“EFFICACY OF PLATELET AND RETICULOCYTE PARAMETERS FOR EARLY DIAGNOSIS OF SEPSIS IN INTENSIVE CARE UNIT PATIENTS”

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Dissertation submitted to BLDE (Deemed to be University), Vijayapura.

In partial fulfilment of the requirements for the award of the degree of

DOCTOR OF MEDICINE

IN

PATHOLOGY

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2019

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LIST OF ABBREVIATIONS

ABBREVIATION	PARAMETER
Hb	Hemoglobin
RBC	Red blood cell
WBC	White Blood Cell
IPF%	Immature Platelet Fraction
RET%	Reticulocyte Percentage
ICU	Intensive Care Unit
DLC	Differential Leucocyte Count
IG	Immature Granulocytes
nRBC	Nucleated RBC
RNA	Ribonucleic Acid
DNA	Deoxyribonucleic acid
EDTA	Ethylene Diamine Tetraacetic Acid
PLT	Platelet
SD	Standard Deviation
PPV	Positive Predictive Value
NPV	Negative Predictive Value
MAP	Mean Arterial Pressure
GCS	Glasgow Coma Scale
SOFA	Sequential Organ failure Assessment
qSOFA	Quick SOFA

ABSTRACT

INTRODUCTION

Sepsis is a clinical condition characterized by a strong inflammatory response to infections and is associated with a high mortality rate. Early diagnosis of sepsis and the establishment of appropriate and timely treatment may both considerably improve the outcome. Emerging parameters include, Immature Platelet Fraction and Reticulocyte Count.

OBJECTIVES

To analyze the role of Immature Platelet Fraction (IPF%) and Reticulocyte percentage (RET%) for early diagnosis of sepsis & to correlate these with blood culture sensitivity test in adults during Intensive care unit (ICU) stay.

METHODS

A hospital based observational study was carried out on of 138 patients, aged >18years, who were admitted to the ICU, for more than 48hours. Baseline data- Complete Blood Count (CBC), IPF% and RET% were repeated along with blood culture sensitivity for confirmation and association of sepsis with IPF% and RET%.

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RESULTS

Out of the 138 patients, 120 patients had increased IPF%, 90 patients had decreased RET% and 39 patients showed positive blood cultures. There was a statistically significant association between IPF% and Blood culture positivity (p = <0.001) and between RET% and Blood culture positivity (p= <0.001).

CONCLUSION

The combination of both IPF% and RET% values in ICU patients may be considered as an early, rapid, inexpensive and widely available measure of sepsis allowing more efficient and timely patient management.

KEYWORDS

Sepsis, Immature platelet fraction, Reticulocyte, Blood Culture.

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“EFFICACY OF PLATELET AND RETICULOCYTE PARAMETERS IN EARLY DIAGNOSIS OF SEPSIS IN INTENSIVE CARE UNIT PATIENTS”

INTRODUCTION

Sepsis is a clinical condition characterized by a strong inflammatory response to infections and is associated with a high mortality rate.[1] Sepsis represents a substantial health care burden, and there is limited epidemiologic information about the demography of sepsis or about the changes in its incidence and outcome.[2] Evidence has been provided that nearly one-third of patients die after admission to intensive care unit (ICU).[1-3] The incidence of sepsis has considerably increased in recent years due to immunosuppressive states like diabetes mellitus, chronic kidney disease, retro positive status, use of immunosuppressive drugs and a growing number of surgical procedures.[1] Severe sepsis occurs as a occurs most commonly because of infections acquired from the community and infections which are acquired from hospitals. [1-3] Pneumonia is the most common cause, accounting for about half of all cases, followed by intra-abdominal and urinary tract infections. Blood cultures are essentially positive in only 1/3rd of cases. The most common gram positive organisms are Staphylococcus aureus and Streptococcus pneumoniae, and the most common gram negative organisms are Klebsiella species, Escherichia coli, and Pseudomonas aeruginosa. Therefore, early diagnosis of sepsis and the establishing an appropriate and in-time treatment may considerably improve the outcome. [3]

There is great interest about innovation in hematological parameters used in sepsis, which can also be used as diagnostic biomarkers and patient monitoring. [1]

These modern hematological analyzers offer a great spectrum of the usual traditional and the latest innovative parameters besides the ones included in the routine CBC (complete blood count) and TC/DC (Total and differential leucocyte count). [1,3] The new emerging parameters like, IG's (Immature Granulocytes), RET% (Reticulocyte count) and IPF% (Immature platelet fraction) are expressed both in absolute values and percent.[4] These are majorly being used due to the relatively low cost, widespread accessibility/applicability and very short turnaround time. [1]

IPF% assessment provides ample information for diagnosis and follow-up of patients with sepsis. [3] It is noted that, IPF% correlates with positive of blood cultures, and tends to increase before the onset of sepsis being the only parameter whose values vary independently as those of conventional coagulation tests. [1]

It is reported that risk of developing ICU sepsis reduces by 68% for each unit rise in RET%. [3] Therefore, reticulocyte percent (RET%) provides meaningful clinical information for depicting the risk of ICU sepsis.[1]

Therefore, this study aims to exploit the interactions between Platelet parameters and Reticulocyte parameters with the risk of developing sepsis. [5]

OBJECTIVES OF THE STUDY

To analyze the role of Platelet (PLT) and Reticulocyte (RET%) parameters, for early diagnosis of sepsis & to correlate these parameters with blood culture sensitivity test in adult patients during intensive care unit (ICU) stay.

REVIEW OF LITERATURE

HISTORY

Sepsis is one of the oldest and most evasive syndromes in medicine [8]. About 2 millennia ago, Hippocrates addressed sepsis as rotting flesh and festering wounds. [1] Hundreds of years later, Galen described sepsis as an important part of wound healing [1,8]. When Semmelweis, Pasteur and others proposed the Germ Theory in the 19th century, sepsis was again redefined as a generalized systemic infection, and they named it 'Blood poisoning'. [1] It was believed to be caused due to invasion of pathogen and its spread in the host's bloodstream. [1] However many patients suffering from sepsis died, in spite of successfully removing of the causative pathogen, thereby not fully complying with the germ theory. [1,2]

Bone and colleagues, in 1992 put forward that, it is the not the germ but the host, which is responsible for the causation of sepsis. [1] Thus, Sepsis was defined as a Systemic Inflammatory Response to infection [1,2].

As many pathogens contributed in the pathogenesis of Sepsis, the term severe sepsis was coined to describe cases where sepsis was complicated by an acute organ dysfunction and the term septic shock was coined for a subset of sepsis cases that were further complicated by low blood pressure (despite adequate fluid resuscitation) and perfusion abnormalities [1,2].

DEFINITION

The description of sepsis has undergone many improvisations, still, with a lack of complete agreement as to what the exact wording would best reflect this complex, multisystem process. [4]

Pre requisites for Sepsis are- Presence of an infection resulting in a systemic inflammatory state and organ dysfunction (as a result of infection and inflammation). [2].

There have been varied definitions for severe sepsis. [1,114] The American College of Chest Physicians/Society of Critical Care Medicine (ACCP/SCCM) consensus conference of 1992 had distinguished between the systemic inflammatory response syndrome (SIRS), sepsis, and severe sepsis [1,14].

ACCP/SCCM Consensus Conference Definitions of the Systemic Inflammatory Response Syndrome (SIRS), Sepsis and Severe Sepsis [14] are-

SYSTEMIC INFLAMMATORY RESPONSE SYNDROME (SIRS)

SIRS includes, but is not limited to, more than one of the following:

1. Temperature- More than 38 degree Celsius OR less than 36 degree Celsius.

2. Heart rate- More than 90 beats per minute.

3. Respiratory rate- More than 20 breaths per minute OR Partial pressure of Carbon Dioxide of less than 32 mm Hg.

4. WBC count- More than 12000 cells per mm^3 OR less than 4,000 cells per mm^3 OR more than 10% immature granulocytes.

These changes represent an acute alteration from baseline in the absence of other known causes for such abnormalities. [1]

SEPSIS

An infectious process which results in systemic inflammatory response syndrome (SIRS) is termed Sepsis [14].

SEVERE SEPSIS

Sepsis is with super added organ dysfunction, hypo perfusion, or hypotension is termed Severe Sepsis. [14] Lactic acidosis, oliguria, or an acute alteration in mental status comprise the criteria for hypo perfusion and perfusion abnormalities [5,14].

CONDITION	DEFINITION	COMMON CLINICAL FEATURES	CRITERIA IN 1991/2003 ("SEPSIS-1"/"SEPSIS-2")	CRITERIA IN 2016 ("SEPSIS-3")
Sepsis	A life-threatening organ dysfunction caused by a dysregulated host response to infection. [1]	Include signs of infection, with organ dysfunction, plus altered mentation; tachypnea; hypotension; hepatic, renal, or hematologic dysfunction. [1]	Suspected (or documented) infection plus ≥2 systemic inflammatory response syndrome (SIRS) criteria. [1]	Suspected (or documented) infection and an acute increase in ≥2 sepsis-related organ failure assessment (SOFA) points. [1]
Septic shock	A subset of sepsis in which underlying circulatory and cellular/metabolic abnormalities lead to substantially increased mortality risk. [1]	Signs of infection, plus altered mentation, oliguria, cool peripheries, hyper lactemia. [1]	Suspected (or documented) infection plus persistent arterial hypotension (systolic arterial pressure, 40 mmHg from baseline. [1]	Suspected (or documented) infection plus vasopressor therapy needed to maintain mean arterial pressure at ≥65 mmHg and serum lactate >2.0 mmol/L despite adequate fluid resuscitation. [1]

Table 1- Definitions and Criteria for Sepsis and Septic shock.

To comprehend the complex details, the clinicians developed a simple bedside criteria. [1] With this ratification, the Sepsis Definitions Task Force recommended that, at any suspicion of infection, clinicians should assess the possibility of organ dysfunction by determining SOFA score. [1] SOFA score- A 24 point measure of organ dysfunction which uses 6 organ systems-

1. CNS,
2. CVS,
3. RS,
4. Hepatic,
5. Renal
6. Hematologic, where 0–4 points are assigned per organ system. [1]

The SOFA score is routinely studied in the ICU among patients with infection, sepsis, and shock. [1] With more than OR equal to TWO new SOFA points, the infected patient is considered septic and may be at more than 10% risk of in-hospital death. [1]

SOFA score requires multiple laboratory tests and is an expensive test to measure repeatedly, the "quick SOFA" (qSOFA) score was proposed as a clinical tool to identify patients at high risk of sepsis outside the ICU, either in the medical ward or in the emergency department. [1]

The qSOFA score ranges from 0 to 3 points, with 1 point each for-

1. Systolic hypotension (≤100 mmHg),
2. Tachypnea (≥22 breaths/min),
3. Altered mentation. [1]

A qSOFA score of ≥2 points has a predictive value for sepsis similar to that of more complicated measures of organ dysfunction. [1]

SYSTEM	SCORE				
	0	1	2	3	4
Respiration Pao2/Fio2, mmHg (kPa)	≥400 (53.3)	<400 (53.3)	<300 (40)	<200 (26.7) with respiratory support	<100 (13.3) with respiratory support
Coagulation Platelets, $\times 10^3$/ml	>150	<150	<100	<50	<20
Liver Bilirubin, mg/dL (μmol/L)	<1.2 (20)	1.2-1.9 (20-32)	2.0-5.9 (33-101)	6.0-11.9 (102-204)	>12.0 (204)
CVS	MAP >70 mmHg	MAP <70 mmHg	Dopamine < 5 or dobutamine (any dose)	Dopamine 5.1–15 or epinephrine <0.1	Dopamine >15 or epinephrine >0.1 or norepinephrine >0.1
CNS Glasgow Coma Scale	15	13-14	10-12	6-9	<6
Renal Creatinine, mg/dL (μmol/L)	<1.2 (110)	1.2-1.9(110-170)	2.0-3.4 (171-299)	3.5-4.9 (300-440)	>5.0 (440)
Urine output, mL/dL	-	-	-	<500	<200

[1]

Table 2- Calculation of SOFA Score

EPIDEMIOLOGY

Sepsis is a life threatening syndrome following a dysregulated host response to infection [13]. It represents a substantial health care burden, and there is limited epidemiologic information about the demography of sepsis and changes in its incidence and outcome [9]. Septic shock is a subset of sepsis in which underlying circulatory and cellular abnormalities are associated with substantially increased ICU mortality rates >40% [14]. Sepsis is one of the most common causes of multi-organ failure [2]. Sepsis is often lethal, killing 20-50% of severely affected patients. [9] It is the second leading cause of death among patients in non-coronary ICUs and the tenth leading cause of death overall in the US. Furthermore, sepsis substantially reduces the quality of life of those who survive [9].

