Original Article

Serum Ferritin as a Predictor of Disease Severity in Dengue: A Prospective Observational Study

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Abstract

Background: Dengue is a major mosquito-borne viral illness affecting children in tropical and subtropical countries. Although most infections are self-limiting, a proportion of patients progress to severe dengue with complications such as plasma leakage, shock, bleeding, and organ dysfunction. Early identification of children at risk of severe disease remains a clinical challenge. Serum ferritin, an acute-phase reactant and marker of immune activation, has emerged as a potential biomarker for predicting disease severity. The aim is to evaluate whether serum ferritin can be used as an early predictor of dengue severity in pediatric patients. The objective is to predict early complications and identify children requiring close monitoring using serum ferritin as a prognostic marker, and to determine the correlation between serum ferritin levels and dengue severity. **Material and Methods:** This prospective observational study was conducted over 18 months in the Department of Paediatrics, Bokaro General Hospital, Bokaro, Jharkhand. A total of 100 children aged 2 months to 12 years with NS1 antigen-positive dengue infection were enrolled. Clinical details, laboratory investigations, and serum ferritin levels measured by particle-enhanced immunoturbidometric assay on Day 3 and Day 4 of illness were recorded. Patients were classified into non-severe and severe dengue. Statistical analysis was performed using SPSS version 27.0. Continuous variables were compared using the independent Student's t-test, while categorical variables were analyzed using the Chi-square or Fisher's exact test. A p-value <0.05 was considered statistically significant. **Results:** Among the 100 patients, 79% had non-severe dengue and 21% had severe dengue. The mean age was 7.00 ± 3.18 years, with a slight male predominance (52%). Shock was observed in 15% of patients, bleeding in 3%, and organ involvement in 5%. The mean serum ferritin level on Day 3 was significantly higher in severe dengue than in non-severe dengue (842.33 ± 106.61 ng/mL vs. 385.35 ± 87.71 ng/mL; p<0.0001). Similarly, the mean Day 4 serum ferritin level was significantly higher in severe dengue (1307.24 ± 111.08 ng/mL) compared with non-severe dengue (554.76 ± 149.87 ng/mL; p<0.0001). Serum ferritin categories on both Day 3 and Day 4 showed a highly significant association with dengue severity (χ^2=85.53 and 86.03, respectively; p<0.0001). Children with severe dengue also had a significantly longer duration of fever than those with non-severe disease (5.33 ± 0.73 vs. 4.70 ± 0.70 days; p=0.0004). **Conclusion:** Serum ferritin demonstrated a strong positive association with dengue severity and was significantly elevated in children who developed severe disease. Measurement of serum ferritin during the early phase of illness may serve as a simple, accessible, and reliable biomarker for early risk stratification, enabling prompt identification of high-risk patients and facilitating timely monitoring and appropriate clinical intervention.

Keywords: Dengue, Serum ferritin, Severe dengue, Children, Biomarker, Disease severity, Prognostic marker.

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Introduction

Dengue is one of the most rapidly expanding mosquito-borne viral infections worldwide and continues to pose a major public health challenge, particularly in tropical and subtropical regions. The disease is caused by four antigenically distinct serotypes of the dengue virus (DENV-1 to DENV-4), transmitted primarily by Aedes aegypti mosquitoes. According to the World Health Organization (WHO), nearly half of the world's population lives in areas at risk of dengue, with an estimated 100–400 million infections occurring annually. Clinical manifestations range from asymptomatic infection and uncomplicated dengue fever to severe dengue characterized by plasma leakage, severe bleeding, shock, and multiorgan dysfunction, which may be fatal if not recognized and managed promptly.[1]

Dengue has emerged as one of the most important viral infections affecting children in developing countries, including India. The increasing frequency of outbreaks has been attributed to rapid urbanization, population growth, climate change, globalization, and inadequate vector control measures. The disease places a considerable burden on healthcare systems because of seasonal epidemics and the unpredictable progression

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from mild illness to severe dengue. Therefore, early identification of children at risk for severe disease remains an important clinical priority to facilitate timely intervention and reduce morbidity and mortality.[2]

