Mean Platelet Volume is a Promising Diagnostic Marker for Systemic Inflammation in Cirrhotic Patients with Ascitic Fluid Infection

Abdelmoez Amal 1, Atalla Mahmoud 2, Hefry Zeinab 1, Abdellatif Zeinab 2, Adel Eman 2, Abbas Bahaa 3, Mehrez Mai 4, Guda Mohamed 5, Abouelkhair Mahmoud 6 and Elzahry Mohammad AME*

1Department of Medicine, Ain Shams University, Cairo, Egypt
2Department of Endemic Medicine and Hepatology, Faculty of Medicine, Cairo University, Cairo, Egypt
3Air Force Hospital, Cairo, Egypt
4National Hepatology and Tropical Medicine Research Institute, Cairo, Egypt
5Theodore Bilharz Research Institute, Cairo, Egypt

Abstract

Introduction: Spontaneous bacterial peritonitis (SBP) is the most frequent and life-threatening infection in patients with decompensated liver cirrhosis. Diagnosis should be prompt and treatment must not be delayed until the microbiology results are available.

Aims and Methods: The current study was conducted to assess the potential role that MPV may have in the diagnosis of SBP in cirrhotic patients with ascites. Three groups were included in the study. Group I (SBP group) included 100 patients with liver cirrhosis and ascites complicated by SBP, group II (non-SBP group) included 98 patients with liver cirrhosis and ascites without SBP and group III included 50 healthy subjects to serve as a control group.

Results: ESR, CRP and total leucocytic count (TLC) in ascitic fluid were significantly higher in SBP group compared to non-SBP group (median 37.5 vs. 12, 12 vs. 5 and 530 vs. 60 respectively with p value<0.01). The MPV was significantly higher in SBP group vs. non-SBP group and healthy control group (8.5, 7.9 and 8.3 respectively and P value<0.0001). On constructing ROC curve for the MPV, at a cutoff value of 8.4 ft, MPV had 73% sensitivity and 85.7% specificity for detecting SBP with overall accuracy 79.3%, (AUC=0.84 with negative predictive value (NPV) and positive predictive value (PPV) for MPV of 75.7 and 83.9%, respectively). Regarding ESR, at a cutoff value of 20.

Conclusion: MPV is increased in SBP in cirrhotic patients with ascites than the other inflammatory markers.

Keywords: MPV; Ascites; Infection

Introduction

Spontaneous bacterial peritonitis (SBP) is infection of the ascitic fluid with no intra-abdominal source of infection or malignancy. It is one of the most frequent complications in patients with liver cirrhosis and ascites [1]. Ascitic fluid infection is extremely common at the time of hospital admission of a patient with cirrhosis and ascites that requires a diagnostic paracentesis [2].

For SBP diagnosis, the count of polymorphonuclear leucocytes [PMN] in the ascitic fluid obtained by paracentesis must exceed 250 cells/mm². Because SBP is in most cases an infection with a single organism, the presence of many organisms in the culture, raise the suspicion of secondary peritonitis [3].

Ascidic Fluid Infection (AFI) occurs in 10 to 30% of hospitalized cirrhotic patients. Despite the early start of treatment, which may lead to good results in most cases, the mortality still remains considerably high [4].

For this reason, early detection of inflammation is important for the assessment of AFI and for treatment options. Although the diagnosis is based mainly upon clinical suspicion, several methods have also been studied for early detection of AFI in cirrhotic patients [5].

Additional diagnostic tools, such as leucocyte esterase reagent strips, pH testing, and lactoferrin in ascitic fluid are also considered to be helpful in SBP diagnosis. Fecal calprotectin (FC), ascitic fluid calprotectin, proinflammatory cytokines like interleukin 1b [IL1b], tumor necrosis factor a (TNFa), and interleukin 6 (IL6) were also studied. Another study focused on the diagnostic role of plasma and ascitic fluid procalcitonin for estimating SBP diagnosis [6].

Circulating platelets are a plentiful source of prothrombotic agents which are inflammatory markers that have an important role in the inflammatory cascade [7]. The platelet content of granules increases as the platelet size increases to have their hemostatic and pro-inflammatory actions with more efficiency, for this reason mean platelet volume (MPV) is proposed to be an indicator of platelet function and activation. Some studies have reported that MPV increases in myocardial infarction, cerebrovascular disease, Alzheimer’s disease, hypertension, and celiac disease [8].