Data shows that there is significant inconsistency among races, between males and females in the occurrence of sepsis. [9] There has been a considerable rise in the incidence of sepsis with rise in number of deaths, despite a fall in total in-hospital mortality [9].

Data demonstrates that though sepsis is a significant general public-health burden in first world countries, its impact on the populations of second and third world countries is more likely to be ravaging due to the rise in incidence of infectious diseases and the soaring prevalence of HIV in the developing world [1].

ETIOLOGY

Sepsis arises from community-acquired and health care/hospital-acquired (Nosocomial) infections. [1] Pneumonia being the most common amongst these, accounting for about 50% of cases. Intraabdominal and genitourinary infections are

amongst the lesser common causes for sepsis. [1] Blood cultures being positive in only 1/3rd of the cases, whilst rest 2/3rd are culture negative- At all sites. [1]

The most commonly associated gram positive organisms are- Staphylococcus aureus and Streptococcus pneumoniae, while most commonly associated gram-negative organisms are- Escherichia coli, Klebsiella species, and Pseudomonas aeruginosa. [1,3] Gram positive infections are being more frequently reported than gram-negative infections, in the recent years [1,3].

Risk factors for sepsis are in the dual relation to the predisposition of development of an infection and, after the development of infection, the probability of developing an acute organ dysfunction. [1] The more common risk factors for increased risk of infection include Presence of chronic diseases like- HIV, COPD (chronic obstructive pulmonary disease), carcinomas, Metabolic disorders, Cardiovascular disorders, Autoimmune diseases, and immunosuppression [1].

The process begins when infection in one part of the body triggers a localized inflammatory response. [1] In most cases, containing the inciting pathogen and a healthy and competent immune system will control the infection at this stage. [1] However, in the presence of varied factors, the infection might become systemic. [1] It is difficult to fully illustrate the causative factors, which may include-

• Genetically predisposed to sepsis,

• A large microbiological load,

• A highly virulent organism,

• A delay to control the source (surgical or antimicrobial),

• A highly resistant of the organism,

• Patient factors which include-immune status, nutrition, built, etc. [2]

INFECTION SITE AND MICROBIOLOGIC CONSIDERATIONS

The host response to infection is equally important as the infection site or the microorganism causing sepsis. [4] The lung being the commonest site of infection, is followed by abdomen and genitourinary tract. [4] In 20-30 % of patients, a precise infection site is not found, and even if suspected or found, a large proportion of patients have sterile cultures or indefinite microbiologic isolates. [4] Pleural, paranasal-sinus, and peritoneal infections can be overlooked easily, even by using CT (computed tomography). [4] It is difficult to rule out definitive infection by imaging studies. [4]

PATHOGENESIS

For a long time, the clinical features of sepsis were thought to be the result of an exaggerated inflammatory host response. [1,2] The definitive response of individual patients depends on the load and virulence of the inciting pathogen and the genetic makeup and comorbidities of the host, having varied responses at systemic and local levels. [1-3] The host response progresses over the time with the clinical course of the patient. [1] In general, the pro-inflammatory reactions which are directed to eliminate the inciting pathogen are responsible for "collateral" tissue damage in sepsis, however the anti-inflammatory responses are thought to amplify the susceptibility to secondary infections which occurs later in the clinical course. [1] These mechanisms may be described as an interaction between, direct damage caused to organs by the inciting pathogen and the damage caused to organs from the hosts immune response. [1] The

ability of the host to resist and tolerate both, the direct and immuno-pathologic damage will decide if the uncomplicated infection becomes sepsis [1].

INITIATION OF INFLAMMATION

Imbalance of inflammatory mediators imbalance constitute the most crucial basis in the pathogenesis of sepsis. [1] This occurs throughout the entire process of sepsis. [1] The inciting pathogens which elicit the response are organisms like bacteria, viruses, fungi, and parasites. [17].The immune cells are activated by the pathogen with the help of pattern recognition receptors [TLR's (Toll like receptors), C-type lectin receptors, RIG-I-like receptors, and NOD-like receptors]. [1] The recognition of PAMP's (Pathogen-associated molecular patterns) by these receptors result in the initiation of innate immunity by the upregulation of inflammatory gene transcription. [1] The lipid A moiety of lipopolysaccharide (LPS or endotoxin), is a common PAMP which is transferred to TLR4 and signals through it to produce and release cytokines like TNF (tumor necrosis factor) which grow the signal and alert other tissues and cells. [1] Injured cells release DAMP's (damage-associated molecular patterns) which are sensed by these receptors. [1,2] These inflammatory responses which are implicated in sepsis pathogenesis not only activate the pro-inflammatory cytokines but also activate the complement system, platelet-activating factor, arachidonic acid metabolites, and nitric oxide [1].

COAGULATION ABNORMALITIES

Coagulation disorders like DIC (disseminated intravascular coagulation) is a common finding with sepsis. [1-3] Coagulation abnormalities are considered to isolate inciting

pathogen and to prevent the infection/inflammation from spreading to other cells, tissues and organs. [1] via Tissue factor is a transmembrane glycoprotein which is expressed by various cells which causes excessive fibrin deposition by (i) Coagulation, (ii) Impaired anticoagulant mechanisms, like protein C system and anti-thrombin, (iii) Decreased fibrin removal because of depression of fibrinolytic system. [1] Other proteases and coagulation further intensify inflammation via protease-activated receptors. [1] In infections which have endothelial predominance (meningococcemia), these mechanisms are more common and fatal. [1-3]

ORGAN DYSFUNCTION

Though the mechanisms that cause organ failure in sepsis are not fully known, defective tissue oxygenation plays a vital role. [1] Many factors are responsible for the reduction in oxygen delivery to the tissues and cells in sepsis and septic shock. [1]

These are-

1. Hypotension,
2. Reduction in red-cell deformability,
3. Microvascular thrombosis.

Inflammation causes dysfunction in the vascular endothelium, alongside cell death and loss in barrier integrity, which gives rise to sub-cutaneous and body cavity edema. [1] Excessive release of NO (nitric oxide) causes vasomotor collapse, arteriovenous shunts opening, and shunting of oxygenated blood from susceptible tissues. [1-3] Moreover, there is mitochondrial damage caused by oxidative stress and other mechanisms which impair cellular oxygen utilization. [1] The reduced rate of oxidative metabolism, along

with impaired oxygen delivery, decreases cellular oxygen extraction. [1] Energy (i.e., ATP) is derived from glycolysis and fermentation to support basal, vital cellular functions. [1] With continuous/severe insult, the ATP levels fall below a critical threshold and bioenergetics failure takes place, releasing toxic reactive oxygen species, and apoptosis which leads to irreversible cell death and organ failure. [1]

There are complex morphologic changes in sepsis-induced organ failure, [1] where, organs such as lungs undergo substantial microscopic changes, whereas other organs may undergo only few histologic changes. [1] Few organs, like kidney may even lack significant structural damage but still have significant tubular-cell changes which impairs their function [1].

ANTI-INFLAMMATORY MECHANISMS

The immune system comprises of humoral, cellular, and neural mechanisms which might exacerbate the harmful effects of the pro-inflammatory responses. [1] Phagocytes may switch to anti-inflammatory phenotype which promotes tissue repair, whereas regulatory T cells and myeloid-derived suppressor cells reduce inflammation. [1]

The neuro-inflammatory reflex causes relay of sensory input via the afferent vagus nerve to the brainstem, from which the efferent vagus nerve activates the splenic nerve in the celiac plexus, with consequent norepinephrine release in the spleen and acetylcholine secretion by a subset of CD4+ T cells. [1,2] The acetylcholine release targets α7 cholinergic receptors on macrophages, which reduce the pro-inflammatory cytokine release. [1]

IMMUNE SUPPRESSION

Patients who survive sepsis but still remain dependent on prolonged intensive care, sometimes show evidence of a repressed immune system. [1,2] These patients may harbor an ongoing infection in spite of specific antimicrobial therapy or might undergo the reactivation of latent infections/viruses. [1] Many studies have reported decreased response of blood leukocytes to inciting pathogens in patients with sepsis. [1] Recently, these findings were tallied by PM (Post-mortem) studies, suggesting strong impairment in function of splenocytes which were harvested from ICU patients with fatal sepsis. [1] Both, lungs and spleen show significant immune suppression, [1] by the increased expression of parenchymal cell ligands for T cell inhibitory receptors. [1-3] Enhanced apoptosis of B cell, CD4+ T cells and follicular dendritic cells, has been indicated in sepsis associated suppression of immune system and death [1].

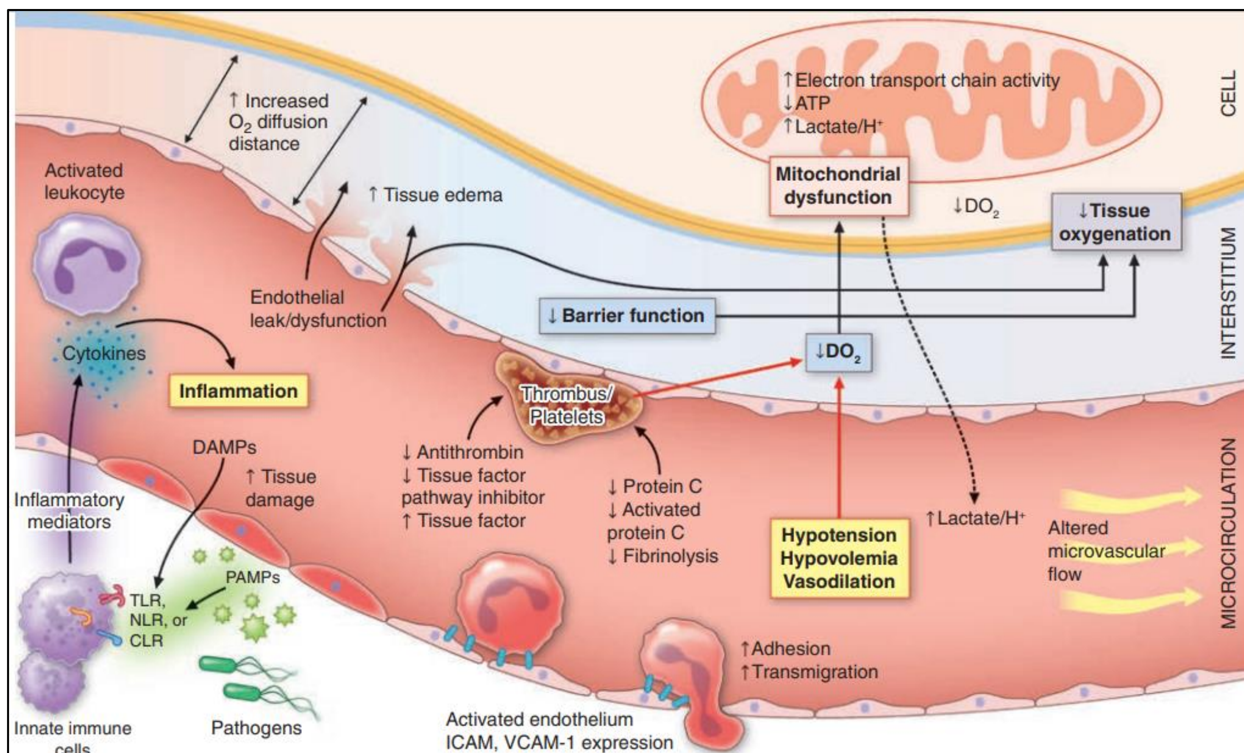


Fig.1- Mechanisms implicated in the pathogenesis of sepsis-induced organ and cellular dysfunction [1].

