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Significance of Ascitic Fluid C-Reactive Protein in Differentiating Malignancy Related Ascites from Benign Ascites

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Abstract
Introduction: *Simple test(s) on ascitic fluid or serum which can help differentiate between Malignancy and Non-Malignancy Related ascites is a challenge that is not always met satisfactorily.*
Aim: *The purpose of this study is to assess the usefulness of Ascitic Fluid C-Reactive Protein (CRP) in differential diagnosis of Ascites.*
Methodology: *This prospective study included 80 patients with Ascites, admitted to Medical Gastroenterology Department of Madras Medical College, from January 2018 to January 2019. The patients were divided into two groups – Malignant ascites (n=30) and Non-malignant Ascites (n=50). The modalities for selecting malignant group were either cytology/ peritoneal biopsy positive cases. Complete Blood Count, LFT, Ascitic Fluid Analysis including Total Protein, albumin, cholesterol, CRP, culture & sensitivity, Total and differential counts were done in all patients. USG abdomen, upper GI endoscopy, CECT abdomen, ADA, FNAC of peritoneal nodules and liver biopsy were performed in selected cases. Bacterial peritonitis was excluded in all these patients.Serum and ascitic fluid CRP were analyzed in all cases. Data were entered in Microsoft Excel and analyzed using IBM SPSSS (Ver. 20.0)*
Results: *Mean value of Ascitic Fluid CRP were significantly higher in Malignancy group than Non-malignancy group (16.6±13.4 vs 2.1±3.2 ng/ml) (p value 0.001). Serum CRP was also significantly higher in malignant ascites than benign ascites patients [10.8±6.3 mg/ml vs. 6.2±4.9 mg/ml;p<0.001].*
Conclusion: *Elevated ascitic fluid and serum CRP values can be used to differentiate malignancy related ascites from benign ascites.*
Keywords: *Ascites; Ascitic Fluid; CRP; C-reactive protein; Malignant.*

Introduction

Usually, Malignant ascites is caused by lung, breast, ovarian, endometrial, colorectal, pancreatic, hepatobiliary, and primary peritoneal carcinomas; and it accounts for about 10% of all cases of ascites [6]. For further diagnostic and therapeutic procedures, it is important to differentiate between malignancy-related ascites (MRA) and non-malignant ascites (NMA)[6]. Due to poor sensitivity, Cytology is not a good screening tool for malignant ascites. Furthermore, reactive mesothelial cells in the ascitic fluid are lookalikes of malignant cells. Hence, it is difficult to distinguish between the two, based on morphology alone[7]. So, simple tests on ascitic fluid or serum, which can be used to differentiate between malignancy-related ascites (MRA) and

non-malignant ascites (NMA) will be a blessing in solving this diagnostic predicament.

Identification of the etiology of the ascites depends on ascitic fluid analysis, physical examination, and history taking. Clinically, an alternative predominant beneficial tool is the difference between serum-ascites albumin gradient (SAAG)[1]. However, the use of SAAG measurements or the exudate/transudate separation in etiological diagnosis is a matter of debate[1]. Therefore, we try to find out laboratory markers which could be used as indicators in these situations and in the differential diagnosis of benign and malignant ascites.

CRP stands for c-reactive protein, which is a classical member of pentraxin family[8]. It is acute phase protein that is synthesized by liver & increases in inflammatory process. Because of synthesis in liver, production of CRP in patient with chronic liver disease is expected to be lower than in patients without liver disease. This may result in difference in interpreting CRP levels in patient with portal and non-portal hypertension ascites[9]. In malignant ascites a common cause of non-portal hypertension ascites with intensive inflammation process, values of CRP could be guide in discriminating the underlying the cause of ascites[2]. Therefore, we performed this study to establish the usefulness of serum & ascitic fluid C-reactive protein (CRP) in differential diagnosis of malignant, non-malignant and tubercular ascites.

Aims & Objectives

To study the usefulness of serum & ascitic fluid C-reactive protein (CRP) in differential diagnosis of malignant and non-malignant Ascitis.

Methodology

This prospective study included 80 patients with Ascites admitted to Medical Gastroenterology Department from January 2018 to January 2019. The patients were divided into two groups – Malignant (n=30) and Non-malignant (n=50). The modalities for selecting malignant group were either cytology/ peritoneal biopsy positive cases or cases with liver secondaries. Complete Blood Count, LFT, Ascitic Fluid Analysis including Total Protein, Albumin, culture & sensitivity, Total and differential counts, Cytology, Ascitic Fluid CRP and serum CRP levels were done in all patients. USG abdomen, upper GI endoscopy, CECT abdomen, ADA, FNAC of peritoneal nodules and liver biopsy were performed in selected cases where it was indicated. Bacterial peritonitis was excluded in all these patients.