The current study was conducted to assess the potential role that MPV may have in the diagnosis of SBP in cirrhotic patients with ascites together with other inflammatory markers; ESR and CRP.

Patients and Methods

The current study is a case control study that took place during...
the period from May 2015 to March 2016 at Tropical medicine department, Faculty of Medicine, Ain Shams University, Egypt. Patients with ascites due to liver cirrhosis who were admitted to the hospital were included after signing an informed consent. History taking and physical examination with baseline Complete blood count (CBC), Erythrocytes Sedimentation Rate (ESR), C-reactive protein (CRP) and liver and kidney function tests and abdominal ultrasonography were performed. Ascitic fluid samples were aspirated under complete aseptic precautions. All areas of scarring were avoided since they are often the site of collateral vessels formation or adherent bowel loops in patients with portal hypertension [9]. The aspirated samples were checked for total and differential white blood cells (WBC). Culture of ascitic fluid was done by inoculating 10 ml of ascitic fluid at the bedside in two blood culture bottles one for aerobic and the other for anaerobic bacteria under complete aseptic precautions. Incubation was done at 37°C for 48 to 72 hours. Any colony (growth) or turbidity appearing on the media was subjected to microscopic examination of film stained by Gram stain, biochemical reaction and serological examination. Patients were then classified into two groups:

1. Group I (AFI group) included 100 patients with PMNs count ≥ 250 cell/cmm.

2. Group II (Non-SBP group) included 98 patients with PMNs count <250 cell/cmm.

3. Control group of 50 healthy subjects were also included to serve as a control group (Group III). They were 35 males and 15 females, their age ranged from (23-61 years) with a mean of 44.93 ± 12.73 years.

All group I patients received intravenous third generation cephalosporins antibiotics for at least five days and followed up clinically and laboratory to confirm resolution of infection. Antibiotic response was assessed by another ascitic fluid sample after 48 hours of initiation, and patients were considered responsive if a 25% decrease or more was achieved in PMNs count.

Patients with ascites due to local causes (such as tuberculous and malignant ascites), or those who have recent abdominal surgery were excluded. Also, patients with medical diseases affecting platelets (e.g. heart failure, hypertension, diabetes, hyperlipidemia, peripheral vascular disease, hematological and neoplastic disorders) were excluded as well as patients who received antibiotics or anti-platelet medications prior to hospital admission.

**Statistical analysis**

Data analysis was done using Statistics/Data Analysis (STATA) version 13.1 software. Continuous variables were tested for normality by the Shapiro-Wilk normality test. Values are presented as mean ± standard deviation, or in the case of non-normally distributed data as median and inter-quartile range. The Chi-squared test was used to compare percentages between different groups of patients. Normally distributed data were analyzed using independent samples T-test. Data found to be non-normally distributed were analyzed using the Mann-Whitney U test. Non-normally distributed paired samples were analyzed using the Wilcoxon signed-rank test. Kruskal-Wallis equality-of-populations rank test was used to compare non-normally distributed data in the 3 groups. Spearman’s correlation analysis was done between MPV & inflammatory markers; ESR, CRP, WBCs and platelets. Receiver operating characteristic [ROC] curve analysis was used to identify the best cut-off value of MPV with maximum sensitivity and specificity for differentiation of cirrhotic patients with SBP from those without SBP.

**Results**

The age of the studied population ranged from 26 to 61 years with a mean of 50.43 ± 7.49 years in group I (SBP group); and 75% of them were males. In group II (Non SBP group), the age ranged from 35 to 75 years with a mean of 53.8 ± 8 years; and 73 (74%) were males. In group III (control group), the age ranged from 23 to 61 years with a mean of 44.93 ± 12.73 years. Thirty-five persons (70%) were males. Baseline laboratory characteristics of the studied groups are shown in Table 1. The results of ascitic fluid culture in group I (SBP group), *Escherichia coli* was detected in 15 patients (15%), *Klebsiella* in one patient (1%) and *Acinetobacter* in one patient (1%) while no bacterial growth had occurred in 83 patients (83%).

As shown in Table 2; MPV is significantly higher in group I more than group II and more than the control subjects (P value in group I vs group II<0.0001, P value healthy controls vs group II<0.0001 and P value group I vs healthy controls=0.05). Also, in group I patients, MPV was significantly reduced after treatment of AFI (Table 3).

