BULLETIN OF STOMATOLOGY AND MAXILLOFACIAL SURGERY
Volume 21, Issue 8

DOI: 10.58240/1829006X-2025.21.8-126

ORIGINAL RESEARCH

THE DIFFERENCES IN SALIVARY SECRETORY IMUNOGLOBULIN A (SIGA) LEVELS BEFORE AND AFTER CONSUMPTION OF LACTOBACILLUS REUTERI PROBIOTICS IN CHILDREN WITH THALASSEMIA MAJOR EXPERIENCING GINGIVITIS

Afifa Syafiqoh[1], Heriandi Sutadi[2], Mochamad Fahlevi Rizal[2], Pustika Amalia Wahidiyat[3]

[1]Pediatric Dentist Specialist Study Program, Department of Pediatric Dentistry, Faculty of Dentistry, Universitas Indonesia, Jakarta, Indonesia
[2]Department of Pediatric Dentistry, Faculty of Dentistry, Universitas Indonesia, Jakarta, Indonesia
[3]Department of Pediatric, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia
Jalan Salemba Raya No.4, Jakarta 10430 Faculty of Dentistry, Universitas Indonesia, Jakarta Pusat, DKI Jakarta 10430, Indonesia
*Corresponding Author**: Mochamad Fahlevi Rizal[2]** Department of Pediatric Dentistry, Faculty of Dentistry, Universitas Indonesia, Jakarta, Indonesia: mochamad.fahlevi@ui.ac.id

Received: *Jun 11, 2025;* ***Accepted:*** *Jul 21, 2025;* ***Published:*** *Aug 31, 2025*

ABSTRACT

Thalassemia is an inherited blood disorder caused by genetic mutations that prevent the body from producing normal haemoglobin. A common oral manifestation in patients with thalassemia is a high incidence of gingivitis, caused by iron overload and a decreased immune response to infection. A decrease in salivary sIgA levels can increase the risk of periodontal tissue infection. Probiotics can stimulate the immune system, thereby increasing sIgA production. Evaluate the differences in salivary sIgA levels before and after Lactobacillus reuteri probiotic consumption in children with thalassemia major. The study was conducted in the Blood Transfusion Room at Dr. Cipto Mangunkusumo General Hospital (RSUPN Dr. Cipto Mangunkusumo), Jakarta, Indonesia. This research employed an experimental test design. This research involved 16 children aged 6–12 years diagnosed with thalassemia major with gingivitis. The study commenced with unstimulated saliva sampling before probiotic administration. After 14 days of probiotic consumption, unstimulated saliva was collected again. The sIgA levels in the saliva samples were quantitatively assessed by the enzyme-linked immunosorbent assay (ELISA) method. Statistical analysis used: Statistical analysis was performed using a paired t-test. The study showed that the mean salivary sIgA level before probiotic consumption was 0.97 ± 0.54, and the mean after was 1.31 ± 0.73. Paired t-test results yielded a p-value of 0.100 (>0.05), indicating no statistically significant difference. Fourteen days of Lactobacillus reuteri probiotic consumption increased salivary sIgA levels in children diagnosed with Thalassemia major with gingivitis, though the increase was not statistically significant.

Keywords: Thalassemia major; Gingivitis; Lactobacillus reuteri; sIgA.

INTRODUCTION

In thalassemia there is a defect in the polypeptide chain that makes up the globin molecule in haemoglobin.[1] Haemoglobin is the oxygen-carrying component of red blood cells. This abnormality makes red blood cells easily damaged, resulting in anaemia. The World Health Organization (WHO) estimates that about 7% of the world's population are carriers of the Thalassaemia trait. Every year around 300,000 - 500,000 newborns are born with severe haemoglobin disorders, and 50,000 to 100,000 children die from beta-thalassaemia, 80% of whom are from developing countries. Indonesia is one of the countries in the world's Thalassemia belt, which is a country with a high number of carriers of the Thalassemia trait.[2] Prevalence is most

prevalent around the Mediterranean sea, such as Italy, Greece, Turkey, West Asia, North Africa, South Asia, and Southeast Asia such as Thailand and Indonesia.[3]

