

RESEARCH ARTICLE

Evaluation of platelet indices as early biomarkers for bacterial infections in pediatric emergency departments

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ABSTRACT

Discriminating bacterial from viral etiology of infectious diseases can be challenging in pediatric age, especially during the first year of life and even more in an emergency setting. Recent research has identified in platelet indices a potential biomarker for bacterial infections, nonetheless current results remain inconsistent. The primary objective of this retrospective observational study was to assess the ability of platelet indices to distinguish bacterial from viral infections in infants presenting to the emergency department. As secondary endpoints, it aimed to evaluate these indices in differentiating upper from lower urinary tract infections and bacterial from viral pneumonias. The present study included 236 patients younger than 12 months consecutively admitted for urinary tract infections, bronchiolitis, pneumonia or gastroenteritis to our Pediatric Emergency Department. Blood cells count was performed in each patient at admission and PLT indices (platelet count, mean platelet volume-MPV and platelet distribution width) were extracted from medical charts. Children with suspected bacterial diseases had slightly lower PLT and PLT/MPV ratio compared to those with suspected viral infections. None of the platelet indices showed significant differences between upper and lower urinary tract infections. A slight decrease in PLT, MPV and PLT/MPV was recorded in infants with bacterial pneumonias compared to those with viral forms. Platelet indices have not proved effective in defining

the bacterial etiology of an infectious in children younger than 12 months. They do not discriminate between lower and upper urinary tract infections, nor between bacterial and viral pneumonias.

HIGHLIGHTS BOX

What is already known about this topic? Discriminating bacterial from viral etiology of infectious diseases can be challenging. Increasing evidence points out the role of platelet indices as biomarkers of bacterial infections. **What does this article add to our knowledge?** Platelet indices do not discriminate between lower and upper urinary tract infections, nor between bacterial and viral pneumonias. **How does this study impact current management guidelines?** The accuracy of the available biomarkers in discriminating the etiology of an infectious process in the paediatric population remains limited. More studies are needed to fill this gap.

KEYWORDS

Bacterial infection; children; infection biomarkers; platelet indices.

Abbreviations

ANC: Absolute Neutrophil Count

AUC: Area Under the Curve

CRP: C-Reactive Protein

LP: Lymphocyte percentage

MPV: Mean Platelet Volume

NP: Neutrophil Percentage

PCT: Procalcitonin

PDW: Platelet Distribution Width

PLT: Platelet

UTI: Urinary tract infection

WBC: White Blood Cells

INTRODUCTION

Pediatric infectious diseases can develop through heterogeneous and mostly non-specific clinical pictures. This is particularly true and critical in infants younger than 12 months, whose clinical presentation is often unclear and who are at higher risk of invasive diseases. It is even more relevant in an emergency setting, where the presence of a temporal constraint to the diagnostic evaluation enhances the need for a timely etiological framework. The distinction between viral and bacterial infections defines the entire diagnostic and therapeutic path, since the latter correlate with a greater tendency to evolve into severe forms and require antibiotic treatments, ineffective and inappropriate in the management of viral diseases (1).

Several authors have identified multiple biomarkers and evaluated their efficacy in discriminating the etiology of an infectious process. Nevertheless, in pediatric age bacterial-viral co-infections are common and complicate the diagnostic path in the absence of a specific biomarker. Despite plenty of evidence suggesting the effectiveness of C-Reactive Protein (CRP) and procalcitonin (PCT) in identifying bacterial diseases (2, 3), their diagnostic accuracy in the pediatric population remains limited, especially regarding their negative predictive value. Furthermore, their routine dosage is limited due to its expensiveness (4, 5). Most recent research has identified promising alternatives to traditional biomarkers. An assay combining 3 circulating proteins (TRAIL, IP-10, and CRP), differently produced by the host in response to bacterial or viral infections, demonstrated superior effectiveness than CRP and PCT in detecting bacterial pathologies among children aged under 60 months (6). Preliminary studies have furthermore shown the ability of RNA biosignatures to distinguish between patients with or without bacterial infections (7). The applicability of such tools in the clinical practice of emergency departments remains debatable, both in terms of costs and availability.