On constructing ROC curve for the sensitivity and specificity of MPV: At a cutoff value of 8.4 fl, MPV had 73% sensitivity and 85.7% specificity for detecting SBP with overall accuracy of 79.3%, (AUC=0.84 with negative predictive value (NPV) and positive predictive value (PPV))for MPV of 75.7 and 83.9%, respectively. On the other hand, at a cutoff value of 20, ESR had 78% sensitivity and 63.4% specificity for detecting SBP with overall accuracy 70.7, AUC= 0.845 with negative predictive value NPV and positive predictive value of 73.8 and 68.5%, respectively). However, at a cutoff value of 7, CRP had 66% sensitivity and 67.4% specificity for detecting SBP with overall accuracy 66.6%.
AUC= 0.71 with negative predictive value NPV and positive predictive value of 66% and 67.3%, respectively (Figure 1).

Spearman correlation analysis showed that there was a significant correlation between MPV and ESR (r=0.3720, P<0.001), MPV and CRP (r=0.275, P=0.0001), MPV and Platelets (r=-0.146, P=0.039). However, no correlation was observed with WBC (r=0.0218, P=0.76).

As shown in Table, Logistic regression model was performed to assess the relationship between SBP as an outcome variable and the MPV, CRP, and ESR as predictor variables and showed that MPV, ESR and CRP were good predictors with best performance for MPV.

Discussion

Diagnostic paracentesis, which is the gold standard for diagnosing AFI, is sometimes associated with minor complications e.g. bleeding, visceral perforation, local infection and persistent leakage after paracentesis. Therefore, a simple, rapid, non-invasive and inexpensive diagnostic test to make a presumptive diagnosis of AFI in cirrhotic patients is needed.

Many tests have been subjected to investigation for this purpose and some proved to be helpful in diagnosing ascitic fluid infection in cirrhotic patients but their use is confined to research purposes or limited by high cost. Examples include leucocyte esterase reagent strips, pH testing, lactoferrin in ascitic fluid, serum and ascitic fluid procalcitonin [6].

Platelets are considered an important source of prothrombotic agents associated with inflammatory markers, and play a role in the initiation and propagation of vascular and inflammatory diseases [7]. MPV is one of the most widely used surrogate markers of platelet activation, in which larger platelet volume means both an enzymatically activated, in which larger platelet volume means both an enzymatically

Platelets are considered an important source of prothrombotic agents associated with inflammatory markers, and play a role in the initiation and propagation of vascular and inflammatory diseases [7]. MPV is one of the most widely used surrogate markers of platelet activation, in which larger platelet volume means both an enzymatically and metabolically more active platelet compared to a smaller one [10,11].

The role of platelets as a marker of systemic inflammation in patients with liver cirrhosis and ascites complicated with SBP has not yet been clearly evaluated.

A statistically significant elevation was noted about MPV, CRP and ESR in the SBP group as compared to the non-SBP group. The statistically significant difference in MPV (P<0.0001), between SBP and non-SBP groups was independent of the severity of liver cirrhosis as determined by the Child-Turcotte-Pugh score. A statistically significant difference was noted between SBP group and healthy controls (P<0.05) and between non-SBP group and healthy controls as well (P=0.0001). Similar results were obtained by Suvak and colleagues who reported a significant difference between MPV levels between cirrhotic patients with AFI compared to cirrhotic patients without AFI (P<0.001) and healthy controls (P<0.001) (8.79 ± 1.01 fl, 8.05 ± 0.83 fl and 7.88 ± 0.47 fl, respectively). No statistically significant difference was observed between cirrhotic patients without AFI and healthy controls (P=0.368) [5].

Data obtained by Suvak and colleagues showed a significant correlation between MPV and CRP (r=0.335, P ≤ 0.001). No correlation was observed with WBC (r=0.049, P=0.714), ESR (r=0.105, P=0.524) [5]. On the other hand, our study showed a significant correlation between MPV and CRP (r=0.275, P=0.0001), ESR (r=0.3720, P<0.0001), but no correlation was observed with WBC (r=0.0218, P=0.76). The positive correlation between MPV and other inflammatory markers may support the hypothesis that increased MPV could reflect ongoing systemic inflammatory responses in cirrhotic patients with SBP.

