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Pseudoeosinophilia Associated with Blue-Green Neutrophilic Inclusions: An Eight-Case Series with Poor Prognostic Significance

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ABSTRACT

Background: Pseudoeosinophilia is an uncommon analytical phenomenon in which automated hematology analyzers report increased eosinophil counts despite the absence of eosinophilia on peripheral blood smear examination. Blue-green cytoplasmic inclusions in neutrophils and monocytes are rare morphological findings associated with severe systemic illness and poor clinical outcomes.
Aim: To describe the clinicopathological characteristics, hematological findings, analyzer abnormalities, and prognostic implications of pseudoeosinophilia associated with blue-green cytoplasmic inclusions.
Materials and Methods: A retrospective case series was conducted on eight critically ill patients who demonstrated pseudoeosinophilia on an automated hematology analyzer along with abnormal white blood cell scattergram patterns. Clinical data, biochemical parameters, complete blood counts, analyzer findings, and peripheral blood smear features were reviewed.
Results: The patients ranged in age from 37 to 73 years and presented with diverse underlying conditions, including malignancy, severe sepsis, renal failure, hepatic dysfunction, dengue infection, acute pancreatitis, and post-transplant immunosuppression. Automated hematology analysis reported eosinophil counts ranging from 7.0% to 25.0%. Peripheral blood smear examination consistently failed to demonstrate true eosinophilia and instead revealed toxic neutrophilic changes with characteristic blue-green cytoplasmic inclusions in neutrophils and monocytes. Most patients showed elevated liver enzymes, increased lactate and lactate dehydrogenase levels, metabolic acidosis, thrombocytopenia, and evidence of multiorgan dysfunction. Six of the eight patients died despite intensive supportive treatment.
Conclusion: Pseudoeosinophilia associated with blue-green cytoplasmic inclusions represented an important diagnostic pitfall in critically ill patients. Recognition of characteristic scattergram abnormalities and careful peripheral blood smear review prevented misinterpretation of eosinophil counts and served as an early indicator of severe systemic injury and adverse clinical outcome.

Keywords: Pseudoeosinophilia; Blue-green cytoplasmic inclusions; Neutrophils; Automated hematology analyzer; Peripheral blood smear; Critical illness.

INTRODUCTION

Automated hematology analyzers have substantially improved the efficiency and accuracy of leukocyte differential counting. Nevertheless, abnormal cellular morphology may occasionally interfere with automated classification algorithms, resulting in erroneous leukocyte population identification. One such phenomenon is pseudoeosinophilia, in which automated analyzers report elevated eosinophil counts despite the absence of eosinophilia on peripheral blood smear review. Alterations in cellular volume, internal complexity, conductivity, and light-scatter characteristics caused by toxic leukocytic changes can lead to misclassification of neutrophils or immature granulocytic forms as eosinophils, generating high eosinophil counts. [1,2]

Blue-green cytoplasmic inclusions within neutrophils and monocytes represent an uncommon but noteworthy hematologic finding encountered predominantly in critically ill patients.[3] Since their initial description, these inclusions have been reported in association with severe systemic disorders including septic shock, acute liver injury, lactic acidosis, acute pancreatitis, and multiorgan dysfunction syndrome.[3–6] Owing to their frequent association with rapidly progressive clinical decline, they have attracted considerable attention as potential indicators of adverse prognosis. [4,5]

The precise origin of these inclusions has not yet been fully established. Available evidence suggests that they may consist of lipofuscin-like pigments or lipid-rich cellular debris released during extensive tissue destruction and subsequently phagocytosed by circulating leukocytes. [4,6] Histopathologic studies have demonstrated morphological similarities between these inclusions and pigments identified in necrotic hepatocytes. On peripheral blood smear examination, they typically appear as refractile blue-green coarse cytoplasmic granules within neutrophils and, less commonly, monocytes. [3,4]

Peripheral blood smears from patients with severe infections commonly demonstrate toxic neutrophilic alterations such as toxic granulation, Döhle bodies, cytoplasmic vacuolization, and a left-shifted granulocytic series. In contrast, blue-green inclusions are encountered far less frequently and are generally associated with profound metabolic disturbances and advanced organ dysfunction.[5] Recent reports have documented their occurrence in patients with severe coronavirus disease 2019 (COVID-19), fulminant hepatic failure, and acute pancreatitis, further reinforcing their association with critical illness and high short-term mortality. [6–8]

Eosinophils constitute a small proportion of circulating leukocytes, typically accounting for less than 7% of peripheral blood white cells. Beyond their established role in defense against parasitic infections, eosinophils participate in a broad range of immunological processes, including modulation of inflammatory responses, maintenance of tissue homeostasis, wound healing, and antitumor immunity. [9,10] Despite extensive research on eosinophilia and its clinical implications, decreased eosinophil counts have received comparatively less attention in routine diagnostic practice.

