The Importance of Computed Tomography Guıded Transthorasic Fine Needle Aspiration Biopsy / Cell Block Method in Early Diagnosis and Treatment of Lung Lesions: Cytopathologic Analysis in 354 Cases

Akciğer Lezyonlarının Erken Tanı ve Tedavisinde Bilgisayarlı Tomografi Eşliğinde Transtorasik İnce İğne Aspirasyon Biopsisi/Hücre Bloğu Yönteminin Önemi: 354 Vakada Sitopatolojik Analiz

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Abstract

Objective	The aim of this study was to evaluated the effectiveness of computed tomography(CT) guided transthorasic fine needle aspiration biopsy in the diagnosis of lung mass lesions. **(Sakarya Med J 2019, 9(3):513-521)**.
Materials and Methods	In this study, we are presenting 354 CT guided TTFNAB materials. The 348 of aspirations were performed from lung directly and 6 of them were performed from masses with mediastinal location. After the lesion was localized by CT, aspiration biopsy was performed by thoracic surgeon with 20-22 gauge Chiba needle by using 20 ml. If TTFNAB material is sufficient, it was divided into two equal parts for conventional smear cytology and the cell block technique. The results were classified in following catogories: 1. Benign/non-neoplastic, 2. suspicious of malignancy, 3. neoplastic-diagnostic, 4. inadequate for interpretation.
Results	Among the 220 patients who underwent TTFNAB (354 aspiration biopsy), 163 (46.04%) were diagnosed as tumor, 74(20.90%) as suspicious lesion, 71 (20.05) as benign lesion and 46 (12.99%) as aspirate nondiagnostic. 140 (39.54%) aspirata cell blocks were applied to 134 cases. Immunohistochemical and histochemical studies were performed in these cases and type differentiation could be made in tumor. Suspicious for neoplasia and neoplastic cases formed major group with 237 cases (66.94%). Among 163 (46.04%) neoplastic aspirates; there were the diagnoses of EC in 58 (35.58%) aspirates, NSSC/non-specific in 54 (33,12 %) aspirates, AC in 23 (14.11%) aspirates, SCC in 14 (8.58%), malignant tumor in 8 aspirates, neuroedocrine tumor in 3 aspirates, thymoma in 1 aspirate, plasmacytoma in 1 aspirate and schwannoma in 1 aspirate.
Conclusion	TTFNAB-CB in expert hands and experienced pathologists is fairly accurate for lung masses and can be regarded as an acceptable procedure for early diagnose and treatment planning purposes in most lung lesions.

Keywords lung; computed tomography; transthoracic fine needle aspiration biopsy; cell block

Öz

Amaç	*Bu çalışmanın amacı, akciğer kitle lezyonlarının tanısında bilgisayarlı tomografi (BT) kılavuzluğunda transtorasik ince iğne aspirasyon biyopsisinin (TTİİAB) etkinliğini değerlendirmektir.* ***(Sakarya Tıp Dergisi 2019, 9(3):513-521)***
Gereç ve Yöntemler	*Bu çalışmada 354 BT eşliğinde TTİİAB materyalini ve tecrübelerimizi geriye dönük literatür eşliğinde sunmaktayız. Aspirasyonların 348'i direkt akciğer, kalan 6'sı mediasten lokalizasyonlu kitleden yapılmışdı. Lezyonlar BT ile lokalize edildikten sonra göğüs cerrahı tarafından 20ml. 20-22 gauge Chiba iğnesi kullanılarak aspirasyonlar gerçekleştirildi. TTİİAB materyalleri yeterli ise konvansiyonel yayma sitolojisi ve hücre bloğu tekniği için iki eşit bölüme ayrıldı. Sonuçlar, aşağıdaki kategorilerde sınıflandırıldı: 1. Benign / neoplastik olmayan, 2. Neoplazi şüpheli, 3. Neoplastik tanısal, 4. Tanımlama için yetersiz.*
Bulgular	*TTİİAB (354 aspirasyon biopsisi) yapılan 220 hastanın 163 (%46.04)'ü tümör, 74 (%20.90)'ü şüpheli lezyon, 71 (%20.05)'i benign lezyon tanısı almış olup, 46 (%12.99) aspirat nondiagnostik özelliktedir. 134 vakadan 140 (%39.54) aspirata hücre bloğu uygulanmış olup bu vakalara immünohistokimyasal ve histokimyasal çalışmalar uygulanabilip tümörlerde tip ayrımı yapılabilmiştir. Neoplazi ve neoplazi şüpheli aspiratlar, 237 (% 66.94) vaka ile ana grubu oluşturdu. Neoplastik 163(46.04%) aspiratın 58'i (35.58%) EC, 54 (33,12 %) aspirat NSSC/spesifiye edilemeyen, 23 (14.11%) aspirat AC, 14'ü (8.58%) SCC, 8 aspirat malign tümör, 3 aspirat nöroendokrin tümör, 1 aspirat timoma, 1 aspirat plasmasitom ve 1 aspirat schwannomdu.*
Sonuç	*Tecrübeli patologların uzman ellerinde TTİİAB–hücre bloğu incelemesi çoğu akciğer lezyonunun erken tanı ve tedavi planlaması için uygun bir yöntem olarak kabul edilebilir. Pnömotoraks gibi basit ve tedavi edilebilir komplikasyonları olduğundan popülerliği klinisyenler, radyologlar ve patologlar arasında artmaktadır*