The pathogenesis of severe dengue is complex and involves an exaggerated host immune response rather than direct viral cytopathic effects alone. Secondary dengue infection, antibody-dependent enhancement, activation of T lymphocytes, complement pathways, and excessive release of inflammatory cytokines contribute to endothelial dysfunction and increased vascular permeability. These immunological events lead to plasma leakage, thrombocytopenia, coagulopathy, shock, and organ impairment. Although platelet count, hematocrit, and liver enzymes are routinely used to monitor disease progression, these parameters often become abnormal only after significant pathological changes have occurred, thereby limiting their value as early predictors of disease severity.[3]

Several investigators have therefore explored novel biomarkers capable of identifying patients likely to develop severe dengue during the early febrile phase. Among these, serum ferritin has attracted considerable attention because of its role as both an intracellular iron-storage protein and an acute-phase reactant. During systemic inflammation, activated macrophages, hepatocytes, and monocytes release ferritin into the circulation in response to pro-inflammatory cytokines such as interleukin-6, tumor necrosis factor-α, and interferon-γ. Consequently, elevated serum ferritin levels reflect the intensity of immune activation and inflammatory response, making ferritin a potential biomarker for severe infectious diseases, including dengue.[4,5]

Hyperferritinemia has been reported in several inflammatory and hypercytokinemic conditions such as sepsis, macrophage activation syndrome, hemophagocytic lymphohistiocytosis, and severe viral infections. In dengue, elevated ferritin levels are believed to result from macrophage activation and cytokine-mediated inflammatory pathways that accompany severe disease. Excessive ferritin production may therefore mirror the underlying immune dysregulation responsible for capillary leakage, thrombocytopenia, hepatic dysfunction, and circulatory collapse. These observations suggest that serum ferritin could serve as an early laboratory indicator of disease severity before overt clinical deterioration becomes evident.[5]

Van de Weg and colleagues demonstrated that patients with severe dengue exhibited significantly higher serum ferritin concentrations than those with uncomplicated dengue, with ferritin levels closely correlating with immune activation markers and disease severity. Their study suggested that hyperferritinemia may represent an important surrogate marker of the cytokine storm associated with severe dengue infection, supporting its potential utility in early risk stratification and clinical decision-making.[6]

Similarly, Soundravally and Hoti reported that serum ferritin concentrations were markedly elevated in patients with severe dengue and were significantly associated with plasma leakage, thrombocytopenia, hepatic involvement, and adverse clinical outcomes. Their findings indicated that ferritin possesses good prognostic value for predicting disease progression and may help clinicians identify patients requiring intensive monitoring during the early stages of illness.[7]

Despite growing evidence supporting the role of serum ferritin as a prognostic biomarker, prospective pediatric data from eastern India remain limited. Regional differences in host immunity, circulating viral serotypes, and disease epidemiology may influence ferritin responses and disease severity. Therefore, the present prospective observational study was undertaken to evaluate the usefulness of serum ferritin as an early predictor of dengue severity in children admitted to the Department of Paediatrics, Bokaro General Hospital, Jharkhand, and to determine its association with clinical outcomes and disease severity.

Materials and Methods

Study Design: The present study will be conducted as a prospective observational hospital-based study to evaluate the usefulness of serum ferritin in predicting the severity of dengue among pediatric patients. The study aims to determine whether serum ferritin measured during the early phase of illness can identify children at increased risk of developing severe dengue and its associated complications.

Study Area: The study will be conducted in the Department of Paediatrics, Bokaro General Hospital, Bokaro, Jharkhand, a tertiary care teaching hospital catering to pediatric patients from Bokaro district and surrounding regions.

Study Duration: The study will be carried out over a period of 18 months, following approval from the Institutional Ethics Committee.

Study Population: The study population will include children admitted to the Department of Paediatrics who are diagnosed with dengue infection and satisfy the predefined eligibility criteria.

Sample Size: A total of 100 pediatric patients will be included in the study.

