

Clinical Presentation of COVID-19 and Correlation of Severity with Laboratory Parameters among Children Admitted with Severe Acute Respiratory Infection- An Observational Study

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ABSTRACT

Introduction: On March, 2020, the World Health Organisation (WHO) declared COVID-19 a pandemic caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). In adults it causes mild to severe infections, but in children it usually causes asymptomatic or mild illness. Being a new pandemic, it is prudent to analyse the clinical profile, laboratory parameters and severity interpreters in children to formulate optimal management protocols.

Aim: To determine the clinical and laboratory profile of children hospitalised for Severe Acute Respiratory Infections (SARI) and to evaluate the correlation between clinical severity and laboratory parameters- C-Reactive Protein (CRP), Neutrophil to Lymphocyte Ratio (NLR) and thrombocytopenia.

Materials and Methods: The present study was a prospective observational study which was undertaken for the duration of seven months from 1st April to 30th November 2020. The study included children aged 1 month to 12 years with the criteria of SARI and who were Coronavirus Disease-2019 (COVID-19) positive. Test parameters such as Complete Blood Count (CBC),

CRP, serum electrolytes and kidney function tests were performed at local laboratory as per standard guidelines. The correlation of laboratory parameters (thrombocytopenia, CRP and NLR) with disease severity was done with Pearson's Rho correlation coefficient. Chi-square test was used for statistical analysis.

Results: A total of 118 (10.2%) children were tested positive for COVID-19, 71 (60.17%) were boys and 47 (39.83%) were girls. The commonest clinical symptoms were fever and tachypnea. Gastrointestinal symptoms were found in 54 (45.76%) of the cases. Myocarditis and shock were noticed in 10 (8.47%) children. In 25 (59.52%) of severe COVID-19 cases, NLR was >3.5 and 18 (42.86%) had thrombocytopenia. Seventeen children died of the disease with a mortality rate of 14.41%. Platelet count and NLR ratios were significantly correlated with disease severity ($p \leq 0.05$).

Conclusion: Markers such as NLR and thrombocytopenia which can be used efficiently to assess the severity even in low resource settings, are relevant to a developing country like India. The ratio of neutrophils to lymphocytes can be used as a prognostic marker in resource-constrained settings.

Keywords: Coronavirus disease-2019, C-reactive protein, Infections, Paediatrics, Thrombocytopenia

INTRODUCTION

The SARS-CoV-2 is a virus that causes COVID-19 and has emerged as a deadly pandemic. As per the available surveillance data, children account for upto 5% of cases of the total disease burden that are confirmed in the laboratory [1]. Children have a relatively mild disease with SARS-CoV-2 infection and have a better prognosis than adults [2,3]. This is postulated to be because of lesser expression of Angiotensin Converting Enzyme-2 (ACE-2) receptors in the lung and intestinal epithelium, frequent vaccinations and viral infections leading to trained innate immunity in children and absence of associated co-morbidities as in adults [4]. There are no clearly defined biomarkers to indicate the severity of infection in children [5]. To confirm the diagnosis of COVID-19, nasopharyngeal and oropharyngeal swabs with Reverse Transcriptase-Polymerase Chain Reaction (RT-PCR) are required for the detection of SARS-CoV-2 nucleic acids [6].

As it is a new disease, a widespread compilation of the data from centres across the world would help to propagate the knowledge of epidemiology and clinical presentation in children. This would further facilitate establishing optimal management protocols. Research was conducted to understand the laboratory and clinical presentation and the correlation of the severity with laboratory parameters in children who were admitted in a tertiary care centre. Null hypothesis

was established with no correlation existing between severity and inflammatory biomarkers. This was the first study conducted in Hyderabad, which was useful for low-resource settings to know the severity of illness and to correlate the clinical severity of COVID-19 with the laboratory parameters (thrombocytopenia, CRP and NLR). Primarily, the study objectives were to understand the clinical manifestations and test parameters of COVID-19 in children with SARI and secondarily to correlate the clinical severity of COVID-19 with the laboratory test parameters NLR, CRP and thrombocytopenia.

MATERIALS AND METHODS

This was a prospective observational study conducted for seven months from April 1, 2020 to November 30, 2020 at Niloufer Hospital, Institute for Women and Children's Health, Hyderabad, Telangana, India. Prior to the start of the study, the approval of the Organisational Ethics Committee was obtained (IEC/OMC/M.NO.49 (Acad)/62).

Inclusion criteria: Children aged 1 month to 12 years, hospitalised with SARI and tested for COVID-19 RT-PCR positive were selected for the study.

Exclusion criteria: Neonates, children with congenital respiratory tract infections, other co-morbidities like asthma, cardiac cases

and other bacterial and viral pneumonia cases were excluded from the study.