Although platelets have been traditionally evaluated for their central role in the hemostatic process, several studies have supported their involvement in the acute phase response, in which IL-6 production underlies the increase in thrombopoietin levels (8). Moreover, the identification of toll-like receptors on platelet surface and their ability to release antibacterial molecules has suggested thrombocytes involvement in the response to infectious pathogens (9). This highlights the potential role of platelet

indices (platelet count, mean platelet volume, platelet distribution width) as biomarkers of inflammatory conditions and even of bacterial infections, especially in the emergency setting, where blood count is rapidly and routinely evaluated.

Since their clinical significance remains uncertain and given the absence of comparative analyses in this regard, the aim of our study was to evaluate the efficacy of platelet indices in discriminating between the bacterial or viral etiology of an infectious disease in infants younger than 12 months. Furthermore, we aimed to characterize their diagnostic accuracy in differentiating between upper and lower urinary tract infections and between bacterial and viral pneumonias.

MATERIALS AND METHODS

We performed a retrospective observational study evaluating 236 patients younger than 12 months admitted to our Pediatric Emergency Department with a diagnosis of urinary tract infection (UTI), bronchiolitis, pneumonia or gastroenteritis. Such age range corresponds to the population group whose clinical presentation is more nonspecific and which, at the same time, is at a higher risk of invasive forms. Platelet indices were extracted from medical charts, specifically referring to the blood sample collected at admission. Platelet count and volume can be altered by an inadequate blood sample collection (e.g., haemolysis). Being ours a retrospective analysis, it was not possible to certify the quality of the pre-analytical phase; however, patients whose laboratory tests displayed signs of haemolysis and/or extremely altered platelet counts (PLT $<50000/\mu\text{l}$ or $>1000000/\mu\text{l}$) were excluded by the study. Additional exclusion criteria were prematurity (gestational age <37 weeks) and comorbidities (chronic diseases, syndromes, respiratory and/or urinary malformations, congenital heart diseases). Infants with UTIs (single bacterial species >100.000 CFU/ml in urine culture) (10), have been divided into two subgroups: lower (cystitis, urethritis) and upper UTIs (pyelonephritis). Upper UTIs were defined in presence of fever (body temperature ≥ 37.5 °C) and a CRP value \geq median +1 quartile (5.58 mg/dl in our statistic). In children with respiratory illnesses, nasopharyngeal secretions were collected through a nasal tube after the injection of 3 ml of isotonic saline solution into each nostril. A panel of either reverse transcriptase PCR or nested PCR assays allowed to detect 14 respiratory viruses (RSV, RV, Influenza A

and B, Adenovirus, Coronavirus OC43-229E, NL63 and HKUI, Parainfluenza 1-3, MPV and BoV) (11). *M. pneumoniae* and common bacteria DNA were detected by Real-Time PCR reactions on samples extracted from throat swab (12). An additional blood sample was collected to evaluate *M. pneumoniae* and *C. pneumoniae* serologies. Bronchiolitis was defined as the first episode of lower respiratory tract infection in previously healthy infants with spread crackles on chest auscultation (13). Pneumonia diagnosis was established when fever was associated with clinical or radiological evidence of a new pulmonary consolidation. Further criteria differentiated two subgroups:

- bacterial pneumonias. Defined by the presence of all the following: lobar consolidation at chest X-ray; elevated white blood cells count; elevated CRP values;
- viral pneumonias. Defined by the presence of all the following: radiological evidence of perihilar peribronchial thickening, interstitial infiltrates or non-lobar pulmonary consolidation; detection of a virus in the nasopharyngeal aspirate; absence of *M. pneumoniae* and common bacteria in throat swab; negativity of *M. pneumoniae* and *C. pneumoniae* serologies.

Gastroenteritis diagnosis was made in the presence of fever, diarrhea and/or vomiting, thence a stool sample was collected to perform a coproculture and to detect viral antigens. Children with documented presence of Rotavirus and/or Adenovirus antigens in stools and a negative coproculture were included in the study. To carry out a comparative analysis, we grouped enrolled patients in two categories as follows: bacterial diseases (UTIs and bacterial pneumonias) and viral diseases (bronchiolitis, viral pneumonias and gastroenteritis).