The sample size has been calculated using the formula:

[n=\frac{4pq}{L^2}]

Where:

- n = Required sample size
- p = Prevalence of dengue = 21.2% (0.212)
- q = 1 − p = 0.788
- L = Allowable error (8.17%)

Calculation:

- p = 0.212
- q = 0.788
- 4pq = 4 × 0.212 × 0.788 = 0.6682
- L^2 = 0.00668

Therefore,

n = 0.6682 / 0.00668 = 100

Thus, the calculated sample size is 100 patients, providing approximately 89% study power with 95% confidence interval.

Sampling Technique

A consecutive sampling technique will be employed. All eligible patients admitted during the study period who satisfy the inclusion criteria and provide informed consent from parents or legal guardians will be recruited consecutively until the required sample size is achieved.

Inclusion Criteria
- Children aged 2 months to 12 years.
- NS1 antigen-positive dengue infection.
- Patients presenting with fever with or without classical clinical features of dengue.
- Parents or legal guardians willing to provide written informed consent.

Exclusion Criteria
- Patients without serological evidence of NS1 antigen positivity.
- Fever duration greater than five days at presentation.
- Hemoglobin level below 10 g/dL.
- Patients with transfusion-dependent chronic diseases.
- Patients diagnosed with hemochromatosis.
- Parents or guardians unwilling to participate.

Study Procedure: After obtaining informed consent, all eligible patients will undergo detailed clinical evaluation.

The following information will be recorded using a predesigned structured proforma:
- Demographic characteristics
- Detailed clinical history
- Duration of fever
- Warning signs
- Bleeding manifestations
- Shock
- Hepatomegaly
- Ascites
- Pleural effusion
- Duration of hospitalization
- Requirement of intensive care
- Clinical outcome

Patients will be classified according to the WHO 2009 Dengue Classification into:
- Dengue without warning signs
- Dengue with warning signs
- Severe dengue

Blood Sample Collection: Approximately 4 mL of venous blood will be collected under aseptic precautions in a clot activator vacutainer.

The collected blood sample will be centrifuged to separate serum.

Approximately 500 µL of serum will be utilized for serum ferritin estimation.

Serum Ferritin Estimation: Serum ferritin concentration will be measured using Particle Enhanced Immunoturbidometric Assay (PEITA) on an automated clinical chemistry analyzer according to the manufacturer's instructions.

Other Laboratory Investigations

The following investigations will also be performed as part of routine patient management:
- Complete blood count
- Hemoglobin
- Hematocrit
- Total leukocyte count
- Differential leukocyte count
- Platelet count
- Liver function tests
- Renal function tests
- Serum electrolytes
- Coagulation profile (where indicated)
- NS1 antigen test
- Dengue IgM/IgG antibody (where clinically indicated)
- Chest radiograph (if indicated)
- Ultrasonography for ascites or pleural effusion (if indicated)

Outcome Measures

Primary Outcome
- Early prediction of severe dengue and complications using serum ferritin levels.

Secondary Outcomes
- Correlation of serum ferritin with dengue severity.
- Correlation with thrombocytopenia.
- Correlation with plasma leakage.
- Correlation with duration of hospitalization.
- Requirement of intensive care.
- Clinical outcome.

Data Collection Tool: A standardized, pretested case record form will be used to collect all demographic, clinical, laboratory, treatment, and outcome variables.

Statistical Analysis: The collected data will be entered into Microsoft Excel and analyzed using SPSS software version 26.0. Continuous variables will be expressed as mean ± standard deviation (SD) or median with interquartile range, depending on data distribution.

Categorical variables will be presented as frequency and percentage.

Statistical tests will include:
- Independent Student's t-test
- Mann–Whitney U test
- Chi-square test
- Fisher's exact test
- One-way ANOVA
- Pearson's correlation coefficient
- Spearman's rank correlation
- Receiver Operating Characteristic (ROC) curve analysis for determining optimal serum ferritin cutoff values
- Multivariate logistic regression to identify independent predictors of severe dengue

A p-value <0.05 will be considered statistically significant.

Ethical Considerations: Prior approval will be obtained from the Institutional Ethics Committee before commencement of the study. Written informed consent will be obtained from the parents or legal guardians of all participating children. Patient confidentiality will be maintained throughout the study, and all collected data will be used solely for research purposes.

