Section: Paediatrics

Original Research Article

EVALUATING SERUM FERRITIN AS A DUAL MARKER OF JOINT DISEASE ACTIVITY AND IRON STATUS IN PATIENTS WITH HEMOPHILIA

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ABSTRACT

Background: Serum ferritin's role as an acute phase reactant complicates its interpretation in patients with hemophilia. This study aimed to evaluate serum ferritin as a dual marker of joint disease activity and iron status in this population. **Materials and Methods:** This cross-sectional study was conducted in August 2025 among 27 male patients with hemophilia at a tertiary care centre in Tamil Nadu, India. An invalidated provisional composite joint disease activity score, based on clinical parameters of Annual Joint Bleeding Rate, presence of target joints and deformities was developed to categorize joint disease activity. Blood samples were analysed for complete hemogram and serum ferritin. Statistical significance was analysed at p-value < 0.05. **Result:** The mean age of the 27 participants was 27.21 (SD 14.52) years, with 70.37% having Hemophilia A and 29.63% Hemophilia B. The mean serum ferritin was 115.44 (SD 86.17) ng/ml and was significantly higher with increasing age. Anemia was present in 33.33% of patients, but only one case was associated with low ferritin (<30 ng/ml), pointing more towards anemia of inflammation in the study group. No statistically significant correlation between serum ferritin level and joint disease activity was found in this study. **Conclusion:** Serum ferritin is an unreliable marker of joint disease activity and iron status when used in isolation. Its interpretation requires a complete iron panel and other inflammatory markers for accurate clinical assessment. Considering the limitations of the present study including small sample size, use of an invalidated provisional composite joint disease activity score and lack of joint imaging findings, these findings should be considered preliminary. Studies in a larger cohort would be required to make definite conclusions.

INTRODUCTION

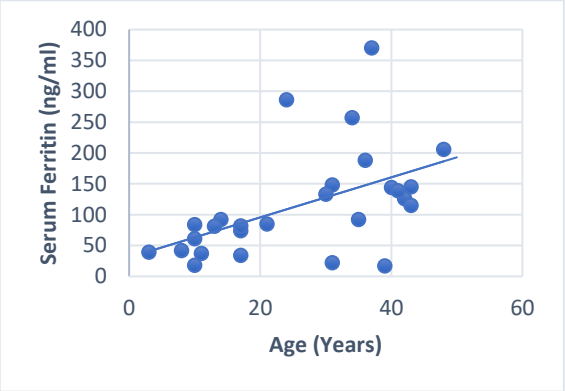
Hemophilia is an X linked congenital disorder of clotting factor deficiency characterised by multiple bleeding episodes particularly in the major joints. According to World Federation of Hemophilia Annual Global Survey, the estimated number of males worldwide with hemophilia is 1,125,000.[1] India has only 20,778 registered patients with hemophilia which is less than 15% of the estimated number of patients.[2] Recurrent hemarthrosis and resulting arthropathy are major causes of disability and long-term morbidity in patients with hemophilia, resulting in loss of quality of life and financial constraints especially in resource limited settings. Joints with three or more bleeding in a six month period are target joints and are prone to permanent long term arthropathy if not treated adequately. Recurrent joint bleeding can lead to pain, limitation of range of movements, synovitis, synovial hyper trophy, cartilage changes, bone remodelling, muscle atrophy, osteoporosis and debilitating osteoarthritis. Patients with hemophilia are also prone to anemia due to iron deficiency due to chronic iron loss from

recurrent bleeding from multiple sites. Serum ferritin has a dual role in patients with hemophilia for assessing iron status as well as marker for inflammation, particularly subclinical synovitis. Serum ferritin's role as an acute phase reactant leads to challenges in interpretation of serum ferritin levels in patients with hemophilia. The clinical utility of serum ferritin to simultaneously perform the dual role as marker of inflammation and iron status particularly in resource limited settings is not well established. This study was done with a primary objective of assessing the correlation of serum ferritin level with joint disease activity and iron status in patients with hemophilia. The secondary objective was to estimate the prevalence of anemia among patients with hemophilia and categorize them based on serum ferritin levels.

