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Source / Izvornik: **Collegium antropologicum, 2014, 38, 611 - 616**

**Journal article, Published version**

**Rad u časopisu, Objavljena verzija rada (izdavačev PDF)**

Permanent link / Trajna poveznica: <https://um.nsk.hr/um:nbn:hr:239:934339>

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Download date / Datum preuzimanja: **2024-12-23**



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# Value of Cytology in Small Cell Lung Carcinoma Diagnostic – Single-Center Study

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## ABSTRACT

*Small cell carcinoma of the lung (SCLC) together with the large cell neuroendocrine carcinoma (LCNEC), typical carcinoid (TC), and atypical carcinoid (AC) make a group of morphologically identifiable neuroendocrine tumors. The differential diagnosis of SCLC includes, first of all, other neuroendocrine tumors, and primary or metastatic non-small cell carcinomas. Although the criteria for the morphologic separation from other tumors of the lung are defined, in everyday practice it can be a problem, both in cytology and with histological samples. Accurate and early differentiation of the SCLC is important because it exhibits aggressive behavior, rapid growth, early spread to distant sites, but also exquisite sensitivity to chemotherapy and radiation. The study included 127 patients who underwent bronchoscopic examination or percutaneous transthoracic fine-needle aspiration (PTTFNA) during the period from early 2003 to 2007 in University Hospital Center Osijek whose cytological diagnosis was SCLC. The value of cytological diagnosis was determined by comparing it with histological findings obtained from a biopsy sample during bronchoscopy or on a resection specimen in 50 patients. In the remaining 77 patients, histological verification of cytological diagnosis was not made and the patients were treated based on cytological diagnosis of small cell carcinoma. In 76% of cases (38/50) cytological diagnosis of small cell lung carcinoma was also confirmed histologically. In 8% of cases (4/50) adenocarcinoma was histologically confirmed, in 10% (5/50) of the cases the squamous carcinoma was confirmed, and there was one case of urothelial carcinoma, one case of sarcoma and one undifferentiated carcinoma. Cytological diagnosis of SCLC was made in all cases in a brush smear while the catheter aspirate was positive in only 32 cases (25.8%). Median survival in the group of patients with histologically confirmed small cell cancer was 238 days, for women 250 days, and for men 237 days. Cumulative survival was 63.2% for 6 months, 26.3% for 12 months, 13.2% for 18 months and 7.9% for two years. In conclusion, cytology is a reliable and relatively non-invasive method for patients. Our results confirm that there is a good correlation between cytology and histology diagnoses, especially when it comes to malignant lesions. In determining the type of tumor cytology must be supported with additional methods, especially in cases when it is not possible to take samples for histological verification.*

**Key words:** lung cancer, fine needle aspiration cytology, histology, cell morphology, small cell lung carcinoma, neuroendocrine carcinoma, non-small cell lung carcinoma, bronchoscopy, Kaplan Meier survival curve, mean survival time

## Introduction

Small cell carcinoma of the lung (SCLC) together with the large cell neuroendocrine carcinoma (LCNEC), typical carcinoid (TC), and atypical carcinoid (AC) make a group of morphologically identifiable neuroendocrine tumors. Neuroendocrine tumors of the lung are a distinct subset of tumors, which share morphologic, ultrastructural, immunohistochemical and molecular characteris-

tics, however these tumors are classified into different morphologic categories within the WHO classification<sup>1</sup>.

The incidence of small cell lung cancer (SCLC) has declined over the last few years. SCLC once accounted for 20–25% of all newly diagnosed lung cancers; it now comprises only about 15% of all lung cancers. Separate worldwide data for small cell carcinoma are not available<sup>2</sup>.

Small cell lung cancer (SCLC) is usually centrally located and can be approached easily with a bronchoscope. The advantage of endoscopy is direct visualization of the tumor, allowing direct biopsy as well as cytological examination of bronchial washings and brushings. For tumors that cannot be diagnosed with transbronchial cytology or biopsy, a transthoracic percutaneous fine-needle aspiration (PTTFNA) carried out under computed tomography (CT) scan guidance is a reasonable alternative.