Statistical Analysis: Data were entered in Microsoft Excel and analyzed using IBM SPSS Software Version 20.0. Percentage Analysis was used for categorical variables (Gender, Etiology). Mean with Standard Deviation or Median with Inter-quartile range (IQR) were used for continuous variables. Comparison of Parametric Data and Non-Parametric data between two groups were done by using Student's t test (Unpaired t test). Comparison of categorical variable between the groups (gender with type of Ascites) was done by using Chi-Square test. Discriminatory performance of variables was determined by area under the receive operating characteristic (ROC) curve, and best cut-off values were calculated based on operating characteristic (ROC) curve, and best cut-off values were calculated based on the High Sensitivity and Specificity. A p value of <0.05 was considered statistical significance with 95% Confidence Interval.

Results

Our study included 80 cases - 50 cases with non-malignant ascites cases and 30 with malignant ascites.

Table 1: Demographic Analysis

Demographic Details	Malignant Ascites		Benign Ascites		t Test	P value
	Mean	SD	Mean	SD		
Age	56.7	10.5	49.2	11.9	t value= 2.877	**0.005***

The mean age of the patients in malignant ascites group was 56.7±10.5 years and in benign ascites group was 49.2±11.9 years.

Table 2: Gender-wise Analysis

Gender	Malignant Ascites N (%)	Benign Ascites N (%)	Total N (%)	Chi-square test (df)	P value
Male	12 (40)	41 (82)	53 (66.3)	14.8 (1)	**<0.001 ***
Female	18 (60)	9 (18)	27 (33.8)		
Total	30 (100)	50 (100)	80 (100)		

Among 80 cases, 53 (66.3%) were males and 27 (33.8%) were females.

Table 3: Distribution of Ascites based on Etiology

Etiology	**Number**	**Percentage**
Cirrhosis	46	57.5
TB Ascites	2	2.5
Cardiac Ascites	1	1.3
Pancreatic Ascites	1	1.3
CA Ovary	12	15
CA Stomach	7	8.8
CA Bladder	2	2.5
CA Gall Bladder	2	2.5
CA Colon	2	2.5
CA Pancreas	1	1.3
CA Prostate	1	1.3
HCC	1	1.3
Cholangio Carcinoma	1	1.3
Pancreatic NET	1	1.3
Total	**80**	**100**

Out of 50 cases in benign group, 46 cases (57.5%) were having cirrhotic ascites, 2 cases (2.5%) with TB ascites and remaining ones with cardiac and pancreatic ascites (1.3%). In the malignancy group, out of 30 cases, 12 cases were carcinoma of ovary.

Table 4: Ascitic fluid Protein Analysis

Variable	Malignant Ascites		Benign Ascites		t Test	P value
	Mean	SD	Mean	SD		
Ascitic fluid Protein (g/dl)	3.73	1.3	1.5	0.8	8.57	**0.001***

For the malignancy group mean value of protein is 3.73±1.3 vs 1.5±0.8 in benign group. Student t test is applied, p value is 0.001.

Table 5: Serum-ascites Albumin Gradient (SAAG) Analysis

Variable	Malignant Ascites		Benign Ascites		t Test	P value
	Mean	SD	Mean	SD		
SAAG (g/dl)	0.7	0.22	1.88	0.52	-14.1	**0.001***

The mean value of SAAG was higher in non-malignant group 1.88+-0.52 vs 0.7+-0.22 in malignant group. Students t test is applied, and p value is 0.001.

Table 6: Ascitic Fluid CRP

Variable	Malignant Ascites		Benign Ascites		t Test	P value
	Mean	SD	Mean	SD		
Ascitic fluid CRP (mg/l)	16.6	13.4	2.1	3.2	5.85	**0.001***

T test= Unpaired t test *- Significant

The mean value of ascitic fluid CRP was significantly higher in malignancy related ascites group.16.6 ± 13.4 vs 2.1 ± 3.2 and p value is 0.001.

Table 7: Ascitic Fluid CRP

Varaiable	**AUC**	**Level ≥5**	
		Sensitivity	**Specificity**
Ascitic Fluid CRP	0.897	76.7%	92%

Ascitic fluid CRP cut-off>-5 mg/l had a sensitivity of 76.7% and specificity of 92%.