MATERIALS AND METHODS

This cross-sectional study was undertaken in August 2025 at the Integrated Centre for Hemophilia and Hemoglobinopathies functioning at Kanyakumari Government Medical College and Hospital, Tamil Nadu, India. A total of 27 patients with hemophilia registered and treated in the centre were included in the study. Patients with hemophilia who had acute infections, chronic kidney disease, chronic liver disease, iron supplements intake in the past 6 months, blood transfusions in the past one year, life threatening bleeding or hospitalisation in the past four weeks were excluded from the study. Demographic, clinical and bleeding details were collected in a predesigned questionnaire. Socioeconomic status was classified as per Modified Kuppusamy's Socioeconomic Status Scale. Blood samples were collected by phlebotomy and analyzed in the laboratory for complete hemogram and serum ferritin. Complete hemogram was performed using automated analyzer and serum ferritin tested by immunoturbidimetry based on latex agglutination reaction. Anemia was defined by age appropriate WHO cut off. [children under 5 years: <11 g/dL. 5-11 years: <11.5 g/dL, 12-14 years: <12 g/dl. men ≥ 15 years: <13 g/dL). VanderMeulen (2018) indicates that employing a serum ferritin cut off of less than 30 ng/ml in patients with inherited bleeding disorders to define iron deficiency enhances the sensitivity to 92% while maintaining a specificity of 98%.(3) On this basis, serum ferritin cut off of less than 30 ng/ml was used to define iron deficiency in this study to minimize the risk of missing iron deficiency in this population with underlying chronic inflammation.

For the purpose of this study, a provisional Composite Joint Disease Activity Score was developed to evaluate joint disease activity based on simple clinical parameters (Annual Joint Bleeding Rate, Target Joints and Deformities). Joint disease activity was categorized as Mild, Moderate or Severe based on the score (Table 1). This provisional composite score was created for this study as it relies on simple, readily available clinical parameters that do not require specialised equipment, training or imaging skills. This was designed to be a simple alternative suitable for the study's specific setting and context and allowed for a standardised method of classifying joint disease activity among the study participants. The score is currently invalidated, which is acknowledged as a major limitation of this study. The validation of the score against standard joint health scores and imaging findings is planned for future studies. Data collected were analysed by suitable statistical methods using SPSS 25 software. Categorical data were summarized as frequencies and percentages. Continuous variables were presented as mean and standard deviation. Independent samples t-test was used to compare the means between two groups, and a one-way ANOVA was used for comparisons across three or more groups. When the assumption of normality was not met, the non-parametric equivalents, the Mann-Whitney U test (for two groups) and the Kruskal-Wallis test (for three or more groups), were employed. Pearson correlation was used to assess linear association between continuous variables. A p-value of less than 0.05 was considered statistically significant.



Pearson correlation coefficient (r) = 0.38, p-value = 0.0485 (<0.05) (statistically significant)
Figure 1 - Scatter Plot of Age and Serum Ferritin

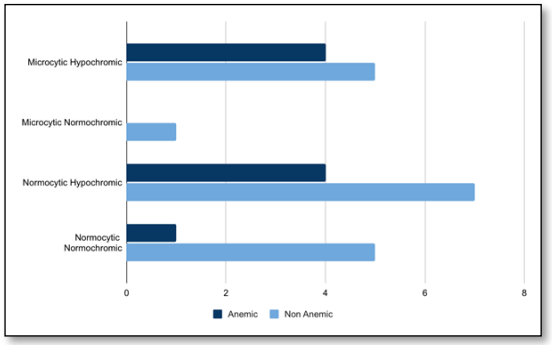


Figure 2 - Distribution of RBC morphology among anaemic and non anaemic patients

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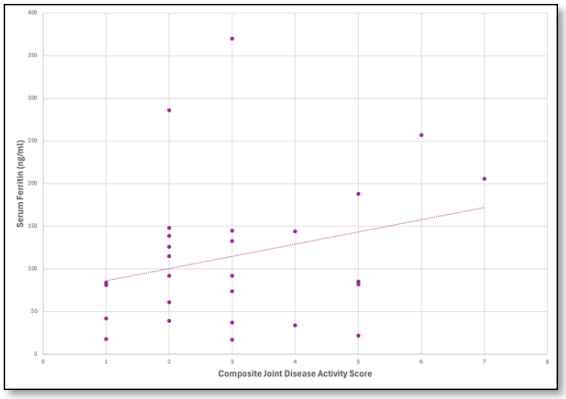