Results

A total of 100 pediatric patients with NS1 antigen-positive dengue infection were enrolled in the present study. The demographic profile, clinical characteristics, serum ferritin levels, and their association with dengue severity were analyzed. Among the study participants, 79 (79%) had non-severe dengue, whereas 21 (21%) developed severe dengue according to the study criteria.

Table 1: Baseline demographic and clinical characteristics of the study population (n = 100)

Variable	Frequency (%) / Mean ± SD
Age group	
<5 years	30 (30.0%)
>5 years	70 (70.0%)
Mean age (years)	7.00 ± 3.18
Median age (years)	7
Age range (years)	1–12
Gender	
Male	52 (52.0%)
Female	48 (48.0%)
Mean duration of fever (days)	4.83 ± 0.75
Fever present	100 (100%)
Dengue severity	
Non-severe dengue	79 (79.0%)
Severe dengue	21 (21.0%)

The study included 100 children with laboratory-confirmed dengue. The majority (70%) were older than five years, while 30% were below five years of age. The overall mean age was 7.0 ± 3.18 years, indicating that school-aged children formed the largest proportion of the study population. Male children (52%) marginally outnumbered females (48%). Fever was the presenting symptom in all participants with an average duration of 4.83 ± 0.75 days. Nearly one-fifth (21%) progressed to severe dengue, whereas 79% had non-severe disease.

Table 2: Clinical complications observed among study participants

Clinical variable	Frequency	Percentage
Shock		
Present	15	15.0%
Absent	85	85.0%
Bleeding manifestations		
Present	3	3.0%
Absent	97	97.0%
Fluid accumulation with respiratory distress		
Present	1	1.0%
Absent	99	99.0%
Organ involvement		
Liver	4	4.0%
CNS	1	1.0%
None	95	95.0%

Most children recovered without major complications. Shock was the commonest severe manifestation, affecting 15% of patients. Bleeding manifestations were relatively uncommon and were observed in only 3% of cases. Fluid accumulation associated with respiratory distress occurred in 1% of patients, indicating that respiratory compromise was rare. Organ involvement was identified in 5% of children, with hepatic involvement (4%) being more frequent than central nervous system involvement (1%). Overall, 95% of patients had no clinically evident organ dysfunction.

Table 3: Distribution of serum ferritin levels on Day 3 and Day 4

Serum ferritin category (ng/mL)	Day 3 n (%)	Day 4 n (%)
<300	9 (9.0%)	0 (0%)
301–600	66 (66.0%)	61 (61.0%)
601–800	10 (10.0%)	15 (15.0%)
801–1200	—	2 (2.0%)
>800 (Day 3) / >1200 (Day 4)	15 (15.0%)	22 (22.0%)
Mean ferritin (ng/mL)	481.32 ± 208.21	712.78 ± 339.23
Median (ng/mL)	412	576

Serum ferritin concentrations increased as the illness progressed. On Day 3, two-thirds (66%) of children had ferritin values between 301 and 600 ng/mL, whereas 15% already demonstrated markedly elevated levels exceeding 800 ng/mL. By Day 4, ferritin concentrations showed a substantial upward shift. No child had ferritin below 300 ng/mL, while 22% exhibited levels exceeding 1200 ng/mL. The mean ferritin concentration increased from 481.32 ± 208.21 ng/mL on Day 3 to 712.78 ± 339.23 ng/mL on Day 4, suggesting progressive inflammatory activation during dengue infection.

Table 4: Association of serum ferritin and fever duration with dengue severity

Variable	Non-severe dengue (n=79)	Severe dengue (n=21)	p-value
Fever duration (days)	4.70 ± 0.70	5.33 ± 0.73	0.0004
Serum ferritin Day 3 (ng/mL)	385.35 ± 87.71	842.33 ± 106.61	<0.0001
Serum ferritin Day 4 (ng/mL)	554.76 ± 149.87	1307.24 ± 111.08	<0.0001
Chi-square (Day 3 ferritin categories vs severity)	—	—	χ^2=85.53, p<0.0001
Chi-square (Day 4 ferritin categories vs severity)	—	—	χ^2=86.03, p<0.0001

Children with severe dengue demonstrated significantly higher serum ferritin concentrations than those with non-severe disease. The mean Day 3 ferritin level in severe dengue (842.33 ± 106.61 ng/mL) was more than twice that observed in non-severe dengue (385.35 ± 87.71 ng/mL) ($p < 0.0001$). Likewise, the Day 4 ferritin concentration increased dramatically to 1307.24 ± 111.08 ng/mL among severe cases compared with 554.76 ± 149.87 ng/mL in non-severe cases ($p < 0.0001$). Children with severe dengue also experienced a significantly longer duration of fever (5.33 ± 0.73 days) than those with non-severe dengue (4.70 ± 0.70 days) ($p = 0.0004$).

