Original Research Article

A Study of Cancer Antigen-125 as a Diagnostic Marker in Tuberculous and Malignant Pleural Effusion

Abstract

Background: CA-125 was first known as a specific tumor marker of the ovary but gradually it was found that inflammation even without polymorphism (the early stage of pregnancy, menstrual cycle, PID, and endometriosis) causes this tumor marker to increase. Tuberculosis in various sites of body cause increase in serum antigen level. The aim of this study was to evaluate the diagnostic value of CA-125 and to determine the cut-off value, distinguishing tuberculous from malignant pleural effusion.

Methods: This is a prospective analytic case control study that was carried out on 90 patients above 18 years old and with a confirmed diagnosis of tuberculous, malignant and parapneumonic pleural effusion. Patients were classified equally into 3 groups: group I "Tuberculous": patients with a confirmed diagnosis of tuberculous pleural effusion, group II "Malignant": patients with a confirmed diagnosis of malignant pleural effusion and group III " Control": patients with a confirmed diagnosis of parapneumonic effusion.

Results: There was positive significant correlation between CA-125 in serum and pleural fluid in tuberculous effusion group (group I) and in malignant effusion group (group II).

Conclusions: Serum and pleural fluid CA-125 may be added to the diagnostic workup of pleural fluid for accurate diagnosis of pleural effusion cause.

Keywords: Cancer antigen-125, Tuberculous, Malignant pleural effusion

Introduction:

Pleural effusion is the accumulation of liquid in the pleural cavity and is considered a result of a systemic or intrathoracic process. This pathological entity is a frequent problem in pulmonology and, though its incidence varies with clinical background, 90% of all pleural effusions are attributed to congestive heart failure, malignant processes, and pneumonia [1].

The main issues regarding pleural effusions are the differentiation of exudates and transudates and the accurate determination of effusion etiology (i.e., whether the pleural effusion is malignant or non-malignant). The differentiation of exudates and transudates requires the evaluation of various biochemical parameters and their comparison in pleural fluid and serum. When differentiating transudates from exudates using classical Light's criteria, it is helpful to recognize the pathogenic mechanism resulting in the pleural effusion. Recognizing the correct pathogenic mechanism is also useful for the purpose of differential diagnosis [2].

CA-125 is a 200 KD glycoprotein, which exists on the surface of ovarian, and some inflammatory and non-inflammatory cells. Proliferation of these cells causes this antigen to be released in serum. CA125 was first known as a specific tumor marker of the ovary but gradually it was found that inflammation even without polymorphism (the early stage of pregnancy, menstrual cycle, PID, and endometriosis) causes this tumor marker to increase. Later it was revealed that tuberculosis in various sites of body cause increase in serum Antigen level [3-5]. Increased serum CA-125 level in both malignancy and tuberculosis, caused some limitations in the use of this tumor marker in a way that the role of CA-125 was decreased as an acute phase reactor and a factor for observing the response to treatments [6, 7].

The aim of this study was to evaluate the diagnostic value of CA-125 and to determine the cut-off value, distinguishing tuberculous from malignant pleural effusion.

Patients and Methods:

This is a prospective analytic case control study that was carried out on 90 patients above 18 years old with a confirmed diagnosis of tuberculous, malignant and parapneumonic pleural effusion in Chest Department, Tanta University Hospitals and Tanta Chest Hospital.

An informed written consent was obtained from all patients. The study was done after approval from the Ethical Committee Tanta University Hospitals.

Exclusion criteria were patients with hepatic cirrhosis, heart failure, or any other non-infectious diseases, female patients with endometriosis and pregnancy and age less than 18 years.

Patients were classified equally into 3 groups: group I "Tuberculous": patients with a confirmed diagnosis of tuberculous pleural effusion, group II "Malignant": patients with a confirmed diagnosis of malignant pleural effusion and group III " Control": patients with a confirmed diagnosis of parapneumonic effusion.

All patients were subjected to cancer antigen-125 "CA-125" measured in both serum and pleural fluid using ELISA kit.