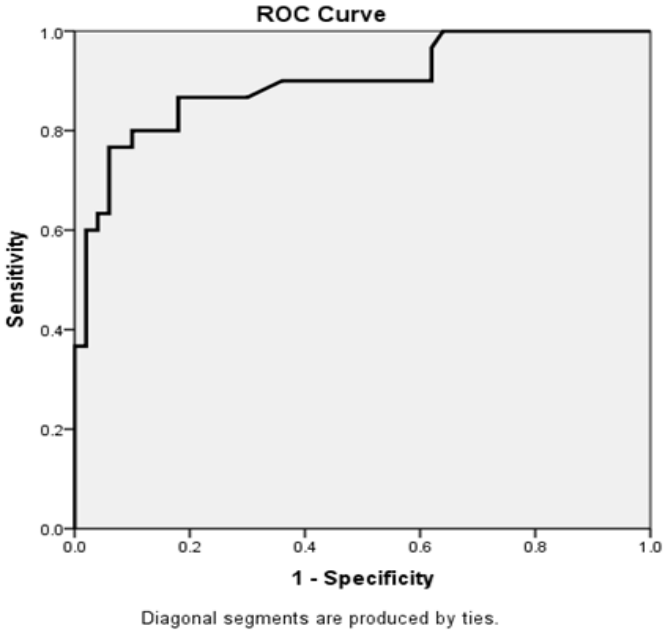

Fig. 1: Receiver operating characteristics (ROC curve) of Ascitic Fluid CRP

Serum CRP was also significantly higher in malignant ascites than benign ascites patients [10.8±6.3 mg/ml vs. 6.2±4.9 mg/ml;p<0.001

Discussion

The search for novel biochemical markers in the serum and/or ascitic fluid for differentiating malignant and non-malignant ascites is still under investigation. To identify malignant effusions, various biochemical markers have been employed in the past, because it is difficult to demonstrate malignant cells in effusions [10]. A simple, quick and reliable test on ascitic fluid is essential, if the diagnosis is not obvious from the clinical presentation. Some ascitic or serum tumour markers such as CA15-3, CA19-9, CA125, α-fetoprotein, soluble aminopeptidase N/CD13, soluble interleukin-2 receptor α, tissue polypeptide-specific antigen, carcinoembryonic antigen (CEA), and insulin-like growth factor-1 (IGF-1) may be used for additional diagnostic purpose [11]. None of them were not specific enough to differentiate the benign ascites from the malignant ones. There is no golden standard method for the diagnosis of malignant and benign ascites; the clinical follow-up could represent a definitive confirmation for this type of diagnosis.

CRP was emitted predominantly by the liver and other possible sites of production, including adipose tissue, alveolar macrophages, human neurons, and coronary artery smooth muscle cells, in response to external stimuli such as coffee intake, smoking, and alcohol [12]. Generally, CRP levels are decreased in cirrhotic patients than without cirrhosis and elevated in different inflammatory disorder. The possible mechanism responsible for the association between tumours and CRP level is imputed to severe inflammatory processes. The production of CRP by the liver is stimulated by tissue injuries and inflammatory conditions due to the emission of proinflammatory cytokines e.g. Interleukin-6 (Il-6)[13]. Moreover, CRP could be a marker of an immune response to various antigenic stimulation of tumour cells[13]. Some previous studies reported that tumour cells can enhance the synthesis of inflammatory proteins, that subsequently explain the increased CRP levels in patients with different malignancies. Some malignant cells have been shown to produce CRP[13].

The levels of CRP may be helpful in determining the effectiveness of treatments and disease progress. Moreover, elevated levels of CRP were stated as a predictor of lower survival rates in patients with many tumours, including esophageal, pancreatic, colorectal, renal, ovarian, cervical cancer, and urinary bladder [4]. According to these data, in agreement with our results on CRP, elevated levels of CRP were linked to malignant ascites.

In our study we included 80 patients. The patients were divided into malignant ascites (n=30) and benign ascites group (n=50).

In our study, mean ascitic CRP (mg/L) level was significantly higher in malignancy related ascites group(16.6 ± 13.4 vs 2.1 ± 3.2)and p value is 0.001,which is statistically significant (p value <0.05).Ascitic fluid CRP at a cut-off≥5 mg/l had a sensitivity of 76.7% and specificity of 92%. Serum CRP was also significantly higher in malignant ascites than benign ascites patients [10.8±6.3 mg/ml vs. 6.2±4.9 mg/ml;p<0.001.