Categorical analysis further demonstrated a highly significant association between elevated ferritin levels and disease severity. On Day 3, 71.4% of severe dengue cases had ferritin values exceeding 800 ng/mL, whereas 83.5% of non-severe cases had values between 301 and 600 ng/mL ($\chi^2 = 85.53$; $p < 0.0001$). By Day 4, 95.2% of severe dengue patients exhibited ferritin levels above 1200 ng/mL, while 77.2% of non-severe patients remained within the 301–600 ng/mL range ($\chi^2 = 86.03$; $p < 0.0001$).

Discussion

Early identification of children at risk of developing severe dengue remains one of the greatest challenges in pediatric practice. Although clinical warning signs and conventional laboratory parameters such as thrombocytopenia and hematocrit are routinely used for monitoring, they often become abnormal only after significant disease progression. The present prospective observational study evaluated the role of serum ferritin as an early biomarker for predicting dengue severity among children. Our findings demonstrated that serum ferritin levels measured on both Day 3 and Day 4 were significantly higher in severe dengue than in non-severe dengue, indicating that hyperferritinemia is closely associated with disease severity.

In the present study, 21% of patients developed severe dengue, while 79% had non-severe disease. The mean serum ferritin level on Day 3 was 842.33 ± 106.61 ng/mL in severe dengue compared with 385.35 ± 87.71 ng/mL in non-severe dengue ($p<0.0001$). Similarly, on Day 4, mean serum ferritin levels increased to 1307.24 ± 111.08 ng/mL in severe dengue compared with 554.76 ± 149.87 ng/mL in non-severe dengue ($p<0.0001$). Moreover, on Day 3, 71.4% of the severe dengue case had high ferritin levels of more than 800 ng/mL, and on Day 4, 95.2% had high level of more than 1200 ng/mL on Day 4, which showed strong association between the progressively higher levels of ferritin and the severity of clinical disease.

The observations are in line with the systematic evaluation of various clinical studies by Shukla et.al, who found that high levels of serum ferritin was a reliable indicator of severe dengue and that patients with plasma leakage, hemorrhage, shock and organ dysfunction (fundamental clinical categories) had higher levels of ferritin. The authors highlighted that ferritin has a good ability to discriminate the cases in the early phase of severe disease, and suggested that it should be used as a component of the risk stratification model in clinical practice.[8]

A prospective observational study by Golhar et al. in children showed the serum ferritin level to be independently associated with the severity of the disease and in-hospital outcomes. Patients with a higher level of ferritin had longer hospital stays, more warning signs, thrombocytopenia and more cases of severe dengue than patients with a lower level of ferritin. The data is similar to the present study, which indicated that serum ferritin level was significantly and highly increased in severe dengue and fever duration was significantly longer.[9]

Similar results were also seen in Goyal et al. who found that serum ferritin levels were significantly raised in the patients with severe dengue compared to the uncomplicated dengue cases. They found that ferritin levels were highly predictive of severe illness in their study and estimated that measuring ferritin levels as early as the beginning of fever may help doctors in selecting who might need intensive care. The level of association found in our study between ferritin categories and dengue severity ($p<0.0001$) further reinforces these observations.[10]

This prospective cohort study, by Jha et al., specifically analysed children who developed severe dengue and found that markedly high serum ferritin was linked to poor clinical outcomes, such as shock, intensive care admission, longer hospitalisation and higher risk of death. The authors suggested that ferritin was a useful biomarker in the diagnosis of dengue in children due to its association with macrophage activation and cytokine mediated inflammation. In our study, 15% of the patients experienced shock, and high serum ferritin concentrations were more common among patients who experienced shock, which is indicative of the biologic plausibility of this association.[11]