Pearson correlation coefficient (r) = 0.27, p-value = 0.1 (> 0.05) (not significant)

Figure 3 - Scatter Plot of Composite Joint Disease Activity Score and Serum Ferritin

Table 1: Composite Joint Disease Activity Score

Parameter	Score
Annual Joint Bleeding Rate	
< 15	1
15 to 30	2
> 30	3
Target Joint	
None	0
One Joint	1
≥ 2 Joints	2
Joint Deformity	
None	0
One Joint	1
≥ 2 Joints	2
Joint Disease Activity	**Total Score**
Mild	1 to 2
Moderate	3 to 4
Severe	5 to 7

RESULTS

27 patients with hemophilia registered at the Integrated Centre of Hemophilia and Hemoglobinopathies functioning at Kanyakumari Government Medical College and Hospital were included in the study. All the study participants were male with mean age 27.21 (SD 14.52) years. All the study participants belonged to nuclear family and were non-vegetarian. The demographic and clinical characteristics of the study participants are as shown in Table 2.

Of the 27 patients with hemophilia enrolled, 19 patients (70.37%) had hemophilia A while 8 patients had hemophilia B (29.63%). Three of the patients (11.11%) were inhibitor positive and were on treatment with bypassing agents. Of the 27 patients, six patients (22.22%) were enrolled in the hemophilia prophylaxis programme while the rest were receiving episodic therapy.

Table 2. Demographic and Clinical Characteristics of the Study Participants

	Number (n = 27)	Percentage
Age		
1 to 15 years	8	29.63%
15 to 30 years	6	22.22%
31 to 50 years	13	48.15%
> 50 Years	0	0%
Socioeconomic Status		
Class I	0	0%
Class II	5	18.52%
Class III	14	51.85%
Class IV	8	29.63%
Class V	0	0%
Hemophilia Type		
A	19	70.37%
B	8	29.63%
Severity		

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Mild	4	14.81%
Moderate	2	7.41%
Severe	21	77.78%
Inhibitor		
Absent	24	88.89%
Present	3	11.11%
Type of treatment		
Prophylaxis	6	22.22%
Episodic	21	77.78%

The mean serum ferritin of the 27 study participants was 115.44 (SD 86.17) ng/ml. Mean serum ferritin according to demographic and clinical details of study participants are shown in Table 3. The mean serum ferritin was observed to be significantly higher with increasing age (Figure 1). There was no statistically significant correlation of serum ferritin with other parameters.

Table 3: Mean Serum Ferritin among Demographic and Clinical Groups

	Number (n = 27)	Mean Serum Ferritin (SD) (ng/ml)	p-value
Age			
1 to 15 years	8 (29.63%)	56.75 (26.78)	0.04338 (Significant)
15 to 30 years	6 (22.22%)	115.67 (89.21)	
31 to 50 years	13 (48.15%)	151.46 (93.02)	
> 50 Years	0	-	
Socioeconomic Status			
Class I	0		0.23 (Not Significant)
Class II	5 (18.52%)	105.40 (103.39)	
Class III	14 (51.85%)	119.50 (69.56)	
Class IV	8 (29.63%)	114.63 (111.27)	
Class V	0	105.40 (103.39)	
Hemophilia Type			
A	19(70.37%)	126.26 (98.78)	0.32 (Not Significant)
B	8 (29.63%)	89.75 (37.65)	
Severity			
Mild	4(14.81%)	91.75 (85.25)	0.64 (Not Significant)
Moderate	2(7.41%)	79.00 (7.07)	
Severe	21(77.78%)	123.43 (90.82)	
Inhibitor			
Absent	24(88.89%)	121.25 (87.55)	0.37 (Not Significant)
Present	3 (11.11%)	69.00 (68.83)	
Type of treatment			
Prophylaxis	6 (22.22%)	69.67 (27.31)	0.13 (Not Significant)
Episodic	21(77.78%)	128.52 (93.05)	

Of the 27 patients, 9 (33.33%) were anemic according to WHO cut off. Only one patient (3.70%) had iron deficiency anemia while the rest of the anemic patients (29.63%) had serum ferritin more than 30 ng/dl. Similarly, 2 patients (7.41%) had iron deficiency without anemia. RBC morphology findings of the patients in the anemic and non anemic groups are presented in (Figure 2).The details of Annual Joint Bleeding Rate (AJBR), Target Joints and Deformities among the study participants with mean ferritin levels are shown in Table 4. No statistically significant association of serum ferritin with the joint disease parameters were observed. Overall mean ferritin levels did not show any statistical significance with Composite Joint Disease Activity Score (Table 4).