CA 125 ELISA

The CA-125 ELISA-kit is a solid phase enzyme-linked immunosorbent assay (ELISA) based on the sandwich principle. The microtiter wells were coated with a monoclonal [mouse] antibody directed towards a unique antigenic site of the CA-125 molecule. An aliquot of patient sample containing endogenous CA-125 was incubated in the coated well with enzyme conjugate, which is a monoclonal anti CA-125 antibody conjugated with horseradish peroxidase.

Statistical analysis

Statistical analysis was done by SPSS v26 (IBM Inc., Chicago, IL, USA). Quantitative variables were presented as mean and standard deviation (SD) and compared between the two

groups utilizing ANOVA (F) test. Qualitative variables were presented as frequency and percentage (%) and were analysed utilizing the Chi-square test or Fisher's exact test when appropriate. Linear Correlation and ROC-curve (receiver operating characteristic curve analysis) were utilised. A two tailed P value < 0.05 was considered statistically significant

Results:

Age was significantly higher in group II when compared to group III (P3 = 0.009) with no statistically significant difference between groups I and II (P1 = 0.092) or groups I and III (P2 = 0.343). Sex was insignificantly different between the three studied groups. Table 1

Table 1: Age and sex in the three studied groups

		Group I	Group II	Group III	P. value		
Age		54.60 ± 14.79	60.80 ± 15.84	51.13 ± 11.17	0.031*	**P1**	0.092
						P2	0.343
						P3	0.009*
Sex	**Male (%)**	22 (73.3%)	16 (53.3%)	20 (66.7%)	0.257	**P1**	0.107
	Female (%)	8 (26.7%)	14 (46.7%)	10 (33.3%)		**P2**	0.573
						P3	0.292

Data are presented as mean ± SD or frequency (%). P1: Group I & Group II; P2: Group I & Group III; P3: Group II & Group III, *: statistically significant as P value ≤ 0.05

X ray findings were significantly different in the three studied groups (P value= 0.001). TLC and CRP were significantly higher in group III when compared to groups I and II (P2=0.024, P3=0.011, P2 = 0.001, P3 = 0.001 respectively) with no statistically significant difference between groups I and II. ESR was significantly higher in group I when compared to group II and III (P1=0.001) and (P2=0.001) respectively, with no statistically significant difference between groups II and III. Table 2

Table 2: X ray and routine laboratory findings in the three studied groups

		Group I (N=30)	Group II (N=30)	Group III (N=30)	P-value
X ray findings	**Unilateral obliteration of costophrenic angle rising to axilla**	9 (30%)	9 (30%)	19 (63.3%)	0.001*

	Unilateral upper lobe infiltration + unilateral obliteration of costophrenic angle rising to axilla	17 (56.7%)	0 (0%)	0 (0%)	
	Unilateral upper lobe cavitary lesion + unilateral obliteration of costophrenic angle rising to axilla	4 (13.3%)	0 (0)	0 (0%)	
	Unilateral well defined opacity + unilateral obliteration of costophrenic angle rising to axilla	0 (0%)	10 (33.3%)	0 (0%)	
	Bilateral multiple opacities + unilateral obliteration of costophrenic angle rising to axilla	0 (0%)	11 (36.7%)	0 (0%)	
	Unilateral consolidation with air bronchogram + unilateral obliteration of costophrenic angle rising to axilla	0 (0%)	0 (0%)	11 (36.7%)	
Laboratory findings	**TLC (mm^3)**	8386.67 ± 2081.73	8153.33 ± 3656.05	10160.00 ± 3020.57	0.021* P1= 0.763 P2= 0.024* P3= 0.011*
	ESR (mm/h)	108.67 ± 21.29	17.07 ± 18.23	42.33 ± 17.99	0.001* P1= 0.001* P2= 0.001* P3= 0.372*
	CRP (mg/L)	15.33 ± 11.86	11.67 ± 14.35	112.93 ± 70.81	0.001* P1= 0.738 P2= 0.001* P3= 0.001*

Data are presented as mean ± SD or frequency (%). P1: Group I & Group II; P2: Group I & Group III; P3: Group II & Group III, TLC: total leucocytic count, ESR: Erythrocyte Sedimentation Rate, CRP: C-reactive protein, *: statistically significant as P value ≤ 0.05

Positive tuberculin test and turbid yellow appearance were significantly higher in group I compared to group II and group III (P value <0.05). Cloudy appearance, growth of culture and sensitivity of pleural fluid were significantly higher in group III compared to group I and group II. (P value= 0.001). Bloody turbid appearance, serum CA-125 and pleural CA-125 were significantly higher in group II compared to group I and group III (P value <0.05).

