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Case Report

Serum Procalcitonin and C-Reactive Protein in Prediction of Spontaneous Bacterial Peritonitis

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Abstract

Objectives: To compare between Procalcitonin and C-reactive protein regarding diagnosis of spontaneous bacterial peritonitis (SBP).

Design: case control cross sectional study.

Setting: Hepatology and Gastroenterology unit, Specialized Medical Hospital, Mansoura University Hospitals.

Participants: 162 patients admitted to Hepatology and Gastroenterology unit from january 2014 to january 2015, complaining of liver cirrhosis and ascites.

Methods: Blood sample were withdrawn from all subjects involved in the study and used for complete blood count, INR, serum creatinine, albumin, bilirubin, liver enzymes, serum CRP and PCT. Ascitic fluid samples (20ml) were withdrawn under complete aseptic technique and analyzed for polymorph nuclear leukocyte count, glucose, protein, lactate dehydrogenase (LDH), and culture (causative organism, antibiotic susceptibility testing).

Results: The cut-off point of serum PCT at which SBP can be diagnosed was 495 pg/ml with sensitivity and specifity of 90% and 92% respectively. The cut-off point of serum CRP at which SBP can be diagnosed was 10.5 mg/L with sensitivity and specifity of 91% and 97% respectively.

Main outcome measures: Specific diagnostic impact of tests.

Conclusion: Serum procalcitonin and CRP are good indicators of SBP but serum CRP is more sensitive and specific in comparison with PCT as a predictor for SBP.

Keywords: *Procalcitonin (PCT), C-reactive protein (CRP), Spontaneous bacterial peritonitis (SBP).*

Spontaneous bacterial peritonitis (SBP) is defined as an infection of the previously sterile ascitic fluid after exclusion of perforation of viscus, intra-abdominal inflammatory focus like abscess, acute cholecystitis, or acute pancreatitis (1).

Introduction

Spontaneous bacterial peritonitis (SBP) is defined as an infection of the previously sterile ascitic fluid after exclusion of perforation of viscus, intra-abdominal inflammatory focus like abscess, acute cholecystitis, or acute pancreatitis [1].

The bacterial inoculation mechanism of ascites has been the subject of argument and debate since Harold Conn first identified the disorder in the 1960s. Enteric organisms have been isolated from more than 90% of ascitic fluid in patients with SBP, suggesting that the gastrointestinal tract is the source of bacterial contamination [2].

The suspicion of SBP which is always present in patients with liver cirrhosis and ascites. Suggestive signs and symptoms are abdominal pain, fever, and/or altered mental status, although some patients are asymptomatic and the infection is detected when they undergo paracentesis after the admission to the hospital for another purpose like hematemesis and melena, hepatorenal syndrome and/or hepatic encephalopathy [3,4].

Diagnosis of spontaneous bacterial peritonitis (SBP) is dependent upon a manual count of ascetic fluid polymorph nuclear leukocytes (PMNs). This procedure is operator-dependent, lysis of PMNs can occur during transport to the laboratory, and that explains the presence of false-negative results. Furthermore, ascetic fluid culture is insensitive and consumes much time to give results so there is need to research about new diagnostic tools of SBP [5,6].

This study was conducted to assess the role of serum procalcitonin and CRP in diagnosing spontaneous bacterial peritonitis, and to identify the Cut-off value of procalcitonin and CRP that can be used as early predictors for SBP.

Patients and Methods

This is a case control cross sectional study included 162 patients with liver cirrhosis and ascites. The patients were classified into two groups:

Group 1: Patients with ascetic PMNs count above or equal to 250 cells/mm^3 and/or positive ascetic fluid culture (SBP group) (No: 82).

The group 1 was subdivided into three subgroups:

I. Culture positive neutrocytic ascites (CPNA): Patients with ascetic

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PMNs count ≥ 250 cells/mm³ and positive ascetic fluid culture

II. Culture negative neutrocytic ascites (CNNA): Patients with asceticPMNs count ≥ 250 cells/mm³ and negative ascetic fluid culture.

III. Monomicrobial non-neutrocytic Bacterascites (MNNB): Patients with asceticPMNs count < 250 cells/mm³ and positive ascetic fluid culture.

Group 2: patients with ascetic PMNs count below 250 cells/mm³ and negative ascetic fluid culture (Non SBP group) (No: 80).