Anahtar Kelimeler *akciğer; bilgisayarlı tomografi; transtorasik ince iğne biopsisi; hücre bloğu*

INTRODUCTION

Lung cancer remains one of the leading causes of cancer mortality world wide. Much of the cases are at the advanced stage on diagnosis.[1] Early diagnosis have been shown to increase survival in localized lung cancer. Transthoracic fine needle biopsy was used for the first time by Leyden in order to take bacteriological samples at the end of the19th century, by Menetrier soon afterwards for the diagnosis of lung cancer.

Computed tomography (CT) guided transthorasic fine needle aspiration biopsy (TTFNAB) is a commonly used diagnostic method with the help of developing radiological methods in lung disease especially lung cancer.[2] All intra-thoracic lesions are now routinely and safely using TTFNAB under CT.[3-6] It is a rapid diagnostic method which can be selected firstly in particular for the lesions smaller than a few centimeters localized at mediastinum, pulmonary apex, medial upper lobe or periphery. However, the use of this method often results in pneumothorax development.[4] Recognition of the accuracy of TTFNAB and simpler methods of treating pneumothorax has brought this method within the each of most hospital radiologists and pathologists.[7-12]

It is known as a reliable method with limited morbidity risc and simple procedure with good patient acceptance.[7] The value of the diagnosis decreases in cases of inadequate sampling and taking necrotic material.

The diagnosis rate and accuracy in the making of differentiation of small cell carcinom a(SCC) and nonsmall cell carcinoma (NSCC) in tumor typing increased more with the cell block method applied to aspirates in recent years with the implementation of histochemical and immunohistochemical staining.

As the majorty of lung cancer is the diagnosed on aspiration cytology, or small biopsy samples, often obtained by increasingly experienced diagnostic procedures, the pathologist must obtain maximal diagnostic yield from cytology specimens. In about 90 percent of patients, needle biopsy provides enough tissue fort he pathologist to determine the cause of the abnormality.[13,14]

While differentiation of SCC and NSCC is certainly important for radical surgical procedure, now subtyping should be done in NSCC. Because, the specific subtypes have different answers from each other to various chemotherapeutic agents.[14]

The purpose of this retrospective study, determination of early diagnosis value, reliability, success of type determination in tumors and various lung lesions of CT guided TTFNAB and cell blocks was intended.

Materials and Methods

In this study, we are presenting 354 CT guided TTFNAB materials and our experience with literature. Results were evaluated retrospectively. The 348 of aspirations were performed from lung directly and 6 of them were performed from masses with mediastinal location. The lesion was localized by CT aspiration biopsy was performed by thoracic surgeon with 20-22 gauge Chiba needle by using 20 ml. injector. The patient was watched carefully for signs of pneumothorax and a follow-up X-ray performed four hours after the TTFNAB to look for any such signs. Fresh TTFNAB was received. If the material is sufficient, it was divided into two equal parts. One part was subjected to conventional smear cytology (CS) and the other part for the cell block (CB) technique.

