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CLINICOPATHOLOGICAL ASSESSMENT OF PLEURAL FLUID CYTOMORPHOLOGY IN A SAMPLE OF IRAQI PATIENTS

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ABSTRACT

Introduction: The International System for Reporting Serous Fluid Cytopathology was applied to assess pleural effusions. Because pleural cavity can be affected by a variety of distinct processes ranging from benign (autoimmune, infectious) to malignant (primary or metastatic neoplasms), effusion Cytopathological diagnosis can be challenging. **The aim of the study:** This study aims to assess pleural fluid cases in a sample of Iraqi patients according to the International System for Reporting Serous Fluid Cytopathology (TIS) in correlation with age, sex, presenting symptoms, type of pleural fluid, radiological findings, laterality and associated diseases. **Subjects and methods:** A retrospective study included 153 randomly chosen pleural fluid samples that were sent to the Teaching Laboratories of Al-Emamain Al-Kadhmain Medical City (AS) between January 2022 and June 2024. **Results:** The mean age of the patients in this study was 56.20 ±19.19 years. 80 patients (53.3%) were male, and 73 patients (47.7%) were female. 141 patients (92.15%) exhibiting shortness of breath. Regarding fluid cytological diagnosis according to the International System For Reporting Serous Fluid Cytopathology, non-diagnostic (ND) was found in 14 patients (9.2%), negative for malignancy (NFM) in 83 patients (54.3%), atypia of undetermined significance (AUS) in 21 patients (13.7%), suspicious for malignancy (SFM) in 25 patients (16.3%) and malignant (MAL) in 10 patients (6.5%). Eighty-six patients (56%) with pleural effusion had exudate fluid, transudate was found in 53 patients (35%). Regarding radiological finding 38 patients (25%) had pleural effusion with finding suggesting benign disease of them (24%) had pulmonary infection including 8 patients (5%) had TB, 22 patients (14%) with finding suggesting malignant disease of them 7 patients (5%) had solitary lung mass suggesting primary origin. Eighty four patients (55%) of these effusions were unilateral, while 69 patients (45% were bilateral. Associated malignant disease was found in 18 patients (12%); GIT origin (3%) and breast (2.6%) were the most encountered cases. Diagnostic categories were found to be significantly correlated to the type of fluid, radiological finding and associated diseases (p value < 0.00001). **Conclusion:** Pleural effusion patients are commonly encountered in clinical practice and in cytopathological laboratories, tend to occur more in middle age men, shortness of breath was the most encountered presenting symptom, the majority of patients were classified as negative for malignancy (NFM) 54%, other categories: suspicious for malignancy (SFM) 16%, atypia of unknown significance (AUS) 14%, nondiagnostic (ND) 9%, and malignant (MAL) 7%. Diagnostic categories were found to be significantly correlated with the presence of associated diseases, type of pleural fluid and radiological findings.

INTRODUCTION

The pleural cavity, which covers both lungs and often contains a tiny amount of fluid, is a small space that lies between the parietal and visceral pleurae. An ongoing pathology is indicated by the buildup of fluid in these cavities, and an examination of the latter might reveal crucial details regarding the etiology and course of the disease. They demonstrate ingenuity in distinguishing the main mesothelium neoplasms from their subsequent involvement.[1]

Numerous conditions, such as infections, autoimmune and metabolic disorders, trauma, and malignancy, are among the many causes of pleural effusions. The diagnosis of a malignant pleural effusion had an average survival of three to nine months and an 80% one-year mortality rate, bears a bad prognosis. Furthermore, effusions could be the first sign of undiagnosed malignancy.[2,3]

Cytology is a crucial technique for the preliminary assessment of effusions; it is an easy, safe, minimally

invasive, and reasonably priced procedure that can assist in determining the stage and prognosis of a cancer as well as its presence and origin. Clinical management greatly benefits from a precise cytological evaluation. The sensitivity and specificity values reported in literature for cytological evaluation of malignant pleural effusion range between 40% to 90% and 90% to 100%, respectively.[4]

Finding out if an effusion contains malignant cells is the most frequent reason to send it to cytopathology. Metastatic disease of the pleura/mediastinum lymph nodes is a common cause of exudative pleural effusions. Pleural effusions are caused in 75% of instances by breast malignancies, and lung tumors. A malignant pleural effusion is seen in 23.1% of lung cancer cases. In serous effusions, accurately identifying cells as reactive or mesothelial, or benign or malignant, is a frequent diagnostic problem.[5]