DIAGNOSTIC CRITERIA

There is not one specific test for sepsis, nor is there a gold-standard method for determining whether a patient is in sepsis or not. [1]

A simplified definition of sepsis can be written as a statement as-

Sepsis = f (infection ⟶ dysregulated host response ⟶ organ dysfunction ⟶ threat to life). [1]

Where sepsis is the dependent variable, which in turn is a function of four independent variables linked in a causal pathway, with one condition upon the other. [1]

MODERN HEMATOLOGY ANALYZERS AND HEMATOLOGICAL PARAMETERS

The essential components of the modern analyzers include hydraulics, pneumatics, and electrical systems. The hydraulics system includes an aspirating unit, dispensers, diluters, mixing chambers, aperture baths or flow cells or both, and a hemoglobinometer. The pneumatics system generates the vacuums and pressures required for operating the valves and moving the sample through the hydraulics system. The electrical system controls operational sequences of the total system and includes electronic analyzers and computing circuitry for processing the data generated. A data display unit receives information from the analyzer and prints results, histograms, or cytograms [36].

In Sysmex XN-1000 automated hematology analyzer (Sysmex Corporation, Kobe, Japan), which gives CBC with six-part differential: neutrophils, lymphocytes, monocytes, eosinophils, basophils, and immature granulocytes. It also gives fully automated reticulocyte count, including nRBCs, Ret-He, and IRF. Fluorescent flow cytometry is used for the WBC count, WBC differential, and to detect nRBCs. [36].

The WBC, RBC, platelet counts, hemoglobin, and HCT are measured directly. Three hydraulic subsystems are used for determining the hemogram: the WBC channel, the RBC/platelet channel, and a separate hemoglobin channel [36].

In the RBC/platelet channel, a sheathed stream with hydrodynamic focusing is used to direct cells through the aperture, which reduces the co-incident passage, particle volume distortion, and recirculation of blood cells around the aperture; and in the WBC and RBC/platelet channels, floating thresholds are used to discriminate each cell population. As cells pass through the aperture, signals are transmitted in sequence to

the analog circuit and particle volume distribution analysis circuits for conversion to cumulative cell volume distribution data. The lower platelet threshold is automatically adjusted in the 2 to 6fL volume range, and the upper threshold is adjusted in the 12 to 30fL range, based on particle volume distribution. Likewise, the RBC lower and upper thresholds may be set in the 25 to 75 fL and 200 to 250fL volume ranges. This floating threshold circuitry allows for discrimination of cell populations on a specimen-by-specimen basis. Cell counts are based on pulses between the lower and upper auto-discriminator levels, with dilution ratio, volume counted, and coincident passage error accounted for in the final computer-generated numbers. In the RBC channel, the floating discriminator is particularly useful in separating platelets from small RBCs. [36].

PLATELETS

Platelets were first identified by Max Schultze in the year 1865. [18] Later on, a German Anatomist Bizzozero described platelets as disc shaped, having parallel surfaces, round to oval structures with diameter 2–3 times smaller than the diameter of the red cells [18].

Platelets play a pivotal role in antimicrobial host defense, linking the processes of inflammation and coagulation. Substantial evidence suggests that platelets detect and respond to bacterial infections with specific receptors such as Toll-like receptors and release cytokines and chemokines. [7]

QUANTIFICATION OF PLATELETS: -

Platelets can be counted either by manual methods or by automated methods. Manual methods include counting the number of platelets using a Neubauer chamber, or on a Romanowsky stained peripheral blood smear [24].

Till 2007, the manual method of counting platelets on a stained peripheral smear was considered Gold standard [24]. Platelets are now counted by automated methods using Automated hematology analyzers. Many methods are employed like Impedance platelet counting, optical scattering, and fluorescence [24].

IMMATURE PLATELET FRACTION-

"Immature platelet fraction is usually expressed as a proportional value of the total optical platelet count to indicate the rate of platelet production, although an absolute count can also be obtained." [28] The IPF% can be measured easily during routine blood sample analysis and results can be obtained immediately. The normal range of IPF% is 1-7% with a mean of 3.4% [29].

Some studies take into account the absolute IPF% values to determine the underlying etiology. The absolute IPF% is the total number of immature platelets per unit volume (IPF% x Platelet count). The absolute IPF% reflects the number of immature platelets in circulation [32].

The ideal time for determination of IPF % is within 1-12 hours of collection of the sample [33]. The analysis should not be done within the first hour after sampling. A reduction in the IPF% is seen during the first hour due to the swelling of the platelets caused by the anticoagulant EDTA [33].

The parameter IPF% reflects the severity of damage to the platelets and indicates the rate of production of platelets in bone marrow. In patients with bone marrow dysfunction, there is decreased production of platelets. In such patients the IPF% was found to be low [34]. And in thrombocytopenia due to increased destruction of platelets, The IPF% remains high and a fall in increased destruction if followed by fall in IPF% to normal or near normal values. Therefore, IPF% estimation is useful in differentiating these conditions [34]

Even though the reference intervals obtained for IPF% differed in different studies, it is still considered to be a better indicator of thrombopoiesis as compared to other platelet indices. [47] The difference in reference intervals is due to the use of different analyzers for estimation of IPF% like SYSMEX XE 2100, XE 5000 and XN series. SYSMEX XN series utilizes different principles of IPF% measurement from its older versions, so it is considered to be better [35].

IMMATURE PLATELET FRACTION QUANTIFICATION

In this haematology analyser, the blood cells are classified using a DC (Direct current) detection method and flow cytometry using a semiconductor laser. A specific channel (PLT-F) is used for the measurement of IPF% and it is measured using fluorescence method using oxazine dye, which binds specifically to the nucleic acid -rich platelet organelles like ribosomes and mitochondria [26, 27]. The platelets are irradiated using a semiconductor laser beam, and are plotted on a 2-D scatter gram. PLT-F channel improves the gating of the platelets by depicting side fluorescence (based on RNA content of platelets), side scatter (based on intracellular content of platelets) and forward scatter (based on size of the platelets). Since, the reticulated platelets or the

immature platelets have larger size and more RNA content as compared to the mature platelets, so they are easily differentiated in the scatter plot. The mature platelets are detected by the Impedance method (PLT- I) [26, 27]

The graph plotted against forward scatter and fluorescence defined platelets showing higher intensity of fluorescence and these are termed as "Immature platelets" [28]. The mature platelets are seen as "Blue dots". The immature ones are seen as "Green dots".

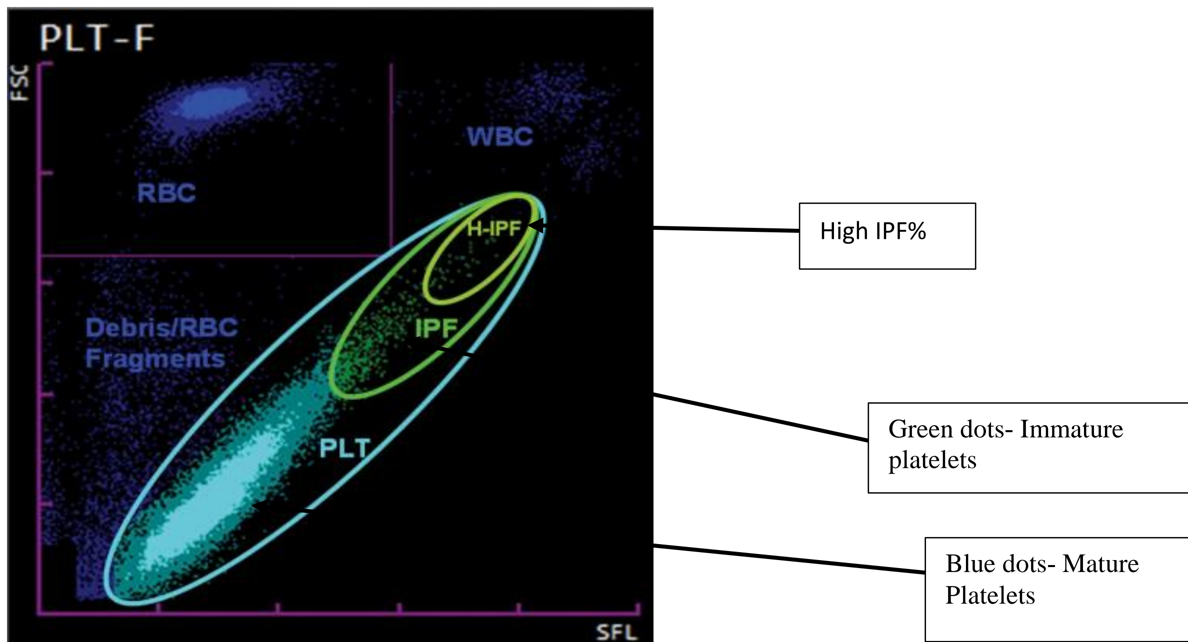


Fig.2- Scattergram from a patient with high IPF% related parameters.

RETICULOCYTE MEASUREMENTS

To accurately measure reticulocyte counts, automated counters use a combination of laser excitation, detectors, and a fluorescence marker that labels RNA and DNA (such as thiazole orange or polymethines). The sample is incubated with an RNA-binding fluorescence marker and counted by flow cytometry. Automated reticulocyte counters use objective thresholds for the classification of cells. This ensures a high level of reproducibility of the results. In automated counts, the measurement signals of up to 30,000 red blood cells are evaluated. This results in both high count rates and a high degree of precision [37].

In the Reticulocyte channel, fluorescence flow cytometry in conjunction with a nucleic acid staining dye to measure the amount of hemoglobin obtained within the reticulocytes. Sysmex XN series analyzer uses a Laser beam of wavelength = 633nm for analysis [37,38,39]. In the reticulocyte scatter gram, forward scatter a measure of individual cell size, on the y-axis is plotted against fluorescence intensity, a measure of RNA content, on the x-axis [40,41].

The reticulocyte count provides an initial assessment of whether the cause of anemia is due to impaired RBC production or increased loss in the peripheral circulation [42]. Previously, the reticulocyte count was done via microscopic examination of a smear prepared from fresh blood stained with a supravital stain, such as new methylene blue. The normal reticulocyte count by light microscopy is 0.5% - 1.5% of the total red cells [42,43]. Automated methods count a larger number of cells, and exhibit a greater degree of reproducibility compared to manual methods [42].

TOTAL WBC COUNT MEASUREMENTS

In the WDF channel, RBCs are lysed, WBC membranes are perforated, and the DNA and RNA in the WBCs are stained with a fluorescent dye. Plotting side scatter on the x-axis and side fluorescent light on the y-axis enables separation and enumeration of neutrophils, eosinophils, lymphocytes, monocytes, and immature granulocytes. In the WNR channel, the RBCs are lysed, including nucleated RBCs, and WBC membranes are perforated. A fluorescent polymethine dye stains the nucleus and organelles of the WBCs with high fluorescence intensity and stains the released nuclei of the nucleated RBCs with low intensity. Plotting side fluorescent light on the x-axis and forward scatter on the y-axis enables separation and enumeration of the total WBC count, basophils, and nucleated RBCs. The WBC count is automatically corrected when nucleated RBCs are present in the specimen. A WPC channel similarly detects blasts and abnormal lymphocytes using a lysing agent and fluorescent dye and plotting side scatter on the x-axis and side fluorescent light on the y-axis [36].

MATERIALS AND METHODS

SOURCE OF DATA

The study population consisted of adult patients admitted to the ICU of BLDE (Deemed to be University) Shri B. M. Patil Medical College, Hospital and Research Centre, Vijayapura.

STUDY PERIOD: 1st December, 2019 to 31st July, 2021.

INCLUSION CRITERIA:

All the adult(>/=18 years of age)[3] patients admitted in the intensive care unit in BLDE (Deemed To Be University) Shri B.M. Patil Medical College, Hospital and Research Centre, Vijayapura.

EXCLUSION CRITERIA:

All patients with a positive medical history for hematological disorders and/or hospitalized for less than 48 hours and/or already with a diagnosis of sepsis at ICU admission will be excluded.

SAMPLE COLLECTION:

After informed consent, venous blood samples under aseptic precautions were collected in K2 EDTA anticoagulated vacutainer to analyze hematological parameters.

METHODS OF COLLECTION OF DATA:

The study included a total of 138 patients who were admitted to the ICU. Peripheral venous blood samples were collected in K2 EDTA blood tubes and were analyzed using the Sysmex XN1000 (Kobe, Japan). The parameters that were analyzed in this study are platelet and reticulocyte parameters.

On ICU admission, all patients were evaluated for Complete blood count, Reticulocyte parameters and Platelet parameters along with general physical examination and systemic examination. Baseline data was collected and recorded. Patients whose baseline parameters were abnormal, were excluded from the study. After 48 hours of ICU admission, Complete blood count, Reticulocyte parameters, and Platelet parameters were repeated to check for any deviation from the baseline parameters or for the onset of sepsis and were recorded and in case of any deviation blood culture sensitivity was sent for confirmation of sepsis. If there was no deviation, investigation findings were documented until any deviation in parameters / or until ICU stay.

In our study, out of 138 patients, 132 patients had deviation in baseline parameters within 48 hours (2 days) of ICU stay, whereas 4 patients showed deviation from

baseline parameters after 72 hours (3 days) of ICU stay and 2 patients showed deviation from baseline parameters after 96 hours (4 days) of ICU stay. These 6 patients were young patients (aged between 22-28 years) and were kept in the Surgical ICU for observation.

STATISTICAL ANALYSIS

The data obtained was entered in a Microsoft Excel sheet, and statistical analysis was performed using a statistical package for the social sciences (Version 17). Results are presented as drawings, Mean ±standard deviation (SD), counts, and percentages. Results were compared using independent t-test, Chi-square test, the correlation between variables will found using correlation coefficient, Positive predictive value (PPV), Negative predictive value (NPV), ROC Curve, Sensitivity, and Specificity is be used, significance was achieved at $p < 0.001$ using IBM SPSS statistics version 23 and Microsoft Excel 2016.

REFERENCE RANGE [40]

PARAMETERS	REFERENCE RANGE
PLT count	1.5-4 lakhs/mm^3
IPF(%)	1-7 %
Reticulocyte count (RET%)	Adults- 0.5-2.5%

Table 3: Reference values [40]

RESULTS

Our study was done at the Department of Pathology, B.L.D.E (Deemed to be University), Shri B. M. Patil Medical College, Hospital and Research Centre, Vijayapura, Karnataka. In our study, we studied 138 patients who were admitted to the ICU for more than 48 hours. Peripheral whole blood samples were analyzed for CBC, Immature Platelet Fraction and Reticulocyte Percentage, along with SOFA score and Blood Cultures and were statistically analyzed.

Here, we present an evaluation of the results of our study.

AGE DISTRIBUTION

In this study, the minimum age was 18 years and maximum was 77 years and the mean age of presentation in this study was 41.3 years.

Among all the patients (N = 138) in the study, the majority of patients were in age group 41 to 50 years comprising of 45 cases (32.6% of study population). The detailed representation is shown below.

AGE (YEARS)	NO. OF PATIENTS	PERCENTAGE (%)
18 - 20	07	5.07
21 - 30	09	6.52
31 - 40	22	15.9
41 - 50	45	32.6
51 - 60	25	18.1
61 - 70	22	15.9
71 - 80	08	5.79
TOTAL	**138**	**100**

Table 4- Age of all the patients and the number of patients in each group with percentage

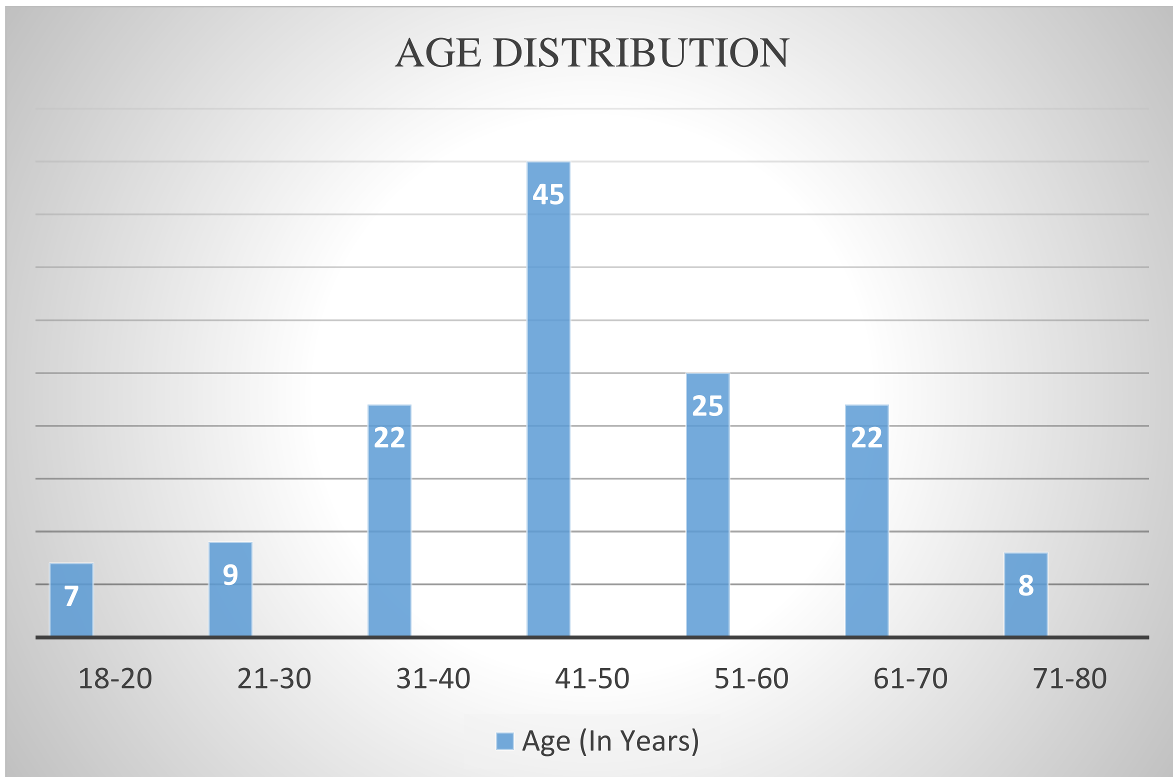


Fig. 3- Distribution of patients according to Age

GENDER DISTRIBUTION

Among all the patients included in this study, 77 were females and 61 were males comprising 55.8% and 44.2 % of total cases respectively.

SEX	NUMBER	PERCENTAGE (%)
FEMALE	77	55.8
MALE	61	44.2
TOTAL	**138**	**100**

Table 5- Gender distribution of all the patients and the number of patients in each group with percentage.

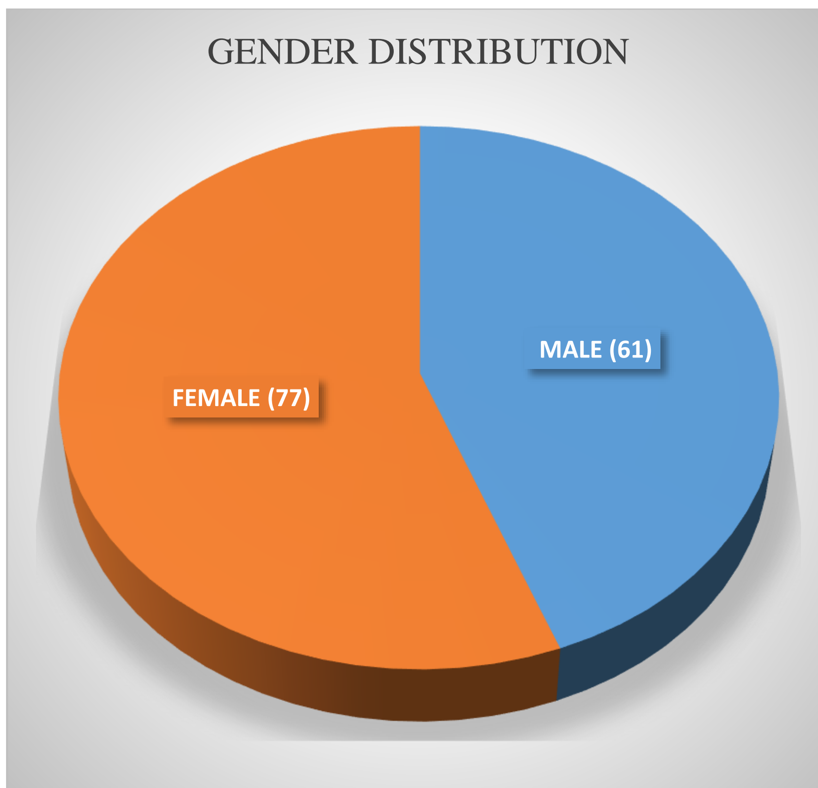


Fig. 04- Distribution of patients according to gender.

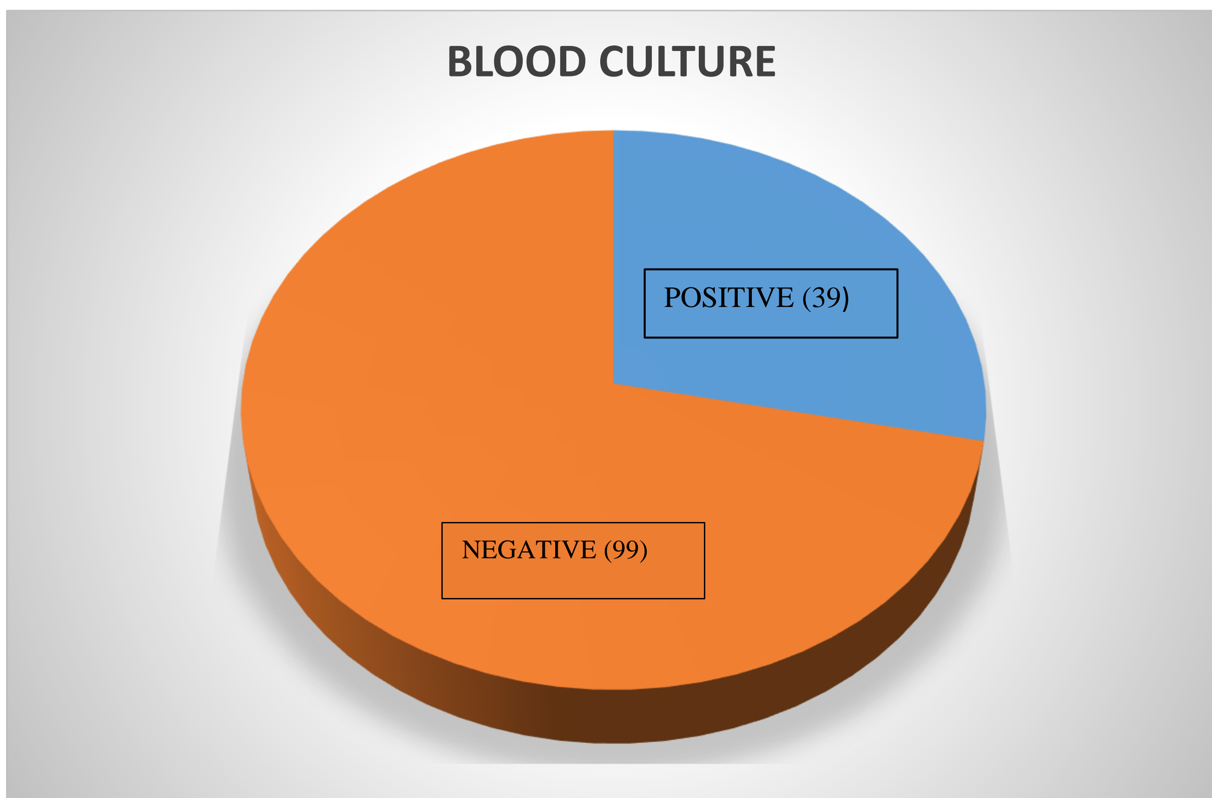
DISTRIBUTION OF PATIENTS ACCORDING TO BLOOD CULTURE

Among all the patients included in this study, 39 patients showed positive blood cultures, whereas 99 patients showed negative blood cultures, comprising 28.3% and 71.7 % of total cases respectively.

BLOOD CULTURE	NUMBER	PERCENTAGE (%)
POSITIVE	39	28.3
NEGATIVE	99	71.7
TOTAL	**138**	**100**

Table 06- Blood culture positivity of all the patients and the number of patients in each group with percentage.

Fig.05 – Distribution of patients according to Blood culture positivity.



COMPARISON OF PATIENTS ACCORDING TO PLATELET COUNT AND BLOOD CULTURE POSITIVITY

Among all the patients included in this study, the minimum PLT was 0.1 lakh/cumm and maximum PLT was 4.99 lakh/cumm. The mean PLT being 2.4 lakh/cumm on admission and 1.46 lakh/cumm after 48 hours of ICU stay.

PLT (lakh/cumm)	BLOOD CULTURE		TOTAL	p value
	NEGATIVE	POSITIVE		
<0.5	15	06	21	**<0.001***
0.51 – 1.0	33	14	47	
1.01 – 1.50	14	02	16	
1.51 – 2.0	19	06	25	
2.01- 2.50	02	04	06	
2.51 – 3.0	08	03	11	
3.01 – 3.50	02	01	03	
3.51 – 4.0	01	02	03	
4.01 – 4.50	03	0	03	
4.51- 5.0	02	01	03	
TOTAL	**99**	**39**	**138**	

Table 07- PLT of all the patients and the number of patients in each group.

Note: p value* significant at 5%level of significance ($p<0.05$)

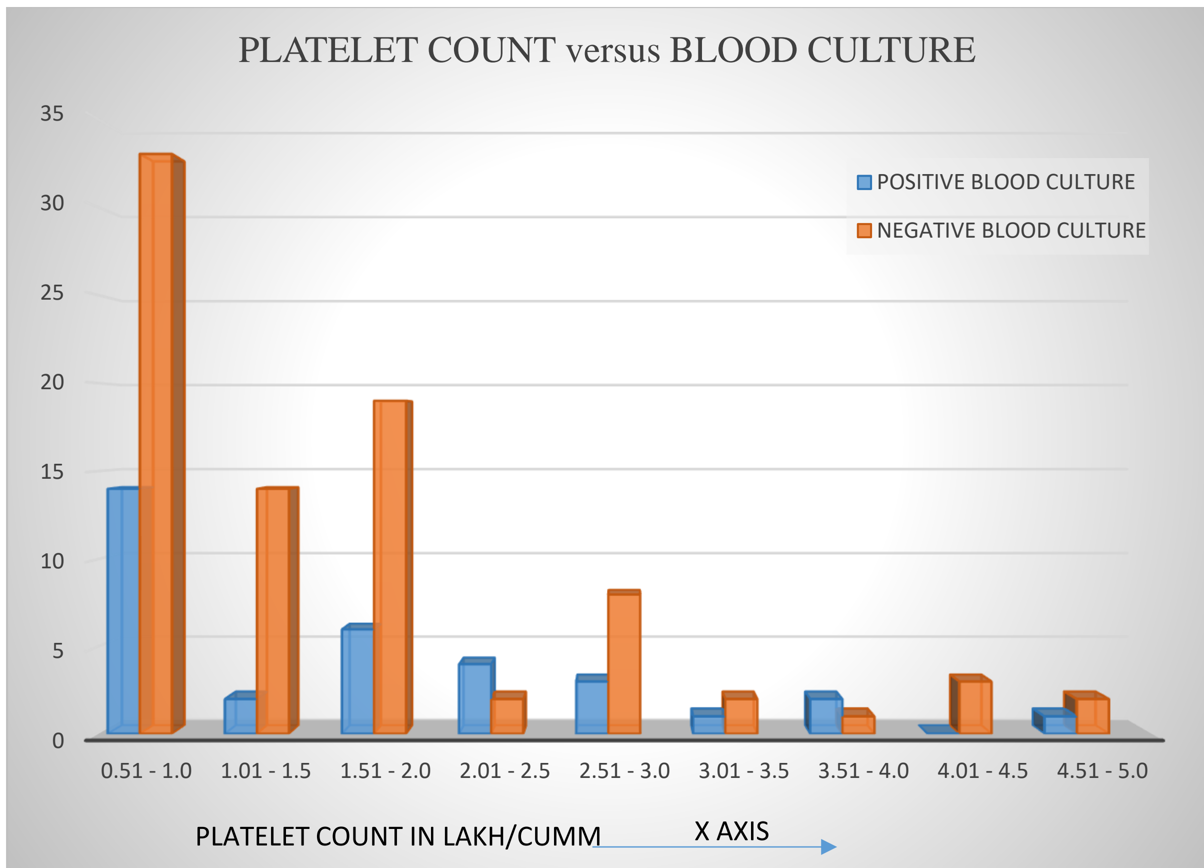


Fig. 06- Platelet count versus Blood Culture

COMPARISON OF PATIENTS ACCORDING TO IMMATURE PLATELET FRACTION AND BLOOD CULTURE POSITIVITY

The normal range of IPF% is 1-7%. [45]

Among all the patients included in this study, the minimum IPF% 0.9% and maximum IPF% was 13.6%. The mean IPF% being 3.9% on admission and 9.1% after 48 hours of ICU stay.

IPF %	BLOOD CULTURE		TOTAL	p value
	NEGATIVE	POSITIVE		
<1	0	0	0	**<0.001***
1 - 7	10	08	18	
>7	89	31	120	
TOTAL	**99**	**39**	**138**	

Table 08- IPF% of all the patients and the number of patients in each group.

Note: p value* significant at 5% level of significance ($p<0.05$)

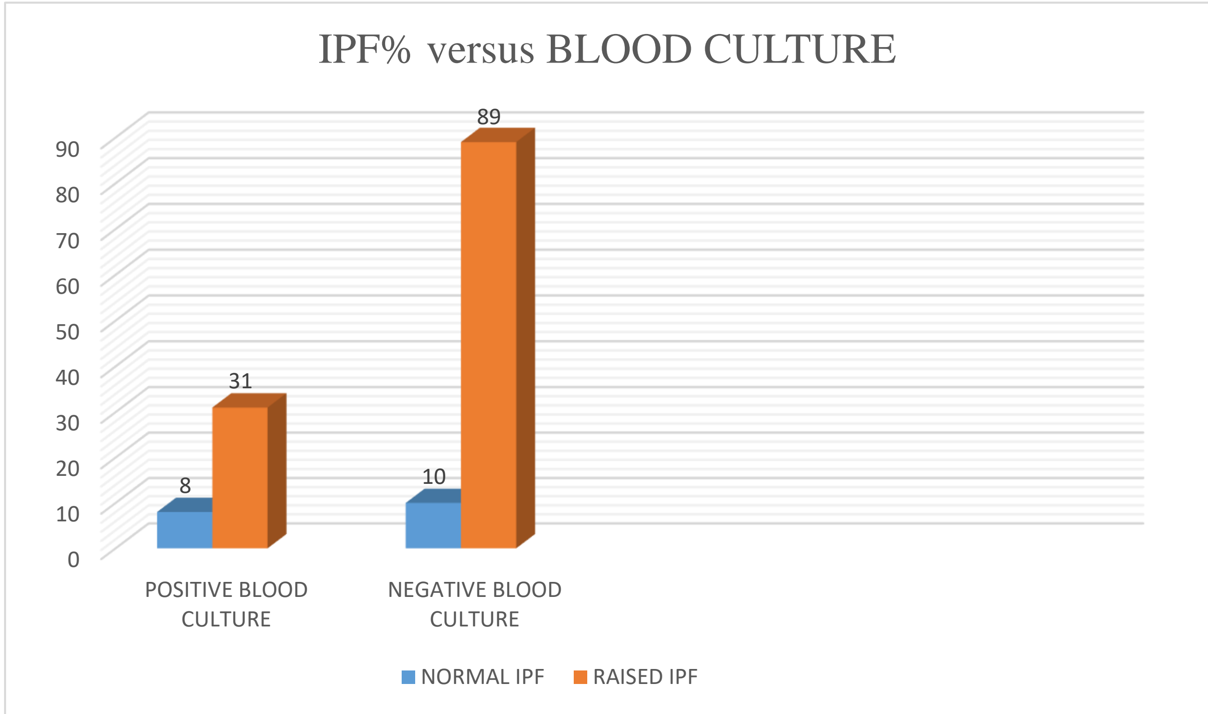


Fig. 07- IPF% versus Blood Culture

ROC CURVE FOR THE COMPARISON BETWEEN IMMATURE PLATELET FRACTION AND BLOOD CULTURE

ROC Curve when IPF% is compared against blood culture at 95% confidence interval, the lower bound value is 0.546 and the upper bound value is 0.771 with an area of 0.658.

- Sensitivity- 66.7%
- Specificity- 68.7%

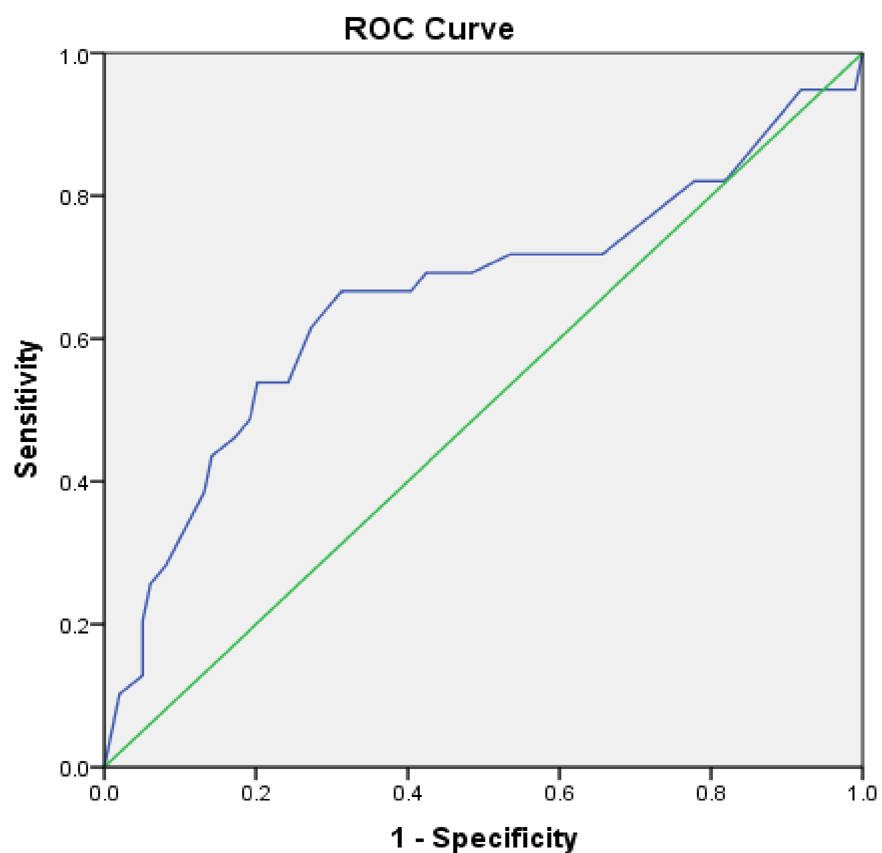


Fig. 08- ROC Curve for comparison between IPF% and Blood Culture

COMPARISON OF PATIENTS ACCORDING TO RETICULOCYTE PERCENT AND BLOOD CULTURE POSITIVITY

The normal range of RET% is 0.5-2.5%. [45]

Among all the patients included in this study, the minimum RET% was 0.4 %and maximum RET% was 2.5%. The mean RET% being 1.52% on admission and 0.6% after 48 hours of ICU stay.

RET %	BLOOD CULTURE		TOTAL	p value
	NEGATIVE	POSITIVE		
<0.5	57	33	90	**<0.001***
0.5 – 2.5	42	06	48	
>2.5	0	0	0	
TOTAL	**99**	**39**	**138**	

Table 09- RET% of all the patients and the number of patients in each group.

Note: p value* significant at 5% level of significance (p<0.05)

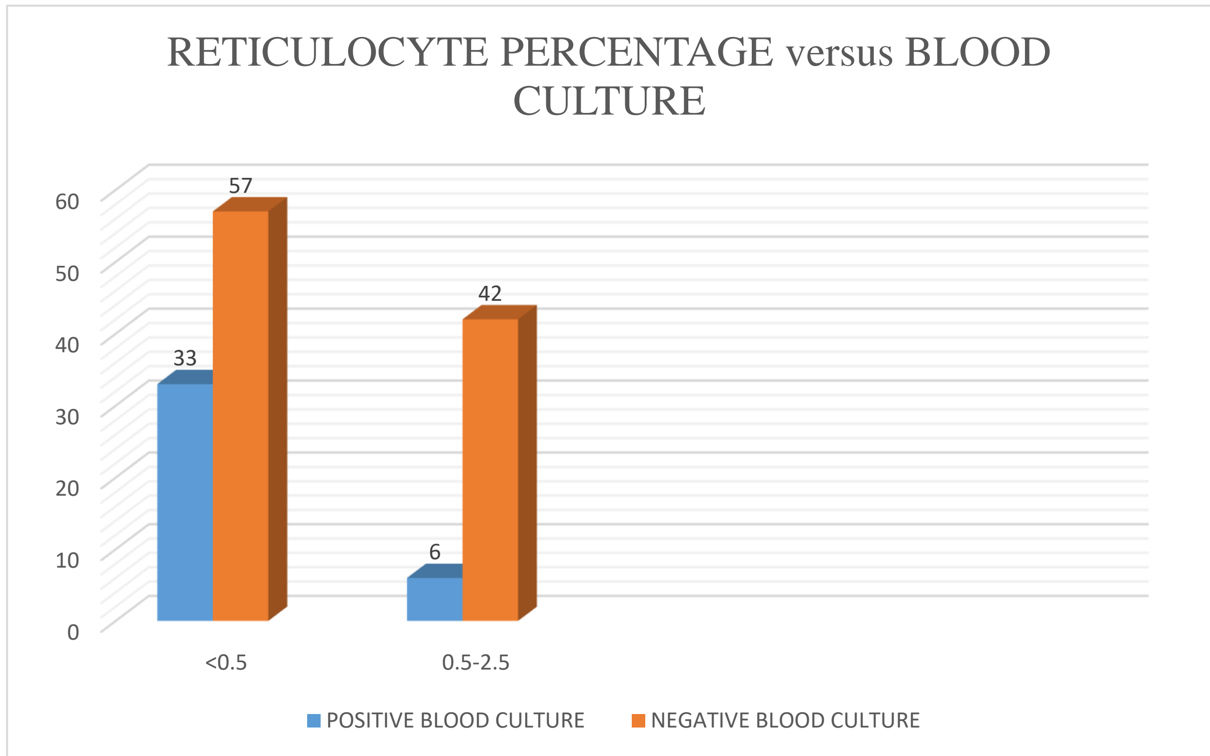


Fig. 09- Reticulocyte Percentage versus Blood Culture

ROC CURVE FOR THE COMPARISON BETWEEN RETICULOCYTE PERCENT AND BLOOD CULTURE

ROC Curve when RET% is compared against blood culture at 95% confidence interval, the lower bound value is 0.584 and the upper bound value is 0.776 with an area of 0.680.

- Sensitivity- 61.5%
- Specificity- 71.7%

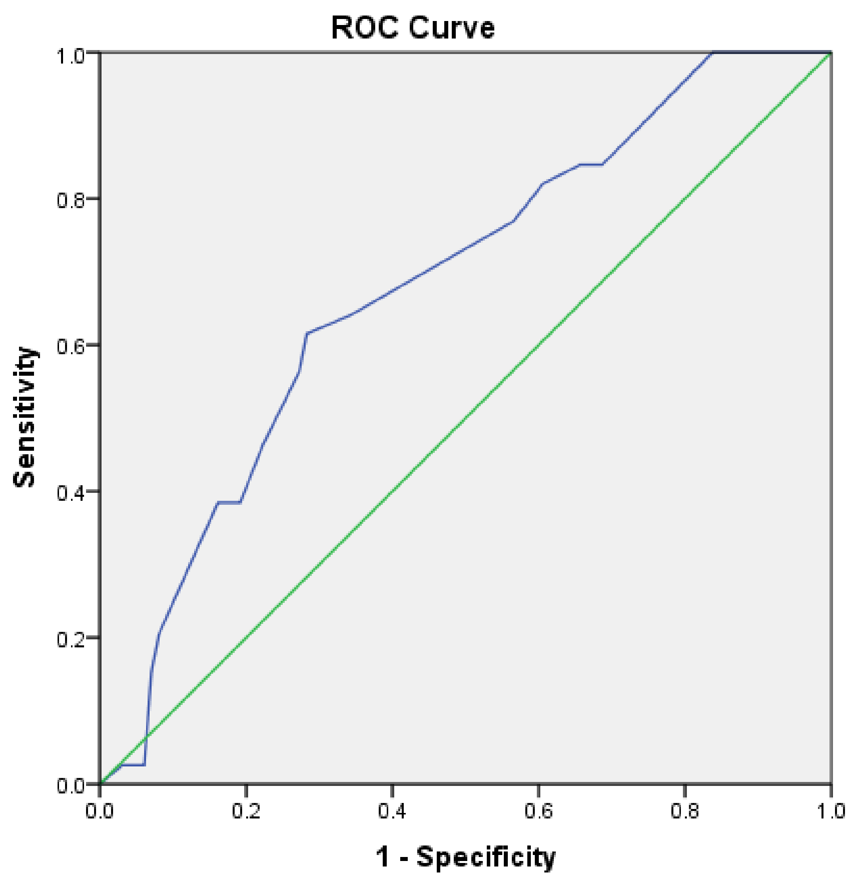


Fig. 10- ROC Curve for comparison between RET% and Blood Culture

COMPARISON OF PATIENTS ACCORDING TO SOFA SCORE AND BLOOD CULTURE POSITIVITY

The normal SOFA score is 0 and the maximum SOFA score is 24.

Among all the patients included in this study, the minimum SOFA score was 0 and maximum SOFA score was 8.

SOFA Score	BLOOD CULTURE		TOTAL	p value
	NEGATIVE	POSITIVE		
0	31	12	43	**<0.001***
1	48	4	52	
2	2	4	6	
3	3	3	6	
4	5	2	7	
5	2	4	6	
6	3	3	6	
7	1	5	6	
8	4	2	6	
TOTAL	**99**	**39**	**138**	

Table 10- SOFA score of all the patients and the number of patients in each group.

Note: p value* significant at 5%level of significance ($p<0.05$)

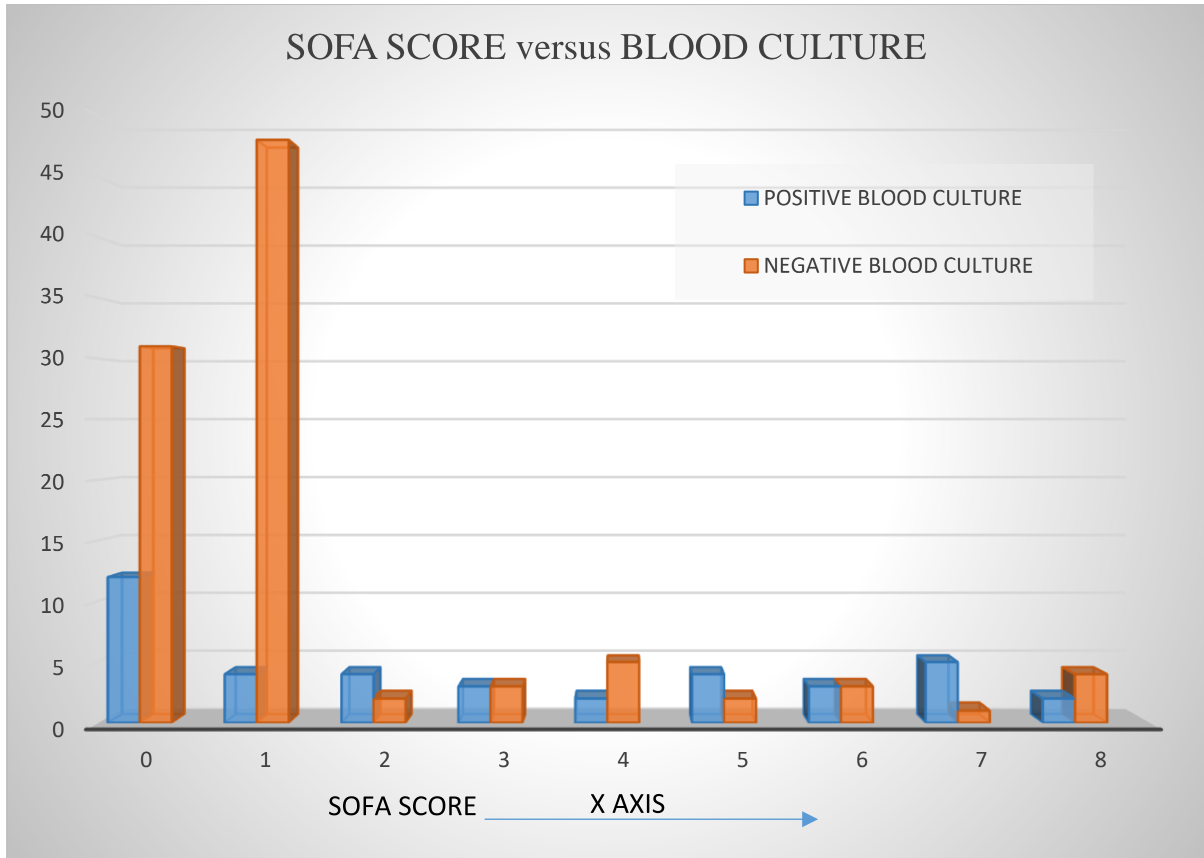


Fig. 11- SOFA Score versus Blood Culture

Sl. No	PARAMETER	p value	SIGNIFICANCE	SENSITIVITY	SPECIFICITY
01	Platelet Count	<0.001*	Significant	-	-
02	IPF%	<0.001*	Significant	**66.7%**	**68.7%**
03	RET%	<0.001*	Significant	**61.5%**	**71.7%**
04	SOFA Score	<0.001*	Significant	-	-

Table 12- Comparison table of various parameters

DISCUSSION

"Sepsis" is a syndrome described by generalized/systemic inflammation, dysregulated immune system/host response, and end-organ damage which coincides with a suspected or a proven infection. [49] The incidence of morbidity and mortality are high in sepsis, especially in newborns, infants, and others suffering from an underlying chronic ailment or immunocompromised status. [49] Though the diagnosis of sepsis is based majorly on clinical data, laboratory confirmation is equally important. [49] Blood culture being the gold standard method for sepsis diagnosis is a time consuming and costly affair. [49] Thus, narrowing down to a biomarker which could identify sepsis at a prior stage is of immense significance, [48] and immature platelet fraction fulfils this criteria.

The occurrence of sepsis is increasing nearly at a rate of 10 % per year for the past few years. [45] Thus, worldwide, sepsis is still a major cause of ICU deaths. [6] The probable reasons could be due to the improvements in life expectancy, use of immuno suppressive agents and invasive procedures. With the increasing knowledge about the pathogenesis of sepsis, high mortality rates of up to 30% are still being observed, even with best supportive care. [45] Most important challenge in sepsis management is early diagnosis, because any delay in sepsis recognition increases sepsis-related mortality. [45] Heterogeneous nature of sepsis, is another limiting cause since it restricts the effectiveness of a versatile treatment strategy for these patients. [45]

Ideally, a sepsis biomarker should be able to:

(i) Differentiate between sepsis and other causes of sterile inflammation,

(ii) Permit risk stratification, and

(iii) Identify sepsis complications and enable target-specific treatments. [45]

Sepsis clinically manifests in a highly variable manner, which depends on the causative organism, the site where the infection first began, the organ dysfunction pattern, the patient's immune status, and the time taken to initiate the treatment. [8] Infection and organ dysfunction may present with subtle signs, which raises a need for warning signs of incipient sepsis which is issued by the International consensus guidelines.

Early diagnosis of sepsis is the mainstay for improving patient outcomes and has contributed greatly to boost the research for innovative biomarkers. [3] On time diagnosis and an apt management of septic patients is a challenge. [6] Thus the inexpensive parameters generated by modern hematological analyzers should are valuable. [6]

It is known that platelets play a pivotal role in antimicrobial host defense, linking the processes of inflammation and coagulation, [7] and immature platelets are a proportion of circulating platelets containing RNA. [48] IPF% is a newer variable that is being measured using automated hematology analyzers in the diagnosis of sepsis. [48] the findings of our study conclude that IPF% is a dependable biomarker which accurately discriminates between patients who developed sepsis and who did not develop sepsis. [50] This effortlessly measurable cellular variable reflects the rate of platelet production which is infrequently used in general ICUs. [47] In our study IPF% has shown to have a sensitivity of 66.7% and specificity 68.7 % as an early marker in predicting the onset of sepsis.

Our study results are in concordance with the study done by Buoro S, et al. [3] Their study results show that the risk of developing sepsis in ICU is significantly associated with raised IPF% values 48 hours before the index date and decreased RET% values 24 hours before the index date, where index date was defined as the date of onset of sepsis. [3] Association of raised immature platelets with disease severity and mortality in sepsis may indicate an increased platelet turnover due to increased platelet consumption in sepsis and septic shock. [46] As immature platelets are more hemostatically active than mature platelets, an increased number of immature platelets might contribute to the formation of microthrombi and organ failure, increasing risk of severity and mortality in sepsis. [46] This easily measurable cellular variable reflects the thrombopoietic rate, which is rarely used in general ICUs, has high diagnostic specificity (90.0 %) as an early marker predicting the onset of sepsis. [47]

Our study results are in concordance with the study done by De Blasi et al, [3] who reported that an increased value of IPF%, predicted the development of sepsis in ICU patients. However, other significant associations between additional hematological parameters and sepsis were not conclusive. [3] Most studies confirm that IPF% can predict development of sepsis and assess severity of sepsis. [48]

There is proven evidence that diagnosing sepsis early is important for better patient outcomes, which has led to the boost for the research for innovative biomarkers which are by affordable and possess optimal diagnostic accuracy. [3] The study conducted by Buoro S. et al prove the diagnostic performance of two innovative parameters- IPF% and RET%, which can be measured with Sysmex XN module. [3]

As per the study conducted by Kaukonen K. et.al, [5] from 2000 to 2013, of the total admissions in 172 ICU's with a total of 11,71,797 patients, 1,062,134 (90.6%) patients did not have sepsis, and 109,663 (9.4%) had infection and organ dysfunction. Of the patients with infection and organ dysfunction, 86,394 (87.9%) patients had Severe Inflammatory Response Syndrome (SIRS) positive severe sepsis and 23,269 (12.1%) patients had SIRS-negative severe sepsis. Patients with SIRS-positive severe sepsis were younger, were more severely ill, and had higher mortality than those with SIRS-negative severe sepsis. [5]

SUMMARY

This study was done at the Department of Pathology, B.L.D.E (Deemed to be University), Shri B. M. Patil Medical College, Hospital and Research Centre, Vijayapura, Karnataka.

In our study, we studied 138 patients who were admitted to the ICU and who fulfilled inclusion and exclusion criteria.

We studied hematological parameters processed in the Sysmex XN1000 (Sysmex Corporation, Kobe, Japan) analyser.

Salient observations from our study are, Out of the 138 patients-

- 120 patients had increased IPF% of which 31 had positive blood cultures and 89 had negative blood cultures;
- 90 patients had decreased RET% of which 33 patients had positive blood cultures and 57 patients had negative blood cultures;
- 39 patients showed positive blood cultures and 99 patients showed negative blood cultures.

There was a statistically significant association between IPF% and blood culture positivity ($p = <0.001$) and between RET% and blood culture positivity ($p = <0.001$).

Early diagnosis of sepsis is the mainstay for improving patient outcomes and has contributed greatly to boost the research for innovative biomarkers. [3]

LIMITATIONS

- IPF% cut-off and reference values for different study population needs to be standardized and established; as the reference ranges taken in our study are of adults.
- Follow up with treatment was not done in our study, which would have given valuable inputs as how high or low IPF% and RET% responded to therapy.

CONCLUSION

The immature platelet fraction (IPF%) reflects the degree of reticulated platelets. [50] IPF% values obtained after 48 hours of ICU stay are higher in patients with sepsis compared to patients who did not develop sepsis. [45] The combination of an increased IPF% and decreased RET% (that is 41 patients in our study) in ICU patients may hence be considered as an early, rapid, inexpensive and widely available measure of sepsis prediction, so allowing a more efficient and timely patient management by means of strengthened monitoring and more aggressive treatment. [3] Our results confirm and extend a recent report of IPF% as an informative sepsis biomarker, in an independent and clinically representative population. [45] Studies on a larger population are required to define how this readily accessible parameter could be incorporated in sepsis management protocols. [45]

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ANNEXURE – I

IEC/131/19
22/11/2019

B.L.D.E. (DEEMED TO BE UNIVERSITY)

(Declared vide notification No. F.9-37/2007-U.3 (A) Dated. 29-2-2008 of the MHRD, Government of India under Section 3 of the UGC Act, 1956)

The Constituent College

SHRI. B. M. PATIL MEDICAL COLLEGE, HOSPITAL AND RESEARCH CENTRE

INSTITUTIONAL ETHICAL CLEARANCE CERTIFICATE

The ethical committee of this college met on 13-11-2019 at 3-15 pm to scrutinize the synopsis of Postgraduate students of this college from Ethical Clearance point of view. After scrutiny the following original/corrected and revised version synopsis of the Thesis has been accorded Ethical Clearance

Title: Efficacy of platelet and reticulocyte parameters for early diagnosis of sepsis

Name of PG student: Dr. Umme Romaan, Department of Pathology

Name of Guide/Co-investigator: Dr. Surekha.B.Hippargi, Professor Department of Pathology

DR RAGHVENDRA KULKARNI
CHAIRMAN
Institutional Ethical Committee
BLDEU's Shri B.M. Patil
Medical College,BIJAPUR-586103

Following documents were placed before Ethical Committee for Scrutinization:

1. Copy of Synopsis / Research project
2. Copy of informed consent form
3. Any other relevant documents.

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ANNEXURE-II

B.L.D.E (DEEMED TO BE UNIVERSITY) SHRI B.M.PATIL MEDICAL COLLEGE HOSPITAL AND RESEARCH CENTRE, VIJAYPURA-586103

RESEARCH INFORMED CONSENT FORM

TITLE OF THE PROJECT: "EFFICACY OF PLATELET AND RETICULOCYTE PARAMETERS FOR EARLY DIAGNOSIS OF SEPSIS IN INTENSIVE CARE UNIT PATIENTS"

PRINCIPAL INVESTIGATOR: DR. UMME ROMAAN
P.G. DEPARTMENT OF PATHOLOGY

P.G GUIDE **:** DR. SUREKHA B. HIPPARGI MD
PROFESSOR, DEPT OF PATHOLOGY.

PURPOSE OF RESEARCH:

I have been informed that the present study is a study of Efficacy of Platelet and Reticulocyte parameters in early diagnosis of sepsis in Intensive Care Unit patients.

PROCEDURE:

I understand that I undergo detailed history and after which necessary investigations will be done.

RISK AND DISCOMFORTS:

I understand that, there is no risk involved for me being a part of the study.

BENEFITS:

I understand that my participation in the study will help to know the Efficacy of Platelet and Reticulocyte parameters in early diagnosis of sepsis in Intensive Care Unit patients.

CONFIDENTIALITY:

I understand that the medical information produced by the study will become a part of hospital record and will be subjected to confidentiality and privacy regulations of the hospital. If data is used for publications the identity of patient will not be revealed.

REQUEST FOR MORE INFORMATION:

I understand that I might be asked for more information about my disease at any time.

REFUSAL FOR WITHDRAWL OF PARTICIPATION:

I understand that my participation is voluntary and that I may refuse to participate or may withdraw from the study at any time

INJURY STATEMENT:

I understand that in the unlikely event of injury to me during the study I will get medical treatment but no further compensations.

I have read and fully understood this consent form. Therefore, I agree to participate in the present study.

____

Participant/Guardian   Date:

____

Signature of Witness   Date:

I have explained the patient/patients attender the purpose of study, the procedure required and possible risk and benefit of my ability in the vernacular language.

