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IRON OVERLOAD, COMPROMISED SALIVARY FUNCTION, AND DENTAL CARIES RISK IN BETA-THALASSEMIA MAJOR: A CROSS-SECTIONAL STUDY

Amilia Ramadhani[1], Hafizh Zufar Pramuditya[2], Putri Silvia Nurcahyani[2], Ali Taqwim[3], Muhammad Basalamah[4], A. Haris Budi Widodo[1], Lantip Rujito[5]

1. *Department of Oral Biology, School of Dentistry, Faculty of Medicine, Jenderal Soedirman University, Central Java, Indonesia*
2. *School of Dentistry, Faculty of Medicine, Jenderal Soedirman University, Central Java, Indonesia*
3. *Department of Pediatric Dentistry, School of Dentistry, Faculty of Medicine, Jenderal Soedirman University, Central Java, Indonesia*
4. *Department of Pediatric, Banyumas District General Hospital, Central Java, Indonesia*
5. *Department of Molecular Biology, Faculty of Medicine, Jenderal Soedirman University, Central Java, Indonesia*

Correspondence: amilia.ramadhani@unsoed.ac.id

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ABSTRACT

Background: β-thalassemia major patients exhibit higher dental caries prevalence due to systemic complications including iron overload from regular blood transfusions. This study aimed to assess dental caries status, salivary pH, and buffer capacity in β-thalassemia major patients and investigate correlations with ferritin blood concentration.
Method: A cross-sectional analytical study was conducted on 24 β-thalassemia major patients aged 12-17 years. Salivary function was assessed by salivary pH and buffer capacity. Dental caries status was measured using the DMF-T index. Ferritin blood concentrations were correlated with salivary function and dental caries using Pearson analysis.
Outcome: Mean salivary pH was 5.74±0.63 (acidic), buffer capacity was 3.98±0.21, DMF-T score was 6.17±1.97, and ferritin level was 5830.54±2823.91 ng/mL. Ferritin levels showed moderate negative correlations with salivary pH ($r=-0.718$, $p<0.001$) and buffer capacity ($r=-0.737$, $p<0.001$), and positive correlation with DMF-T scores ($r=0.696$, $p<0.001$). Strong negative correlations were found between DMF-T scores and salivary pH ($r=-0.915$, $p<0.001$) and buffer capacity ($r=-0.913$, $p<0.001$).
Conclusion: Iron overload in patients with β-thalassemia major is associated with impaired salivary parameters and increased dental caries experience. These findings suggest that elevated ferritin levels may contribute to an altered oral environment, potentially elevating caries risk. The results underscore the importance of integrating oral health monitoring into comprehensive thalassemia management protocols.

INTRODUCTION

Beta-thalassemia represents an inherited hematological disorder resulting from mutations that affect β-globin chain synthesis, leading to reduced or absent β-hemoglobin chain in erythrocytes. The main forms of β-thalassemia, based on genotype, include β-thalassemia major, β-thalassemia inter-

media, and β-thalassemia minor.[1]

While global β-thalassemia prevalence varies significantly by geographic region, with the highest rates observed in Mediterranean and Asian population,[2] Indonesian data indicates carrier rates of 3-10% resulting in approximately 2500 annual with β-thalassemia major.[3]

Severe anemia, caused by ineffective erythropoiesis, leads to oral manifestations commonly seen in patients with β-thalassemia major. The increased production of erythrocytes causes bone marrow expansion, which enlarges the maxilla. This can result in malocclusion, maxillary protrusion, dental diastema, overbite, and open bite.[4,5] Epidemiological investigations consistently demonstrate that the prevalence of dental caries in thalassemia patients is higher.[5–11] However, the contributing factors to dental caries in β-thalassemia major vary significantly across studies. Transfusion-induced iron accumulation promotes oxidative damage within salivary gland tissues, compromising secretory function and altering oral environmental parameters.[1]

Iron overload from transfusion therapy may compromise oral health in β-thalassemia major patients, yet these relationships remain poorly characterized. We examined the status of dental caries, as well as the salivary pH and buffer capacity, and ferritin blood level in patients with β-thalassemia major. This study investigates the correlation between ferritin blood concentration, salivary pH, buffer capacity, and dental caries in these patients. The findings will provide insights into the mechanisms underlying oral health deterioration in this vulnerable population and inform evidence-based preventive strategies.