Effusion cytopathology must be assessed in conjunction with clinical and radiologic data and, if necessary, correlated with ancillary procedures (immunostains, molecular, flow cytometry) in order to provide a sufficient diagnosis.[6]

Mesothelioma makes up the bulk of primary (MAL-P) neoplasms; however primary lymphoma and primary mesenchymal tumors can also arise. Metastatic adenocarcinoma makes up the bulk of secondary (MAL-S) neoplasms; they can also include squamous cell carcinoma, neuroendocrine tumors, melanoma, lymphoma, mesenchymal, and germ cell tumors. Immunostains can help with the differential diagnosis in most situations.[7,8]

The application of the International System for Reporting Serous Fluid Cytopathology serves as a template for improving the communication of cytology reports and decreasing reporting variability. This framework provides a meaningful correlation with follow-up cytology and surgical pathology specimens, thereby enhancing patient management and the quality of clinical care.[1]

There are five categories in the international system for reporting cytopathology diagnosis of serous fluids:[9,10,11]

1. Nondiagnostic (ND): The fluid's cellular components are insufficient to make a definitive diagnosis. Only once an adequate amount of fluid has been processed may the nondiagnostic category be utilized. Risk of malignancy for this category is 17%.
2. Negative for malignancy (NFM); the lack of any indication of mesothelial or non-mesothelial malignancy, only inflammatory and reactive mesothelial cells seen. Risk of malignancy for this category is 21%.
3. Atypia of undetermined significance (AUS): There are atypical cells in the smear. Nevertheless, there is insufficient data, both quantitative and qualitative, to classify these cells as neither malignant nor benign. Risk of malignancy for this category is 66%. In general, the cells mimic reactive, benign cells. The atypical cells could be malignant cells with comparatively bland monomorphic nuclei or mesothelial macrophages.
4. Suspicious for malignancy (SFM): Atypical cells that are highly indicative of malignancy are visible in the smears. Nevertheless, there is little quantitative or qualitative data to draw firm conclusions about malignancy. In such a group, the likely form of cancer, such as carcinoma, lymphoma, or mesothelioma, should be indicated. According to newly documented cases, the probability of malignancy in SFM can reach 82%.
5. Malignant (MAL) primary and secondary: In this category, the diagnosis of malignancy is confirmed by the cytological findings either by themselves or in conjunction with additional ancillary investigations. Risk of malignancy in this category is 99%.

This study aims to assess pleural fluid cases in a sample of Iraqi patients according to the International System for Reporting Serous Fluid Cytopathology (TIS) in correlation with age, sex, presenting symptoms, type of pleural fluid, radiological findings, laterality and associated diseases.

MATERIALS AND METHODS

A retrospective study included 153 randomly selected pleural fluid samples sent to the Teaching Laboratories of Al-Emamain Al-Kadhmain Medical City (AS), received Between January 2022 and June 2024. The International System for Reporting Serous Fluid Cytopathology (TIS) guided the Cytopathological assessment. We gathered the following clinic-cytological information from the cytological reports of the patients:

1. Age
2. Sex
3. Presenting symptoms
4. Chest Radiological findings
5. Associated diseases
6. Laterality
7. Fluid appearance
8. Diagnosis.

Exclusion Criteria

Incomplete radiological or clinical information provided by the referral doctors.

Two cytopathologists revised the diagnosis after all samples had previously been fixed in 95% ethyl alcohol and stained with hematoxyline and eosin.

SPSS version 26 was used for all statistical analysis, which included calculating the mean, standard deviation, frequency, and percentage using Yates Chi square. A p-

value of less than 0.05 was deemed statistically significant.

RESULTS

This study enrolled 153 patients with pleural effusion, of them 80 patients (53.3%) were male and 73 patients (47.7%) were female. Mean age in the study population was 56.20 ±19.19 years. Eighteen patients (12%) were younger than 30 years, 61 patients (40%) belonged to the 30 years to 60 years age group and 74 patients (48%) were older than 60 years. Frequencies of different age groups in the sample are shown in figure 1.