____

Investigator/P.G Date:

____

Witness to Signature Date:

ANNEXURE - III

PROFORMA

NAME	:	OP/IP No.:
AGE	:	
SEX	:	Date of Admission:
RELIGION	:	Date of Discharge:
OCCUPATION	:	
RESIDENCE	:	

PRESENTING COMPLAINTS :

PAST HISTORY :

PERSONAL HISTORY :

FAMILY HISTORY :

TREATMENT HISTORY :

GENERAL PHYSICAL EXAMINATION**:**

Pallor	Present/Absent
Icterus	Present/Absent
Clubbing	Present/Absent
Lymphadenopathy	Present/Absent
Edema	Present/Absent
Built	Poor/Average/Well

VITALS: PR: RR:

BP: TEMPERATURE:

ADDITIONAL DATA:

MAP (Mean Arterial Pressure)

Urine Output

GCS (Glasgow Coma Scale)

SOFA (Sequential Organ Failure Assessment) Score

SYSTEMIC EXAMINATION:

Cardiovascular system:

Respiratory system:

Per Abdomen:

Central nervous system:

Clinical Diagnosis:

INVESTIGATIONS:

Red blood cell count (RBC count):

Hemoglobin (Hb):

Total Leucocyte Count (TLC):

Platelet Count (PC):

Immature Platelet Fraction (IPF%):

Reticulocyte count (RET%):

Bilirubin:

Creatinine:

Blood Culture:

Other Investigations:

KEY TO MASTER CHART

Sl No.	Serial Number
IP No.	In Patient Number
PLT	Platelet Count (Lakh per mm^3)
IPF%	Immature Platelet Fraction
RET%	Reticulocyte Count
On ADM	On Admission
GCS	Glasgow Coma Scale
SOFA	Sequential Organ Failure assessment Score
Bld Cul	Blood Culture

SL. NO	AGE	SEX	PLT	IPF%		RET%		SOFA	BLD CUL
				On ADM	AFTER 48 Hours	On ADM	AFTER 48 Hours		
1	60	F	1.56	4.6	8	0.8	0.5	1	NEGATIVE
2	62	M	3.11	3.9	11.2	1.8	0.7	1	POSITIVE
3	40	M	1.5	5.3	10	0.8	0.5	0	NEGATIVE
4	20	F	0.71	4	13	1.5	0.5	4	POSITIVE
5	52	F	0.66	4	8.3	0.6	0.5	1	NEGATIVE
6	37	F	2.7	4	9.1	1	0.7	1	NEGATIVE
7	22	F	0.98	3.8	8	1.4	1	1	NEGATIVE
8	65	M	0.8	3	8.6	1.5	1	1	NEGATIVE
9	45	M	1.96	3.8	9	1.4	1	1	NEGATIVE
10	65	M	4.99	3.3	9	1.7	0.5	0	NEGATIVE
11	38	F	0.95	3.7	9	2	0.5	0	NEGATIVE
12	58	F	1.17	6	10	2	0.6	0	NEGATIVE
13	45	M	0.4	4	9	1.6	0.5	0	NEGATIVE
14	28	M	0.58	4	10.3	1.6	0.6	8	POSITIVE
15	71	M	4.33	3.5	8	0.9	0.5	1	NEGATIVE
16	75	M	0.19	3	8.2	2	0.5	0	NEGATIVE
17	28	F	0.97	4.6	9	1.8	0.6	0	NEGATIVE
18	75	F	2.47	4	11	1.5	0.5	2	POSITIVE
19	65	M	0.83	5	11.8	1.8	0.6	6	POSITIVE
20	62	M	1.24	3	8.5	0.7	0.5	0	NEGATIVE
21	25	M	0.84	3	8	1.8	0.5	0	NEGATIVE
22	25	F	2.5	5	10	1.5	0.9	0	NEGATIVE
23	70	F	0.97	5	13.1	0.7	0.5	5	POSITIVE
24	75	M	1.89	5	10	1.3	0.5	0	NEGATIVE
25	65	F	2.52	4	13.6	1.9	0.5	2	POSITIVE
26	35	F	1.78	5	9.1	1.5	0.7	1	NEGATIVE
27	25	M	0.89	7.1	10.5	1.3	0.5	1	NEGATIVE

28	40	M	0.76	3	9	2	0.6	1	NEGATIVE
29	20	F	1.06	5.7	10	2	0.5	1	NEGATIVE
30	35	F	0.1	4	12.6	1.1	0.5	8	POSITIVE
31	65	F	3.55	5	11.6	1.3	0.9	5	POSITIVE
32	87	M	0.24	4.2	9	1.5	0.5	1	NEGATIVE
33	45	M	1.78	6	12.7	1.4	0.5	3	POSITIVE
34	22	M	0.21	3	7.6	0.5	0.5	1	NEGATIVE
35	80	M	0.78	4	10.2	0.7	0.5	7	POSITIVE
36	23	F	0.43	3	8	0.9	0.5	1	NEGATIVE
37	70	M	0.98	4	10.5	0.8	0.5	6	POSITIVE
38	65	F	2.96	4.2	11.8	1.3	0.5	3	POSITIVE
39	22	F	0.52	3	8	1.3	0.5	1	NEGATIVE
40	26	F	0.41	4	12.9	1.1	0.5	7	POSITIVE
41	27	M	1.9	3	8	1.6	0.5	0	NEGATIVE
42	72	M	1.55	2	7	1.5	0.7	0	NEGATIVE
43	78	F	1.61	3	11.8	1.7	0.5	4	POSITIVE
44	50	M	1.5	3	7.2	1.9	0.5	0	NEGATIVE
45	51	F	1.56	4	8.6	1.1	0.9	1	NEGATIVE
46	45	M	3.11	3.5	8	1.8	0.8	1	NEGATIVE
47	33	M	1.5	5.3	10	1.3	0.5	0	NEGATIVE
48	70	M	0.71	3	8	1.4	0.5	4	NEGATIVE
49	84	F	0.66	0.9	7	1.2	0.5	1	NEGATIVE
50	35	M	2.7	4	9.1	1.5	1	1	NEGATIVE
51	60	F	0.98	3.8	8	1.3	0.5	1	NEGATIVE
52	32	F	0.8	3	8.3	0.8	0.5	1	NEGATIVE
53	40	F	1.96	2	8	0.7	0.5	1	NEGATIVE
54	67	F	4.99	3	7	1.6	0.5	0	NEGATIVE
55	25	F	0.95	2	6	1.8	1	0	NEGATIVE
56	35	M	1.17	4	8	2	0.5	0	NEGATIVE
57	35	M	0.4	2.1	7	1.3	0.5	0	NEGATIVE

58	57	M	0.58	4	9	1.6	0.5	8	NEGATIVE
59	80	M	4.33	5.5	10	1.2	1	1	NEGATIVE
60	23	F	0.19	3	8	2	0.5	0	NEGATIVE
61	23	F	0.97	4.6	10.2	1.8	0.5	0	POSITIVE
62	24	F	2.47	5	9.9	1.3	0.5	2	POSITIVE
63	30	F	0.83	3	7	1.2	0.5	6	NEGATIVE
64	26	F	1.24	3	8.5	0.7	1	0	NEGATIVE
65	21	F	0.84	4.5	11	0.8	0.5	0	POSITIVE
66	41	F	2.5	6	9.8	1.5	0.5	0	POSITIVE
67	21	M	0.97	5	13.1	1.3	0.5	5	POSITIVE
68	42	F	1.89	5	10.2	1.3	0.5	0	POSITIVE
69	62	M	2.52	4.3	12	1.8	0.5	2	POSITIVE
70	25	M	1.78	6	9	1.5	0.5	1	NEGATIVE
71	30	F	0.89	7.5	9.4	1.3	0.5	1	NEGATIVE
72	19	F	0.76	3.7	8	2	0.9	1	NEGATIVE
73	29	F	1.06	3.2	10	2.5	0.5	1	NEGATIVE
74	30	F	0.1	1.5	8	2	0.5	8	NEGATIVE
75	42	M	3.55	1.9	7.8	2	0.6	5	NEGATIVE
76	35	F	0.24	4.2	11.2	1.3	0.5	1	POSITIVE
77	21	F	1.78	4	12.7	2	0.6	3	POSITIVE
78	63	M	0.21	3.2	9.5	1.8	0.5	1	NEGATIVE
79	40	F	0.78	2.1	8.7	2	0.7	7	NEGATIVE
80	35	M	0.43	3	8	1.9	0.5	1	NEGATIVE
81	44	M	0.98	3.6	9.1	2	0.6	6	NEGATIVE
82	26	F	2.96	2.2	7.8	2	0.7	3	NEGATIVE
83	30	F	0.52	4	8.8	1.1	0.5	1	POSITIVE
84	26	F	0.41	4	12.9	2	0.5	7	POSITIVE
85	30	M	1.9	3	9.4	1.5	0.5	0	POSITIVE
86	40	M	1.55	3	8.6	1.3	0.5	0	POSITIVE
87	24	F	1.61	4	8.1	1.8	0.5	4	NEGATIVE

88	40	F	1.5	3	9.1	1	0.5	0	NEGATIVE
89	36	F	1.56	4.6	8	0.5	0.7	1	NEGATIVE
90	70	M	3.11	3.9	8.4	1.8	0.5	1	NEGATIVE
91	21	M	1.5	7.3	8.4	0.5	0.8	0	NEGATIVE
92	30	F	0.71	4	7.5	1.4	0.5	4	NEGATIVE
93	65	F	0.66	3.2	8	1.4	0.7	1	NEGATIVE
94	23	M	2.7	4	7.1	1.3	0.5	1	NEGATIVE
95	52	M	0.98	3.8	9	1.4	0.6	1	NEGATIVE
96	52	F	0.8	6.8	8.9	0.5	0.5	1	NEGATIVE
97	27	M	1.96	5	7.8	1	0.5	1	NEGATIVE
98	23	F	4.99	4.2	11.8	1.6	0.5	0	POSITIVE
99	30	F	0.95	3.7	12.1	1.8	0.5	0	POSITIVE
100	30	F	1.17	4.2	9.8	2	0.5	0	POSITIVE
101	27	M	0.4	2.7	9	1.4	0.5	0	NEGATIVE
102	60	M	0.58	0.8	6.8	1.3	0.5	8	NEGATIVE
103	46	F	4.33	5.5	8.8	1.2	0.5	1	NEGATIVE
104	30	M	0.19	5.9	8	2	0.8	0	NEGATIVE
105	65	M	0.97	4.6	7	1.8	1	0	NEGATIVE
106	27	M	2.47	3.2	9	2	1	2	NEGATIVE
107	30	F	0.83	4	11.8	2	0.5	6	POSITIVE
108	20	M	1.24	3	11.5	0.7	0.5	0	POSITIVE
109	24	M	0.84	4.5	11.3	0.8	0.5	0	POSITIVE
110	24	F	2.5	6	11.2	0.5	0.5	0	POSITIVE
111	32	M	0.97	2.2	7	2	0.9	5	NEGATIVE
112	40	F	1.89	5	8	1.3	0.5	0	NEGATIVE
113	20	F	2.52	2	7	2	0.9	2	NEGATIVE
114	25	F	1.78	3	8	1.5	0.5	1	NEGATIVE
115	25	F	0.89	5.8	8.5	1.3	0.7	1	NEGATIVE
116	60	F	0.76	3.8	7	2	0.5	1	NEGATIVE
117	27	F	1.06	5.7	8	2	0.6	1	NEGATIVE

118	27	M	0.1	3	8	2.5	0.5	8	NEGATIVE
119	27	F	3.55	3.1	11.6	1	0.5	5	POSITIVE
120	21	M	0.24	4.2	11.8	2.1	0.5	1	POSITIVE
121	22	F	1.78	3	7.3	2	0.6	3	NEGATIVE
122	40	F	0.21	4.2	7.2	1.8	0.5	1	NEGATIVE
123	62	M	0.78	3	10.2	2.5	0.5	7	POSITIVE
124	24	F	0.43	6	8.1	1.9	0.6	1	NEGATIVE
125	24	M	0.98	2.2	5.7	2.2	0.5	6	NEGATIVE
126	18	F	2.96	5.1	9	2.3	0.9	3	NEGATIVE
127	60	F	0.52	4	6.2	2	1	1	NEGATIVE
128	20	F	0.41	7.8	12.9	2.1	1.2	7	POSITIVE
129	23	F	1.9	4	9	2.2	1	0	NEGATIVE
130	26	F	1.55	3.2	9	2.5	1.1	0	NEGATIVE
131	62	M	1.61	4.1	6.6	2.4	1	4	NEGATIVE
132	26	F	1.5	4	6.2	1	0.8	0	NEGATIVE
133	75	M	1.56	4.6	8	1.1	0.6	1	NEGATIVE
134	60	M	3.11	3.9	6.1	1.8	0.5	1	NEGATIVE
135	26	M	1.5	5.3	8	1.5	0.5	0	NEGATIVE
136	60	F	0.71	5	7	2.1	0.5	4	NEGATIVE
137	55	F	0.66	4.2	9	2.5	0.6	1	NEGATIVE
138	60	F	2.7	4	7	1.4	0.5	1	NEGATIVE