RESEARCH METHODS

This cross-sectional analytical descriptive study was conducted in November 2020 at Banyumas District General Hospital, with approval from the Ethical Board of Banyumas District General Hospital (No. 105/KEPK-RSUDBMS/X/2020). A total 24 β-thalassemia patients of Banyumas District General Hospital were selected through purposive random sampling as participants in this study.

The inclusion criteria consisted of participants who met the following conditions: a confirmed diagnosis of β-thalassemia major, aged between 12 and 17 years, receiving regular blood transfusions and iron chelation therapy, and a written confirmation of informed consent provided by the participant and their legal guardians. The exclusion criteria included participants with salivary gland disorders, systemic conditions known to affect caries risk (such as diabetes mellitus or congenital heart disease), recent antibiotic use (within the past two weeks), or inability to comply with study procedures.

To standardize baseline salivary conditions and reduce dietary confounders, participants underwent a two-hour fasting period prior to sample collection. After rinsing the oral cavity with distilled water to remove any food debris, unstimulated whole saliva was collected using passive

drooling technique. Participants were instructed to maintain a standardized seated posture, with a slight head declination, to facilitate gravity-assisted collection. The saliva was collected over a five-minute protocol, with intervals of one minute documented to ensure an adequate sample volume while promoting compliance among the pediatric participants.[12]

Immediate post-collection pH determination was performed using calibrated digital pH meter to prevent CO_2-mediated pH drift. Salivary buffering capacity assessment followed modified Ericsson protocols, with sample volumes adjusted based on collection yield (0.5-1.0 mL). For acid challenge testing, a 5mM hydrochloric acid was used, with standardized 10-minute equilibration periods implemented to ensure complete CO_2 degassing and accurate buffering capacity quantification.[13]

The Decayed, Missing, and Filled Teeth (DMF-T) index was evaluated based on 28 permanent teeth by calibrated observers. Serum ferritin concentrations were obtained as secondary data from pre-transfusion routine blood examinations within 3 months prior to study enrollment and documented in medical records.

Statistical analysis utilized SPSS 22.0 with Shapiro-Wilk normality testing for our sample (n=24). Pearson correlation coefficients quantified linear relationships between ferritin levels, salivary parameters (pH, buffer capacity), and caries indices (DMF-T). Statistical significance was set at $\alpha=0.05$ for all analyses, with correlation coefficients and their corresponding p-values reported to assess the clinical relevance of observed relationships in this β-thalassemia cohort.

RESEARCH FINDINGS

This research investigated the relationship between salivary parameters, dental caries status, and ferritin blood levels in β-thalassemia major patients. Twenty-four subjects aged 12-17 (mean age 14.54 ± 1.53 years) were included in the analysis. The descriptive statistics of all parameters are presented in Table 1.

The mean salivary pH was 5.74 ± 0.63, ranging from 4.14 to 6.56, indicating a tendency toward acidic saliva among the study participants. The salivary buffer capacity averaged 3.98 ± 0.21, with values ranging from 3.58 to 4.31. The dental caries status, represented by DMF-T scores, showed a mean of 6.17 ± 1.97, with minimum and maximum values of 3.00 and 11.00, respectively. The ferritin blood levels were markedly elevated, with a mean of 5830.54 ± 2823.91 ng/mL, ranging from 851.00 to 12000.00 ng/mL, confirming iron overload in these patients.

Pearson correlation analysis revealed significant correlations between ferritin blood levels and all studied parameters (Table 2). There was a moderate negative correlation between ferritin blood levels and both salivary pH ($r = -0.718$, $p < 0.001$) and buffer capacity ($r = -0.737$, $p < 0.001$), as well

as a moderate positive correlation with DMF-T scores ($r = 0.696$, $p < 0.001$). These results indicate that iron overload is associated with impaired salivary function and increased dental caries.