Based on its clinical severity, thalassemia can be classified into thalassemia minor, thalassemia intermedia, and thalassemia major.[4] In Thalassaemia major, patients require regular blood transfusions for life with an average of two to four weeks of blood transfusions throughout their life by maintaining haemoglobin levels above 10 g/dl.[1] One of the major complications in routinely transfused patients is iron overload resulting in irregular antibody development.[3] Excess free iron can accumulate in tissues and blood, catalysing the formation of Reactive Oxygen Species (ROS). ROS in the oral cavity can increase the risk of biofilm formation and affect the quality and quantity of saliva. This also has an effect on increasing the risk of periodontal disorders gingivitis.[5]

sIgA acts as a first line of defence against periodontal pathogens and inhibits bacterial attachment.[6] Salivary sIgA levels in children with Thalassemia major who experience gingivitis are lower.[7] If salivary sIgA levels are decreased it can lead to an increased risk of periodontal tissue infection.[6] Probiotic bacteria can stimulate the immune system which will increase sIgA production.[8] A study has proven that the administration of probiotic strain Lactobacillus reuteri for 14 days in healthy people can reduce gingival inflammation.[9] Previously there was also research with the consumption of probiotic supplements for 14 days able to increase salivary sIgA levels.[6] However, there has been no research on salivary sIgA levels in children with Thalassemia major who experience gingivitis before and after probiotic consumption. Based on this, the author is interested in examining the difference in sIgA levels before and after probiotic consumption in the saliva of children with Thalassaemia major who experience gingivitis.

METHOD

This research employed an experimental test design involving 16 children aged 6–12 years diagnosed with Thalassemia major. The study was conducted in the Blood Transfusion Room at RSUPN Dr. Cipto Mangunkusumo. The study analyzed changes in sIgA levels in children with gingivitis before and after a 14-day intervention with Lactobacillus reuteri probiotics DSM 17938 and Lactobacillus reuteri ATCC PTA 5289, using the BioGaia Interlac Pro-D brand (POM SI214591521). Ethical approval was obtained under letter number KET 1346/UN2.F1/ETIK/PPM.00.02/2024 from the Research Ethics Committee of the Faculty of Medicine, Universitas Indonesia – RSUPN Dr. Cipto Mangunkusumo.

The inclusion criteria were: 1. Subjects diagnosed with thalassemia major with blood transfusion every two to four weeks; 2. Aged 6-12 years; 3. Having gingivitis. Exclusion criteria are: 1. Subjects have other systemic diseases (Tuberculosis, Human Immunodeficiency Virus); 2. Subjects took antibiotics and other probiotics one month before the study; 3. Girls who have begun menstruation.

Sampling was conducted through purposive sampling. The sample size was calculated using the G*Power application version 3.1.9.6 for a paired t-test (two-tailed) with an effect size of 0.75, based on previous studies, an α of 0.05, and a power of 0.80. The total sample size required was 16 subjects.

After obtaining informed consent from the subjects and their parents, the study commenced with unstimulated saliva sampling before probiotic administration. Participants and their parents were educated on oral hygiene and instructed to brush their teeth using fluoride toothpaste twice daily, in the morning after breakfast and at night before bed. Probiotics were administered for 14 days, with instructions for the subjects to dissolve probiotic lozenges in their mouths for approximately two minutes after brushing their teeth at night before sleeping. Subjects were instructed not to eat or drink for one hour after consuming the probiotics and to refrain from using other probiotic products during the testing period. After 14 days of probiotic consumption, unstimulated saliva was collected again. Two millilitres of saliva was collected using a centrifugator tube. The sIgA levels in the saliva samples, both before and after the intervention, were measured using an ELISA method with Bioenzy brand.

RESULTS

The research subjects, aged 6–12 years, had an average sIgA level of 0.97 ± 0.54 microliter (µl) before consuming Lactobacillus reuteri probiotics and 1.31 ± 0.73 µl after 14 days of probiotic consumption. A Shapiro-Wilk normality test ($n \leq 50$) was conducted on the sample size of 16 before performing the mean difference analysis. The normality test confirmed a normal distribution of sIgA levels before and after treatment. A paired t-test was then used to compare the mean sIgA levels before and after 14 days of consuming Lactobacillus reuteri probiotics, with a significance level of $p<0.05$. While the mean sIgA levels increased after the intervention, the paired t-test results showed that this difference was not statistically significant, with a p value = 0.100) (Table 1).

Table 1. Comparison of sIgA levels before and after consuming the probiotic Lactobacillus reuteri for 14 days

Variable	Time	n	Mean ± SD	*P* Value
sIgA	Before	16	0,97 ± 0,54[a]	0,100*
	After	16	1,31 ± 0,73[a]	

[a] Normal distribute data, presented in mean ± SD

* Paired t-test, P>0,05

DISCUSSION

All subjects in this study were children aged 6-12 years. The consideration of choosing this age is the dental phase mixed with exfoliation of primary teeth and eruption of permanent teeth so that the possibility of finding gingival inflammatory conditions is quite high.[7] Children with Thalassaemia major have a high prevalence of gingivitis. According to research studies, children aged 5-18 years with Thalassaemia major experience gingivitis as much as 92.2% ranging from mild to severe.[10] Another consideration for choosing this age is that in thalassaemia major children with a routine history of blood transfusions, their growth and development will have minimal complications until they reach the age of 12 years. After this age, complications can arise due to the accumulation of excess iron deposits due to annual transfusions.[11]

The method of measuring gingivitis in this study was the Modified Gingival Index (MGI) according to Lobene. This method was chosen because it is a modification of the Löe and Silness Gingival Index measurement with changes in criteria through a non-invasive way using clinical images without the use of probes. This is important because in thalassaemia children the use of periodontal probes for the measurement of gingivitis should be avoided.[12] As the literature states in the field of dentistry, Thalassaemia minor, major or intermedia differs in its treatment plan. Before starting treatment, attention should be focused on the type of Thalassemia, haemoglobin level, degree of iron overload, relative organs involved, and the overall prognosis of the patient. Any kind of invasive procedure should be performed immediately after blood transfusion followed by antibiotic prophylaxis as these patients have anaemia, haemolysis and ineffective erythropoiesis and thus have a greater risk of infection.[1]

This study used saliva samples because sIgA is abundant in saliva, which is 90 - 98%.[13] Saliva samples were taken because they are in the oral cavity and are in direct contact with the gingiva. Because this study took subjects with gingivitis, the sIgA used was taken from saliva. Saliva sampling in this study was carried out in the morning between 08.00 and 11.00. The reason for sampling at this hour is to avoid bias in the results of salivary sIgA levels due to the circadian rhythm of the body. Circadian rhythm is the physiological behaviour of the body to adapt to changes in time over 24 hours and is regulated by the hypothalamus. Circadian rhythms can affect salivary flow rates and cause changes in the concentration of salivary sIgA levels. Salivary sIgA levels have a diurnal rhythm, where salivary sIgA levels are highest in the morning and decrease until the evening. In addition, at this hour it is estimated that the research subjects have had breakfast and there is a break of about 90 minutes before sampling. Saliva sampling was carried out in unstimulated saliva conditions. Saliva sampling is done without stimulation because stimulated saliva has higher sIgA levels than unstimulated saliva.[7]

The results of this study showed that the mean sIgA level before consumption of probiotic Lactobacillus reuteri was 0.97 ± 0.54 µl, and the mean sIgA level after 14 days of consumption of probiotic Lactobacillus reuteri was 1.31 ± 0.73 µl. The average results showed an increase in salivary sIgA levels. The increase is in line with research on gargling with probiotic solution for 14 days can increase salivary sIgA levels in the elderly age group. Factors that can affect salivary sIgA levels include oral conditions, systemic conditions, physical activity and nutrition.[6] This is associated with the systemic conditions of children with major thalassaemia have been studied and found that the salivary sIgA levels of children with major thalassaemia are lower at 111,541 ± 71,000 micrograms (µg/ml) and in normal children at 186,136 ± 92,342 µg/ml.[7] These results indicate that there is a significant difference in salivary sIgA levels in beta major thalassaemia children and normal children who experience gingivitis. Gingivitis indicates an infection in the oral cavity, so the body will respond by activating the humoral immune response. Until now, several studies have been conducted to determine the condition of humoral and cellular immunity in thalassaemia major children. One study suggested changes in serum

immunoglobulin levels and changes in the number and function of B cells and T cells which may be caused by repeated antigen exposure due to blood transfusion treatment in thalassaemia major children.[14]

Probiotic therapy has recently been widely used as an adjunctive therapy to restore dysbiosis in the oral cavity such as periodontal disease. However, there has been no research on salivary sIgA levels of children with thalassaemia major who experience gingivitis before and after consumption of Lactobacillus reuteri. The results of this study showed an increase in salivary sIgA levels after the use of probiotic Lactobacillus reuteri but the difference was not statistically significant. This could be attributed to indirect mechanisms of probiotics in the oral cavity, including modulation of innate and adaptive immune function. Lactic acid bacteria can interact with immunocompetent cells, such as macrophages and T cells, leading to changes in cytokine production and B cells that will differentiate into plasma cells. Plasma cells will produce sIgA.[15] This is consistent with the literature that probiotics can activate and modulate the host immune system for a longer time under certain systemic conditions literature that probiotics can activate and modulate the host immune system for a longer time under certain systemic conditions.[16]

All subjects in this study reported no complaints or side effects during probiotic use. This is consistent with previous studies that daily probiotic use has been shown to be safe and well tolerated. The Center for Disease Control (CDC) supports the conclusion that the incidence of Lactobacillus reuteri bacteraemia is inconsistent and its oral use is safe.[17] To the best of the researchers' knowledge, this study is the first study conducted to see the difference in salivary sIgA levels of children with thalassemia major who experience gingivitis after probiotic Lactobacillus reuteri consumption.

CONCLUSION

This study concluded that consumption of probiotic L.reuteri for 14 days can increase salivary sIgA levels in children with thalassaemia major who experience gingivitis but the increase is not statistically significant.