Serum CA-125 and pleural CA-125 were significantly higher in group I when compared to group III (P2 <0.05). Table 3

Table 3: Comparison between the three studied groups as regard to Tuberculin test, gross appearance, culture and sensitivity of pleural fluid, serum levels of CA125 and Pleural CA-125 U/ml

		Group I	Group II	Group III	P-value
Tuberculin test	**+ve**	18 (60.0%)	10 (33.3%)	8 (26.7%)	**0.020***
	-ve	12 (40.0%)	20 (66.7%)	22 (73.3%)	
Gross appearance	**Cloudy**	0 (0%)	0 (0%)	6 (20.0%)	**0.001***
	Turbid yellow	22 (73.3%)	8 (26.7%)	18 (60.0%)	
	Bloody turbid	8 (26.7%)	22 (73.3%)	6 (20.0%)	
Culture and sensitivity of pleural fluid	**Growth**	0 (0%)	0 (0%)	26 (86.7%)	**0.001***
	No growth	30 (100%)	30 (100%)	4 (13.3%)	
Serum CA-125 U/ml		120.07 ± 112.77	363.67 ± 221.40	29.40± 19.32	0.001* P1= 0.001* P2= 0.017* P3= 0.001*
Pleural CA-125 U/ml		138.83 ± 149.43	780.87 ± 427.27	44.83 ± 29.02	0.001* P1= 0.001* P2= 0.034* P3= 0.001*

Data are presented as mean ± SD or frequency (%). P1: Group I & Group II; P2: Group I & Group III; P3: Group II & Group III, CA: cancer antigen, *: statistically significant as P value ≤ 0.05

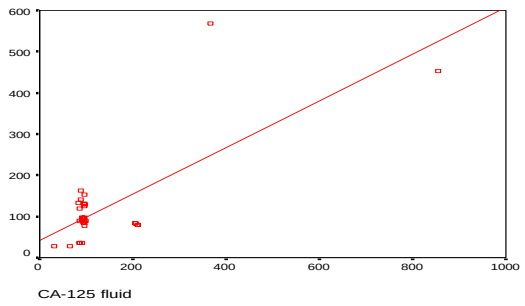
Specific gravity and pleural fluid protein / serum protein ratio were insignificantly different in the three studied groups. PH was significantly lower in group I than group II (P1=0.001), and in group III than groups I and II (P <0.05). Protein and LDH were significantly higher in groups II and III than group I (P <0.05), with no statistically significant difference between groups II and III. Glucose was significantly higher in groups I and II than group III (P <0.05), with no statistically significant difference between groups I and II. Pleural fluid LDH / serum LDH ratio was significantly higher in group II than groups I and III (P1 <0.05), and in group III than group I (P2=0.001). ADA was significantly higher in group I than groups II and III (P <0.05), with no statistically significant difference between group II and III. Table 4