Ahmed Abdel-Razik et al,[17,18] also reported that ascitic and serum CRP were significantly elevated in malignant ascites than benign ascites group respectively. In their study Serum and ascitic CRP were significantly higher in malignant ascites than benign ascites patients [12.8±6.3 mg/ml vs. 6.1±4.9 mg/ml;p<0.001 and 5.1±2.2 mg/ml vs. 1.6±1.3 mg/ml;p<0.001] respectively .At a cut-off value of 7.3 ng/ml, serum CRP levels had specificity of 77.3% and sensitivity of 84.7% for detecting malignant ascites respectively. Similarly, Yuskel et al,19 also showed that mean baseline serum and ascites levels of CRP were significantly higher in Group 2 (low gradient ascites) compared to those in Group 1(high gradient ascites) (p = 0.021, p = <0.0001, respectively). In mayank et al study, mean ascitic CRP (mg/L) level in malignant group (n=20) is 3.16 vs 0.29 in non-malignant group which is statistically significant (p value <0.01). For the non-malignant group mean value of Serum CRP (mg/l) is 4.40 +

2.18 vs 8.5 + 3.4 in malignant group. p value is <0.01. According to these data, in agreement with our results on CRP, elevated levels of CRP were linked to malignant ascites

Conclusion

This study elucidates a significantly increased serum and ascitic fluid CRP levels in patients with malignant ascites compared to benign ascites. CRP, which isa cheap, simple, non-invasive, widely distributed marker of inflammation can be used for differentiation of malignant ascites from benign ascites.

References

1. Abdel-Razik A, Mousa N, Elalfy H, et al. A Novel Combination of C-Reactive Protein and Vascular Endothelial Growth Factor in Differential Diagnosis of Ascites. Journal of Gastrointestinal Cancer. 2017 Mar;48(1):50-57. DOI: 10.1007/s12029-016-9873-x.
2. Yuksel, I., Karaahmet, F., Coskun, Y. et al. Significance of Serum and Ascitic Fluid C-Reactive Protein in Differential Diagnosis of Benign and Malignant Ascites. Dig Dis Sci 59, 2588–2593 (2014). https://doi.org/10.1007/s10620-014-3205-4
3. Rana SV, Babu SG, Kocchar R. Usefulness of ascitic fluid cholesterol as a marker for malignant ascites. Med Sci Monit 2005;11(3):CR136-42.
4. Wang CS, Sun CF. C-reactive protein and malignancy: clinico-pathological association and therapeutic implication. Chang Gung medical journal. 2009;32:471-82
5. Bijoor AR, Venkatesh T. Value of ascitic fluid cholesterol and serum-ascites albumin gradient in differentiating cirrhotic and malignancy related ascites. Indian Journal of Clinical Biochemistry 2001;16(1):106-9.
6. Sangisetty SL, Miner TJ. Malignant ascites: A review of prognostic factors, pathophysiology and therapeutic measures. World J Gastrointest Surg 2012; 4(4): 87-95
7. Schofield J, Krausz T. Metastatic disease and lymphomas. In: Diagnostic Cytopathology, 2nd ed, Gray W, McKee GT, ed. Elsevier, 2003:153–204
8. Sproston NR and Ashworth JJ (2018) Role of C-Reactive Protein at Sites of Inflammation and Infection. Front. Immunol. 9:754. doi: 10.3389/fimmu.2018.00754
9. Jain S, Gautam V, Naseem S. Acute-phase proteins: As diagnostic tool. J Pharm Bioall Sci 2011;3:118-27
10. Volarić D, Flego V, Žauhar G, Bulat-Kardum L. Diagnostic value of tumour markers in pleural effusions. Biochem Med (Zagreb). 2018;28:010706
11. Ramona C. Dolscheid-Pommerich, Steffen Manekeller, Gisela Walgenbach-Brünagel, Jörg C. Kalff, Gunther Hartmann, Birgit Stoffel Wagner, And Stefan Holdenrieder. Clinical Performance of CEA, CA19-9, CA15-3, CA125 and AFP in Gastrointestinal Cancer Using LOCI™-based Assays Anticancer Res January 2017 37 (1) 353-359
12. Westhuyzen, Justin & Healy, Helen. (2000). Review: Biology and relevance of C-reactive protein in cardiovascular and renal disease. Annals of clinical and laboratory science. 30. 133-43.
13. Sproston NR and Ashworth JJ (2018) Role of C-Reactive Protein at Sites of Inflammation and Infection. Front. Immunol. 9:754. doi: 10.3389/fimmu.2018.00754.