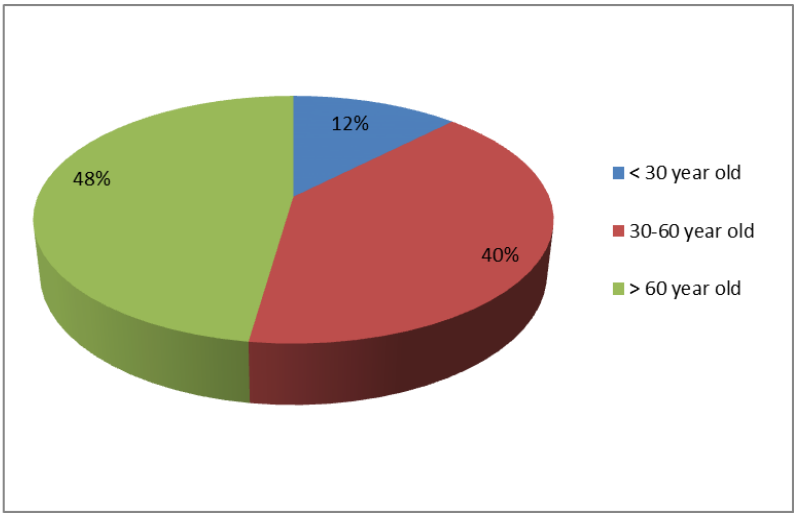

Figure 1: Distribution of age groups in the study sample.

The presenting symptoms of the enrolled patients are listed in table (1). Shortness of breath and cough were recorded in 141 patients (92%) and 13 patients (8.49%) of patients respectively.

Table 1: The presenting symptoms in the study sample.

Symptom	Patients No.	Percentage
Shortness of breath	141	92.15%
Cough	13	8.49%
weight loss	7	4.57%
Fever	7	4.57%
Hemoptysis	3	1.96%
Chest pain	2	1.30%
Orthopnea	2	1.30%
Night sweating	1	0.65%
Dysphagia	1	0.65%
total	153	100%

Regarding the diagnosis according to the International System for Reporting Serous Fluid Cytopathology, non-diagnostic (figure 5) was found in 14 patients (9.2%), negative for malignancy (figure 6) was found in 83 patients (54.3%), atypia of undetermined significance (figure 7) was found in 21 patients (13.7%) , suspicious for malignancy (figure 8) was found in 25 patients (16.3%) and Malignant (figure 9, 10, 11) was found in 10 patients (6.5%) as shown in table (2).

Table 2: Frequency of pleural fluid Diagnostic categories according to the International System for Reporting Serous Fluid Cytopathology in the study sample.

Diagnostic category	Patients No.	Percentage
Non-diagnostic	14	9.2%
Negative for malignancy	83	54.3%
Atypia of undetermined significance	21	13.7%
Suspicious for malignancy	25	16.3%
Malignant	10	6.5%
total	153	100%

Eighty-six patients (56%) with pleural effusion had exudate fluid, transudate was found in 53 patients (35%) and undetermined pleural fluid were recorded in 14 patients (9%) as shown in figure (2).

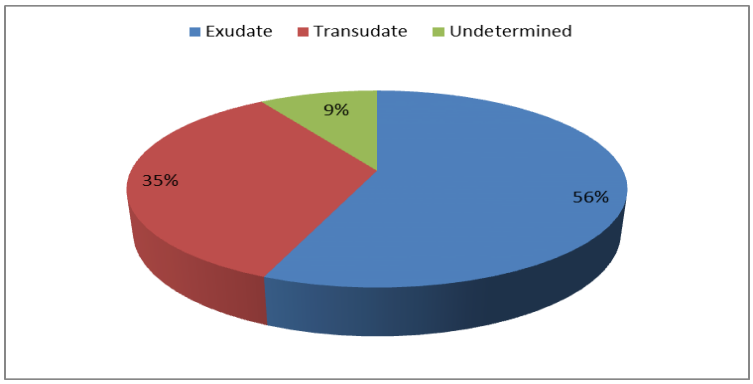

Figure 2: Types of pleural fluid in the study sample.

Ninety-three patients (61%) presented with only pleural effusion on chest imaging while 38 patients (25%) had pleural effusion with radiological findings suggesting benign diseases and 22 patients (14%) had pleural effusion with radiological findings suggesting malignant diseases as illustrated in figure 3.

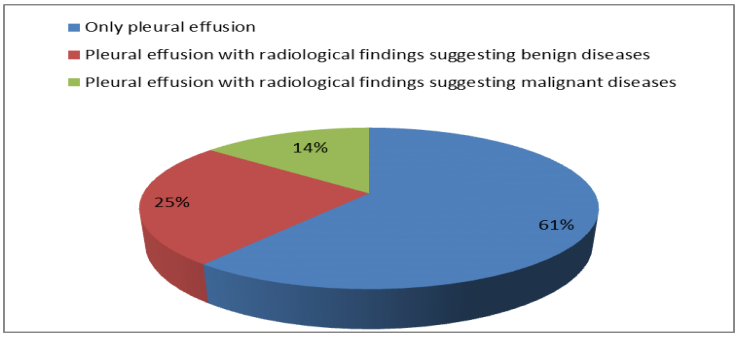

Figure 3: Radiological findings in the study sample.