In conventional smear technique; the sent material was centrifuged at 2500 rpm for 10-15 minutes. A minimum of two thin smear slight were prepared from the precipitate. One slight was prepared after air drying and stained with the May-Grünwald-Giemsa stain (MGG). The other slight was urgently fixed in 95% alcohol and stained with the Papanicolaou stain (Pab). In CB tecnique; the adaquate material that remained was subjected to fixation for one

hour by mixing with alcohol-formalin. After fixation, this fluid material was centrifuged at 2500 rpm for 10-15 minute. Tube taken from the bottom sediments of blotting paper soaked in alcohol. On the following day, this cell button sediment sample was processed along with routine histopathological specimens. CB sections of 5–6 µ thickness were prepared and stained with the hematoxylin and eosin stain (HE). Special stains like the periodic acid Schiff (PAS), mucin, pas-alcian blue and immunohistochemycal stain were performed wherever necessary. Classification of lung tumors was made according to the 2004 World Health Organization (WHO). The results were classified in following catogories: 1. Benign/non-neoplastic, 2. suspicious of malignancy, 3. neoplastic-diagnostic, 4. inadequate for interpretation (Table I).

Table I- Cytological diagnosis of pulmonary mass lesions

Cytological categories	Number of cases (n:354)	Percentage (%)
Non-neoplastic	71	20.06
Suspicious malignancy	74	20.90
Neoplastic-diagnostic	163	46.05
Inadequate	46	12.99

Necrotic materials were also included into the suspicious aspirates. Neoplastic materials were divided into NSCC / non-specific, epidermoid carcinoma (EC), adenocarcinoma (AC), small cell carcinoma (SCC) and other tumors. Cell block was prepared for 140 TTFNAB. Histopathologic examinations of the 71 cases could be done, cytologic diagnoses were compared with tissue diagnoses (Table II).

This study is a descriptive research.

The study was approved by the Ethical Committee of the institution.

There are no conflict interest.

TABLEII- The histopathological results

Cytologic diagnosis	Histopathologic diagnosis	Number of cases (n:354)
Non-diagnostic/inadequate	Nonspesific inflamation	4
	Cyst hydatic	2
	Timic cyst	1
	Hodgkin Lymphoma	1
	Bronchial Tissue	1
	Squamous cell carcinoma	1
Suspicious for malignancy	High grade dysplasia	1
	Nonspesific inflamation	3
	Malign mesothelioma	2
	PNET	1
	Atypical Carcinoid	1
	Malign tumor	1
	Small cell carcinoma	2
	NSCC	3
	Epidermoid carcinoma	6
Benign conditions	Nonspesific inflamation	5
	Aspergillosis	1
	Thymoma	1
	Adenocarcinoma	1
	Malign lymphoma	1
	Epidermoid carcinoma	2
	Undifferantiated carcinoma	1
Small Cell Carcinoma	Necrotic tissues	1
	Small Cell Carcinoma	1
Epidermoid Carcinoma	Moderate dysplasia	1
	Transitional cell carcinoma	1
	Epidermoid carcinoma	7
	Adenocarcinoma	1
	Small round-cell malignancies	1
Adenocarcinoma	Adenocarcinoma	2
	Inflammatory events	1
NSCC / unclassified	Adenocarcinoma	4
	NSCC	3
	Inflammatory events	3
Plasmacytoma	Plasmacytoma	1
Schwannoma	Schwannoma	1
Thymoma	Thymoma	1

NSCC: non small cell carcinoma
PNET: primitive neuroectodermal tumor

RESULTS

Total number of 220 patients-354 TTFNABs were selected for the study. The age ranged was from 31 to 89 years, with an average of 63.78 years. One hundred forty CBs from 354 aspirates brought to our laboratory were prepared for 134 patients. Only 11 of the CB (7.85%) were non-diagnostic, the failure rate was decreased more by applying the CB. One hundred twenty five CB preparations with adequate cellularity were subjected to histochemical and / or immunohistochemical staining. Forty-six (12.99%) of the 354 TTFNAB materials examined were non-diagnostic aspirates, the remaining 308 (87.00%) aspirates were adequate and could be diagnosed. The distribution of the 354 aspirated diagnosed with needle aspiration and CB.

Seven cyst hydatid, 1 aspergillus fungal infection, 1 granulomatous reaction were available in 71 (20,05%) aspirates with benign diagnosis, the remaining benign lesions were constituted with abscesses and inflammatory events(Table III).

Table III- Sub-categorization of cytologically non-neoplastic lesions

Non-neoplastic lesion(n:71)	Number of cases
Cyst hydatid	7
Aspergillus	1
Granulomatous inflamation	1
Abscess	10
Chronic non-spesific inflamation	52

Fourteen (18.91%) of 74 (20.90%) suspicious aspirates were necrotic material. As expected, suspicious for neoplasia and neoplastic cases formed major group with 237 cases (66.94%). Among 163 (46.04%) neoplastic aspirates; there were the diagnoses of EC in 58 (35.58%) aspirates (Figure 1,2), NSSC/non-specific in 54 (33,12 %) aspirates, AC in 23 (14.11%) aspirates(Figure 3), SCC in 14 (8.58%), malignant tumor in 8 aspirates, neuroedocrine tumor in 3 aspirates, thymoma in 1 aspirate, plasmacytoma in 1 aspirate and schwannoma in 1 aspirate (Table IV).