Statistics

Data statistical analysis was performed using SPSS software (version 25.9; SPSS Inc., Chicago, Illinois, USA). Qualitative variables were expressed as absolute values and percentages, and then compared with Chi-squared test. Normally distributed quantitative variables (PLT, PLT/MPV) were expressed considering average and standard deviation and Student t-test was used for their comparison. Quantitative variables with no-normal distribution (MPV) were expressed using median and the relative range (minimum and maximum value), then compared by Mann-Whitney U test. Concerning PLT, MPV

and PLT/MPV ratio, the normality of their distribution curve was established using the Kolmogorov-Smirnov test. A p-value ≤ 0.05 was considered as statistically significant. The statistical correlation between quantitative variables was evaluated using Pearson's correlation coefficient. A Receiver Operating Characteristic (ROC) curve was constructed to evaluate the diagnostic accuracy of platelet indices and to compare it to the diagnostic value of white blood cell (WBC), neutrophil percentage (NP), lymphocyte percentage (LP) and CRP.

RESULTS

We performed a retrospective observational study involving 236 infants. The median age was 3.0 months (age range: 0.3-11.9 months), and 124 infants (52.5%) were males. The sample was grouped as follows: 145 patients (61%) with bronchiolitis; 41 (17%) with urinary tract infection, 28 of whom were lower UTIs; 29 (13%) with pneumonia, 18 of whom were bacterial; 21 (9%) with gastroenteritis.

We divided patients based on the microbial etiology: 59 children with bacterial infections (31 males) and 177 with viral infections (93 males). We documented a non-significant slightly lower value of PLT and PLT/MPV ratio in bacterial diseases, while MPV values were completely overlapping (**Table 1**).

We used ROC curves to evaluate the diagnostic accuracy of platelet profile. The Area Under the Curve (AUC) of PLT, MPV e PLT/MPV demonstrates that platelet indices do not differentiate between viral and bacterial infections, unlike already validated markers (WBC, NP, LP, and CRP), whose AUC indicates proper accuracy (**Table 2** and **Figure 1**).

Urinary infections were subgrouped into lower (cystitis, urethritis) and upper (pyelonephritis) tract infections. PLT, MPV and PLT/MPV did not differ between the groups (**Table 3**).

Considering bacterial and viral pneumonias, we found slightly lower PLT, MPV and PLT/MPV in bacterial pneumonias, albeit statistically non-significant (**Table 4**).

We documented a positive correlation between white blood cells (WBC) and PLT ($r = 0,198$; $p < 0,05$) as well as WBC and PLT/MPV ($r = 0,176$; $p < 0,05$). The absolute neutrophil count (ANC) positively correlates with PLT ($r = 0,238$; $p < 0,05$) and PLT/MPV ($r = 0,239$; $p < 0,05$). Conversely, we found no statistical association between CRP values and platelet indices (PLT, MPV, PLT/MPV) (**Table 5**).

DISCUSSION

In this retrospective observational study involving 236 infants younger than 12 months, we aimed to evaluate the efficacy of platelet indices in discriminating the bacterial or viral etiology of infectious diseases.

The involvement of thrombocytes in bacterial infections has been evidenced by the correlation between platelet indices and procalcitonin, a well-known bacterial biomarker (14). Significantly higher MPV values were demonstrated in sepsis patients, the emblematic paradigm of a bacterial disease (15, 16). The physiopathological rationale for this increase would lie in the release of larger newly formed platelets, aimed at the replacement of the thrombocytes destroyed by peripheral consumption. Tamelyte *et al.* pointed out that the increase of the PLT/MPV ratio in children presenting to the emergency department resulted more effective than leukocytosis, neutrophilia and CRP in detecting sepsis or bacteriemia (17).

Conversely, MPV reduction under inflammatory conditions has been interpreted as consequence of the seizure of larger platelets inside the mesenteric district during acute appendicitis as well as in rotavirus gastroenteritis, resulting in a lower mean platelet volume (18, 19). Analogous mechanisms have been hypothesized to explain the MPV decrease in infants suffering from acute bronchiolitis (20). As far as most recent findings are concerned, a reduction in MPV has also been documented in influenza and COVID-19 patients, the latter showing significantly lower values (21).

In the present study, none of platelet indices has proved effective in discriminating bacterial from viral infections (non-significant differences between the two groups; smaller AUC than WBC, NP, LP, and CRP). The slightly positive association that correlates WBC and ANC to PLT and PLT/MPV does not allow to draw definitive conclusions concerning platelet indices' ability to detect bacterial infections.