In the present study, we found that MPV is statistically significant (AUC=0.84, P<0.0001) in the diagnosis of SBP with overall accuracy 79.3% and we recommend MPV cutoff value of 8.4 fl as an optimal cutoff value for diagnosis, with 73% sensitivity and 85.7% specificity, independent of the severity of liver disease as determined by CTP score. Suvak and colleagues recommended MPV cutoff value of 8.45 fl, with a sensitivity 70.7% and specificity 67.5%, (AUC=0.768) [5]; while Abdel-Razik and colleagues suggested MPV cutoff value of 8.77 fl, for the diagnosis with 95.9% sensitivity and 91.7% specificity, (AUC=0.964) [12]. Gálvez-Martínez and colleagues supported that MPV can be a useful predictor of systemic inflammatory response syndrome in cirrhotic patients with AFI, particularly culture-negative neutrocytic ascites and found that the best cutoff value of MPV was 8.3 fl, with sensitivity 84% and specificity 82%, (AUC=0.9) [13].

The ROC curve for sensitivity and specificity of MPV for detection of SBP was found to be comparable to CRP and ESR while Suvak and colleagues reported that the increase in MPV in cirrhotic patients with AFI is comparable to CRP, but superior to ESR [5]. Abdel-Razik and colleagues concluded similar results but did not include ESR [12].

To the best of our knowledge, no data exists regarding the role that MPV may have in the follow-up of patients with SBP after treatment up to the time of writing this manuscript. Our results showed a significant decrease in MPV (8.1 ± 0.8 fl) after receiving antibiotic therapy Vs before receiving therapy (8.5 ± 0.6 fl) (P value<0.0001).

Conclusion

MPV was significantly elevated in cirrhotic ascitic patients with SBP compared to non-SBP patients regardless of the severity of liver disease. MPV measurement can be considered an accurate, simple, noninvasive test in the diagnosis of SBP and can be useful in the follow-up in assessing the response to treatment.

References

1. Rimola A, Garcia-Tsao G, Navasa M (2000) Diagnosis, treatment and prophylaxis of spontaneous bacterial peritonitis: A consensus document. International Ascites Club. J Hepatol 32: 142-53.
2. Borzio M, Salerno F, Piantoni L (2001) Bacterial infection in patients with advanced cirrhosis: A multicentre prospective study. Digestive and liver disease: Official journal of the Italian Society of Gastroenterology and the Italian Association for the Study of the Liver 33 :41-48.
3. Guarner C, Soriano G (2005) Bacterial translocation and its consequences in patients with cirrhosis. Eur J Gastroenterol Hepatol 17: 27-31.
4. Perumalswami PV, Schiano TD (2011) The management of hospitalized patients with cirrhosis: The Mount Sinai experience and a guide for hospitalists. DigesDis Sci 56: 1266-1281.

5. Suvak B, Torun S, Yildiz H (2013) Mean platelet volume is a useful indicator of systemic inflammation in cirrhotic patients with ascitic fluid infection. Ann Hepatol 12: 294-300.

6. Koulouzidis A, Bhat S, Saeed AA (2009) Spontaneous bacterial peritonitis. W J Gastroenterol 15: 1042-1049.

7. Kilciler G, Genc H, Tapan S (2010) Mean platelet volume and its relationship with carotid atherosclerosis in subjects with non-alcoholic fatty liver disease. Upsala J Med Sci 115: 253-259.

8. Golebiewska EM, Poole AW (2015) Platelet secretion: From haemostasis to wound healing and beyond. Blood Rev 29: 153-162.

9. Runyon BA (2009) Management of adult patients with ascites due to cirrhosis: an update. Hepatology (Baltimore, Md) 49: 2087-2107.

10. Wiest R, Krag A, Gerbes A (2012) Spontaneous bacterial peritonitis: recent guidelines and beyond. Gut 61: 297-310.

11. Gasparyan AY, Sandoo A, Stavropoulos-Kalinoglou A (2010) Mean platelet volume in patients with rheumatoid arthritis: The effect of anti-TNF-alpha therapy. Rheumatol International 30: 1125-1129.

12. Abdel-Razik A, Eldars W, Rizk E (2014) Platelet indices and inflammatory markers as diagnostic predictors for ascitic fluid infection. Eur J Gastroenterol Hepatol 26: 1342-1347.

13. Galvez-Martinez M, Servin-Caamano AI, Perez-Torres E (2015) Mean platelet volume as a novel predictor of systemic inflammatory response in cirrhotic patients with culture-negative neutrocytic ascites. World J Hepatol 7: 1001-1006.