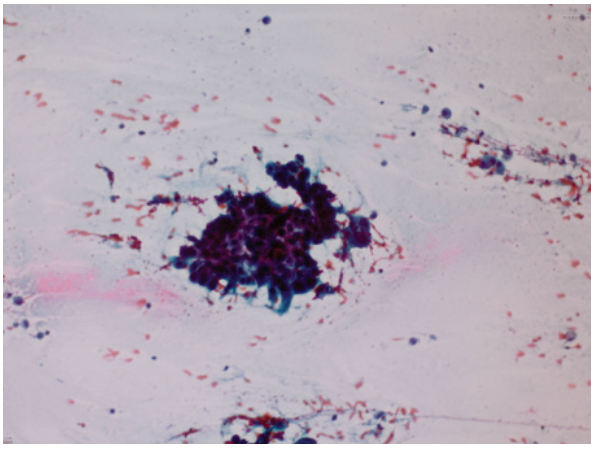

Figure I. Epidermoid carcinoma, neoplastic cells with keratinized cytoplasm MGGx100

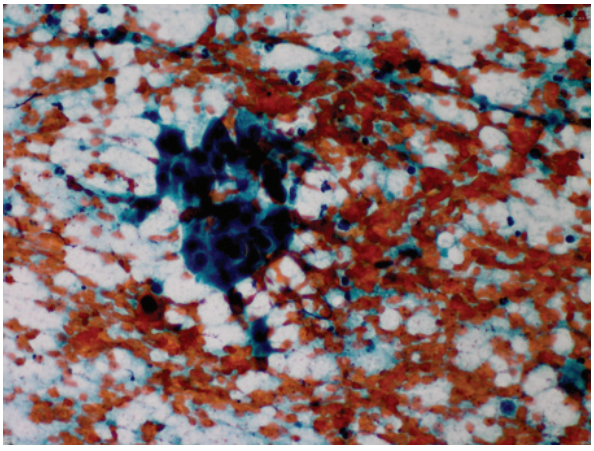

Figure II. A cluster of neoplastic squamous cells with pleomorphic cells having eosinophilic cytoplasm Papx200

Table IV-Types of neoplasia diagnosed in TTFNAB

Type of neoplasia	Number of cases(n:163)	Percentage(%)
Epidermoid carcinoma	58	35.58
NSCC/non-spesific	54	33.13
Adenocarcinoma	23	14.12
Small cell carcinoma	14	8.59
Malign tumor	8	4.90
Neuroendocrin tumor	3	1.85
Tymoma	1	0.61
Plasmacytoma	1	0.61
Schwannoma	1	0.61

NSCC:non small cell carcinoma
TTFNAB:transthorasic fine needle aspiration biopsy

Tissue diagnoses of 71 aspirates were available histopathologically. The tissue diagnoses of 10 inadequate aspirates were: inflammatory events in 4 of them, hydatid cysts in 2 of them and the rest were thymic cysts, Hodgkin's lymphoma, EC, nonspesific bronchial biopsy. Tissue diagnoses of 20 suspicious aspirates were available; inflammatory event in 3 of them, EC in 6 of them, SCC in 2 of them, malignant tumor in 1 of them, atypical carcinoid in 1 of them, PNET in 1 of them, malignant mesothelioma in 2 of them, inflammatory event in 1 of them. Tissue diagnoses of 30 neoplastic aspirates were available; tumor was not observed in 6 of them, 4, 1 and 1 of them were reported to be inflammatory event, tumor necrosis and moderate dysplasia, respectively and they were reflecting the neighboring area of the tumor.

DISCUSSION

It has been reported since 1883 in publications on this subject in the literature that TTFNAB was a safe method with high diagnostic value in benign and malignant masses and infectious diseases of lung.[15-20]

Different imaging modalities such as ultrasonography, fluoroscopy, and computed tomography have been used by various authors.. Computed tomography is widely used.[7,12,15,24] In our study we have used CT which is the most commonly used method today.

The most important indications are lung nodules and masses, mediastinal and hilar lesions, metastatic lesions, chest wall invasion and consolidation and infiltration which may also be caused by infection. The diagnosis rates has been reported above 80% for malignant lesions in various studies.[3] This rate decreases to 12-68% in benign lesions.[4,5] The main cause of becoming a common diagnostic procedure of this method is that the lesion can be reached easily with especially with accompaniment of CT, which was began to be widely used especially in recent years, and the material is taken directly from the lesion with minimum artefacts and more cellular and adequate sample is provided, unlike other pathological and cytological methods.[7]

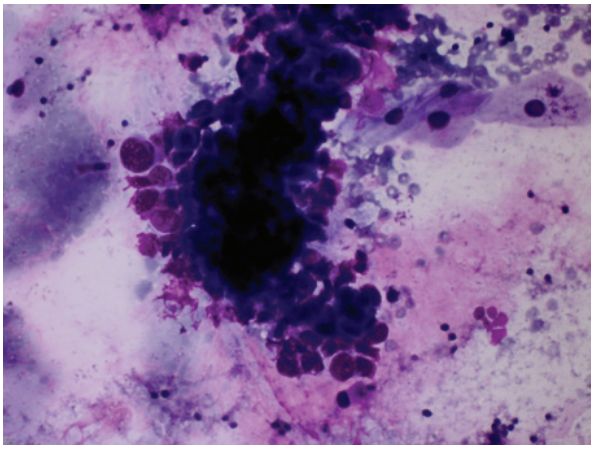

Figure III. Adenocarcinoma, neoplastic cells with delicate cytoplasm and pleomorphic nucleus-prominent nucleoli MGGX400

Inadequate sampling rate in TTFNAB in the literature varies between 8.8% and 25.4% [23,25]. In our study this rate was 12.99% (46/354), this shows that entering into the mass accurately accompanied by CT is important which is compatible with the literature. It draws attention that this rate in our study decreased to 7.88% (11/354) with the cell block application and had higher adequacy.

CB method increased high cellularity (Figure 4), more obvious morphological features, accuracy of cytological diagnosis in the identification of malignant cells and the rate of early diagnosis.[26] In 1928, Zemansky concluded that the CB method was superior to the CS technique and that examination of materials other than pleural and ascitic fluids was unreliable.[27]

In addition, performing cell block on the materials taken provides significant advantages. Re-evaluation with serial sections when hematoxylin and eosin staining, which is a routine method, was required in patients with tumor suspicion and performing auxiliary pathological additional methods such as histochemical-immunohistochemical

staining when necessary. It is possible to make a diagnosis equivalent to histopathological examination and subtyping of tumors with this method.

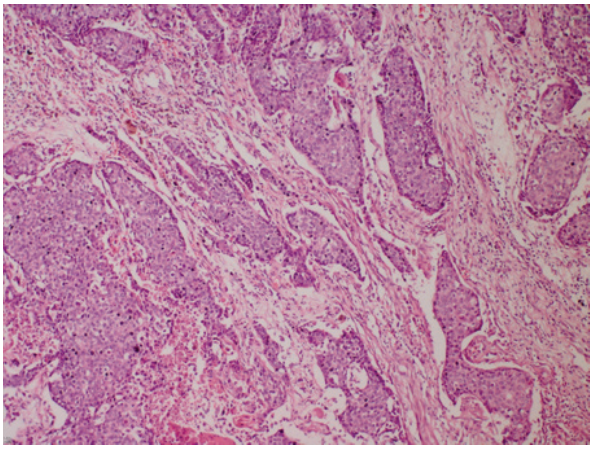

Figure IV. Cell block with high cellularity HEx200

More reliable diagnosis can be made earlier in the sections with less cellularity with cell block method. In this study, 140 CB have been prepared in 354 aspirates for 134 patients. Only 11 of the CBs (7.8%) were non-diagnostic, the failure rate has been reduced even further with the application of CB. The histochemical and / or immunohistochemical staining were applied to 125 CB preparations with adequate cellularity. While cytokeratin7 (CK7), cytokeratin 5-6, p63 were present in our panel for EC (Figure 5,6), one or more antibodies were selected such as TTF1, CK7, mucin, pas-alcian blue histochemistry for AC and CK7, CD45, CD56, synaptophysin and chromogranin for SCC (Figure 7,8). While making the discrimination of SCC and NSCC in lung tumors has importance for surgery, today pathologists should give the specific types of NSCC as much as possible to oncologists and radiation oncologists. Because, spesific subtypes of NSCC display varying responses to different chemotherapeutic agents. Tumor classification and accurate staging to allow treatment planing and the identification of patients who will benefit from potentially curative redical treatment). In our study, 58 (42.96%) and 23 (17.03%) of 135 NSCC were reported to be EC and AC respectively.

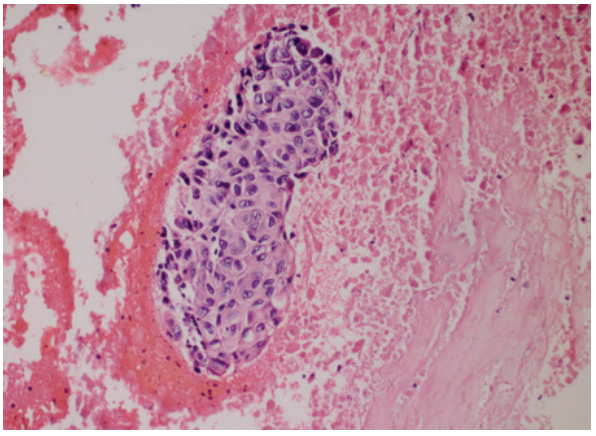

Figure V. Epidermoid carcinoma, cell block HEx200

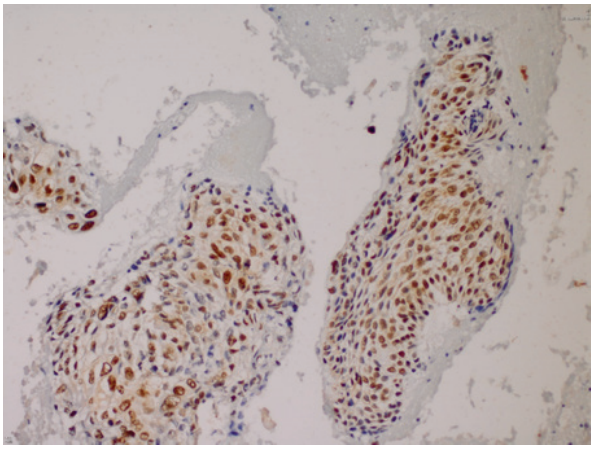

Figure VI. Epidermoid carcinoma, p63 immunohistochemicalstainingx400

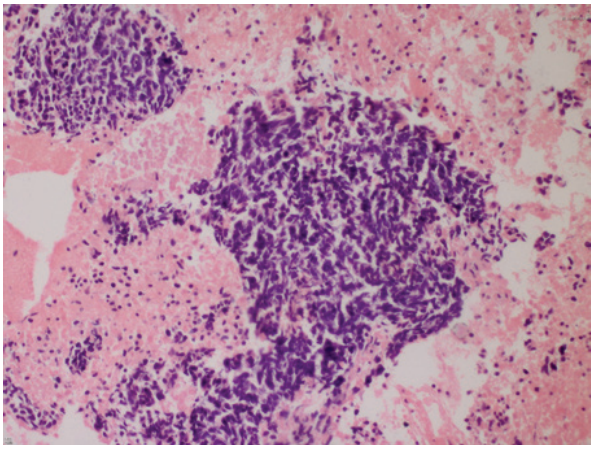

Figure VII. Neoplastic small cell with scant cytoplasm and crush artefact HEx400

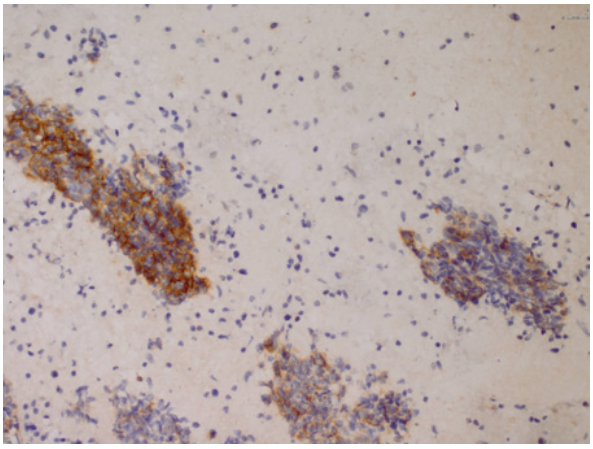

Figure VIII. Small cell carcinoma, CD56 immunohistochemical stainingx200

The sub-typing could not be performed in 54 cases and they were diagnosed as NSCC. Literature reveals that 70% of the primary lung cancers are diagnosed as NSCC, whereas SCC are found to comprise around 20% of all primary lung tumors. In our study, 135 (90.6%) and 14 (9.3%) of 149 primary lung cancer were reported to be NSCC and SCC, respectively. We thought that the difference with the literature was caused due to SCC cases which involve extensive areas of necrosis were considered among the suspicious lesions as necrotic material. The diagnostic value of TTFNAB, wich is quite diagnostic for malignant lesions, is lower for benign lesions. In benign lesions; the diagnosis rates of TTFNAB is reported to be 12-68%.[4] In our study, 6 (50%) of the 12 cases with tissue diagnosis and diagnosed as benign with TTFNAB were diagnosed as benign. The other six cases, although cytology was reported as benign, were diagnosed as malign histopathologically. Two of these cases were malignant lymphoma that lymphoid cells on the ground were interpreted as inflammatory events. One of them was thymoma that lymphoid cells were on the ground and epithelial area. was not found in the smear. The tissue diagnosis of other 4 cases were 2 EC, 1 AC, 1 undifferentiated carcinoma. When the smears and CBs were re-evaluated, it has been seen that the smears reflected the changes of tumor-adjacent areas and the actual mass was not fully entered. The evaluation was difficult for 2 cases due to smear and staining artifacts in the preparatşons caused by technical reasons.