Findings suggesting benign diseases were found in 38 patients (25%), of them 36 patients (24%) had findings of pulmonary infection (8 patients (5%) were tuberculosis), 1 patient (0.6%) had findings of pleuritis and 1 patient (0.6%) had bronchopleural fistula.

Findings suggest malignant diseases were found in 22 patients (14%), of them 7 patients (5%) had solitary lung mass, 6 patients (4%) had multiple lung nodules, 3 patients (2%) had mediastinal mass, 2 patients (1%) had breast mass and 1 patient (0.6%) had peribranchial mass.

Eighty four patients (55%) were presented with unilateral pleural effusion while 69 patients (45%) were presented with bilateral pleural effusion that was identified in chest radiology **as shown in figure 4.**

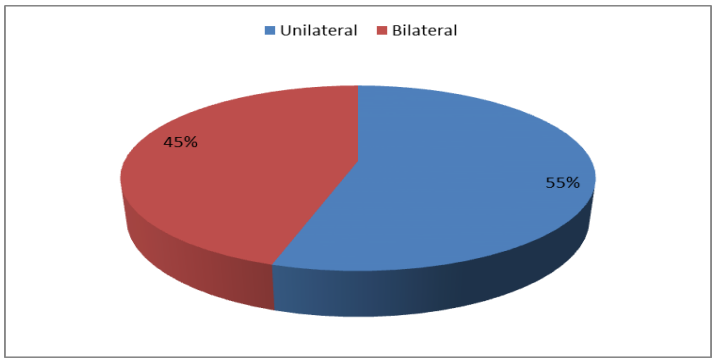

Figure 4: Laterality of pleural effusion in the study sample.

Regarding past medical history 18 patients (12%) with pleural effusion reported associated previous malignant diseases, of them 5 patients (3%) had GIT origin (3 patients colorectal carcinoma and 2 patients with gastric carcinoma), 4 patients (2.6%) had breast cancer, 3 patients (2%) had sarcoma and 1 patient (0.6%) for each; bronchogenic carcinoma, prostatic adenocarcinoma,

ovarian carcinoma, renal cell carcinoma, multiple myeloma and lymphoma.

Diagnostic categories of pleural fluid according to International System of Reporting Serous Fluid Cytopathology were found to be significantly correlated with presence of associated malignant diseases (***P-value*** less than 0.00001), type of the pleural fluid (exudate type exhibited higher frequency of higher diagnostic categories in contrast with transudate fluid) **(*P-value*** less than 0.00001) and radiological findings (findings suggesting benign diseases was found to be higher frequency in lower diagnostic categories while findings suggesting malignancy was found to be higher frequency in higher diagnostic categories) (***P-value*** less than 0.00001)(P-value less than 0.05 was regarded significant.) Other parameters did not show significant correlation with diagnostic categories of the pleural fluid as shown in (table 3).

Table 3: Correlation of diagnostic categories according to the International System for Reporting Serous Fluid Cytopathology with clinicopathological and radiological parameters.

Parameter	R	*P-value*
Age	0.0155	0.85
Sex	-0.0401	0.64
Clinical presentation	-0.1306	0.13
Presence of associated malignant disease	0.5797	< 0.00001
Laterality of pleural effusion	-0.0444	0.64
Radiological findings	0.4781	< 0.00001
Type of pleural fluid	-0.4989	< 0.00001

*R; Correlation Coefficient, *P-value* less than 0.05 was regarded

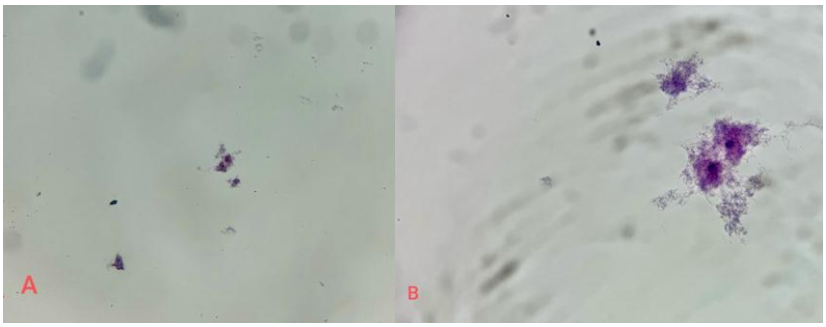

Figure 5: non diagnostic (ND) Smear show no cells for evaluation and contamination by artifact.