Mahendranath and Langade also found that serum ferritin in early stage of dengue was also predictive of progression to severe dengue. Patients who develop complications had significantly higher ferritin levels than patients with uncomplicated dengue fever, reported they. Their results agree with our observation that children with ferritin > 800 ng/mL (Day 3) and < 1200 ng/mL (Day 4) had a much higher risk of developing severe dengue.[12]

In the same way, Sekhar compared ferritin in the blood with severity of dengue in children, and concluded that increased concentration of ferritin in the blood was highly correlated to severe dengue. The study made it recommendable that ferritin determination should be a part of a routine clinical assessment and should be simple, inexpensive and widely available and can be used as a tool to identify high-risk patients prior to the onset of any overt complications. Our observations are supported by

the results and thus confirmed the clinical usefulness of ferritin estimation in dengue in children.[13]

Ghosh et al. have recently validated serum ferritin as a reliable marker of severity of dengue. The ferritin levels were observed to be progressively higher in the worsening clinical stage, high inflammatory activity and also with an increase in the frequency of complications. As in our study, a progressive rise in ferritin was seen from Day 3 to Day 4 with a highly significant correlation of ferritin with severe dengue.[14]

Iramain et al. studied serum ferritin as an additive marker to help to identify dengue severity in the pediatric emergency department and found that ferritin level measured early was helpful for determining children who need close monitoring and aggressive supportive treatment. They concluded that using ferritin alongside clinical assessment provides improved early triage and management choices especially during seasonal outbreaks when hospital resources are restricted. In a similar study, high levels of ferritin were also shown to indicate children at higher risk for severe disease in the early febrile stage.[15]

Deora et al. summarized the importance of hyperferritinemia regarding severe pediatric dengue, its correlation with cytokine storm, activation of macrophages, myocardial dysfunction and circulatory shock. They suggested that ferritin is not just a marker of inflammation, but a marker of the underlying immune dysregulation, that may explain the severity of the disease symptoms. In our study, occurrence of shock, liver involvement, and elevated ferritin level adds extra clinical evidence confirming this proposed pathophysiological pathway.[16]

Last but not least, de Azeredo et al. explained the intricate relationship between inflammatory mediators, thrombocytopenia, endothelial dysfunction and coagulation abnormalities that occur during dengue infection. Their research showed the links between excessive inflammatory activation and platelet destruction and vascular leakage, which are closely associated with the serum ferritin concentrations. Our study showed that very high ferritin levels correlate well with the severity of dengue and this is in line with this concept, as ferritin may be used to reflect the degree of systemic inflammation during the process of the disease.[17]

The present study shows that serum ferritin level in early phase of illness is strongly correlated with the severity of dengue in children overall. The marked differences between severe and non-severe dengue and the progressive increase in ferritin levels along with its correlation with the occurrence of complications like shock suggest that serum ferritin could be a useful prognostic biomarker. Routine estimation of serum ferritin is therefore helpful to identify high-risk patients early, thus providing opportunity for timely referral, closer monitoring and timely supportive management until the onset of life-threatening complications.

Conclusion

The finding of the present study is that serum ferritin is a useful early marker, which would help to predict the severity in children with dengue. The serum ferritin level was significantly higher at Day 3 and 4 in patients with severe dengue vs non-severe patients. Duration of fever, shock, organ involvement and severity of clinical symptoms were all highly correlated with increased ferritin levels. This is congruent with recent evidence showing increased immune activation and inflammatory response during the course of dengue infection, which is reflected by an elevated ferritin level. Since the measurement of serum ferritin is simple, readily available and relatively inexpensive, it can be used as part of the clinical routine to stratify the early risk of children with dengue. Recognition of children at higher risk for severe disease would allow for more frequent monitoring, timely intervention at the appropriate time, and support appropriate referral and therefore, better clinical outcomes. More multicenter studies with larger patient populations are suggested to define standard ferritin cut-off levels for routine clinical use and to test its prognostic value in various pediatric populations.

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Conflicts of interest

There are no conflicts of interest.

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