Additionally, a strong negative correlation was observed between salivary pH and DMF-T scores ($r = -0.915$, $p < 0.001$), suggesting that lower salivary pH values are linked to a higher prevalence of caries. Similarly, salivary buffer capacity exhibited a strong negative correlation with DMF-T scores ($r = -0.913$, $p < 0.001$), indicating that reduced buffering ability is related to increased caries experience (Table 3).

Table 1. The minimum, maximum, mean and standard deviation of age, salivary pH, buffer capacity, dental caries status (DMF-T score) and iron serum concentration

	N	Minimum	Maximum	Mean	Std. Deviation
Age	24	12	17	14.54	1.532
Salivary pH	24	4.14	6.56	5.74	0.63
Salivary Buffer Capacity	24	3.58	4.31	3.98	0.21
DMF_T	24	3.00	11.00	6.17	1.97
Ferritin blood level (ng/mL)	24	851.00	12000.00	5830.54	2823.91

Table 2. Pearson correlation of salivary pH, buffer capacity, dental caries status (DMF-T score), and iron serum concentration

		Salivary pH	Salivary Buffer Capacity	DMF_T
Ferritin Blood Level	Pearson Correlation	-.718**	-.737**	-.696
	Sig. (2-tailed)	<0.001	<0.001	<0.001
	N	24	24	24

Table 3. Pearson correlation of salivary pH, buffer capacity, and dental caries status (DMF-T score)

		Salivary pH	Salivary Buffer Capacity
DMF_T	Pearson Correlation	-.915**	-.913**
	Sig. (2-tailed)	<0.001	<0.001
	N	24	24

DISCUSSION

The present study reveals that patients with β-thalassemia major exhibit significantly high prevalence of dental caries which correlate positively with elevated serum ferritin levels and associated with reduced salivary pH and buffering capacity. These findings indicate a strong interplay between systemic iron overload and oral health deterioration.

Patients with thalassemia require regular red blood cell transfusions (once or twice a month) to maintain a hemoglobin concentration of 9.0-10.5 g/dL. Multiple transfusions over time result in considerable iron overload because the human body lacks an excretion mechanism for excess iron. Iron overload exceeded transferrin's capacity to bind iron within cells and in the plasma compartment.

The presence of 'free iron' in cells or plasma destroys lipid membranes, organelles, and DNA, leading to cell death and fibrosis.[1]

The mean ferritin level of 5830.54 ng/mL observed in our study is substantially elevated, exceeded 500 ng/mL, indicating significant iron overload despite chelation therapy.[14] The association between serum ferritin level and excessive dental caries index related to previous studies[6–9] that showed correlation between β-thalassemia major and dental caries due to increase salivary and serum iron level.

Several mechanisms may elucidate the relationship between elevated ferritin levels and dental caries. Iron overload promotes the formation of non-transferrin-bound iron (NTBI), a highly reactive species that is readily taken up by the acinar and ductal cells of salivary gland via transporter Divalent Metal Transporter 1 (DMT1), ZRT/IRT-like Protein 14 (ZIP14), and calcium channels[15,16]. This cellular uptake appears to contribute to the positive correlation between serum and salivary ferritin concentrations.[17] Excess intracellular iron promotes oxidative stress through the Fenton reaction, generating reactive oxygen species that can damage oral tissues, including salivary glands.[4,18,19] The iron overload causes degeneration of acinar cells of the submandibular gland, characterized by cytoplasmic vacuolation and nuclear pyknosis of acinar cells and interacinar hemorrhage. The ductal cells of submandibular gland also appeared pyknotic and flattened. The iron deposition in salivary glands may cause chronic inflammation, impair the salivary flow rate, and decrease the production and secretion of bicarbonate ions.[6,18,20,21]