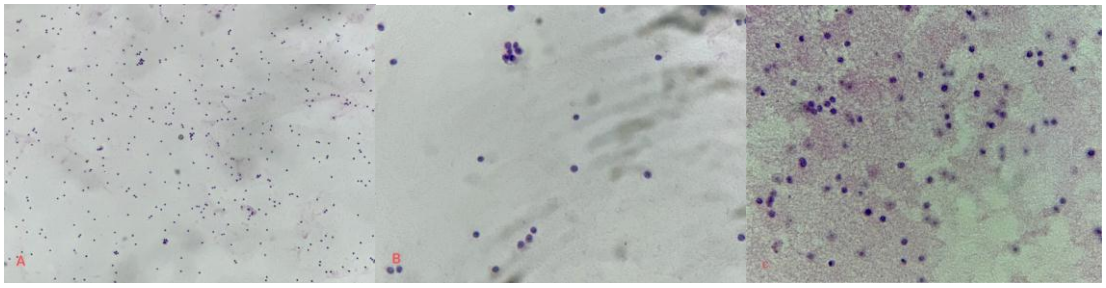

Figure 6: negative for malignancy (NFM) Smear show mostly mesothelial cells as single, small clusters, flat sheets with window, occasional binucleation no multinucleation and no atypia Variable histiocyte and lymphocyte.

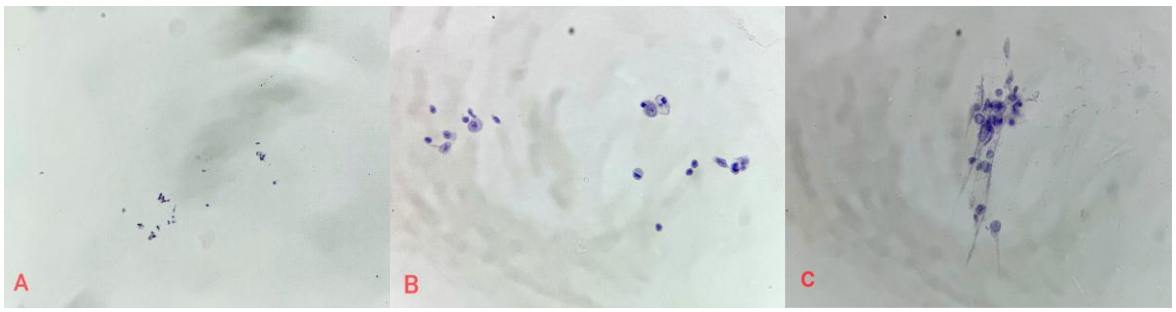

Figure 7: atypia of undetermined significance (AUS) Smear show mild to moderate nuclear enlargement with prominent or variable nucleoli, slight membrane irregularities however scanty in number.

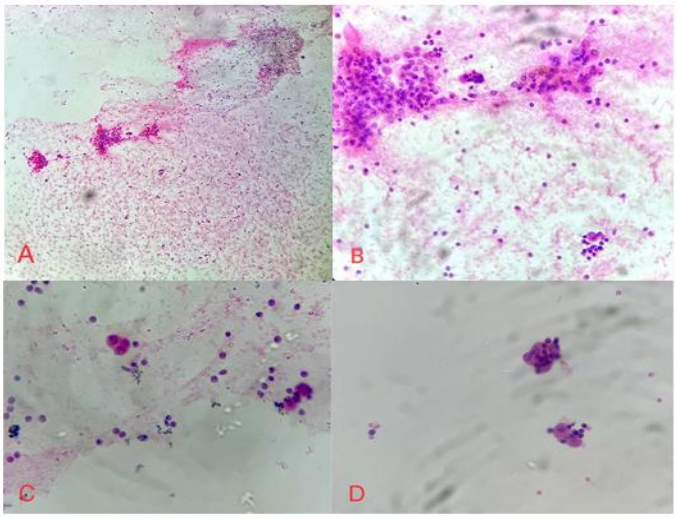

Figure 8: suspicious for malignancy (SFM) Smear show scattered clusters of reactive mesothelial cells with multiple atypical cells, inflammatory cells mainly lymphocyte with hemorgic background need other ancillary tests for conformation. Patient had clinical suspicion of primary lung malignancy.