Sputum cytology is a non-invasive test and, if positive, can provide an accurate diagnosis of central lung cancers. Although small cell lung cancer (SCLC) usually presents as a large, central tumor, tumor cells frequently involve the submucosal layer of the bronchus with little or no exophytic endobronchial extension. Therefore, sputum cytology is not as useful for diagnosing SCLC as it is for the diagnosis of squamous cell carcinoma<sup>1,3</sup>.

The differential diagnosis of SCLC includes, first of all, other neuroendocrine tumors, and primary or metastatic non-small cell carcinomas. Although the criteria for the morphologic separation from other tumors of the lung are defined, in everyday practice it can be a problem, both in cytology and with histological samples.

LCNEC are separated from SCLC using a constellation of criteria, which include larger cell size, abundant cytoplasm, prominent nucleoli, vesicular or coarse chromatin and less prominent nuclear molding<sup>1,4</sup>.

Morphologic separation of SCLC from NSCLC (non small cell lung carcinoma) can be difficult. The distinction does not rest on a single feature but incorporates cell size, nuclear: cytoplasmic ratio, nuclear chromatin, nucleoli, and nuclear molding<sup>5,6</sup>. Cytological specimens may show much better-preserved tumor cell morphology than the pathological ones.

Almost 50% of lung carcinomas exhibit more than one major histological type<sup>1</sup>. This fact has important implications on lung tumor classification and must be kept in mind, especially when interpreting small biopsies or cytological samples.

The staging system most commonly used for SCLC is the Veterans Administration Lung Group (VALSG), a 2-stage system, which defines a limited-stage and an extensive-stage disease<sup>7</sup>. Patients with disease confined to one hemithorax, with or without involvement of the mediastinal, contralateral hilar or ipsilateral supraclavicular, or scalene lymph nodes are considered to have limited-stage disease, whereas those with a disease involvement at any other location are considered to have extensive-stage disease<sup>7</sup>.

Accurate and early differentiation of the SCLC is important because it exhibits aggressive behavior, rapid growth, early spread to distant sites, but also exquisite sensitivity to chemotherapy and radiation, so the key factor in defining a correct diagnosis especially in a limited-stage disease is the ability to encompass the disease within one tolerably safe radiation therapy port.

Approximately 60–70% of patients with small cell lung cancer (SCLC) have clinically disseminated or ex-

tensive disease at presentation. Extensive-stage SCLC is incurable. When given combination chemotherapy, patients with extensive-stage disease have a median survival longer than 7 months; however, only 2% stay alive over a 5 year period<sup>8</sup>. For individuals with limited-stage disease, which is treated with combination chemotherapy plus chest radiation, survival of 17 months has been reported; 12–15% of patients stay alive over a 5 year period<sup>9</sup>.

Indicators of poor prognosis include relapsed disease, weight loss of more than 10% of baseline body weight, and poor performance status.

## Materials and Methods

The study included all of 127 patients in which during the period from early 2003 to 2007 in University Department of Clinical Cytology, University Hospital Center Osijek small cell lung carcinoma was diagnosed (between 22 and 28 cases per year).

Women make up 20.5% (26/127) of patients, and men 79.5% (101/127). The average age at diagnosis was 62, and according to sex, the average age of diagnosed women was 61 and men 62.

Samples for cytological examination were obtained by bronchoscopy or percutaneous transthoracic fine-needle aspiration (PTTFNA). Total of 124 catheter aspirates and 124 brush smears, 8 imprints of excised mucosa and 10 PTTFNA were done. Smears were stained with May-Grunwald-Giemsa.

The value of cytological diagnosis was determined by comparing it with histological findings obtained from a biopsy sample during bronchoscopy or on a resection specimen in 50 patients. In the remaining 77 patients, histological verification of cytological diagnosis was not made and the patients were treated based on cytological diagnosis of small cell carcinoma. The minimum follow-up period was 60 months. We determined the median and cumulative survival in the group of patients with histologically confirmed small cell.

The time-to-event data were summarized using Kaplan-Meier curves, and statistically compared using the log-rank test. Two-tailed p values of <0.05 were considered significant. All tests were performed using a 2007 NCSS software (v07.1.14, LLC, Kaysville, Utah, USA).