Patients involved in study were admitted to Specialized Medical Hospital, Mansoura University Hospitals in the period from January 2014 till January 2015.

Inclusion criteria: Patients with liver cirrhosis and ascites, Age ≥ 18 years old.

Exclusion criteria: Hepatocellular carcinoma, extra hepatic malignancy, autoimmune diseases, organ transplant, any other infection due to parasites, fungi or bacteria, acute pancreatitis, recent abdominal surgery within three months before the study, hemoperitoneum, antibiotic therapy started 48hrs or more before enrollment, human immune deficiency virus, chronic dialysis, use of vasoactive amines or mechanical ventilation before withdrawal of samples, massive stress (severe burn, trauma).

The study was explained to all patients and control subjects, and an informed written consent was obtained from them or relatives in cases of hepatic encephalopathy before starting the study. They were subjected to the following: (A) Full medical history taking and full clinical examination with stress on mental state, jaundice, palpation of spleen, abdominal rigidity, tenderness and/or paralytic ileus. (B) Electrocardiography: 12-lead resting ECG (Electrocardiography) is performed to exclude roughly coronary heart disease. (C) Radiological investigations: Chest x ray is performed to exclude the infection, and Abdominal ultrasound to assess liver, spleen, kidneys, portal vein diameter in addition to perform ultra sound guided paracentesis for needed cases. (D) Laboratory investigations in the form of complete blood count which was performed via Sysmex system (Roch) using 1 ml EDTA blood sample, prothrombin time which was performed via Sysmex system, Serum creatinine, albumin, bilirubin and liver enzymes which were performed using the automated system Cobas Integra (Roch), Serum CRP which performed via latex agglutination test (Omega), Serum PCT performed via Enzyme-Linked Immunosorbent Assay (ELISA) kit (Ray Bio ® Human Procalcitonin ELISA Kit, Email: info@raybiotech.com), urine analysis (chemical and microscopic), ascitic fluid analysis (glucose, protein, LDH) and culture (causative microorganism, antibiotic susceptibility testing) (20 ml ascetic fluid obtained from each subject involved in the study under complete aseptic technique).

Statistical analysis: Data entry and statistical analyses were performed using SPSS (statistical package of social sciences) version 16.0 (SPSS Inc., Chicago, IL, USA). Roc curve (receiver operator characteristics) was used to evaluate diagnostic accuracy of some laboratory tests. Correlation was performed between PCT and CRP. P value < 0.05 was considered as statistically significant.

Results

SBP group (No=82 (100%), (51 males, 31 females)) subdivided into the following: CPNA (No=34 (41.4%), (21 males, 13 females)), CNNA (No= 45 (54.8%), (28 males, 17 females)), and bacterascites (No=3 (3.6%) (2 males, 1 female)). The causative microorganisms of CPNA subgroup which isolated from the culture were *Escherichia coli* (No=17, Percentage=50.0%), Klebsiella *pneumoniae* (No=4, Percentage=11.8%), Streptococcus *pneumoniae* (No=2, Percentage=5.9%), Staphylococcus aureus (coagulase positive) (No=8, Percentage=23.5%) and Staphylococcus aureus (coagulase negative) (No=3, Percentage=8.8%). The causative microorganisms of MNNB subgroup which isolated from the culture were *Escherichia coli* (No=2, Percentage=66.7%) and Staphylococcus coagulase positive (No=1, Percentage=33.3%)

I. The inflammatory and infectivity markers of the two studied groups via Mann - whitney test:

The table 1 showed the range and median of WBC count, ascetic fluid PMNs count, CRP level, and PCT level in both groups. The values were significantly higher in group 1 in comparison with group 2. In group 1 patients with leucopenia were 17 in number (20.7%), with normal WBC count were 46 (56.1%), and with Leucocytosis were 19 (23.2%) while in group 2 patients with Leucopenia were 24(30%), with normal count were 53(66.2%), and with Leucocytosis were 3(3.75%). In group 1 patients with neutropenia were 19 in number (23.2%), with normal neutrophil count were 26 (31.7%), and with neutrophilia were 37 (45.1%) while in group 2 patients with neutropenia were 20 (25%), with normal count were 56 (70%), and with neutrophilia were 4(5%) (data not shown in the table).

Table 1: Range and median of WBC count, ascetic fluid PMNs count, CRP level, and PCT level in both groups.