Sample size calculation: Sample size was calculated as 118 using the below formula. At 95% confidence interval, Z value was 1.96, precision was taken 5% and p-value was 8% [7].

$$Z(1-\alpha/2)^2(p(1-p)/d^2S$$

Putting the values into formula, $n=(1.96)^2 0.08 (1-0.08)/(0.05)^2$

$n=113$, 5% drop-out rate was added and 118 cases were finally taken into study.

Study Procedure

The structured proforma was designed by the Head of the Paediatric Department, who was not included in the research study to avoid any bias. Data was collected which included demographic data, symptoms, examination findings, and associated co-morbidities. Categorisation of the cases was done as per National Institutes of Health (NIH) 2019 Guidelines for COVID-19 [3]. All cases that met the SARI definition were admitted to the isolation wing and tested for COVID-19. Children who were tested positive received treatment according to the standard protocol of COVID Ward. Test parameters such as CBC, CRP, serum electrolytes and kidney function tests were performed at local laboratory as per standard guidelines. According to WHO (2020), SARI is defined as an acute respiratory infection with a history of fever or a measured fever of 38°C or higher, cough or sore throat, difficulty breathing, onset within the last 10 days, and requires hospitalisation [3]. The data was entered in a predesigned proforma.

STATISTICAL ANALYSIS

Statistical analysis of the data was performed using Statistical Package for the Social Sciences (SPSS) version 25.0. Analysis was done using frequency tables and ratios, and continuous data was presented in terms of mean±standard deviation. The categorical data was presented in the form of a frequency distribution table. The correlation of clinical and laboratory parameters with disease severity was done with Pearson's Rho correlation coefficient. The p-value <0.05 was considered to be statistically significant.

RESULTS

During the study duration, 1156 children were admitted with SARI. Among them, 118 children tested positive for COVID RT-PCR accounting for 10.2% of the cases. The mean±SD age of presentation was 3.3±3.72 years.

Out of the total 118 cases, 71 (60.17%) were boys, and 47 (39.83%) were girls. Among them, 55% of the children were under the age of one year. Out of the 118 cases, 77 (65.25%) cases were from rural areas. Analysis of the nutritional status by weight for age showed that 33 (27.07%) of the children were falling below the 3rd centile, and 53 (44.92%) of the children were between the 3rd and 50th centile [Table/Fig-1].

Characteristics	Number (percentage)	
Age (months)	<12	65 (55.08)
	12-60	21 (17.80)
	61-144	32 (27.12)
Sex	Male	71 (60.17)
	Female	47 (39.83)
Residence	Rural	77 (65.25)
	Urban	41 (34.75)
Weight percentile	<3 rd centile	33 (27.97)
	3 rd -50 th centile	53 (44.92)
	>50 th centile	32 (27.11)

[Table/Fig-1]: Distribution as per demographic data.

All the study cases had tachypnea and fever at the time of admission. The predominant presenting symptom was fever, tachypnea, followed by gastrointestinal symptoms. Seizures were seen in 16.1% of children at the time of presentation [Table/Fig-2]. Throat pain (89%), headache (60%), nasal discharge (65%), sneezing (43%), muscle aches (84%) and generalised weakness (90%) were the other features noticed.

Clinical symptom	Number (Percentage)
Fever	118 (100)
Tachypnea	118 (100)
Cough	60 (50.84)
Gastrointestinal symptoms (vomiting, pain abdomen, diarrhea)	54 (45.76)
Seizures	19 (16.10)
Myalgia	10 (8.47)

[Table/Fig-2]: Distribution as per clinical manifestations.

Oxygen saturation was checked for at admission in all the children. On presentation, 43 (36.44%) of the COVID-19 cases had SpO₂ >95%, 30 (25.42%) had SpO₂ of 85-89%, and 12 (10.17%) cases had SpO₂ <85%. A total of 75 children (63.55%) were presented with severe COVID-19 with SpO₂ of <90%, having severe retractions, grunting, lethargy, and seizures [Table/Fig-3]. Myocarditis and shock were noticed in 10 (8.47%) children, in addition to severe pneumonia. Co-morbidities like congenital heart disease, nephrotic syndrome, and cerebral palsy were present in 18 (15.25%) of the cases.

Variables	Number (Percentage)	
SpO ₂	>95%	43 (36.44)
	90-95%	33 (27.97)
	85-89%	30 (25.42)
	<85%	12 (10.17)
Severe COVID-19	Yes	75 (63.55)

[Table/Fig-3]: Oxygen Saturation (SpO₂) at admission.