Emerging evidence suggests that eosinopenia, commonly defined as an absolute eosinophil count below 50 cells/mm^3, may serve as a useful biomarker of acute bacterial infection and systemic inflammatory stress.[11,12] Because eosinophil counts can decline rapidly in response to endogenous corticosteroid release and inflammatory cytokines, eosinopenia has been investigated as an adjunctive marker for distinguishing infectious from noninfectious causes of inflammation. During the COVID-19 pandemic, several studies further demonstrated an association between persistent eosinopenia and severe disease manifestations, increased need for intensive

care support, and adverse clinical outcomes. [13,14]

In the present study, we describe an unusual phenomenon of analyzer-generated "pseudoeosinophilia," wherein automated differential counts reported elevated eosinophil percentages despite the absence of eosinophilia on peripheral blood smear examination. Morphologic review instead revealed marked eosinopenia, highlighting a discrepancy between automated and microscopic leukocyte assessment.

Analyzer-related pseudoeosinophilia has previously been documented in patients with Plasmodium falciparum malaria, where abnormal leukocyte scatter characteristics resulted in erroneous eosinophil enumeration. The six-part differential system of the Sysmex XN-1000 employs flow-cytometric analysis using semiconductor laser technology to characterize leukocyte populations. Cellular identification is based on multiple optical parameters, including forward-scattered light, which reflects cell size; side-scattered light, which correlates with intracellular complexity; and side fluorescence intensity, which represents nucleic acid content. Alterations in these characteristics secondary to toxic morphological changes may disrupt normal cell clustering patterns and lead to misclassification of leukocyte subsets, thereby generating spuriously elevated eosinophil counts. [15–18]

In this case series, we describe eight patients demonstrating pseudoeosinophilia on DxH 900 hematology analyzer associated with blue-green cytoplasmic inclusions in neutrophils and monocytes. Most patients had severe sepsis, hepatic dysfunction, metabolic acidosis, or multiorgan failure, and the majority showed rapid clinical deterioration with fatal outcomes.

MATERIALS & METHODS

This retrospective case series included eight cases identified during routine hemogram reporting. Cases showing pseudoeosinophilia on the hematology analyzer were selected when peripheral blood smear review revealed characteristic blue-green cytoplasmic inclusions in neutrophils and monocytes in critically ill patients.

In this study, the clinical details, biochemical laboratory results, automated hematology analyzer findings, and peripheral smear morphology were analysed. Haematological analysis was performed using the Beckman Coulter DxH 900 automated hematology analyzer.

The study population included patients aged 37–73 years with severe systemic illnesses including septic shock, malignancy, post-transplant complications, acute pancreatitis, dengue infection, liver dysfunction, renal failure, and multiorgan dysfunction syndrome.

Cases showing apparent eosinophilia on analyzer differential counts without corresponding eosinophilia on peripheral smear examination were categorized as pseudoeosinophilia.

The blood films were stained with Leishman stain and observed for toxic neutrophilic granulation, cytoplasmic vacuolization, left shift, nucleated red blood cells, and blue-green cytoplasmic inclusions in neutrophils and monocytes.

RESULT

The study included five males and three females. Common clinical presentations included septic shock, respiratory failure, metabolic acidosis, acute kidney injury, hepatic dysfunction, and multiorgan failure.

Automated hematology analyzer demonstrated eosinophils ranging from 1.1% to 25%. However, peripheral smear examination consistently failed to demonstrate eosinophilia. Although the analyzer reported eosinophilia in seven cases, one case showed an eosinophil count of only 1.1%, despite the absence of eosinophils on peripheral blood smear examination.

Instead, all cases showed toxic neutrophilic changes with blue-green cytoplasmic inclusions. One case showed inclusions in monocytes.

Common haematological findings observed in these cases are-

RBC (Red Blood Cells) findings- Anemia with most cases have high MCV (Mean Corpuscular Volume) and nucleated red blood cells.