## Results

In 76% of cases (38/50) cytological diagnosis of small cell lung carcinoma was also confirmed histologically. In 8% of cases (4/50) adenocarcinoma was histologically confirmed, in 10% (5/50) of the cases the squamous carcinoma was confirmed, and there was one case of urothelial carcinoma, one case of sarcoma and one undifferentiated carcinoma. Histological diagnosis in 50 patients in whom the cytological diagnosis was small cell carcinoma is shown in Table 1.

**TABLE 1**  
HISTOLOGICAL DIAGNOSIS IN 50 PATIENTS IN WHOM THE CYTOLOGICAL DIAGNOSIS WAS SMALL CELL CARCINOMA

Cytology	Histological diagnosis					
	Small cell carcinoma	Squamous cell carcinoma	Adeno-carcinoma	Papillary urothelial carcinoma	Undifferentiated carcinoma	Sarcoma
Small cell carcinoma	38 (76%)	5 (10%)	4 (8%)	1 (2%)	1 (2%)	1 (2%)

Review of 11 misdiagnosed SCLC was made by two cytologists (in one case of histologically confirmed adenocarcinoma the slides were not available). In just 2 from 11 cases both cytologist who made audit did confirmed initial cytologic diagnosis of SCLC. (Table 2, Figures 3–8).

Bronchoscopic samples consisted of catheter aspirates (124), brush smears (124) and in some cases of imprint of excised mucosa (8). Cytological diagnosis of SCLC was made in all cases in a brush smear while the catheter aspirate was positive in only 32 cases (25.8%). Eight imprints and 10 PTP were performed, which all met the criteria for the cytological diagnosis of SCLC.

Median survival in the group of patients with histologically confirmed small cell cancer was 238 days, for women 250 days, and for men 237 days. Cumulative survival

was 63.2% for 6 months, 26.3% for 12 months, 13.2% for 18 months and 7.9% for two years (Figures 1 and 2).

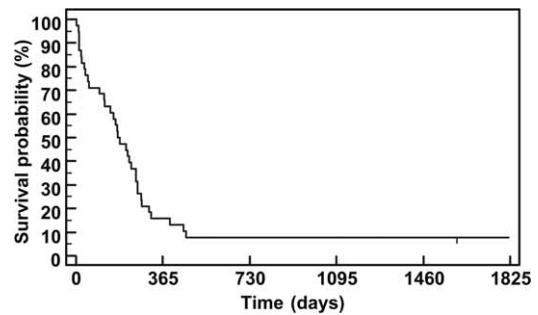


Fig. 1. Survival probability for patients with histologically confirmed small cell carcinoma.

**TABLE 2**  
AUDIT RESULTS OF HISTOLOGICALLY UNCONFIRMED SMALL CELL CARCINOMA

Original cytological diagnosis	Histological findings	Audit 1	Audit 2
Small cell carcinoma	Squamous cell carcinoma	Non small cell carcinoma or lymphoma	Non small cell carcinoma or lymphoma
Small cell carcinoma	Squamous cell carcinoma	Squamous cell carcinoma	Non small cell carcinoma
Small cell carcinoma	Squamous cell carcinoma	Squamous cell carcinoma	Non small cell carcinoma
Small cell carcinoma	Squamous cell carcinoma	Non small cell carcinoma (ddx. Squamous cell carcinoma)	Squamous cell carcinoma
Small cell carcinoma	Squamous cell carcinoma	Non small cell carcinoma (ddx. Squamous cell carcinoma)	Non small cell carcinoma
Small cell carcinoma	Adenocarcinoma	Non small cell carcinoma (Adenocarcinoma)	Non small cell carcinoma
Small cell carcinoma	Adenocarcinoma	Small cell carcinoma	Small cell carcinoma
Small cell carcinoma	Adenocarcinoma	Non small cell carcinoma	Small cell carcinoma
Small cell carcinoma	Adenocarcinoma	No slide	No slide
Small cell carcinoma	Undifferentiated cancer	Non small cell carcinoma	Non small cell carcinoma (ddx. Squamous cell carcinoma)
Small cell carcinoma	Sarcoma	Small cell carcinoma	Small cell carcinoma
Small cell carcinoma	Metastasis of urothelial cancer	Small cell carcinoma	Suspicious bare nuclei with molding