Analysis of the laboratory parameters had shown that 52 (44.07%) of the children with COVID-19 had anaemia, 5 (4.24%) had leukopaenia, and 34 (28.81%) had more than 15000 total leucocyte count. Platelet count was normal in 70 (59.32%) children, thrombocytopenia was seen in 25 (21.19%), and thrombocytosis was noticed in 23 (19.49%) children. Prerenal Acute Kidney Injury (AKI) was observed among 14 (11.86%) children. In 7 (5.93%) children, the serum creatinine was >1 mg/dL. Electrolyte disturbances were observed in a significant number of them, with hyponatraemia in 30 (25.42%), hypokalemia in 16 (13.56%), and hyperkalemia in 9 (7.63%) cases, respectively [Table/Fig-4].

Among the study population, 45 (38.14%) had NLR of >3.5, while 73 (61.86%) cases had NLR <3.5. NLR of >3.5 was seen in 25 (59.52%) of severe COVID-19 cases compared to 20 (26.31%) of non severe COVID-19 cases, which was statistically significant (z score- 2.3075, p-value- 0.0002) [Table/Fig-5]. Pearson coefficient of correlation was r=0.977 with a p-value <0.00001.

CRP was elevated in 82 (69.49%) of the cases. CRP was positive in 28 (66.67%) of cases with severe COVID-19, compared to 54 (71.05%) in non severe cases (z score- 2.8045, p-value- 0.31 and r=0.541) [Table/Fig-6]. However, quantitative CRP could not be done due to resource constraints.

Thrombocytopenia was observed in 18 (42.86%) of severe COVID-19 as compared to 9 (11.84%) of non severe cases (p- 0.0005). Pearson coefficient of correlation for severity and thrombocytopenia was r=0.871 with p-value <0.0005 [Table/Fig-7]. Around 17 (60.7%) of the cases, who presented with raised CRP were under the age of one year.

Investigation	Mean±SD	Range
Hb (Haemoglobin) (gm/dL)	10.1±2.7	<6: 7 (5.93%) 6-8: 20 (16.95%) 8.1-10: 25 (21.19%) 10.1-12: 37 (31.36%) >12: 29 (24.57%)
TLC (Total Leukocyte Count/mm ³)	12,800±3300/mm ³	<5000: 5 (4.24%) 5000-15000: 79 (66.95%) >15000: 34 (28.81%)
PLT (platelet count in lacs/mm ³)	4±2.1	<2 lacs/mm ³ : 25 (21.19%) 2-5 lacs/mm ³ : 70 (59.32%) >5 lacs/mm ³ : 23 (19.49%)
Sodium (meq/dL)	138	<135: 30 (25.42%) 135-145: 78 (66.10%) >145: 10 (8.48%)
Potassium (meq/dL)	4.8	<3.5: 16 (13.56%) 3.5-5.5: 93 (78.81%) >5.5: 9 (7.63%)
Chloride (meq/dL)	104	>105: 48 (40.68%) 95-105: 63 (53.39%) <95: 7 (5.93%)
Blood urea (mg/dL)	28.6	>40 mg/dL: 22 (18.64%)
Serum creatinine (mg/dL)	0.8	>1 mg/dL: 7 (5.93%)

[Table/Fig-4]: Distribution of laboratory findings.

Severe COVID-19 illness	Neutrophil lymphocyte ratio		Total	p-value
	<3.5	≥3.5		
Yes	17 (40.48%)	25 (59.52%)	42	p=0.0002 (Significant)
No	56 (73.68%)	20 (26.32%)	76	
Total	73 (61.86%)	45 (38.14%)	118 (100%)	

[Table/Fig-5]: Neutrophil Lymphocyte Ratio in the study subjects.

Severe COVID-19	CRP levels		Total	p-value
	Raised	Not raised		
Yes	28 (66.67%)	14 (33.33%)	42	p=0.31 (Not significant)
No	54 (71.05%)	22 (28.95%)	76	
Total	82 (69.49%)	36 (30.51%)	118 (100%)	

[Table/Fig-6]: CRP levels in the study subjects.

Severe COVID-19 illness	Platelet count (lacs/cumm)			Total	p-value
	<2	2-5.5	>5.5		
Yes	18 (42.86%)	19 (45.24%)	5 (11.90%)	42	p=0.0005 (Significant)
No	9 (11.84%)	48 (63.16%)	19 (25.00%)	76	
Total	27 (22.88%)	67 (56.78%)	24 (20.34%)	118 (100%)	

[Table/Fig-7]: Platelet count in the study subjects.

Association of the lab parameters (platelet count and NLR) with COVID-19 severity: The laboratory parameters were correlated with the disease severity, out of which, platelet count and NLR showed a statistically significant positive correlation with severe COVID-19 with p-values of less than 0.05 ($r=0.871$), and 0.00029 ($r=0.977$), respectively.

Of the total 118 cases, 101 (85.5%) children were discharged and 17 (14.41%) children died. Out of these 17 children, 10 (58.82%) were under the age of one year and 8 (57.14%), had an NLR of >3.5.