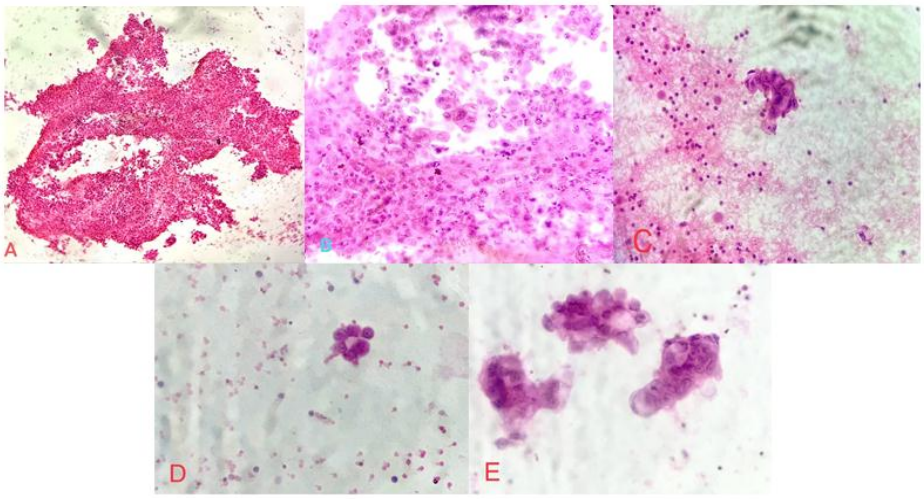

Figure 9: malignant (MAL), patient with history of breast cancer. Smear show forigen population of cells arranged in large 3D clusters, balls with regular outlines, glandular and single cell infiltration; cells are hypercromatic high N/C ratio prominent nucleoli and mitosis.

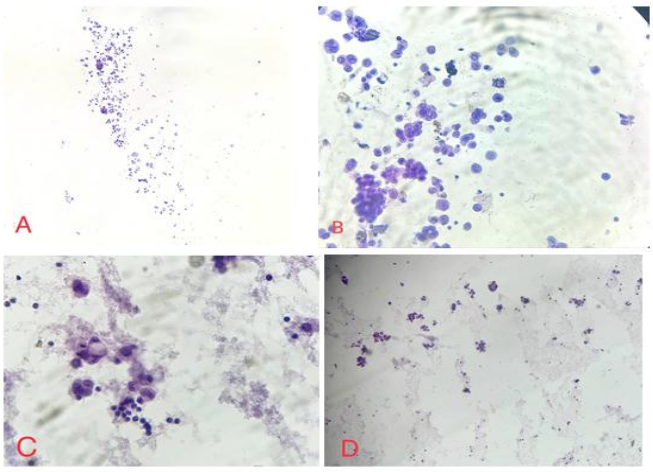

Figure 10: malignant (MAL), patient with history of gastric adenocarcinoma. Smear show numerous forigen glandular and single cell infiltration with intracytoplasmic mucin promoting signet ring appearance. Cells are large, hyperchromatic, high N/C ratio, pleomorphic, irregular nuclear membrane prominent nucleoli.

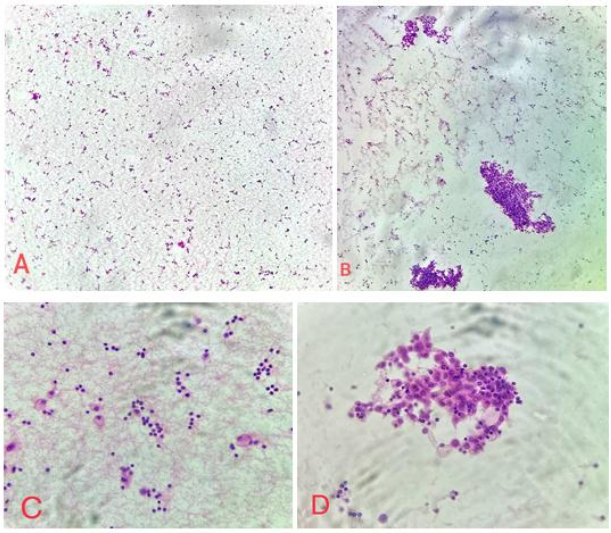

Figure 11: malignant (MAL), patient with history of ovarian cancer. Smear show multiple 3D clusters of forigen malignant cells with single cell and signet ring cell infiltration.

DISCUSSION

In both neoplastic and non-neoplastic diseases, serous effusions can form. Effusion draining is frequently the initial diagnostic procedure when a malignancy is suspected due to its ease of access and low risk of complications.[1] The adoption of the recently suggested International System for Reporting Serous Fluid Cytopathology is assessed in this study.

Different epidemiological patterns are reflected in the mean age of patients receiving pleural fluid cytological assessment, which varies by population and area. The mean age of the patients in this study was 56.20 ±19.19 years. The results are comparable with those of research such as **Biswas et al. (2016, India),** recorded a mean patient age of 51 years[12]; **Loveland et al. (Australia, 2016)**, which found that the mean patient age was 67±16 years[13], and **Kushwaha. et al (2008, India)**, which found that the largest percentage of cases (29.36%) occurred in the sixth decade of life.[14] According to these results, the incidence of pleural pathology appears to peak in older populations, with a high percentage of cases occurring in people over 60.

Regarding sex of patients in this study, male patients were slightly higher in frequency than female patients (53.3% of cases were male), which is in line with multiple prior research. For example, **Loveland et al.'s** Australian study, which found that 59% of cases included were males[13], and **Biswas et al. (2016, India)**, which found that 66.6% of cases were males[12], In contrast to **Kushwaha et al. (2008, India)** who found a female patients were 54.95%[14], These patterns might be the result of environmental or occupational risk factors, such as males' increased exposure to smoking or asbestos, which raises the incidence of pleural diseases in this population.

In 92% of cases in this study, shortness of breath was reported as the most common symptom among patients with pleural effusions. This result is consistent with other research, like **Narayan et al. (2022)**, which found that 93.3% of patients had dyspnea[15], **Gadewad et al. (2017),** which found that 86% of patients presented with shortness of breath[16] and **Biswas et al (2016, India),** which found that this symptom occurred in 95.4% of instances.[12]

The application of the International System for Reporting Serous Fluid Cytopathology serves as a template for enhancing the communication of cytology reports and reducing reporting variability.[1] In this study, the majority of cases (54%) were classified as negative for malignancy (NFM), other categories: suspicious for malignancy (SFM) was 16%, atypia of undetermined significance (AUS) 14%, nondiagnostic (ND) 9% and malignant (MAL) 7%. This distribution underscores the predominance of benign conditions in the present pleural fluid samples. In comparison with other studies, (NFM) category was lower than in other studies; for example, **Pinto et al (2021, Portugal)** found that (NFM) category was reported in 72.29% of cases[17] and **Xuet al (2021, China)** which reported in 68.1% of cases.[18] Similarly, MAL cases constituted 7% in this study, which is lower compared to **Pinto et al (2021, Portugal)** (20.57%) and **Xuet al (2021, China)** (22.4%). Such variations can be the result of regional differences in disease prevalence, sample selection standards, diagnostic techniques and variation in the application of ancillary tests like immunocytochemistry and molecular analysis in addition to pathologist expertise.

ND group made up 9% in this study which was much higher than the rate of **Xuet al (2021, China)** (1.2%) and **Pinto et al (2021, Portugal)** (1.43%). such variation may be due to problems in sample collection and processing (adequate specimen are well preserved, well prepared, well stained and easily visualized).

AUS group made up 14%, which is higher than the percentages given by **Xuet al (2021, China)** (6.2%) and **Pinto et al (2021, Portugal)** (2.00%). Such variation may be due to problems with degeneration or preparation artifacts that can impair morphology (experience and good quality control can help). In addition, nonuse of ancillary tests contributes to the high percentage of the AUS category in this study (ancillary techniques can help decrease the percentage by further classification to SFM or NFM according to immunocytochemistry and molecular analysis results).

Exudative effusions accounted for 56% of the pleural fluid specimens in this study, which is comparable with other research like **Narayan et al**. (83.9%)[15] and **Kushwaha et al.** (82%).[14] This study also found that the frequency of (MAL) and (SFM) diagnostic categories was significantly higher in exudate type effusions than that of transudate effusions (p-value < 0.00001).

The results of **Gadewad et al. (2017),** who found that all malignant pleural effusions were exudative[16], are in line with this study, which has a strong correlation between exudative effusions and higher diagnostic categories. This emphasizes the clinical significance of exudative pleural fluids in pleural cytology examinations; however, rare instances of transudate malignant effusions might arise from concomitant causes, including anemia and hypoproteinemia, even though malignancies are typically linked to exudative effusions.[14,19]

In terms of radiological findings, 55% of pleural effusions cases in this study were presented as unilateral effusions, and 45% of cases were presented as bilateral effusions, This result is comparable to other research such as **Gojiya et al. (2017)**[20] who found that unilateral effusion was found in 88% and bilateral effusion was found in 12%, and **Sandeep et al. (2020)**[21]**,** who found that unilateral effusions accounted for 78.5% and bilateral effusions in 21.5%; the fact that right and left pleural cavities are not connected to each other explains the frequent occurrence of unilateral pleural effusion.[1]