Our findings demonstrated a strong negative correlation between serum ferritin level and salivary buffer capacity ($r= -0.737$, $p<0.001$), and DMF-T scores ($r = -0.913$, $p < 0.001$), highlighting the effect of iron toxicity in buffering systems and caries risk. The mean buffer capacity of 3.98 in our study population is considerably lower than values reported for healthy individuals.[22]

Saliva's buffering capacity is primarily attributed to three systems: bicarbonate, phosphate, and protein systems, with bicarbonate being the most significant contributor.[23] The compromised buffering ability observed in our study has significant clinical implications. Without adequate neutralization the acid-base homeostasis, the prolonged periods of low pH in the oral cavity occur.[23] The mean salivary pH of 5.74 found in our beta-thalassemia major patients is notably lower than the normal salivary pH range of 6.7-7.4 reported in healthy individuals. This finding is consistent with recent research[6,9,20,22] reported significantly lower salivary pH values in thalassemia patients compared to healthy controls.

The acidic salivary environment creates favorable conditions for acidogenic and aciduric bacteria, particularly *Streptococcus mutans* and *Lactobacilli*, which are primary cariogenic pathogens. These microorganisms thrive in low pH environments and further contribute to acid production through fermentation of dietary carbohydrates, establishing a vicious cycle that promotes dental caries

development.[24,25] Excessive iron can be incorporated into bacterial metabolism, enhancing the growth and virulence of cariogenic bacteria. Akbari et al.,[26] demonstrated that high salivary iron concentrations were associated with increased colonization and acid production by *Streptococcus mutans*.

The strong correlations between salivary parameters and dental caries underscore the need for comprehensive oral health assessment as an integral part of thalassemia management protocols. Regular monitoring of oral environment pH and acid-neutralizing ability might provide beneficial prognostic indicators for caries development in affected patients. Collaboration between hematologists and dental professionals is essential for comprehensive patient care.[27] Integration of oral health education into thalassemia management programs could improve patients' awareness and self-care practices.

Evidence-based personalized caries prevention protocols in β-thalassemia patients may minimize invasive dental treatment. A comprehensive preventive protocol may include the following components: (a) improving oral hygiene practices through the use of fluoride toothpaste and dietary counseling that promotes the intake of non-cariogenic and high-antioxidant foods while also considering appropriate iron-rich nutrition[6,19,27,28]; (b) regular dental visits—ideally every three months—for early caries detection via caries risk assessment, professional prophylaxis, topical fluoride applications, and targeted probiotic therapy to restore microbiota balance disrupted by iron overload;[29–31] and (c) salivary stimulation strategies, such as sugar-free chewing gum containing xylitol, to enhance salivary flow and buffering capacity impaired in iron-overloaded individuals.[32,33]

Before treating dental caries in a patient with β-thalassemia major, the dentist should identify several precautions, including: (a) the type of thalassemia; (b) recent levels of haemoglobin and iron; (c) the administration chelating agent; and (d) any systemic complications such as splenomegaly or a history of splenectomy.[19] This integrated approach addresses both the oral and systemic complications of β-thalassemia, supporting better oral health outcomes and reducing the reliance on invasive treatments in this high-risk population.

Multiple constraints must be acknowledged while analyzing the findings from this investigation. The observational nature of this research restricts our capacity to determine causative links among the factors examined. Longitudinal studies offer a superior understanding of how oral fluid alterations and caries development occur in thalassemia patients. The sample size, while adequate for detecting significant correlations, may not capture the full spectrum of variability in this patient population. Larger multi-center studies would enhance the generalizability of the findings. Additionally, inclusion of a healthy control group would allow for more direct comparisons of salivary parameters between thalassemia patients and the general population.

CONCLUSION

This study demonstrates a strong pathophysiological relationship between systemic iron overload and increased dental caries in β-thalassemia major patients. The compromised salivary parameters create an acidogenic oral environment that favors cariogenic bacterial proliferation and enamel demineralization. These findings underscore the critical need for integrating oral health monitoring into routine thalassemia management protocols. Salivary parameters can serve as predictive biomarkers for caries risk assessment in this patient population.

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