Platelets findings- Thrombocytopenia

WBC (White Blood Cells) findings- Neutrophilic leucocytosis, left shift, cytoplasmic toxic granules and vacuoles, blue-green inclusions. One case showed inclusions in monocytes.

Common biochemical laboratory findings include- elevated AST/ALT (Aspartate Aminotransferase /Alanine Aminotransferase), elevated LDH (Lactate Dehydrogenase), hyperlactatemia, metabolic acidosis, renal dysfunction, and coagulopathy.

The haematological and biochemical findings in these cases were shown in table 1.

In true eosinophilia, the histogram demonstrates a distinct, narrow, and steep peak located in the middle region, corresponding to monocytes, eosinophils, and basophils. This is accompanied by a broad, well-defined peak in the neutrophil region, reflecting clear separation of leukocyte populations. The scatterplot in true eosinophilia demonstrated a well-defined and discrete eosinophil cluster distinctly separated from the neutrophil population. [Figure 1A and 1B]

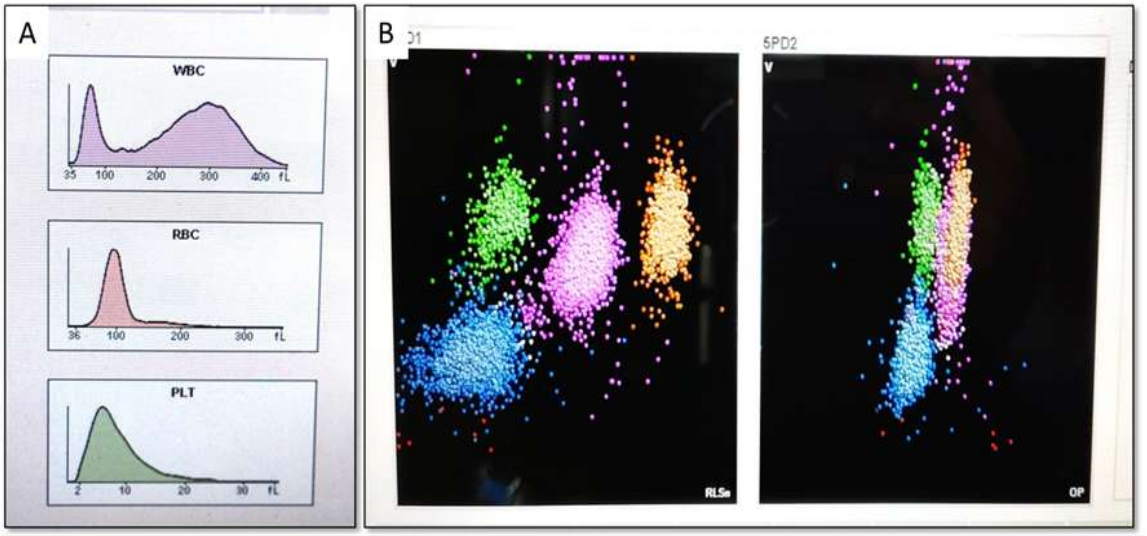

Figure 1A: Histogram of a true eosinophilia case showing a distinct, narrow, and steep peak in the mid region, corresponding to monocytes, eosinophils, and basophils. This is accompanied by a broad, well-defined peak in the neutrophil region.
Figure 1B: Scatterplot of a true eosinophilia case showing orange-colored eosinophils forming a distinct cluster separate from the purple-colored neutrophils.

In contrast, pseudoeosinophilia shows poor differentiation of leukocytes. Instead of distinct peaks, the histogram displays a dome-like or hump-shaped elevation in the neutrophil region, beginning from the mid-zone, without the sharp segregation seen in true eosinophilia. The scatterplot in pseudoeosinophilia showed abnormal overlap and partial merging of neutrophil and eosinophil populations, resulting in erroneous classification of neutrophils as eosinophils by the analyzer. The abnormal neutrophil population displayed increased side scatter, likely related to toxic changes and blue-green cytoplasmic inclusions, producing an elevated eosinophil count on automated analyzer. [Figure 2A and 2B]

