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## [jioh]:Acknowledgment of Online Submission

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Journal of International Oral Health <editor@jioh.org>

21 November 2018 11.25

Balas Ke: symposium@fkg.unair.ac.id

Kepada: m.luthfi@fkg.unair.ac.id

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Dear Dr. Luthfi,

NOTE: This e-mail is sent to you as one of the contributing authors. If you are not corresponding author, please co-ordinate with the author designated by your group as the corresponding author for this manuscript

A manuscript has been submitted to our journal Journal of International Oral Health by Udijanto Tedjosasongko titled 'Analysis of IL-10 Anti-Inflammatory Cytokines Expression in Saliva of Children with Severe Early Childhood Caries and Caries-Free Children'. A copy of the acknowledgment mail is attached here with for your reference.

Thanking you  
Editorial Team  
Journal of International Oral Health

-----  
Dear Dr. Tedjosasongko,

Journal of International Oral Health has received your manuscript entitled "Analysis of IL-10 Anti-Inflammatory Cytokines Expression in Saliva of Children with Severe Early Childhood Caries and Caries-Free Children" for consideration for publication. The reference number for this manuscript is "jioh\_293\_18". Kindly quote this in correspondence related to this manuscript.

The manuscript is being reviewed for possible publication with the understanding that it is being submitted to one journal at a time and have not been published, simultaneously submitted, or already accepted for publication elsewhere either as a whole or in part. Online submission of this article implies that the corresponding author has the written consent from all the contributors to act as corresponding author.

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We thank you for submitting your valuable work to the Journal of International Oral Health.

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21 Desember 2018 17.05

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Status of the manuscript titled 'Analysis of IL-10 Anti-Inflammatory Cytokines Expression in Saliva of Children with Severe Early Childhood Caries and Caries-Free Children' submitted by Dr. Udiyanto Tedjosongko has been changed and a copy of the mail is as;

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Dear Dr. Tedjosongko

With reference to your manuscript entitled 'Analysis of IL-10 Anti-Inflammatory Cytokines Expression in Saliva of Children with Severe Early Childhood Caries and Caries-Free Children', please review the comments of the referees from our site <http://www.journalonweb.com/jioh>. The manuscript would be reconsidered after requisite modifications as per the comments and instructions provided by the journal.

If you wish to continue with the publication process, kindly make the changes according to the comments and upload the revised manuscript along with clarifications for all the comments clearly indicating the areas where the changes have been made. Do check the FAQ regarding replying to the comments and uploading a file. The contributors' form/images should be sent separately to the Administrative Office of the journal.

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We thank you for submitting your valuable research work to Journal of International Oral Health.

With warm personal regards,

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29 Agustus 2019 18.31

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Kepada: m.luthfi@fkg.unair.ac.id

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'jioh\_294\_18'

Dear Dr. Tedjosongko,

As requested, your manuscript entitled 'Effect of Okra Fruit (*Abelmoschus esculentus*) Extract on *Aggregatibacter actinomycetemcomitans* (Aa) Bacteria as the Cause of Aggressive Periodontitis' has been withdrawn from the review process in Journal of International Oral Health.

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23 Desember 2019 17.35

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**Remarks:**

Dear Author

Please add multivariant analysis, means apart from P value, SD, mean regarding other test like ANOVA, multi regression, Post tukey or wilcoxn test which can be applied as per your study type and sample. Please add same in tables and results.

This is job of statastian. Please take help and add tables in manuscripts.

Figure 1 quality need to improve

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1 ANALYSIS OF IL-10 ANTI-INFLAMMATORY CYTOKINES EXPRESSION IN SALIVA OF  
2 SEVERE EARLY CHILDHOOD CARIES

3 Muhammad Luthfi<sup>1</sup>, Aqsa Sjuhada Oki<sup>2</sup>, Retno Indrawati<sup>3</sup>, Priyawan Rachmadi<sup>4</sup>, Muhaimin Rifa'i<sup>5</sup>

4  
5 <sup>1,2,3</sup>Departments of Oral Biology, Faculty of Dental Medicine, Universitas Airlangga, Surabaya-Indonesia

6 <sup>4</sup>Departments of Dental Material, Faculty of Dental Medicine, Universitas Airlangga, Surabaya-Indonesia

7 <sup>5</sup>Department of Immunology and Physiology, Faculty of Sciences, Brawijaya University, Malang- Indonesia

8  
9 *Correspondence:* Muhammad Luthfi, Departement of Oral Biology, Faculty of Dental Medicine, Universitas  
10 Airlangga, Surabaya-Indonesia,.

11  
12  
13 Running title: IL-10 CYTOKINES EXPRESSION IN SALIVA OF CARIES

14 **Abstract:**

15 **Objective:** This study aimed to analyze the expression of IL-10 in children with severe early childhood  
16 caries and caries-free children.

17 **Method:** Saliva taken from preschool aged children (4 to 6 years) was divided into two groups, ie heavy  
18 caries group with dmft > 6 and caries free with dmft = 0. Salivary neutrophils were obtained from  
19 participants by rinsing their oral cavities with 10 mL of sterile and 1.5% NaCl solution while they  
20 gargled without swallowing for 30 s before expectorating the resulting uid into a sterile glass – a  
21 procedure repeated four times. The collected solution was centrifuged (15 min at 450 g) at 4°C and the  
22 pellets then mixed with 2 mL of Roswell Park Memorial Institute medium. For expression test of IL-10,  
23 flow cytometry test was used.

24 **Statistical test used:** The results were listed as the mean ± standard deviation. All statistical analyses  
25 were performed using SPSS 20 (IBM, New York, USA). The statistical difference was analyzed by  
26 *one way anova*. Correlation analyses were performed using tukey HSD with  $P < 0.05$  being considered  
27 to be significant.

28 **Results:** IL-10 expression in saliva of children with severe early childhood caries was  $3.32 \pm 0.79$ .  
29 Whereas, in children with no caries was  $4.04 \pm 0.65$ .

30 **Conclusion:** IL-10 expression in children with severe early childhood caries was significantly lower  
31 than in caries-free children.

32 **Key-Words:** Severe Early Childhood Caries, Il-10, Lymphocyte Cells

33  
34 **Key Messages**

35 Interleukin-10 (IL-10) is an anti-inflammatory cytokine produced by innate immunity secreted because  
36 of the response of pathogen recognition receptors (PRRs) in contact with pathogen associated molculer

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Commented [MOU2R1]: revised

Commented [a3]: Need to elaborate upto 250 words

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Commented [a5]: Need detail method with sampling method and perform test related

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1 patterns (PAMPs). Secretion of IL-10 during bacterial infection is the most important factor in resolution  
2 of infection. ECC has an impact on general health, ranging from local pain, infections, abscesses

### 3 **Introduction:**

4 Caries in early childhood (ECC) is a multifactorial disease resulting from interactions of various factors,  
5 namely: cariogenic microbes, exposure to carbohydrate fermentation and various social variables. ECC  
6 is a condition of health abnormalities found in children living in socially disadvantaged communities,  
7 such as malnourished people with social and health inequalities.<sup>[1]</sup> ECC has an impact on general health,  
8 ranging from local pain, infections, abscesses, difficulty in chewing, malnutrition, indigestion, and  
9 insomnia.<sup>[2]</sup>

10 The body's immune system functions to defend the human body from foreign invaders. A compromised  
11 immune system can cause various diseases, such as infection, aging, allergies, various organ disorders  
12 and other diseases, such as cancer and auto immune deficiency syndrome (AIDS).<sup>[3]</sup> The role of the  
13 body's immune system is becoming increasingly important in understanding the mechanisms of disease  
14 prevention. The effective function of the body's immune system is to immediately eradicate the  
15 infectious agent from the body. This is done by an interactive system of actions, namely innate (very  
16 specific), fast but non-specific and adaptive immune system.<sup>[4]</sup> which function as a pathogenic killer<sup>[5]</sup>,  
17 produce antibodies, and CD8 + T cell cytotoxicity<sup>[6]</sup>.

18 Immunity in the oral cavity is a system that makes a balance by controlling various microbes in the oral  
19 cavity that are fluctuating due to external aggression. The mouth is the entrance and exchange with the  
20 outside environment. Therefore, homeostasis factors must be evaluated and controlled by the immune  
21 system.

22 The immune response to pathogens involves the rapid activation of the secretion of pro-inflammatory  
23 cytokine which functions to initiate host defenses against microbial invasion. However, excessive  
24 inflammatory cytokines in the tissues can cause systemic metabolic and hemodynamic disorders that  
25 are harmful to the host. As a result, the immune system has evolved to form anti-inflammatory functions  
26 to suppress the production of pro-inflammatory cytokines which function to limit tissue damage and to  
27 maintain tissue homeostasis.<sup>[7]</sup> Interleukin 10 (IL-10) is an anti-inflammatory cytokine that plays an  
28 important role in preventing prolonged inflammation.<sup>[8]</sup>

29 Various preventions of dental caries had been carried out, for example by brushing teeth properly,  
30 fluoridating with topical applications, and making vaccines which until now have not shown the  
31 expected results.<sup>[9]</sup> Therefore, this study aimed to analyze the expression of IL-10 in saliva which  
32 functions as an anti-inflammatory. The results of this study are expected to be used as a marker of early  
33 detection of S-ECC.

### 34 **Subjects and Methods:**

1 Study Design:

2 This was an observational analytic study using children with severe early childhood caries and caries-  
3 free children as the objects of research with a cross sectional study design. Ethical clearance test at  
4 Universitas Airlangga, Faculty of Dental Medicine was done with Health Research Ethical Clearance  
5 Commission number of 209/HRECC. FODM/IX/2017.

6 **Sampling Criteria:**

7 eight children with S-ECC and eight free of caries were taken from the saliva of kindergarten children  
8 aged 4 to 6 years in the south Surabaya region which were previously divided into two groups.

9 Group one was children with who were diagnosed with severe early childhood caries (S-ECC) marked  
10 by decay exfoliation and filling (def-t>6). Whereas, group two was kindergarten children diagnosed  
11 with free caries marked with def-t = 0.

12 5ml saliva taken from the kindergarten children with SECC and caries-free . Sampling was carried out  
13 by researchers and trained personnel using protocol standards. Subjects might not eat, drink, chew gum,  
14 or brush their teeth for 60 minutes before sampling. Furthermore, samples were frozen at -80oC for  
15 analysis.

16 **Observational parameters:**

17 Analysis of IL-10 expression was determined using flow cytometry, according to Cherng et al (2008).  
18 Fluorescein isothiocyanate (FITC), phycoerythrin (PE), allophycocyanin (APC), Peridinin chlorophyll  
19 protein (PerCP), PerCP-Cy5.5-conjugated monoclonal antibodies (mAbs) from Becton Dickinson (San  
20 Jose, CA, USA). The optimal concentration of mAbs is determined for each mAb with titration. Flow  
21 cytometry simultaneously measures and analyzes the physical properties of particles such as cells  
22 because it flows through the flow of fluid through a beam of light. The nature of scattering cell light can  
23 be used to analyze changes in size, granularity, internal complexity and relative fluorescence intensity.  
24 Flow cytometry analysis was performed to determine directly the pattern of lymphocyte  
25 immunomodulation, using conjugated monoclonal antibodies (mAbs).

26 **Statistical Analysis:**

27 The results were listed as the mean ± standard deviation. All statistical analyses were performed  
28 using SPSS 20 (IBM, New York, USA). The statistical difference was analyzed by one way anova.  
29 Correlation analyses were performed using tukey HSD with  $P < 0.05$  being considered to be  
30 significant.

32 **Results:**

33

**Commented [a9]:** Add sampling method  
Inclusion and exclusion criteria  
Add how the samples size determined

**Commented [MOU10R9]:** revised

**Commented [a11]:**  
•Add country name of state software with detail of version.  
Statistical tests should be described in more details. P values  
should be accompanied by degree of freedom, and confidence  
interval.

Please add multivariant analysis, means apart from P value,  
SD, mean regarding other test like ANOVA, multi regression,  
Post tukey or wilcoxon test which can be applied as per your  
study type and sample. Please add same in tables and results.  
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Must be started with baseline parameters and any bias, or drop of  
sample must be mentioned

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1 Data from the results of the study before the test using the t test, conducted tests of normality  
2 and homogeneity using the Shapiro-Wilk test. From the results of these tests indicate a value of  $P > 0.05$   
3 which means that all data are normally distributed and homogeneous.

4 The results of statistical analysis one way anova in the S-ECC group showed that IL-10  
5 expression (3.31%) was lower than that of the caries-free group (4.03%) (Table 1). Based on Levene's  
6 test, there was significant difference in IL-10 expression between S-ECC with caries-free.

## Discussion:

The results of the study showed that there was expression of IL-10 decrease in children with S-ECC compared to that in caries-free children. This was probably due to the S-ECC patients responding more antigens in the form of *S. mutans* bacteria which were relatively high in numbers compared to that in children with free caries.<sup>[10]</sup>

An antigen structure called Pathogen Associated Molecular Pattern (PAMPs), which will be recognized by Pattern Recognition Receptors (PRRs), namely Toll-like receptors (TLRs), is very important for triggering the effector phase of the innate immune response.<sup>[11]</sup> TLR2 and TLR4 involved in the introduction of Gram-positive and Gram-negative bacteria that have been detected in odontoblast cell membranes in healthy pulp show that odontoblasts are equipped to recognize these pathogens when they diffuse through the dentinal tubules during carious infection.<sup>[12]</sup> One of the main consequences of TLR activation is an increase in the efficacy of innate immunity, including antimicrobial agents and proinflammatory cytokines and chemokines that recruit and activate immune cells.<sup>[13]</sup> One of the main consequences of TLR activation is increased efficacy of innate immunity, including antimicrobial agents and proinflammatory cytokines and chemokines that recruit and activate immune cells.<sup>[13]</sup> This causes the S-ECC saliva to increase prolonged inflammatory cytokines which eventually can cause tissue damage that affects health in general, starting from local pain, infection, abscess, difficulty in chewing, malnutrition, indigestion, and sleeping difficulty.<sup>[14]</sup>

Based on the results of this study, high expression of proinflammatory cytokines in S-ECC should be balanced by the immune host system by producing anti-inflammatory cytokines, IL-10. As a response to pathogenic microbes, the body's adaptive immune system develops effector cells that function to prevent these threats, namely CD4 + memory T cells which serve as a protective against bacterial infections.<sup>[14]</sup> CD4 + cells participate in responding to secondary infections that have the potential as anti-pathogens<sup>[5]</sup> producing antibodies and CD8 + T-cell cytotoxicity.<sup>[6]</sup> However, this did not occur in S-ECC so IL-10 expression in S-ECC saliva was significantly lower than that in caries-free children. This was probably due to the role of the immune system in S-ECC which was not as good as that in caries-free children.

Acknowledgement: Department of Oral Biology, Faculty of Dentistry, Universitas Airlangga

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**Conflict of interest:**

There is no conflict of interest in this research

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**Author contributions:**

The author contributes to starting to determine the topic of the problem, sampling, research and finally that all the authors approved the final version of the manuscript for publication.

Muhammad Luthfi: Study conception, study design, intellectual content, literature research, data acquisition, data analysis, manuscript review, guarantor

Aqsa Sjuhada Oki: Study concept, clinical studies, experimental studies, data analysis, manuscript review

Retno Indrawati: data interpretation, Statistical analysis, manuscript preparation, manuscript editing,

Priyawan Rachmadi: Statistical analysis, manuscript preparation, manuscript editing,

Muhaimin Rifa'i: manuscript editing, manuscript review

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**Patient declaration of consent:**

Before saliva sampling from children aged 4 to 6 years, parents of the sample had agreed to signed a written informed consent.

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**Abbreviations**

- ECC : Early childhood caries
- S-ECC : Severe early childhood caries
- IL-10 : Interleukin-10
- PRRs : Pathogen recognition receptors
- TLR : Toll like receptors
- PAMPs : Pathogen associated molecular patterns
- CD8 : Cluster differentiation 8
- CD4 : Cluster differentiation 4
- def-t : Decay exfoliation and filling

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## References:

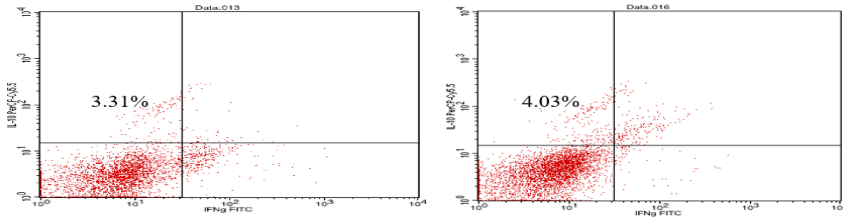
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1. Feldens CA, Giugliani ER, Duncan BB, Drachler Mde L, Vitolo MR. (2010). Long-term effectiveness of a nutritional program in reducing early childhood caries: a randomized trial. *Community Dent Oral Epidemiol.* 38(4):324–32.
2. Finlayson TL, Siefert K, Ismail AI, Sohn W. (2007). Psychosocial factors and early childhood caries among low-income African-American children in Detroit. *Community Dent Oral Epidemiol.* 35(6):439–48.
3. Cherng, J-M., Chiang, W., Chiang, L-C. (2008). Immunomodulatory activities of common vegetables and spices of Umbelliferae and its related coumarins and flavonoids. *Food Chemistry.* 106: 944–950.
4. Volman, J.J., Ramakers, J.D., Plat, J. (2008). Dietary modulation of immune function by  $\beta$ -glucans. *Physiology and Behavior.* 94: 276–284.
5. Zens K.D. and Farber D. L., (2015). Memory CD4 T cells in influenza,” in *Influenza Pathogenesis and Control—Volume II*, vol. 386 of *Current Topics in Microbiology and Immunology.* 399–421.
6. Strutt T. M., Mckinstry K. K., Marshall N. B, Vong A. M. , RDutton. W., and Swain S. L. (2013). Multipronged CD4+ T-cell effector and memory responses cooperate to provide potent immunity against respiratory virus,” *Immunological Reviews.* 255 (1): 149–164..
7. Mosser DM, Zhang X. (2008). Interleukin-10: new perspectives on an old cytokine. *Immunol Rev*226:205–18.
8. Sabat R, Grütz G, Warszawska K, Kirsch S, Witte E, Wolk K, Geginet J. (2010). Biology of interleukin-10. *Cytokine Growth Factor Rev.* 21(5):331–44.
9. Trigg, T.E., et al., (2001). Use of a GnRH analogue implant to produce reversible Longterm suppression of reproductive function in male and female domestic dogs. *J Reprod Fertil Suppl.* 57:255- 61.
10. Luthfi M, Indrawati R, Arundina I and Dachlan Y P. (2015). Correlation of Amount of *Streptococcus mutans* (*S. mutans*) and Expression Rate of Interleukin 8 (IL-8) in Severe Early Childhood Caries. *Maj Ked Gi Ind.* 1(2): 142 – 148
11. Kumar H., Kawai T., and Akira S. (2011). Pathogen recognition by the innate immune system,” *International Reviews of Immunology.* 30 (1): 16–34,
12. Veerayutthwilai O., Byers M. R., Pham T.-T. T., Darveau R. P., and Dale B. A., (2007). Differential regulation of immune responses by odontoblasts,” *Oral Microbiology and Immunology.* 22(1): 5–13,

- 1 13. Turner M. D., B. Nedjai, Hurst T., and Pennington D. J., , (2014). Cytokines and chemokines:  
 2 at the crossroads of cell signalling and inflammatory disease,” *Biochimica et Biophysica Acta—*  
 3 *Molecular Cell Research*. 1843 (11): 2563–2582.
- 4 14. Tubo N. J. and Jenkins M. K.. (2014). CD4+ T Cells: guardians of the phagosome,” *Clinical*  
 5 *Microbiology Reviews*. 27 (2): 200–213,

24 Table 1. Mean and standard deviation of IL-10 expression in caries free and S-ECC analyzed by flow  
 25 cytometry test and t test statistic test

Groups	N	IL-10 Expression (%)		
		Mean	Standard Deviation	Standard error mean
S-ECC	8	3.321	0.787	0.278
Caries-free	8	4.044	0.648	0.229



1  
2 Figure 1. Expression of IL-10 from S-ECC saliva after analyzed by Flow Cytometry test.

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## [jioh]:Article for revision

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