Our findings are in contrast with results emerging from the literature and several reasons might underlie such discrepancy. Most studies involve adults or a wide age range of pediatric patients, resulting in their sample being unavoidably heterogeneous. Furthermore, they take into consideration a specific infectious disease and, above all, the control group is generally composed of healthy subjects. To the best of our knowledge, ours is the first study that evaluated platelet indices accuracy comparing

a pool of bacterial diseases with one of viral infections, regardless of their specific etiology and without a healthy control group. For this purpose, the results obtained document their limited usefulness. Therefore, platelet indices cannot be generalized as markers of bacterial disease; this does not exclude that they may vary significantly from healthy controls, as the latest available evidence shows.

The secondary aim of the present study was to define the ability of platelet indices to distinguish between infections of the upper and lower urinary tract and between bacterial and viral pneumonias. Similarly to Gökçe's *et al.* analysis (22), none of the PLT parameters evaluated in our study showed statistically significant differences between the two subgroups of UTIs. This is in contrast with the hypothesis that the increase in MPV is higher in pyelonephritis, more susceptible to complications (such as bacteremia) (23). To our knowledge, no study has ever assessed platelet indices modifications in pneumonias, far less in the pediatric population. Although our results documented lower PLT, MPV and PLT/MPV values in patients with bacterial pneumonias, no statistically significant differences were found compared to viral forms.

Recent studies have evaluated platelet indices' alterations in pediatric bacterial infections, comparing them to healthy controls. A significant novelty and main strength of the present study is being the first to investigate variations in platelet indices among febrile infants, comparing bacterial to viral diseases. Nevertheless, several limitations should be addressed. Due to its retrospective nature, a prognostic evaluation could not be performed. Furthermore, bacterial and viral pneumonias were differentiated according to laboratory parameters (leukocytes, CRP, serology) and thoracic imaging, without isolating the specific etiological agent. Although the disproportion in the sample size of the two comparison groups is considerable (59 bacterial, 177 viral), it reflects the distribution of admissions in our emergency department. Moreover, the homogeneity of the recruited sample, limited to the first year of life, prevents us from generalizing our conclusions to the entire pediatric population.

CONCLUSIONS

Our results demonstrate the substantial inefficacy and inaccuracy of platelet indices in discriminating the microbiological etiology of an infectious process during the first year of life. It seems more useful

and pragmatic, especially in the emergency setting, to use widely validated markers, such as leukocytosis, neutrophilia and CRP, integrating them with the clinical picture, which always represents the cornerstone of the most appropriate clinical practice.

COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interests

The authors have no conflicts of interest relevant to this article to disclose.

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Authorship

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Author contributions

The authors confirm contribution to the paper as follows. Study conception and design: FV and FM; data collection: FV and GDM; analysis and interpretation of results: FV and EB; draft manuscript preparation: FV and LM; critical revision: RN and LP; supervision: FM. All authors reviewed the results and approved the final version of the manuscript.

Consent to participate

N/A

Ethical approval

The study was approved by the research and ethics committee of the Policlinico Umberto I Hospital (n. 2377/2012).

Human studies and subjects

N/A.

Animal studies

N/A.

Data sharing and data accessibility

N/A.

Publication ethics

Plagiarism

N/A.

Data falsification and fabrication

N/A.

Manipulation of images

N/A.

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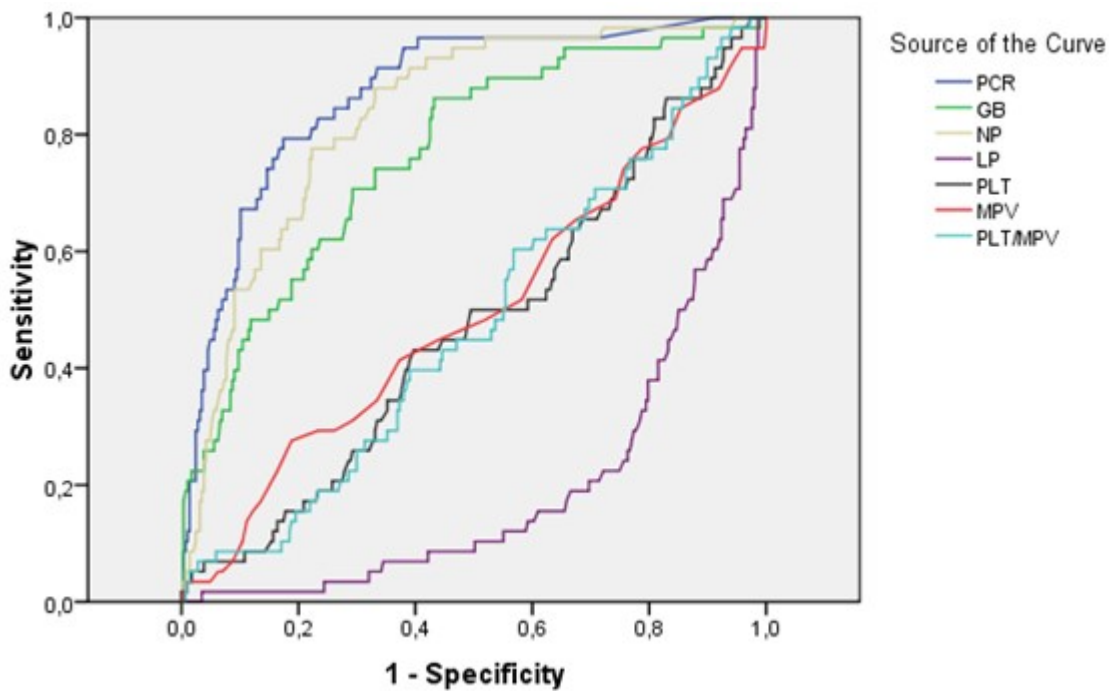