CONCLUSION AND RECOMMENDATIONS

Pleural effusion patients are commonly encountered in clinical practice and in cytopathological laboratories, tend to occur more in middle age men, shortness of breath was the most encountered presenting symptom, the majority of patients were classified as negative for malignancy (NFM) 54%, other categories: suspicious for malignancy (SFM) 16%, atypia of unknown significance (AUS) 14%, nondiagnostic (ND) 9%, and malignant (MAL) 7%. Diagnostic categories were found to be significantly correlated with the presence of associated diseases, type of pleural fluid and radiological findings.

RECOMMENDATIONS

1. Additional research with a larger patient population under long-term follow-up at several centers.
2. Clinician should be encouraged to submit as much of the fluid to the laboratory as reasonable to ensure the greatest possible sensitivity for detection of abnormalities and decreasing ND category.
3. Using ancillary testing to be more certain about cytological diagnosis so increasing the frequency of (NFM), (MAL) and decreasing the use of (AUS) (SFM).
4. Application of quality control management in labs.

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Thank you.

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الخلاصة

المقدمة :يتم الإبلاغ عن الانصباب الجنبي باستخدام النظام الدولي لتقرير علم الخلايا للسوائل المصليّة. نظرًا لأن التجاويف المصليّة يمكن أن تتأثر بمجموعة متنوعة من العمليات المختلفة، منها الحميدة (المناعية الذاتية، الالتهابية) إلى الخبيثة (الأورام الأولية أو النقيلية)، فإن فحص الخلايا الانصبابية قد يكون صعبًا.

هدف الدراسة :تقييم حالات السوائل الجنبية في عينة من المرضى العراقيين وفقًا للنظام الدولي لتقرير علم الخلايا للسوائل المصليّة(TIS) ، بالتوازي مع العمر، الجنس، الأعراض السريرية، النتائج الإشعاعية، الأمراض الحالية أو التاريخ الجراحي، التاريخ الطبي السابق، الجانبية، النتائج التصويرية، مظهر السائل والتشخيص.

المواد والطُرق :دراسة استعادية شملت 153 عينة سوائل جنبية تم اختيارها عشوائيًا، أُرسلت إلى مختبرات التعليم في مدينةالأمامين الكاظمين (ع) الطبية بين يناير 2022 ويونيو 2024.

النتائج :كان متوسط عمر المرضى في دراستنا 56 عامًا، وكان 53.3% منهم من الذكور و 47.7% من الإناث. تم تسجيل ضيق التنفس في 141 مريضًا (92%)، 61% من المرضى كان لديهم فقط انصباب جنبي على التصوير دون وجود نتائج أخرى، 55% من الانصبابات كانت أحادية الجانب و 45% كانت ثنائية الجانب. 56% من الانصبابات كانت إفرازية في الطبيعة. بالنسبة للتشخيص الخلوي للسائل وفقًا للنظام الدولي لتقرير علم الخلايا للسوائل المصليّة، تم العثور على "غير تشخيصي (ND) "في 9.2% من الحالات، و"سلبي للسرطان (NFM) "في 54.3%، و"تغيرات خلوية غير محددة الأهمية (AUS) "في 13.7%، و"مشبوه للسرطان (SFM) "في 16.3%، و"خبيث (MAL) "في 6.5%.

الخاتمة :كان متوسط عمر المرضى في دراستنا 56 عامًا مع وجود تفوق طفيف للذكور على الإناث. تم الإبلاغ عن ضيق التنفس كأكثر الأعراض شيوعًا بين المرضى الذين يعانون من الانصبابات الجنبية. شكّلت الانصبابات الإفرازية 56% من عينات السائل الجنبي، وكانت فئة التشخيصات المشبوهة أو الخبيثة أكثر بكثير في الانصبابات الإفرازية مقارنةً بتلك التي كانت في الانصبابات الانتقالية. كان 55% من الانصبابات الجنبية في دراستنا أحادية الجانب، وكان المرضى في الغالب لديهم انصبابات جنبية فقط دون نتائج إشعاعية أخرى. تم تصنيف أغلب الحالات على أنها "سلبي للسرطان(NFM) "، مع نسب أصغر في الفئات التشخيصية الأخرى.

المجلس العربي للأختصاصات الصحية

المجلس العلمي لعلم الأمراض التشريحي

تقييم سريري ومرضي لخلايا سوائل الانصباب الجنبي لدى عينة من المرضى العراقيين

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