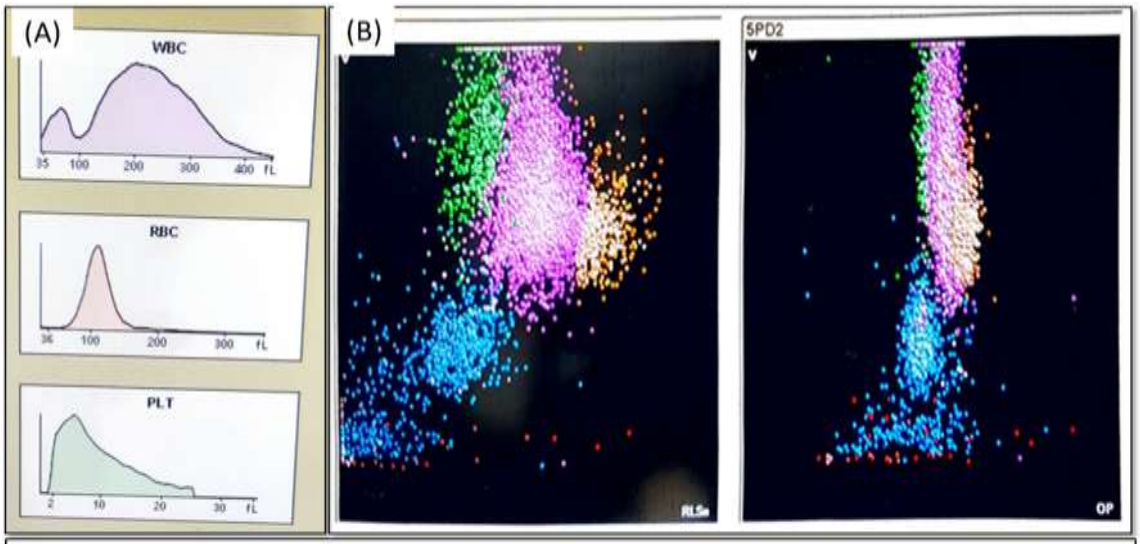

Figure: 2(A): Histogram of a pseudoeosinophilia case showing an indistinct large hump extending from the mid region to the neutrophil region.
Figure: 2(B): Scatterplot of a pseudoeosinophilia case showing orange-colored eosinophils close to purple-colored neutrophils, with no distinct eosinophil cluster.

Peripheral blood smear examination revealed neutrophils showing coarse blue-green cytoplasmic inclusions/granules with associated toxic changes including toxic granulation and cytoplasmic vacuolization. The inclusions appeared refractile, irregular, and variably sized, predominantly within neutrophils and occasional monocytes. Background findings included neutrophilic leucocytosis with left shift and marked thrombocytopenia. No true eosinophilia was identified on microscopy despite eosinophilia on automated hematology analyzer. [Figure 3A and 3B]

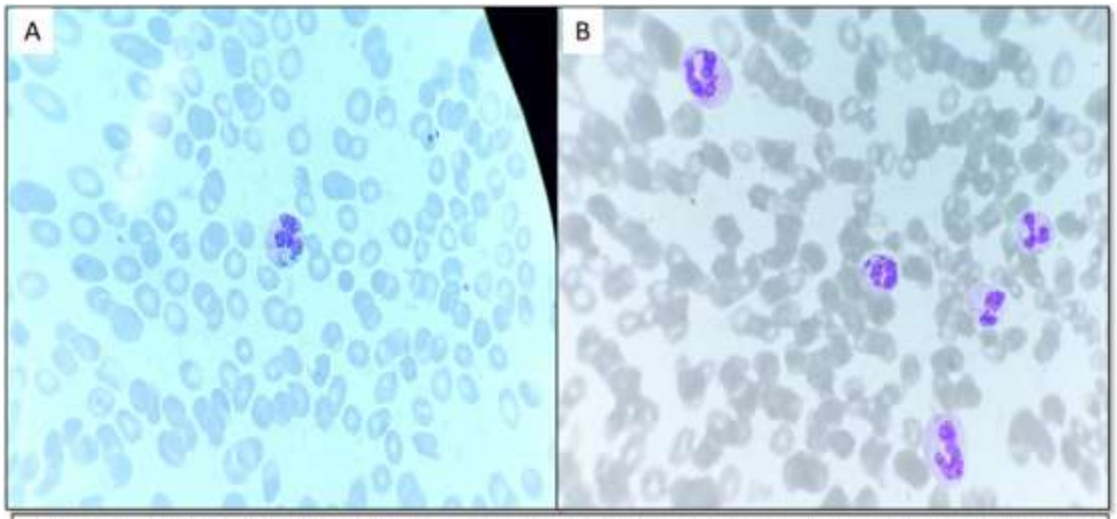

Figure 3(A) and 3(B):Neutrophils showing large green-blue cytoplasmic granules on Leishman-stained peripheral blood smears.(Leishman stain, 1000x)