Figure 1. ROC curves of traditional markers and platelet indices.

Table 1. PLT indices: comparison between bacterial and viral infections.

	Bacterial infections (n = 59)	Viral infections (n = 177)	p-value
PLT	451084 ±137807	454192 ±138701	0.88
MPV	7.9 (6.1-10.9)	7.9 (6.4-10.2)	0.93
PLT/MPV	57291 ±18909	58087 ±19453	0.78

Normally distributed quantitative variables (PLT, PLT/MPV) are expressed as average ± standard deviation. Quantitative variables with no-normal distribution (MPV) are expressed as median (range). Abbreviations: PLT Platelets; MPV Mean Platelet Volume.

Table 2. ROC analysis of traditional markers and platelet indices.

	AUC	Standard Error	Lower Bound	Upper Bound
WBC	0.767	0.034	0.700	0.834
NP	0.832	0.027	0.779	0.886
LP	0.197	0.030	0.137	0.256
CRP	0.871	0.025	0.822	0.920
PLT	0.479	0.041	0.399	0.559
MPV	0.499	0.043	0.414	0.584
PLT/MPV	0.481	0.040	0.402	0.559

Abbreviations: AUC Area Under the Curve; WBC White Blood Cell; NP Neutrophil Percentage; LP Lymphocyte percentage; CRP C-Reactive Protein; PLT Platelets; MPV Mean Platelet Volume.

Table 3. PLT indices: comparison between upper and lower UTIs.

	Lower UTIs (n = 28)	Upper UTIs (n = 13)	p-value
PLT	437607 ±127260	422000 ±109982	0.71
MPV	7.9 (6.1-10.9)	8 (6.4-10.3)	0.81
PLT/MPV	55556 ±17155	54789 ±19451	0.90

Normally distributed quantitative variables (PLT, PLT/MPV) are expressed as average ± standard deviation. Quantitative variables with no-normal distribution (MPV) are expressed as median (range). Abbreviations: PLT Platelets; MPV Mean Platelet Volume.

Table 4. PLT indices: comparison between bacterial and viral pneumonias.

	Bacterial pneumonias (n = 18)	Viral pneumonias (n = 11)	p-value
PLT	493055 ±166495	540000 ±178266	0.48
MPV	7.8 (6.8-9.9)	8.0 (7.1-9.8)	0.41
PLT/MPV	61797 ±21311	65535 ±21347	0.65

Normally distributed quantitative variables (PLT, PLT/MPV) are expressed as average ± standard deviation. Quantitative variables with no-normal distribution (MPV) are expressed as median (range). Abbreviations: PLT Platelets; MPV Mean Platelet Volume.

Table 5. Pearson's correlation coefficient.

	WBC		ANC		CRP	
PLT	0.19815	$p < 0.05$	0.23814	$p < 0.05$	0.00026	$p = 0.99$
MPV	0.00496	$p = 0.93$	-0.08395	$p = 0.12$	-0.03932	$p = 0.47$
PLT/MPV	0.17568	$p < 0.05$	0.23923	$p < 0.05$	0.01102	$p = 0.84$

Abbreviations: WBC White Blood Cells; ANC Absolute Neutrophil Count; CRP C-Reactive protein; PLT Platelets; MPV Mean Platelet Volume.