Table 4: Thoracentesis parameters in the three studied groups

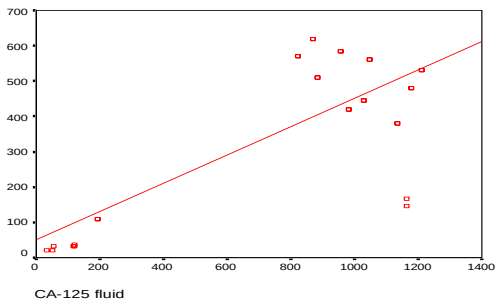
		Mean ± S. D	P. value		
Specific gravity	**Group I**	1026.80 ± 7.68	0.909	P1	0.706
	Group II	1027.53 ± 7.18		P2	0.708
	Group III	1027.53 ± 7.64		P3	0.999
PH	**Group I**	7.17 ± 0.06	0.001*	P1	0.001*
	Group II	7.39 ± 0.06		P2	0.001*
	Group III	7.02 ± 0.14		P3	0.001*
Protein (mg/dl)	**Group I**	4.06 ± 0.63	0.018*	P1	0.022*
	Group II	4.62 ± 1.08		P2	0.009*
	Group III	4.70 ± 1.02		P3	0.753
Glucose (mg/dl)	**Group I**	44.00 ± 10.09	0.001*	P1	0.129
	Group II	40.80 ± 6.83		P2	0.001*
	Group III	27.27 ± 6.92		P3	0.001*
LDH (U/L)	**Group I**	510.07 ± 100.11	0.001*	P1	0.001*
	Group II	1251.13 ± 548.35		P2	0.001*
	Group III	1475.40 ± 639.01		P3	0.080
Pleural Ptn./S. Ptn.	**Group I**	0.71 ± 0.09	0.910	P1	0.776
	Group II	0.70 ± 0.09		P2	0.670
	Group III	0.70 ± 0.09		P3	0.887
Pleural LDH/S. LDH	**Group I**	0.95 ± 0.21	0.001*	P1	0.001*
	Group II	1.91 ± 0.49		P2	0.001*
	Group III	1.60 ± 0.57		P3	0.009*
ADA (U/L)	**Group I**	60.80 ± 9.66	0.001*	P1	0.001*
	Group II	17.37 ± 9.77		P2	0.001*
	Group III	21.57 ± 13.91		P3	0.1543

Data are presented as mean ± SD or frequency (%). P1: Group I & Group II; P2: Group I & Group III; P3: Group II & Group III, LDH: lactate dehydrogenase; Ptn: protein; S: serum, ADA: adenosine deaminase, *: statistically significant as P value ≤ 0.05

There was positive significant correlation between CA-125 in serum and pleural fluid in tuberculous effusion group (group I) and in malignant effusion group (group II). Figure 1



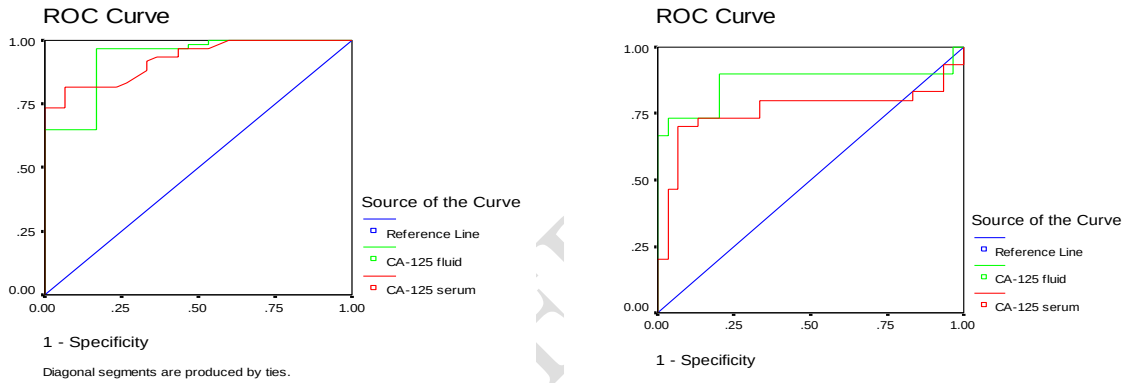
(A)



(B)

Figure 1: Positive significant correlation between CA-125 in serum and pleural fluid in (A) tuberculous effusion group (group I) and (B) malignant effusion group (group II)

Regarding (group I & group II) and group III, CA125 in pleural fluid, at the cut off value of 45 the sensitivity was 93% and the specificity was 83%. While CA125 in serum, at the cut off value of 40 the sensitivity was 82% and the specificity was 77%. Regarding group I & group II, CA125 in pleural fluid, at the cut off value of 110 the sensitivity was 90% and the specificity was 80%. While CA125 in serum, at the cut off value of 100 the sensitivity was 73% and the specificity was 67%. Figure 2



(A) **(B)**

Figure 2: Roc curve show sensitivity and specificity of CA125 in pleural fluid and serum between cases of (A)[Group I & Group II] and Group III and (B) [Group I & Group II]

Discussion

In our work, adenosine deaminase (ADA) (U/L) was significantly higher in Group I when compared to Group II and III (P1 = 0.001) and (P2 = 0.001) respectively, with no statistically significant difference between Group II and III (P3 = 0.1543).