Table 4: Mean Serum Ferritin and Joint Disease Activity

	Number (n = 27)	Mean Serum Ferritin (SD) (ng/ml)	p-value
Annual Joint Bleeding Rate			
< 15	15 (55.55%)	114.07 (97.90)	0.757 (Not Significant)

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15 to 30	5 (18.52%)	94.60 (38.33)	
> 30	7 (25.93%)	133.29 (89.98)	
Target Joint			
None	6 (22.22%)	64.67 (39.65)	0.109 (Not Significant)
One Joint	12 (44.45%)	108.75 (71.71)	
≥ 2 Joints	9 (33.33%)	158.22 (109.64)	
Joint Deformity			
None	22 (81.48%)	114.09 (84.22)	0.889 (Not Significant)
One Joint	5 (18.52%)	121.40 (104.73)	
≥ 2 Joints	0 (0%)	0	
Composite Joint Disease Activity Score			
Mild	12 (44.45%)	102.58 (70.87)	0.7 (Not Significant)
Moderate	9 (33.33%)	116.22 (107.97)	
Severe	6 (22.22%)	140.00 (90.18)	

The mean serum ferritin in the 9 patients with anemia was 70.22 (SD 40.70) ng/ml and in the non-anaemic group was 138.06 (SD 94.62) ng/ml. Five of the non-anaemic patients had serum ferritin levels above 150 ng/ml with a mean serum ferritin level of 261.40 (SD 72.24) ng/ml. All these five patients had target joints with three patients having 2 target joints, one with 1 joint and one with 3 joints.

The mean ferritin levels among anaemic and non-anaemic study participants categorized according to iron status is shown in Table 5. Joint disease activity among anaemic and non-anaemic study participants based on the preliminary Composite Joint Disease Activity Score is presented in Table 6.

Table 5: Mean Serum Ferritin According to Anemia and Iron Status

	Number (n = 27)	**Mean Serum Ferritin (SD) (ng/ml)**
Anaemic (n = 9)		
Serum Ferritin <30 ng /ml	1 (3.70%)	17
Serum Ferritin > ≥30 ng /ml	8 (29.63%)	76.88 (37.92)
Not Anaemic (n = 18)		
Serum Ferritin <30 ng /ml	2 (7.41%)	20 (2.83)
Serum Ferritin > ≥30 ng /ml	16 (59.26%)	152.81 (89.75)

Table 6: Composite Joint Disease Activity Score and Anemia

Composite Joint Disease Activity Score	**Anaemic (n=9)**	**Not Anaemic (n=18)**	**p-value**
Mild (n=12)	4 (44.45%)	8 (44.45%)	1 (Not Significant)
Moderate (n=9)	3 (33.33%)	6 (33.33%)	
Severe (n=6)	2 (22.22%)	4 (22.22)	

An exploratory subgroup analysis was initially done on non-anaemic patients with normal iron status (n=16). This initial analysis revealed a statistically significant correlation between the Composite Joint Disease Activity Score and serum ferritin (p-value 0.0059). However, recognizing that age also significantly correlated with ferritin levels in our study, a partial regression analysis was performed to control for age as a potential confounder. The results of the analysis showed no statistically significant correlation between serum ferritin level and joint disease activity ($r = 0.161$, $p = 0.567$) when adjusted for age. This indicates that the initial significant association observed was due to the influence of age and not the joint disease activity. A similar raising trend of serum ferritin with joint disease severity was observed in anaemic patients with normal iron status, which was not statistically significant (Table 7). Pearson correlation coefficient between Composite Joint Disease Activity Score and Serum Ferritin was 0.27, with a p-value of 0.1 (p-value > 0.05). This indicates a weak positive correlation that is not statistically significant (Figure 3)

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Table 7: Correlation Of Joint Disease Activity and Serum Ferritin in Anaemic and Non Anaemic Subgroups with Normal Iron Status (Serum Ferritin ≥30 ng/ml)

Composite Joint Disease Activity Score	Mean Ferritin (SD) (ng/ml)	p-value
Non Anaemic with Normal Iron Status (Serum Ferritin ≥30) (n= 16)		
Mild (n=7)	94.50 (55.08)	0.567 (Not Significant)
Moderate (n=6)	98.50 (46.97)	
Severe (n=3)	228.75 (56.50)	
Anaemic with Normal Iron Status (Serum Ferritin ≥30) (n= 8)		
Mild (n=4)	112.25 (86.03)	0.2 (Not Significant)
Moderate (n=2)	138.33(135.42)	
Severe (n=2)	166.50 (97.58)	

As shown in Table 8, the mean ferritin level among the patients with hemophilia seemed to rise with increasing physical activity, but this was not statistically significant.