ddx. – differential diagnosis



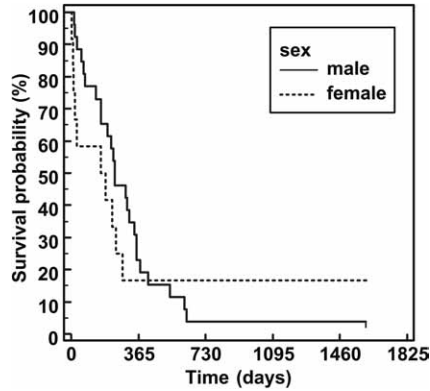


Fig. 2. Survival plots for patients with histologically confirmed small cell carcinoma according to sex.

## Discussion

In lung carcinoma, cell typing of tumor is important in determining prognosis and often in influencing the therapy. Therefore, it is highly desirable to obtain a correct morphological diagnosis. Clinicians are sometimes reluctant to rely on cytological procedures. But many times it is difficult to obtain adequate biopsy material for examination by the pathologist. That could be due to a tumor location (peripheral location) or practical difficulties with bronchoscopy procedure (patient's dyspnoe), and in these cases specimens for histology examination may not be representative or they can even be false negative. Cytological examinations in such cases are very helpful. Thus, when the tumor is not clearly visible by bronchoscopy, the samples may be taken percutaneously

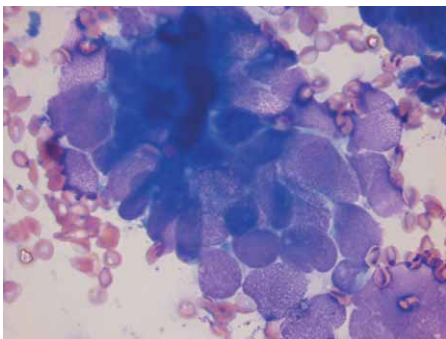


Fig. 3. Metastasis of urothelial carcinoma in lung. May-Grünwald-Giemsa, x1000.

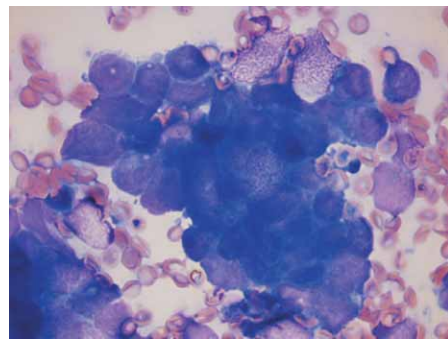


Fig. 4. Metastasis of urothelial carcinoma in lung. May-Grünwald-Giemsa, x1000.

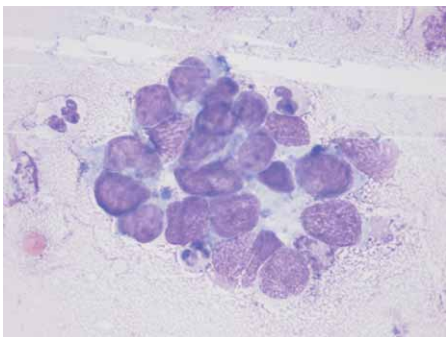


Fig. 5. Undifferentiated carcinoma of lung. May-Grünwald-Giemsa, x1000.

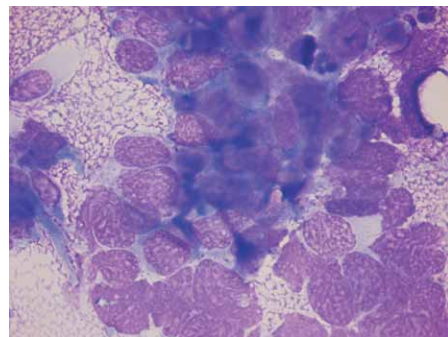


Fig. 6. Undifferentiated carcinoma of lung. May-Grünwald-Giemsa, x1000.

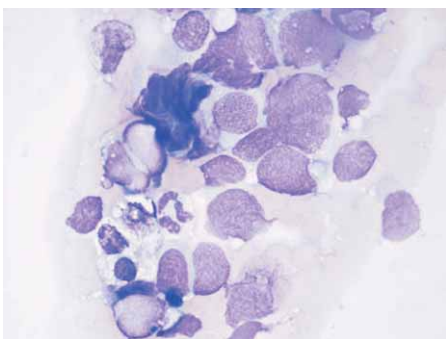


Fig. 7. Sarcoma. May-Grünwald-Giemsa, x1000.

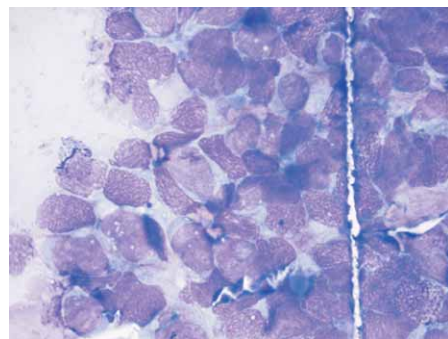


Fig. 8. Sarcoma. May-Grünwald-Giemsa, x1000.

by FNA, guided by CT or ultrasound. Ultrasound guided PTTFNA makes accurate access to lesions in pleura, peripheral lung, anterior mediastinum, bone and soft tissue and thus unnecessary open biopsy (thoracotomy) can be avoided<sup>10,11</sup>. Employing this procedure a false-positive diagnosis of pulmonary malignancy is exceedingly rare, and is estimated to around 1.5%<sup>12,13</sup>.

In our study, from total number of patients with cytological diagnosis of SCLC more than 60% of them (77/127) did not have histological diagnosis and were treated based on clinical and cytological findings.

Total of 124 bronchoscopy was done and malignant tumor was diagnosed in 121 cases. In three cases, bronchoscopy was to be repeated or PTTFNA was performed to obtain correct diagnose. That confirms the value of cytology in diagnostic field of lung cancer, which corresponds to the findings in literature where the sensitivity of cytology for lung cancer rises over 90%<sup>14,15</sup>.

From total number of histologically confirmed diagnoses in 24% of cases cytology recognized existing of malignant tumor but did not recognize the type of tumor. Inadequate evaluation of 4 adenocarcinoma and 5 squamous carcinoma, which together make 18% of the samples, constitutes a problem in further approach to a patient, as the treatment of SCLC and non SCLC is different. From the fact that most of the patients with SCLC get treatment only on the basis of cytological findings, it is necessary to do an ancillary methods, above all immunocytochemistry for making a correct diagnose because our data shows that one quarter of patients did not get correct differential diagnose with only cytomorphological evaluation.

Review of 11 misdiagnosed SCLC showed that the main problems in the diagnosis were degenerative changes on cells, only few preserved cells and lots of bare nuclei. By audit we find out that cells in squamous cell carcinoma were more often present in clusters than expected for SCLC. The nuclei had coarser structure of chromatin and the size of nucleus were 5–6 time of a size an erythrocyte. Metastasis of sarcoma gave totally unexpected morphological picture with some loose groups of cells showing nuclear molding with scant or absent cytoplasm (Figures 7 and 8).

The value of bronchoscopy specimens obtained by brushing and imprinting of excised mucosa is higher than the catheter aspirate. Catheter aspirate was positive in 25.8% of cases, therefore it should not be used as the only material on bronchoscopy, but a suspect place should be located for the brushing or biopsy. SCLC diagnosis made on the imprint was in all cases histologically

confirmed as small cell carcinoma. Tötsch M.<sup>16</sup> cites a much higher accuracy of cytology for the detection of lung cancer: Bronchial Washing: 61 to 76%, while for the Bronchial Brushing: 70 to 77% and FNA: 89%.