Santambrogio et al.[10] have reported the sensitivity, specificity and efficiency for malignant lesions as 98.5%, 100% and 99.1%, respectively in their series with 110 cases; Moultan et al.[11] have reported as 84%. In our study, the tissue diagnoses in 24(70.58%) of 34 cases with tumor diagnosed in 54 TTFNAB with histopathological tissue diagnosis were also reported as tumor.

It was thought that the cases, which were not diagnosed histopathologically, were caused by the problems of biopsy material such as difficulties in taking of tissue from the correct place and adequately, tissue loss and disorders in laboratory procedures. When it was considered that the biopsy diagnosis were necrotic tissue, dysplastic changes and inflammatory reactions, it was seen that the biopsies were taken from adjacent areas of the tumor. Therefore, if clinico-radiological correlation is present in cases diagnosed positively with TTFNAB-CB, we believe that histopathological diagnosis is not required with repeated biopsy.

The discrimination of SCC and NSCC could be performed in 149 cases of 169 smear diagnosed with tumor, 14 of them were SCC and 135 of them were NSCC. Fifty-eight and 23 of the 81 NSCC cases were EC and AC respectively. TTFNAB-CB in expert hands and experienced pathologists is fairly accurate for typing lesions and can be regarded as an acceptable procedure for diagnostic and medical treatment with chemotherapeutic agents planning purposes in most NSCC cases. In poorly differentiated and suspicious cases, the use of ancillary techniques, such as immunohystochemical or hystochemical stains, may be required to improve the diagnostic yield.

In conclusion; when CT guided TTFNAB is applied in the early diagnosis of intrathorasic mass lesions and CB slights can be prepared and auxiliary techniques can be

used, it is a cheap, effective, reliable method which has a high diagnostic value and a reasonable complication rate. Its popularity is increasing among clinicians, radiologists and pathologists due to it is a simple method and has a complications such as pneumothorax, which can be treated. TTFNAB should be used at an early stage and more often in order to shorten the diagnosis time of lung cancer and to ensure a faster treatment.