In the same way, in Lee et al. [10], the median values of pleural fluid ADA levels in patients with TPE and PPE were 82 and 50 U/L, respectively, and were significantly different between the two groups (P < 0.001).

In our study, serum levels of CA-125 (U/ml) was significantly higher in Group II when compared to Groups I and III (P1=0.001) and (P3 = 0.001) respectively, and in Group I when compared to Group III (P2=0.017).

In contrast, Zhai et al. [11] enrolled 174 patients, including 67 malignant pleural effusion (MPE) and 107 benign pleural effusion (BPE) found that, the concentration of serum CA-125, in MPE patients was not significantly higher than those in BPE patients.

Similarly, Mohammad et al. [12] enrolled 80 patients were classified into 3 groups: group I: active pulmonary tuberculosis, group II: pneumonia, group III: apparently healthy subjects as control group. They reported that, there was statistically significant difference among the three studied groups according to serum level of CA 125($P < 0.05$). The serum level of CA125 was significantly higher in group I in their work.

In our study regarding CA-125 between cases of (Group I & Group II) and Group III in serum at the cut off value of 40; the sensitivity, specificity, positive and negative predictive values and diagnostic accuracy were 82%, 77%, 88%, 68%, 80%.

While regarding CA-125 between cases of (Group I & Group II) in serum at the cut off value of 100; the sensitivity, specificity, positive and negative predictive values and diagnostic accuracy were 73%, 67%, 69%, 71%, 70% respectively.

According to sensitivity of CA-125, in Mohammad et al. [12], at a CA-125 serum level of 21.05 U/ml as a cut-off value, CA-125 had sensitivity, specificity, positive and negative predictive values and diagnostic accuracy of 82.5%, 72.5%, 77.3%,83.3% and 80.0% respectively.

Our data showed that pleural fluid levels of CA-125 (U/ml) was significantly higher in Group II when compared to Groups I and III (P1=0.001) and (P3 = 0.001) respectively, and in Group I when compared to Group III (P2=0.034).

Also, in our study regarding CA-125 between cases of (Group I & Group II) and Group III in pleural fluid at the cut off value of 45; the sensitivity, specificity, positive and negative predictive values and diagnostic accuracy were 93%, 83%, 92%, 86%, 90%.

While regarding CA125 between cases of (Group I & Group II) in pleural fluid at the cut off value of 110; the sensitivity, specificity, positive and negative predictive values and diagnostic accuracy were 90%, 80%, 82%, 89%, 85% respectively.

Similarly, in Santotoribio et al. [13], of total, 81 patients had benign pleural effusion (BPE) and 52 had malignant pleural effusion (MPE). The pleural fluid concentrations of CA125 were significantly higher in MPE patients than in BPE patients with P-value (0.037).

Our data showed that correlations between CA-125 in serum and pleural fluid levels was highly positive significant P=0.001 in malignant effusion group (group II) and in tuberculous effusion group (group I).

The studies addressing the level of tumor markers in serum and pleural fluid were reviewed, but only a few described P/S ratio. In Gu et al. [14] study, they found that the Pleural fluid/Serum ratio of CA-125 was lower in TPE than in MPE.

Limitations: this study, however, has several limitations. First, we obtained single center participants in our study. In addition, the sensitivity and specificity of CA-125 was not assessed in combination with other biomarkers.

Conclusions:

Serum and pleural fluid CA-125 levels may be used for differentiation between TB and malignancy-induced effusions. Also, CA-125 may be added to the diagnostic workup of pleural fluid for accurate diagnosis of malignant pleural effusion.

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