DECLARATIONS

Conflict of interest

The authors declare that they have no conflict of interest.

Funding

This study was not funded by any fund.

Acknowledgement

The authors express their gratitude to all members and staff of Pediatric Dentistry Department, Faculty of Dentistry, Universitas Indonesia, staff of Department of Pediatric, Faculty of Medicine, Universitas Indonesia, and staff of Blood Transfusion Room at RSUPN Dr. Cipto Mangunkusumo, Jakarta, Indonesia for helping and supporting the study.

REFERENCES

1. Farmakis D, Porter J, Taher A, et al. 2021 Thalassaemia International Federation Guidelines for the Management of Transfusion-dependent Thalassemia. *Hemasphere*. 2022;6(8). doi:10.1097/HS9.0000000000000732
2. Keputusan Menteri Kesehatan Republik Indonesia. Pedoman Nasional Pelayanan Kedokteran Tata Laksana Thalasemia. Published online January 2018.
3. Shah FT, Sayani F, Trompeter S, Drasar E, Piga A. Challenges of blood transfusions in β-thalassemia. *Blood Rev*. 2019;37. doi:10.1016/j.blre.2019.100588
4. Paloma Canis Natih Ayu Dewa I. Thalassemia : A Literature Review. *BIOCITY Journal of Pharmacy Bioscience and Clinical Community*. 2023;1(2):89-100. doi:10.30.812/biocity.vli2.2525
5. Nuraini P, Wahluyo S, Pradopo S, et al. Effects of Iron Accumulation on Dental Caries, Gingivitis, and Candida albicans Infection in Children with Beta Thalassemia Major: A Narrative Review. *Acta Med Philipp*. 2023;57(3).
6. Indraswary R, Nurenda HY, Faturrahman H. Pengaruh Berkumur Dengan Larutan Probiotik Terhadap Kadar IgA Dalam Saliva. *Mandala Of Health*.2021;14(1):21.doi:10.20884/1.mandala.2021.14.1.4754
7. Pratiwi DT, Sutadi H, Suharsini M. *Perbedaan Kadar SIgA Saliva Pada Penderita Gingivitis Antara Anak Talasemia Beta Mayor Dan Anak Normal*. Universitas Indonesia; 2014. Accessed March 19, 2025. https:lib.ui.ac.id/detail?
8. Vivekananda MR, Vandana KL, Bhat KG. Effect of the probiotic Lactobacilli reuteri (prodentis) in the management of periodontal disease: A preliminary randomized clinical trial. *J Oral Microbiol*. 2010;2(2010). doi:10.3402/jom.v2i0.5344
9. Twetman S, Derawi B, Keller M, Ekstrand K, Yucel-Lindberg T, Stecksén-Blicks C. Short-term effect of chewing gums containing probiotic Lactobacillus reuteri on the levels of inflammatory mediators in gingival crevicular fluid. *Acta Odontol Scand*. 2009;67(1):19-24. doi:10.1080/00016350802516170
10. Faiez NH. Thalassemia Major and related Dentomaxillofacial Complications: Clinical and Radiographic Overview with Reference to Dental Care. *International Journal of Experimental Dental Science*. 2017;6(2):95-104. doi:10.5005/jp-journals-10029-1163
11. Rujito L. *Buku Referensi Talasemia : Genetik Dasar Dan Pengelolaan Terkini*. (Siswandari Wahyu,

Lestrai Dwi Woro Diyah, eds.).; 2019.
12. Mohannad Yasser Z. Periodontal indices. *Department of Periodontology*. Published online 2024.
13. Fábián TK, Hermann P, Beck A, Fejérdy P, Fábián G. Salivary defense proteins: Their network and role in innate and acquired oral immunity. *Int J Mol Sci*. 2012;13(4):4295-4320. doi:10.3390/ijms13044295
14. Marcotte H, Lavoie MC. *Oral Microbial Ecology and the Role of Salivary Immunoglobulin A*. Vol 62.; 1998.
15. Allaker RP, Stephen AS. Use of Probiotics and Oral Health. *Curr Oral Health Rep*. 2017;4(4):309-318. doi:10.1007/s40496-017-0159-6
16. Saraswati H, Si S, Biomed M. *MODUL IMUNOLOGI RESPON IMUN SPESIFIK*.; 2021. http://esaunggul.ac.id0/16
17. Borrell García C, Ribelles Llop M, García Esparza MÁ, Flichy-Fernández AJ, Marqués Martínez L, Izquierdo Fort R. The use of Lactobacillus reuteri DSM 17938 and ATCC PTA 5289 on oral health indexes in a school population: A pilot randomized clinical trial. *Int J Immunopathol Pharmacol*. 2021;35. doi:10.1177/20587384211031107