References

1. *Ferlay J, Shin HR, Bray F, Forman D,Mathers C, Parkin DM. et al. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer 2010;127:2893-2917.*
2. *Ronald S. Winookur, Bradley B. Pua, MD, Brian W. Sullivan, RA, and David C. Madoff. Percutaneous Lung Biopsy: Tecnique, Efficacy, and Complications. Technique, Efficacy, and Complications. Semin Intervent Radiol 2013;30:121–127*
3. *Miller DA, Carrasco CH, Katz RL, Cramer FM,Wallace S,Charnsangavej C. Fine needle aspiration biopsy:the role of immediate cytologic assesment. AJR 1985;144:793-794.*
4. *Kurıyama T, Masago K, Okada Y, Katakamı N. Computed tomography guided lung biopsy: Association between biopsy needle angle and pneumothorax development. Mol Clin Oncol. 2018; 8(2): 336–341.*
5. *Silverman FJ, Finley JL, O'Brien KF, Dabbs DJ, Park HK, Larkin EW et al. Diagnostic accuracy and role of immadiate interpretation of fine needle aspiration biopsy specimens from various sites. Acta Cytol 1989;33:791-796.*
6. *Conces DJ, Schwenk GR Jr, Doering PR, Giant MD.. Thorasic needle biopsy. Improved results utilizing a team approach. Chest 1987;91:813-816.*
7. *Madan M, Bannur H. Evaluation of fine needle aspiration cytology in the diagnosisof lund lesions. Turk jpath 2010;26:1-6.*
8. *Charing MJ, Stutley JE, Padley SP, Hansell DMl. The value of negative needle biopsy in suspected operable lung cancer. Clin Radiol 1991;44:147-149.*
9. *Mack MJ, Hazelrigg SR, Landrenau RJ, Acuff TE. Thoracoscopy for the diagnosis of the indeterminate solitary pulmonary nodule. Ann Thorac Surg 1993;56:825-832.*
10. *Zarbo RJ, Fenoglio-Preiser CM. Interinstitutional database for comparion of performance in in lung fine-needle aspiration cytology. Arch Pathol Lab Med 1992;116:463-470.*
11. *Trail ZC, Gleason FV. Delayed pneumothorax after CT guided percutaneous fine needle aspiration lung biopsy. Thoax 1997;52:575-576.*
12. *Gangopadhyay M, Chackrabarti I, Ghosh N, Giri A. Computed tomography guided fine needle aspiration cytology of mass lesions of lung: our experience. Indian J Med Paediatric Oncol 2011;32:192-196.*
13. *Boskovic T, Stanic J, Pena-Karan S, Zarıgolidis K. Pneumothorax after transthorasic needle biopsy of lung lesions under CT guidance. J Thorac Dis 2014;6: 99-107.*
14. *Davidson MR, Gazdar AF, Clarke BE. The pivotal role of pathology in management of lung cancer. J Thorac Dis 2013;5: 463-478.*
15. *Kocijancic I, Kocijancic K. CT-guided percutaneous transthorasic needle biopsy of lung lesions-2-year experience at the Institute of Radiology in Ljubljana. Radiol Oncol 2007;41: 99-106.*
16. *Westcott JL. Direct percutaneous needle aspiration of localized lesions results in 422 patients. Radiology1980; 137:31-35.*
17. *Kazerooni EA, Lim FT, Mikhail A, Martinez FJ. Risk of pneumothorax in CT-guided transthorasic aspiration needle biopsy of the lung. Radiology 1996;198:371-375.*
18. *Li H, Boiselle PM, Shepared JO, Trotman-Dickenson B,McLoud TC. Diagnostic accuracy and safety of CT-guided percutaneous needle aspiration biopsy of the lung comprasion of small and large pulmonary nodules. AJR Am J Roentgenol 1996;167:105-109.*
19. *Khouri NF, Stitik FP, Erozan YS. Transthorasic needle aspiration biopsy of benign and malignant lung lesions. AJR Am J Roentgenol 1985;144:281-288.*
20. *Stanley JH, Fish GD, Andriole JG. Lung lesions cytologic diagnosis by-fine needle biopsy. Radiology 1987;162:389-391.*
21. *Klein JS, Zarka MA. Transthorasic needle biopsy:an overwiew. J Thorasic Imaging 1997;12:232-249.*
22. *Gupta S, Krishnamurty S, Bromeling LD, Morello FA Jr, Wallace MJ, Ahrar K et al. Small subpleural pulmonary lesions:short-versus long- needle- path CT-guided Biopsy-- comprasion of diagnostic yields and complications. Radiology 2005;234:631-637.*
23. *Kline TS. Handbook of Fine Needle Aspiration Biopsy Cytology, 2nd ed. New York:Churchill Livingstone. 1988;492pp.*
24. *Kowalewski J, Sir J, Dancewicz M, Swiniarska J, Peplinski J. Fine needle aspiration biopsy under CT-guidance-usefulness in diagnosis of malignant lung tumors. Pneumonol Allergol Pol 2004;72:177-180.*
25. *Wu X, Groves FD, McLaughin CC, J emal A, Martin J, Chen VW. Cancer incidence patterns among adholescents and young adults in the United States. Cancer Causes Control 2005;16:309-320.*
26. *Mathew EP, Nair V. Role of cell block in cytopathologic evaluation of image–guided fine needle aspiration cytology. J Cytol 2017; 34:133-138.*
27. *Zemansky AP Jr. Examination of tumour cells; Analysis of 113 cases checked against subsequent examination of tissue. Am J M. Sc 1928;175:489-504.*