Most patients presented with severe sepsis or septic shock (87.5%) and multiorgan dysfunction. Thrombocytopenia and blue-green neutrophilic inclusions were observed in all cases (100%). Eosinophilia on analyzer was present in seven patients despite absence of eosinophilia on peripheral smear examination. One case showed 1.1% eosinophils on analyzer, however peripheral blood smear showed no eosinophils. Hepatic dysfunction and renal impairment were seen in 75% of cases each,

while metabolic acidosis was present in 87.5% of patients. Mortality was high, with six out of eight patients (75%) expiring during hospitalization. [Table 1, 2 and 3]

Most patients showed rapid clinical deterioration with progression to septic shock, metabolic acidosis, hepatic dysfunction, and multiorgan failure shortly before death. Six of the eight patients (75%) expired within a short interval (2-3 days) after these pseudoeosinophilia and peripheral smear findings were identified, while two patients improved with treatment and were subsequently discharged.

Table 1. Clinical profile and hematological–biochemical parameters of patients showing Pseudoeosinophilia associated with blue-green neutrophilic inclusions

Case	Hemoglobin (g/dL)	WBC Count (/µL)	Neutrophils (%)	Analyzer Eosinophils (%)	Platelet Count (/µL)	AST (IU/L)	ALT (IU/L)	Bilirubin (mg/dL)
1	9.6	13,400	73.4	7.5	60,000	3816	1187	Total 1.0, Direct 0.7
2	6.6	20,500	91.5	1.1	12,000	1191	561	Total 2.8, Direct 1.6
3	9.7	49,100	82	25	14,000	Not done	Not done	Not done
4	4.8	28,800	77	15	22,000	4689	733	Total 1.9, Direct 1.8
5	7.6	17,300	88	7	70,000	30	16	Total 1.1, Direct 0.5
6	10.6	20,200	80	9	26,000	174	69	Total 5.8, Direct 4.8
7	8.6	31,400	66	18	32,000	21,211	3673	Total 9.8, Direct 6.6
8	8.8	36,300	71	20	33,000	1409	371	Total 5.3, Direct 3.9

Table 2. Demographic characteristics, clinical diagnoses, and outcomes of the study cases

Case No.	Age (Years)/Sex	Clinical Diagnosis	Outcome
1	37/M	Acute necrotizing pancreatitis with chronic kidney disease post renal transplantation	Expired
2	73/M	Secondary acute myeloid leukemia post-transplant with septicemia	Expired
3	73/F	Metastatic lung carcinoma with septic shock	Expired
4	67/F	Hepatocellular carcinoma with severe sepsis	Expired
5	71/F	Urosepsis with acute kidney injury	Discharged
6	69/M	Metastatic colon carcinoma with septic shock	Expired
7	64/M	Severe dengue with multiple organ dysfunction syndrome	Expired
8	59/M	Urosepsis with cirrhotic liver disease	Discharged

Table 3. Correlation of automated analyzer eosinophil counts with peripheral blood smear findings in patients with Pseudoeosinophilia

Case No.	Analyzer Eosinophils (%)	Peripheral Smear Eosinophils (%)	Blue-Green Inclusions Present	Interpretation
1	7.5	0	Yes	Pseudoeosinophilia
2	1.1	0	Yes	Pseudoeosinophilia
3	25	1	Yes	Pseudoeosinophilia
4	15	0	Yes	Pseudoeosinophilia
5	7	0	Yes	Pseudoeosinophilia
6	9	0	Yes	Pseudoeosinophilia
7	18	0	Yes	Pseudoeosinophilia
8	20	1	Yes	Pseudoeosinophilia

DISCUSSION

Blue-green cytoplasmic inclusions within neutrophils and monocytes are uncommon peripheral blood smear findings that have gained attention because of their strong association with severe systemic illness and adverse clinical outcomes. [3–8] In the current case series, all eight patients exhibited analyzer-generated pseudoeosinophilia on the DxH 900 hematology system, accompanied by abnormal leukocyte histogram and scattergram patterns. Microscopic examination consistently revealed blue-green cytoplasmic inclusions within neutrophils and/or monocytes. Clinically, the majority of patients presented with advanced disease characterized by septic shock, metabolic derangements, hepatic injury, acute kidney dysfunction, or multiorgan failure.