Table 8: Ferritin Level and Physical Activity

Physical Activity	(n=27)	Mean Ferritin (SD) (ng/ml)	p-value
Sedentary	2 (7.41%)	47.50 (19.09)	0.526 (Not Significant)
Mild	11 (40.74%)	118.82 (99.34)	
Moderate to Vigorous	14 (51.85%)	122.50 (80.36)	

DISCUSSION

The prevalence of anemia was 33.33% (9 patients) in our study. The mean serum ferritin among anemic patients was 70.22 (SD 40.70) ng/ml. Only one of the 9 anemic patients had serum ferritin below 30 ng/ml. A study among 50 hemophilia patients at Chennai, Tamil Nadu observed a much higher prevalence of anemia in patients with hemophilia (72%) though the mean ferritin level was within the lower limits of normal in all the patients.[4]

Mutlaq Al-Wataify AS (2022) in a study among 60 hemophilia patients from Iraq aged 6 months to 15 years observed a prevalence of iron deficiency in 58.3% of patients and iron deficiency anemia in 28.3% of the patients.[5] They used a serum ferritin cut off of less than 7 ng/ml to define iron deficiency which is lower than the cut off of 30 ng/ml used in our study. Using similar serum ferritin cut off of less than 7 ng/ml, Kumari et al (2023) in a study among 130 paediatric Hemophilia A patients observed prevalence of iron deficiency and iron deficiency anemia at 24.6% and 53.84%.[6] Using WHO cut off of serum ferritin levels less than 12 µg/l for patients younger than 5 years and less than 15 µg/l for patients older than 5 years, Ahmed S et al observed an overall prevalence of iron deficiency of 48.7% among 39 Hemophilia A patients in northern Nigeria with a higher incidence in those with severe diseases.[7] The prevalence of iron deficiency and iron deficiency anemia were lesser in our study at 7.41% and 3.70% despite using a much higher serum ferritin cut off to define iron deficiency at 30 ng/ml as suggested by VanderMeulen et al (2018).[3] This much lesser incidence than that reported in literature, was probably because all the study participants were non vegetarians with more inclusion of heme iron in diet. They also had easy access to hemophilia treatment and were on regular follow up. Also our study lacks an age and sex matched control group which would have helped to compare the prevalence of anemia among healthy male from the same geographic region. These findings therefore cannot be generalised to all hemophilia patients.

Considering the role of ferritin as an acute phase reactant, care must be exerted in using ferritin as a marker of iron deficiency in these patients as inflammation can coexist in hemophilia patients. The addition of other investigations of the iron panel which are not influenced by inflammation are suggested for confirmation of iron deficiency. VanderMeulen (2018) suggests using transferrin saturation as a more reliable test to distinguish anemia of inflammation from iron deficiency anemia.[3] Mahajan G et al (2017) explored the role of hepicidin in diagnosis of anemia of chronic disease and iron deficiency in children.[8]

The majority of anaemic patients in the study had serum ferritin levels greater than 30 ng/ml with varied RBC morphology. This interpretation guides clinicians in considering other causes of anemia in these patients including anemia of inflammation instead of attributing anemia to iron deficiency alone. For clinicians, this would mean that an anaemic patient with hemophilia cannot be assumed to have normal iron stores based on serum ferritin levels alone. A comprehensive iron panel would help clinicians guide appropriate treatment modalities in these patients and avoid unindicated iron