Median survival of patients after the cytological diagnosis, was 238 days (7.9 months), which is slightly lower than in the literature: Satoshi Igawa and colleagues<sup>17</sup> report an average survival of patients with SCLC 12.3 months, and Demedts<sup>18</sup> 9–10 months, Haque et al.<sup>19</sup> 12 months, Simon and Turris<sup>20</sup> 10 months. Over a period of one year, 26.3% patients survived in our study compared to 48% of patients in the study of Satoshi Igawa et al.<sup>17</sup>. Two years after the diagnosis only 7,9% of patients survived, compared to 12% two-year survival quoted by Merrill et al.<sup>21</sup>. Travis et al.<sup>22</sup> report 9% and 5% five-year and ten-year survival rate of U.S. patients. We noticed significantly shorter survival of patients in our study in compare to the data of other authors. This can probably be explained by a late diagnosis, thus a wider spread of the disease in our patients. Therefore, our efforts in the treatment of patients with SCLC should be focused on improvements in this direction.

In our study the difference in median survival between female and male patients was not statistically significant (8.3 months vs. 7.9 months), in contrast to research of Wolf and colleagues<sup>23</sup> who followed 766 patients in three German multicentre trials. In their study median survival rate for women was 12.1 months, and 9.8 months for men. A 2-year survival rate was significantly higher than in our sample, 19% for women and 8% for men. Wolf and al. conclude that sex constitutes a major prognostic factor in SCLC and is especially useful as a predictor for long-term survival.

Although, patients with histologically confirmed SCLC had a smaller median survival than patients with other types of lung carcinoma, it was not possible to compare median survival statistically between all groups because the number of patients in those specific groups (adenocarcinoma, squamous carcinoma, urothelial carcinoma, sarcoma and undifferentiated carcinoma) was too small.

## Conclusion

In conclusion, cytology is a reliable and relatively non-invasive method for patients. Also, our results confirm that there is a good correlation between cytology and histology diagnoses, especially when it comes to malignant lesions. In determining the type of tumor cytology should be supported with additional methods, especially in cases where it is not possible to take samples for histological verification.

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## CITODIJAGNOSTIKA KARCINOMA MALIH STANICA PLUĆA

### SAŽETAK

Karcinom malih stanica pluća (SCLC) klasificira se u skupinu neuroendokrinih tumora pluća. Iako su kriteriji za njegovo morfološko odjeljivanje od ostalih tumora pluća dobro definirani, u praksi to može predstavljati problem, kako u citologiji tako i na histološkim uzorcima. Važnost točnog i ranog diferenciranja karcinoma malih stanica leži u njegovom agresivnom ponašanju, rapidnom rastu i stvaranju udaljenih metastaza, ali jednako tako i njegovoj osjetljivosti na kemoterapiju i zračenje. U studiju je uključeno 127 pacijenata kojima je u razdoblju od početka 2003. do kraja 2007. godine učinjen bronhoskopski pregled ili transtorakalna punkcija te je citološka dijagnoza bila karcinom malih stanica. Vrijednost citološke dijagnoze karcinoma malih stanica utvrđena je usporedbom s histološkom dijagnozom donesenom na biopsijskom ili resekcijskom materijalu. U 76% slučajeva (38/50) citološka dijagnoza SCLC je potvrđena i histološki. U 8% slučajeva (4/50) histološki je potvrđen adenokarcinom, 10% slučajeva (5/50) pločasti karcinom, a u po jednom slučaju metastaza karcinoma prijelaznih stanica i sarkoma te nediferencirani karcinom. U ostalih 77 pacijenata (77/127; 60,6%) nije učinjena patohistološka verifikacija te su oni liječeni u skladu s kliničkom prosudbom i citološkom dijagnozom SCLC. Citološka dijagnoza SCLC postavljena je kod svih slučajeva na brisu četkicom dok je kateter aspirat bio pozitivan samo u 32 slučaja (26%). Prosječno preživljenje pacijenata od prve citološke dijagnoze u grupi pacijenata s histološki potvrđenim SCLC iznosilo je 238 dana, za žene prosječno 250 dana, a za muškarce prosječno 237 dana. Zaključno, citologija je jednako vrijedna metoda kao i histologija u određivanju prisustva maligne bolesti, ali u određivanju tipa tumora potrebno ju je nadopuniti dodatnim metodama, prije svega imunocitokemijom, osobito u slučajevima kada nije moguće dobiti histološki uzorak.