Variables	Group 1 N=82	Group 2 N=80	P Value
	Range Median		
White blood cell count	1300-22.200 6900	1800 - 11800 6300	0.04*
Ascitic fluid PMNs count	240-14242 1409	18 -232 90	≤0.001*
CRP level	0 -128 24	0-12 0	≤0.001*
PCT level	40 - 5403.5 953.75	32 - 620 115	<0.001

II. Comparisons between 2 groups as regard PCT and CRP: Table 2,3

Table 2: True positive, true negative, false positive and false negative values regarding CRP in both groups.

CRP	Group 1	Group 2	Total
Positive	80 (TP)	4 (FP)	84
Negative	2 (FN)	76 (TN)	78
Total	82	80	162

from the previous table the following results were concluded: Sensitivity = 97.5%, Specificity = 95%, Positive predictive value = 95.2%, Negative predictive value = 97.4%.

Table 3: True positive, true negative, false positive and false negative values regarding PCT in both groups.

Procalcitonin	Group1 (N=82)	Group 2(N=80)	Total
Positive	74(TP)	5(FP)	79
Negative	8(FN)	75(TN)	83
Total	82	80	162

from the previous table the following results were concluded: Sensitivity = 90.2%, Specificity = 93.75%, Positive predictive value = 93.6%, Negative predictive value = 90.4%.

III. Correlations between CRP and PCT: Figure 1 A significant positive correlation was noted between PCT and CRP concentrations (P<0.0001, rs=0.84).

IV. Diagnostic accuracy of PCT, CRP: Figure 2 As determined by ROC curve analysis, the accuracies of PCT concentration (area under the curve [AUC], .95; 95% (confidence interval) [CI], .94 - 1.00) and CRP concentration (AUC, .975; 95%CI, .951- 1.00) for identifying patients with SBP were significantly high. The AUC-ROC for the PCT concentration was not significantly different from that for the CRP

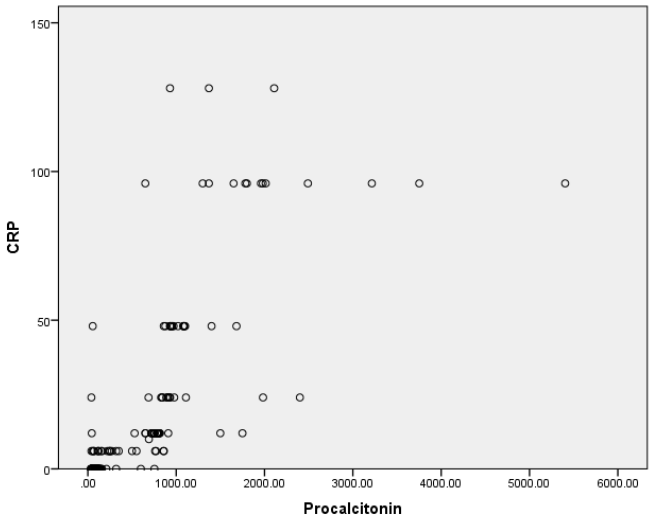

Figure 1: G graph (scatter dot graph) for correlation between PCT and CRP.

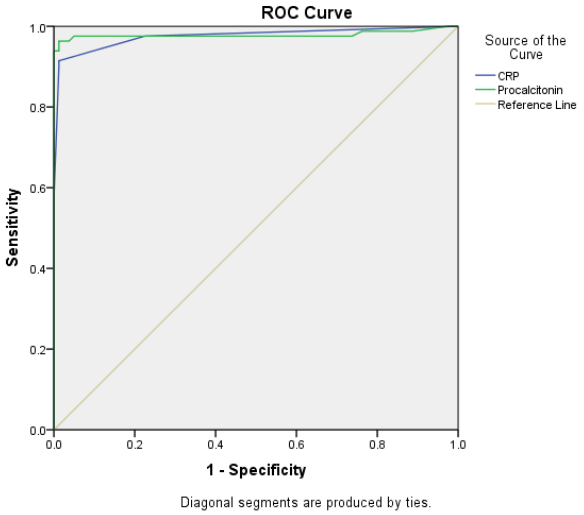

Figure 2: ROC curve for relation between sensitivity and specifity of PCT and CRP in diagnosis of SBP.

concentration. The optimal cut-off point of PCT that can be used for diagnosis of SBP is 495 pg/ml with sensitivity and specifity of 90% and 92% respectively while the optimal cut-off point of CRP that can be used for the diagnosis is 10.5 mg/L with sensitivity and specifity of 91% and 97% respectively.

