers using fine needle aspiration cases. Understanding clinical pitfalls and limitations. Journal of the American Society of Cytopathology, 5(3), 123-132.
7. K. Inamura. (2017). Lung cancer: understanding its molecular pathology and the 2015 WHO classification. Frontiers in oncology, 7, 193.
8. L. Osmani, F. Askin, E. Gabrielson, Q. K. Li. (2018). Current WHO guidelines and the critical role of immunohistochemical markers in the subclassification of non-small cell lung carcinoma (NSCLC): Moving from targeted therapy to immunotherapy. Seminars in cancer biology, 52(Pt 1), 103–109.
9. R. Sharma, Y. Wang, L. Chen, G. T. Gurda, S. Geddes, E. Gabrielson, Q. K. Li. (2016). Utility of a novel triple marker (combination of thyroid transcription factor 1, Napsin A, and P40) in the subclassification of non–small cell lung carcinomas using fine-needle aspiration cases. Human pathology, 54, 8-16.
10. J. P. Gregg, T. Li, K. Y. Yoneda. (2019). Molecular testing strategies in non-small cell lung cancer: optimizing the diagnostic journey. Translational lung cancer research, 8(3), 286–301.
11. F. Kanavati, G. Toyokawa, S. Momosaki, H. Takeoka, M. Okamoto, K. Yamazaki, M. Tsuneki. (2021). A deep learning model for the classification of indeterminate lung carcinoma in biopsy whole slide images. Scientific reports, 11(1), 8110.
12. R. Sareen, CL. Pandey (2016). Lung malignancy: Diagnostic accuracies of bronchoalveolar lavage, bronchial brushing, and fine needle aspiration cytology. Lung India. 2016 Nov-Dec;33(6):635-64

13. B. Efared, G. Ebang-Atsame, S. Rabiou. (2017). The diagnostic value of the bronchoalveolar lavage in interstitial lung diseases. J Negat Results Biomed. 2017; 16: 4.
14. P.H. Patel, M.H. Antoine, A. Sankari, S. Ullah (2024). Bronchoalveolar Lavage. In Stat Pearls. Stat Pearls Publishing.
15. S. Roy-Chowdhuri, S. Dacic , M Ghofrani (2020) Collection and handling of thoracic small biopsy and cytology specimens for ancillary studies. Arch Pathol Lab Med