The biological basis of these inclusions has not been fully elucidated. Available evidence suggests that they may represent intracellular accumulation of lipofuscin-like material or debris derived from extensive tissue destruction, particularly following hepatocellular injury. [4,6] Uptake of such material by circulating phagocytic cells may account for the characteristic cytoplasmic granules observed on peripheral smear examination. The reported association of these inclusions with severe liver dysfunction, lactic acidosis, and widespread tissue injury lends support to this proposed mechanism. Consistent with previous reports, several patients in our series demonstrated markedly abnormal liver function tests, elevated transaminase levels, septicemia-related organ dysfunction, and profound metabolic acidosis, highlighting the close relationship between these inclusions and critical systemic injury. [5–8]

A notable observation in this series was pseudoeosinophilia detected on the DxH 900 hematology analyzer. To our knowledge, this is the first study to demonstrate the utility of hematology analyzer histograms and scatterplots in distinguishing true eosinophilia from pseudoeosinophilia caused by blue-green cytoplasmic inclusions in neutrophils and monocytes, as confirmed on peripheral blood smear review.

Automated differential counts showed elevated eosinophil percentages ranging from 1.1% to 25%, whereas manual peripheral smear examination did not reveal eosinophilia. Instead, neutrophils containing toxic granules, vacuoles, immature forms, and blue-green inclusions were identified. Similar pseudoeosinophilia has been described in malaria and inflammatory states with abnormal leukocyte morphology affecting analyzer scatter characteristics. [15,16]

The Beckman Coulter DxH 900 hematology analyzer utilizes Volume, Conductivity, and multi-angle Light Scatter (VCS) technology for leukocyte differential analysis. Each white blood cell is characterized by measurements of cell size, internal composition, nuclear-cytoplasmic characteristics, granularity, and light-scattering properties. These multiparametric data are processed by clustering algorithms to generate leukocyte scatterplots and differential counts.[19] In cases of pseudoeosinophilia associated with blue-green cytoplasmic inclusions in neutrophils, altered cellular optical and conductivity properties may shift neutrophils toward the eosinophil region on the scatterplot, resulting in elevated eosinophil counts despite the absence of eosinophilia on peripheral blood smear examination.

Characteristic analyzer abnormalities were consistently observed in these patients. WBC histograms demonstrated abnormal dome-shaped elevations in the neutrophilic region starting from mid region, while scatter grams showed eosinophil clusters merging with neutrophilic populations rather than forming distinct eosinophil clusters. These findings correlated with peripheral smear morphology and may serve as an important clue prompting manual smear review.

Most patients in the present study demonstrated additional hematologic

abnormalities including neutrophilic leucocytosis, toxic granulation, cytoplasmic vacuolization, left shift, thrombocytopenia, and nucleated red blood cells. These findings reflect severe systemic inflammatory response, marrow stress, tissue hypoxia, and consumptive coagulopathy commonly encountered in critically ill patients. [17,20-22]

Accumulating evidence suggests that identification of blue-green cytoplasmic inclusions serves as an ominous laboratory finding, frequently accompanying advanced organ dysfunction and a high short-term mortality rate, often within 24–72 hours of recognition. [5,6] Consistent with these observations, six of eight patients in the present study succumbed shortly after appearance of these inclusions, supporting their role as an ominous prognostic marker.

Although pseudoeosinophilia on analyzer and neutrophilic these inclusions are rare, awareness among clinical and intensivists is essential. Peripheral smear examination remains indispensable in critically ill patients showing abnormal scatter grams or unexplained pseudoeosinophilia. Prompt recognition and communication with treating clinicians may help identify patients at high risk of rapid deterioration.

This series highlights the diagnostic importance of recognizing pseudoeosinophilia associated with blue-green cytoplasmic inclusions and correlating analyzer findings with peripheral smear morphology.

CONCLUSION

Pseudoeosinophilia associated with blue-green neutrophilic and monocytic inclusions is an important haematological phenomenon encountered in critically ill patients with severe systemic inflammation, septic shock, hepatic dysfunction, and multiorgan failure. Automated hematology analyzers may classify abnormal neutrophils as eosinophils because of altered cytoplasmic scatter characteristics. Recognition of abnormal WBC scatter grams together with careful peripheral smear examination is essential to avoid misinterpretation and to identify patients with poor prognostic indicators.

Declaration by Authors

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