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supplements. A statistically significant increase in serum ferritin with increasing age has been observed in the study, which could be probably due to the inclusion of the six younger participants in the prophylaxis programme and the cumulative burden of joint diseases in older patients. Six of the study participants between the ages 10 to 17 years were part of the prophylaxis program. Prophylaxis has been associated with lesser bleeds compared to episodic therapy which in turn results in lesser inflammation and ferritin levels. Also, ferritin level might be higher in older individuals due to other age related factors unaccounted for in this study. The earliest complications of hem arthrosis is synovitis, and is characterized by synovial hypertrophy, migration of inflammatory cells, and a high degree of neo-angiogenesis with subsequent bleeding. The pathogenic processes interact ultimately leading to a fibrotic joint.[9] Several bio markers have been investigated as diagnostic and prognostic markers of hemophiliac joint disease process. The systematic review of 220 articles investigating the role of bio markers in evaluating joint health in hemophilia arthropathy by Van Bergen ED (2021) concluded that though Biomarkers may reflect pathophysiological processes, translation into daily clinical practice comes with multiple pitfalls and damages.[10] Toenges R (2021) in a comparative study of immunological and biomarkers parameters in patients with hemophiliac arthropathy and rheumatoid arthritis observed a statistical significant lower ferritin in patients with hemophiliac arthroplasty compared to patients with rheumatoid arthritis and higher ferritin compared to control group.[11]

Primary analysis of the entire group of 27 patients with hemophilia showed no statistical significance of serum ferritin with joint disease activity. On exploratory subgroup analysis, a statistically significant correlation between serum ferritin levels and joint disease activity was observed in the subgroup of non-anaemic patients without iron deficiency. However, a further partial regression analysis controlling for age as a confounder indicated no significant association between serum ferritin level and joint disease activity in this subgroup. This indicated that the initial finding was influenced by age associated changes in serum ferritin observed in the study. No statistically significant correlation was observed in the anaemic patient's subgroup without iron deficiency. All the study participants with serum ferritin above 150 ng/ml had a minimum of one target joint. These findings highlight the limitation of using ferritin as an isolated marker of joint inflammation in patients with hemophilia. Considering the smaller sample size of the sub-group, further research in this cohort may be required to evaluate the possibility of using serum ferritin as a marker of sub clinical joint involvement. Saleh El-Alfy M et al (2024) in a cross-sectional comparative study of 38 hemophiliacs with and without arthropathy, observed no significant difference in serum ferritin levels among hemophilia patients with or without arthropathy. However, TNF alpha levels were significantly higher in hemophilia patients with arthropathy compared to those without arthropathy and hence was suggested as a surrogate bio marker for bleeding episodes.[12] Longitudinal studies with follow up of the same cohort over time would help to understand the dynamics of serum ferritin in homophilic joint disease over time. The study's primary limitation is the small sample size and that all patients were under regular follow up in a tertiary care centre. This limits the generalizability of the findings to a large extent as the majority of the patients with hemophilia in resource limited settings have poor access to hemophilia care. One of the major drawbacks of this study is the use of an invalidated provisional Composite Joint Disease Activity Score created for the purpose of the study to evaluate joint disease activity. This score has not been compared with established joint health scores or radiological findings. Considering the use of an invalidated score, the findings of the study regarding joint disease activity may be considered preliminary and hypothesis generating rather than conclusive. The next critical step would be to validate the provisional Composite Joint Disease Activity Score developed for the study. If validated against standard clinical scores and radiological findings, this score would serve as a valuable tool to evaluate joint disease activity especially in resource limited settings. Also, only clinical parameters related to joint disease activity in hemophilia (Annual Joint Bleeding Rate, Target Joints and Presence of Deformities) have been used in the study without radiological confirmation by ultrasonography or Magnetic Resonance Imaging. Studies including other inflammatory markers and iron study parameters unaffected by inflammation along with serum ferritin level will provide more insights into the clinical utility of serum ferritin in patients with hemophilia.

CONCLUSION

Primary analysis of 27 patients with hemophilia found no statistically significant correlation of serum ferritin with joint disease activity. Significantly higher ferritin levels were observed with increasing age of the participants. While anemia was present in 33.33% of the patients, only one anaemic patient had low serum ferritin pointing more towards anemia of inflammation in this group. The findings of the study underscore the limitations of interpreting serum ferritin in isolation. Serum ferritin should be used along with complete iron studies and other inflammatory markers to assess iron status and joint disease activity. However, considering the small sample size, use of an invalidated provisional Composite Joint Disease Activity Score and lack of joint imaging findings, more longitudinal studies in a larger cohort would be required to make definite conclusions.

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