DISCUSSION

Early in the pandemic, there were few reported cases of COVID-19 in children, but as the time passed, more cases were identified. Little is known about this disease, which has a wide range of clinical manifestations, from asymptomatic to fatal. Despite the few reported deaths in otherwise healthy children, it is reassuring that children have a relatively smooth course of the disease in comparison to adults. Investigators collected the data in 2020 when novel paediatric and neonatal cases were noticed in developing countries.

The present study was performed to verify the clinical presentation, and the correlation between disease severity and laboratory parameters in hospitalised children. In the present study, among 1156 children admitted with SARI during the study period, 118 (10.2%) cases tested positive for COVID-19. The mean age of presentation was 3.3 ± 3.72 years, which was comparable to those of Dong Y et al., and Wei M et al., [6,8], whereas the mean age was higher (6.7 years) in the study by Lu X et al., [9]. Infants were more affected in this study, accounting for 55% of cases and a higher proportion of severe COVID-19 cases. Male preponderance was observed in the present study similar to that of Dong Y et al., and Wei M et al., [6,8]. In the current study, 27.97% of children were malnourished at weights below the 3rd percentile. The most common symptom observed in this study were fever (100%) and tachypnea (100%), followed by cough (50.84%) and stomachache (45.76%), similar to those of Jiehao C et al., and de Souza TH et al., [10,11].

In the present study, 52 (44.07%) of the children with COVID-19 had anaemia, 5 (4.24%) children had reduced leukocyte counts and 25 (21.19%) had thrombocytopenia. Normal leucocyte counts were observed in 79 (66.95%) children, comparable to the study done by Henry BM et al., and where they found in 69.6% of paediatric cases [12].

Thrombocytopenia was observed in 18 (42.86%) cases of severe COVID-19 as compared to 9 (11.84%) cases of non severe cases. The correlation between thrombocytopenia and disease severity was found to be statistically significant, similar to a study by Bashash D et al., [13]. This meta-analysis revealed that non severe cases have a significantly higher number of platelets and showed that the probability of the emergence of thrombocytopenia is significantly higher in the severe cases with the pooled mean difference of -21.5 (95% CI: -31.57, -11.43).

A NLR >3.5 was observed in 45 (38.14%) children and its correlation with disease severity was statistically significant in present study. A meta-analysis done by Lagunas-Rangel FA where patients with COVID-19 who had severe disease were found to have significantly higher NLR values (SMD=2.404, 95% CI=0.98-3.82) [14]. The study done by Sarangi B et al., showed good correlation ($r=0.35$, $p=0.01$) [15].

In the present study, raised CRP was found in 82 (69.49%) children a minimal prevalence of increased CRP when compared to that in adults, suggesting a comparatively milder immunological response in children and less immune damage [16]. CRP was positive in 54 (71.05%) of the non severe cases, and its correlation with severity was not statistically significant, similar to a study by Saleh NY et al., and Chen L et al., [17,18]. This may be because of the lower sample size in both studies. The current study's mortality rate was 14.41% because there was a greater proportion of severe cases compared to other studies where mortality was low due to a greater proportion of less severe cases, 1.4% in Guan WJ et al., study and 5.6% in Sena GR et al., study [19,20].

Limitation(s)

This study was conducted in a tertiary care facility where the incidence of COVID-19 in children could not be calculated because only cases with dyspnea needed to be hospitalised. The duration and sample size of the study was limited. The correlation of other inflammatory markers such as ferritin, D-Dimer, and IL-6 levels with disease severity could not be done due to resource constraints. Quantitative CRP could not be done due to resource constraints.

CONCLUSION(S)

In the present study, the predominant presenting symptom was fever (100%) and tachypnea (100%). About 44.07% children with

COVID-19 had anaemia. Thrombocytopenia was seen in 21.18% and 38.13% had NLR of >3.5. Platelet count and NLR showed a statistically significant positive correlation with severe COVID-19. Markers such as NLR and thrombocytopenia which can be used efficiently to assess the severity even in low resource settings are relevant to a developing country. NLR and thrombocytopenia markers are helpful in early referral. The ratio of neutrophils to lymphocytes can be used as a prognostic marker in resource-constrained settings. More research is required to establish low-cost precise early inflammatory markers to estimate severity for prompt referral.

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PROFORMA

Name:

Case/Control no.:

Address:

Sex: Male / Female Hosp. No.:

Weight:

Phone no.:

Mobile no.:

Clinical Symptoms:

Fever

Tachypnea

Cough

Gastrointestinal symptoms- vomiting, pain abdomen, diarrhoea

Seizures

Myalgia

others

SpO₂ saturation at admission**Investigations**

CRP: CBP: Platelets: N/L Ratio:

Blood culture: Chest x-ray: Band cell ratio:

Renal Parameters

Na, K, CL, Blood Urea Serum Creatine

Treatment Outcome

Discharged home/Died/AMA

Date of discharge: