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ORIGINAL ARTICLE

Haematological parameters and screening tests of haemostasis in children with sepsis: results from a tertiary care centre in India

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Abstract

Sepsis is a common cause of death in infants and children. Haemostatic abnormalities have been reported in such patients. There is scant information on the nature and frequency of these abnormalities in children especially from India. Our aim was to evaluate the nature and frequency of haematological and haemostatic abnormalities in children with sepsis. Fifty children between 1-10 years of age admitted with sepsis and 50 age-matched, healthy controls were included in the study. Complete blood counts, examination of stained peripheral blood film, prothrombin time (PT), activated partial thromboplastin time (APTT), plasma fibrinogen, C-reactive protein, liver function tests and serum creatinine were done in all patients and controls. Prolonged PT and APTT were seen in 9 (18%) and 24 (48%) patients respectively. Plasma fibrinogen was decreased in 6% and increased in 8% patients. One or more haemostatic parameter was abnormal in 35 (70%) patients and in all patients who died.

Keywords: haemostasis, children, sepsis, disseminated intravascular coagulation (DIC), India

INTRODUCTION

Systemic inflammatory response syndrome (SIRS) may have an infectious or non-infectious aetiology. Sepsis is said to be present if an infection is proven/suspected in a patient with SIRS.[1] The manifestations of SIRS include two or more of the following: temperature >38°C or <36°C, heart rate >90/min, respiratory rate >20/min along with a total leucocyte count >12 x 10^9/L or >10% immature band forms.[2] These have been modified for use in children.[1] Sepsis remains the most common cause of death in infants and children worldwide as well as in India.[2,3] A prothrombotic state has been observed in patients with sepsis due to activation of coagulation,[4] low levels of endogenous anticoagulants such as Antithrombin III (ATIII), Protein C (PC) and Protein S (PS) and also suppression of fibrinolysis.[5,6] This leads to enhanced fibrin formation, impaired fibrin degradation and intravascular fibrin deposition. While activation of coagulation depletes platelets and coagulation factors contributing to clinical bleeding, formation of micro-thrombi causes organ dysfunction.[7]

Haemostatic abnormalities contribute to the high mortality observed in patients with sepsis.[4] DIC remains an independent predictor of organ failure and death.[8] Early recognition of these abnormalities is vital. There is scant information on the nature of these abnormalities in children (1-10 yrs) from India. Majority of studies were carried out in neonates or adults.[9,10] This study aimed to evaluate the nature and frequency of haematological and haemostatic abnormalities in children with sepsis at admission in a tertiary care centre in India.

MATERIALS AND METHODS

Subjects

Fifty children 1-10 years of age with clinically suspected acute infection and manifestations of SIRS reporting to the department of paediatrics were included in this prospective study. Patients were evaluated in accordance with criteria for acute infection (i.e. symptoms for less than seven days) which was confirmed in all patients by microbiological methods (blood culture and/

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or examination of any other tissue fluid) and/or radiological evidence and/or cerebrospinal fluid (CSF) examination, as required. Only patients who either had a positive culture/radiological evidence of infection/CSF findings suggestive of meningitis were included in the study. Patients who had received antibiotics prior to admission were excluded from the study. Fifty age- matched controls with no clinical/laboratory evidence of infection were also studied. Informed consent was taken from the parent/guardian of all children.

The study received clearance from the institutional ethics committee for human research.

Methods

The following investigations were done on all patients and controls at admission: complete blood counts (automated haematology analyser LH 500), examination of a stained peripheral blood film (Wright's stain) and screening tests of coagulation including PT (Dade Behring Thromborel S), APTT (Dade Behring Actin FS) and plasma fibrinogen (Siemens based on Clauss technique).[11] C-reactive protein (CRP) (latex agglutination, Bioscientifica SA) was also tested. Relevant biochemical investigations were retrieved from the records. All children were followed up till the time of recovery/death/organ dysfunction. Statistical analysis: Unpaired t-test was used for comparison of blood parameters between cases and controls. Level of significance was taken as 5%.

RESULTS

The age of the patients (range 1-10 y, Mean ± SD 6.4 ± 2.7 y) and controls (range 1-10 y, Mean ± SD 6.4 ± 2.7 y) was not significantly different. Each group comprised 32 (64%) males and 18 (36%) females.

Sepsis was diagnosed on a positive culture in 10 (9 on blood culture and 1 on culture of pleural fluid) patients while 10 patients had radiological evidence of infection (X-ray chest showed features of pneumonia) and in 30 patients, infection was diagnosed on examination of CSF. Of the 10 cases positive on blood culture, *Staphylococcus aureus* and *Klebsiella pneumoniae* were identified in 6 and 4 patients respectively.

Table 1 shows the haematological parameters of patients and controls. Haemoglobin (Hb) concentration, red cell count, MCV and MCH were significantly ($p < 0.01$) lower and TLC significantly ($p < 0.01$) higher in patients as compared to controls. Platelet count was higher in patients as compared to controls, but the difference was not statistically significant.

Anaemia (Hb < 11 g/dl) [12] and leucopenia were detected in 38 (76%) and 1 (2%) patients respectively. Twenty (40%) patients had leucocytosis. Thirteen (26%) patients had thrombocytopenia. Thrombocytopenia was mild (platelet count: 100-150 x 10^9/L) in 7 (14%) patients, moderate (platelet count 50-100 x 10^9/L) in 4 (8%) patients and severe (platelet count <50 x 10^9/L) in 2 (4%) patients. Thrombocytosis was seen in 11 (22%) patients. Peripheral blood smear showed lymphocytosis in five (10%) cases and neutrophilia in 10 (20%) cases (Table 2). A shift to left up to myelocyte stage was seen in 12 (24%) cases. Presence of toxic granules and vacuolations was also identified.

An elevated CRP was observed in 17 (34%) patients; being normal in all controls. Haemostatic parameters: Mean ± SD of screening tests of haemostasis are shown in Table 3. PT and APTT were significantly ($p < 0.01$) higher in patients as compared to controls. A prolonged PT was observed in 9 (18%) patients

TABLE 1: Haematological parameters in patients and controls

Parameter (Mean ± SD)	Patients	Controls	p- value
Haemoglobin (g/dl)	9.9 ± 1.7	12.2 ± 1.0	$p < 0.01$
RBC count (x 10^{12}/L)	3.9 ± 0.6	4.4 ± 0.3	$p < 0.01$
MCV (fl)	79.8 ± 9.0	87.8 ± 4.8	$p < 0.01$
MCH (pg)	25.3 ± 3.2	27.9 ± 1.6	$p < 0.01$
TLC (x 10^9/L)	11.3 ± 4.6	6.6 ± 1.3	$p < 0.01$
Platelet count (x 10^9/L)	270 ± 123	246 ± 48	$p = 0.315$

TABLE 2: Abnormal haematological parameters in children with sepsis

Parameter	Patients (%)
Anaemia	38 (76)
Leucopenia	1 (2)
Leucocytosis	20 (40)
Thrombocytopenia	13 (26)
Thrombocytosis	11 (22)

while APTT was prolonged in 24 (48%) patients. Plasma fibrinogen ranged from 109-450 mg/dl and was significantly ($p < 0.01$) higher than controls. Plasma fibrinogen was increased in 8 (16%) and decreased in 3 (6%) patients; being normal in all controls.

One or more haemostatic abnormality was observed in 35 (70%) children with sepsis. An isolated prolonged APTT was the most frequent abnormality seen in 14 (28%) patients. Both PT and APTT were prolonged in 7 (14%) patients. Eight (16%) patients had thrombocytopenia only. One patient had thrombocytopenia in association with prolonged PT and APTT. All 4 screening tests were abnormal in 1 (2%) patient. A prolonged APTT with thrombocytopenia was seen in 2 (4%) patients. Isolated decrease in fibrinogen was seen in 1 (2%) patient.

Organ dysfunction and mortality: nine (18%) patients had liver dysfunction (total bilirubin >2mg/dl or ALT >2 times upper limit of normal) while 4 (8%) patients showed an increase in creatinine concentration (>2 mg/dl) at the time of admission. The mortality in this study was 10%.

In the five patients who died, PT was prolonged in 2 (40%) and APTT was prolonged in 3 (60%) patients. Three (60%) patients had an elevated plasma fibrinogen. All patients who died showed an abnormality in 1 or more haemostatic parameter.

Laboratory evidence of hepatic dysfunction was seen in all 5 (100%) patients and renal dysfunction was seen in 4 (80%) patients who died.

Of the 50 patients included in the study, 5 (10%) died, 1 (2%) had organ dysfunction and 44 (88%) recovered on day 7.

DISCUSSION

Sepsis is the systemic inflammatory response to infection. The reported incidence of sepsis in India is 11-24.5/1000 live births.[3] A study done on 4209 patients including 171 children admitted to 124 ICU's across India showed that 26% patients developed sepsis, many of whom were children.[3] Worldwide it remains the most common cause of death in infants and children.[2] Haemostatic abnormalities including DIC and multiple organ dysfunction syndrome (MODS) are major causes of mortality in these children.[4] Kim *et al* found that *Klebsiella pneumoniae* was the most frequently detected pathogen on blood culture in adult patients with sepsis.[13] In our study, *Staphylococcus aureus* and *Klebsiella pneumoniae* were identified in 6 and 4 patients respectively.

In this study, haemoglobin concentration was significantly ($p < 0.01$) lower in patients as compared to controls. Anaemia was detected in 38 (76%) patients and 4 (8%) controls, the difference being statistically significant ($p < 0.01$). In a survey of ICU admissions, the mean Hb of patients was reported to be 11.3 g/dl with 29% having a Hb of less than 10 g/dl.[14]

TABLE 3: Screening tests of haemostasis in patients and controls

Parameter	Patients		Controls		p-value
	Range	Mean ± SD	Range	Mean ± SD	
PT (seconds)	10-21	13.7 ± 2.0	11-13	12.4 ± 0.5	$p < 0.01$
APTT (seconds)	27-66	41.2 ± 8.0	34-37	35.6 ± 0.7	$p < 0.01$

Anaemia is a common problem in acutely ill patients, especially those who develop sepsis and has been reported in patients with SIRS. Its aetiology is multifactorial: blood loss due to coagulopathy, decreased production or increased destruction of red cells.[15] Anaemia was mild in the majority (86.8%) of patients in this study. Anaemia of SIRS is generally mild with Hb and haematocrit being greater than 8 g/dl and 24% respectively.[15]

In this study, MCV and MCH were significantly ($p < 0.01$) lower in patients as compared to controls. Microcytosis and hypochromia were observed in 27 (54%) and 36 (72%) patients respectively. A previous study on children with infection reported no significant difference between MCH of patients and controls.[14] Owing to a block in the reticulo-endothelial transport of iron, microcytic red cell indices have been reported in one-third of patients with sepsis.[15] This difference could also be due to iron deficiency among Indian children.

In the present study, TLC was significantly ($p < 0.01$) higher in patients than controls and leucocytosis was observed in 40% patients while 2% patients showed leucopenia. Leucocyte alterations are common in patients with sepsis with elevated counts being observed more frequently. A decreased TLC may be seen in children with sepsis.[16] In a study on 101 adult patients with sepsis, TLC increased in 76% and decreased in 6% patients.[13] An increase/decrease in leucocyte count with left shift and presence of immature neutrophils is one of the SIRS criteria used to diagnose sepsis.[1]

Thrombocytopenia was observed in 13 (26%) patients in this study while thrombocytosis was seen in 11 (22%) patients. In a study on 101 adult patients with sepsis, thrombocytopenia was observed in 65.3% patients while thrombocytosis was observed in 8.9% patients.[13] Thrombocytopenia frequently accompanies critical illness and in the ICU, platelet counts $<100 \times 10^9$/L are observed in 20-40% patients.[17] In a study of an ICU population, sepsis was identified as a major risk factor for thrombocytopenia.[18]

Screening tests of haemostasis revealed significantly ($p < 0.01$) prolonged PT and APTT in patients as compared to controls. Prolonged PT and APTT were observed in 9 (18%) and 24 (48%) patients respectively with results being normal in all controls.

Kim *et al* reported prolonged PT and APTT in 70.4% and 52.7% patients respectively.[13] Other studies have observed prolonged PT in 40-93.4% patients.[19,20] A prolonged APTT was observed in 52.7 - 63% patients with sepsis.[13,20] Coagulopathy is frequently seen in patients with sepsis predisposing these patients to the risk of bleeding. In a study on adult patients with sepsis, prolonged PT was more frequent than APTT.[13]

Plasma fibrinogen was increased in 8 (16%) and decreased in 3 (6%) patients. Increase in the level of fibrinogen in sepsis patients has been observed by other authors.[6,21] Fibrinogen is an acute phase reactant and may be elevated in the early stages of sepsis. Kim *et al* observed an increased plasma fibrinogen in 43.5% patients and decreased fibrinogen in 23.5% adult patients with sepsis.[13] Lower fibrinogen levels have also been reported in neonates with sepsis.[9,22] The reduced fibrinogen in patients with acute infection indicates activation of coagulation leading to its consumption.

To conclude, one or more laboratory parameter of haemostasis was abnormal in 35 (70%) children with sepsis. Of the various parameters, isolated prolongation of APTT was most frequent (14/35; 40%). Both PT and APTT were prolonged in 21/35 (60%) patients. This highlights the frequent occurrence of haemostatic abnormalities in children with infection. MODS is the hallmark of severe sepsis and septic shock and is a major cause of the high mortality in these conditions. Prompt diagnosis of organ dysfunction is crucial in identifying patients who may benefit from appropriate therapeutic intervention. Haemostatic abnormalities contribute to organ dysfunction and hence their evaluation is vital in children with sepsis.

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