V. Multivariate logistic regression using SBP as outcome (Table 4): adjusted for ascitic fluid PMNs, ascitic fluid culture, WBC count, and neutrophil blood count. From the previous table, we can conclude that Serum PCT and CRP are independent detectors of SBP.

Discussion

Liver cirrhosis patients are highly susceptible to bacterial infections because of acquired immune defects of both cell-mediated and humoral immunity and bacterial translocation. Hepatic dysfunction is affiliated strongly with impaired defenses against bacteria, and with structural and functional modifications in the intestinal mucosa that result in an increase in the permeability to bacteria and bacteria-derived products, which worsens over time and with disease progression [7].

Table 4: Multivariate logistic regression using SBP as outcome.

Covariates	Odds ratio	CI	P value
PCT	1.45	1.12 - 1.96	.002
CRP <6: reference category ≥6	 reference category 2.3	 reference category 1.7 - 3.6	 reference category .001

So SBP is a major complication of liver cirrhosis and ascites and is considered the most frequent bacterial infection in patients with liver cirrhosis [8]. The valid diagnostic tools of SBP had high false negative results [6]. That is why there is a necessity to search for a new diagnostic tool. This study aimed to assess the role of serum PCT and CRP in diagnosis of SBP.

In our study CNNA included 45 patients (54.8%). CNNA was the predominant group also in the studies of Bhat et al who lasted for 4 years and performed on 600 patients with suspected SBP. The results showed 70 of patients (11.6%) proved to have SBP, these patients have divided into three groups: CNNA and this involved 40 patients (57,1%), CPNA and this involved 25 patients (35.8%), and bacterascites and this included five patients (7%) [9].

In our study E.coli was the predominant organism (70%), followed by staphylococcus species. E.coli was also the predominant organism found in the studies of Gill et al [10]. The study conducted in Khyber Teaching Hospital, Peshawar 2003, showed E. coli was isolated in 58.13%, *Streptococcus pneumoniae* in 18.60%, Staphylococcus aureus in 9.13%, Klebsiella in 9.13% and Acinectobacter in 4.63% and this difference can be explained by different localities and different sample sizes [11].

In our study the concentrations of PCT and CRP were significantly higher in the SBP group than in those without SBP. Furthermore, there were significant correlations between PCT and CRP. The accuracy of the PCT concentration was not significantly different from that for the CRP concentration.

The diagnostic value of serum PCT level in liver diseases has been evaluated in several studies, with relatively consistent results [12,13]. In patients with decompensated liver cirrhosis, a high PCT concentration showed a high sensitivity and specificity for bacterial infections [14]. In these studies, the serum PCT was significantly elevated in SBP patients compared with those without SBP.

In our study the following results were concluded: Sensitivity = 90.2%, Specificity = 93.75%, Positive predictive value = 93.6%, Negative predictive value = 90.4%. This result was in agreement with Viallon et al as procalcitonin is a good and early diagnostic tool of SBP. Viallon et al found that the cut-off value = 0.76 ng/mL, AUC= 98%, sensitivity= 95%, specificity = 85%, PPV= 77% and NPV= 97%. The different values of viallon et al in comparison with ours can be explained by different localities and different sample size [14].

CRP and PCT have been used as inflammatory markers of bacterial infections, even in liver diseases. However, many questions on the optimal application of CRP and WBC in this patient population have not been addressed. It is vital to assess the determination power of CRP. In previous clinical studies involving 20 to 127 patients, CRP proved to be effective markers of bacterial infections in patients with liver diseases, but they had diverse diagnostic accuracies and cut-off values [15,16]. A possible explanation for this variance may be the differences in the type of liver disease in between patients. In the present study, CRP at the optimal cutoff of 10.5 mg/L had the best sensitivity (91%), specificity (97%), and AUC-ROC (0.975). The optimal cutoff value of PCT was 495 pg/ml, with a lower sensitivity (90%), specificity (92%), and AUC-ROC (0.95). This suggests that the PCT is inferior to the serum CRP for the diagnosis of SBP.

Larger samples and more homogeneous groups of SBP should be used for further studies in order to confirm our results.

Conclusion

From this study, we can conclude that serum PCT alone or

CRP alone or combination of them seems to provide satisfactory diagnostic biomarkers in SBP but CRP is more specific and sensitive in comparison with PCT regard diagnosis of SBP.

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