

Correlation between interleukin 17 (IL-17) levels and malondialdehyde (MDA) levels in patients with chronic suppurative otitis media

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Correlation between interleukin 17 (IL-17) levels and malondialdehyde (MDA) levels in patients with chronic suppurative otitis media

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Abstract

4 **Background:** Otitis media is one of the most common causes of fever in the pediatric population. CSOM or also called chronic otitis media is an ear disease condition where a chronic infection is found in the middle ear without an intact tympanic membrane. Oxygen free radical levels and deficiency of antioxidant defense complexes may be involved in the pathogenesis of chronic suppurative otitis media. The IL-17 pathway plays an important role in the pathogenesis of otitis media by increasing ROS including MDA levels.

Purpose: To analyze the levels of interleukin 17 (IL-17) and malondialdehyde (MDA) in chronic suppurative otitis media patients.

Method: Analytical observational research with a cross sectional approach was conducted in December 2022 at RSUD Dr. Moewardi, the research sample was a patient with chronic suppurative otitis media without active phase cholesteatoma. Sampling used consecutive sampling, namely a non-random sampling method, so that a sample size of 40 participants was obtained, divided into two groups, namely the CSOM group and the healthy group. The data analysis used is univariate and bivariate analysis using parametric tests, namely the product moment correlation test and if it is not normally distributed (p -value < 0.05) then use the Spearman rank correlation test.

Results: The mean IL-17 level in the CSOM group was 78.76 ± 28.01 and the healthy group was 47.68 ± 21.75 with $p = 0.000 < 0.05$. The mean MDA value in the CSOM group was 138.37 ± 123.96 and in the healthy group was 55.84 ± 47.37 with a p value = $0.008 < 0.05$. Because the p value is < 0.05 for the IL-17 and MDA variables, there is a significant difference between CSOM and the group of healthy people. The product moment correlation test obtained a correlation coefficient value of 0.623 with a p value = 0.000. Because the p value is < 0.05 , there is a significant positive relationship between IL-17 levels and MDA in the blood of chronic suppurative otitis media (CSOM) patients.

3 **Conclusion:** There is a significant positive relationship between IL-17 levels and MDA in the blood of chronic suppurative otitis media (CSOM) patients.

Keywords: Chronic Suppurative Otitis Media; Interleukin 17; Malondialdehyde.

4 INTRODUCTION

2 Otitis media is one of the most common causes of fever in the pediatric population. Chronic suppurative otitis media (CSOM) or also called chronic otitis media is an ear disease condition where chronic infection is found in the middle ear without an intact tympanic membrane. This disease is a chronic inflammation of the middle ear and mastoid cavity. The typical

presentation is chronic or persistent otorrhea lasting 2 to 6 weeks through perforation of the tympanic membrane (Head, Chong, Bhutta, Morris, Vijayasekaran, Burton, & Brennan-Jones, 2020; Balasundaram, Phan, Mazzoni, Duong, Sweeny, Del Mar, & Keijzers, 2019; Schilder, Chonmaitree, Cripps, Rosenfeld, Casselbrant, Haggard, & Venekamp,

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4 16). Acquired hearing loss is also typically found in patients with this condition and if untreated, can lead to further morbidity and mortality (Master, Wilkinson, & Wagner, 2018).

The disease begins with an episode of acute otitis media (AOM) with rupture of the amniotic membrane. Mastoid air cells are always involved in these cases. The most common causative organisms are *P. aeruginosa* and *S. aureus*. However, pathogenic bacteria typical of AOM may also be the cause, especially in young children or in winter (Simpson, Duncanson, Oben, Adams, Wicken, Pierson, & Gallagher, 2017). Treatment is guided by the results of microbiological examination. If cholesteatoma is not found, parenteral antimicrobial treatment combined with ear cleaning may successfully clear the infection, but in refractory cases, tympanomastoidectomy may be necessary (Hong, Khampang, Kerschner, Mackinnon, Yan, Simpson, & Kerschner, 2019).

Diagnosis is made through a combination of history taking, physical examination and support. By performing an ear examination, the degree of mobility of the tympanic membrane in response to negative or positive pressure can be evaluated to assess fluid in the middle ear, which is a feature of chronic suppurative otitis media. Other abnormalities in the tympanic membrane that can be found are erythema, protrusion, fullness, or retraction. Even though the diagnosis of CSOM is quite clear, until now the pathogenesis of CSOM is not fully understood (Rosario, & Mendez, 2023; Zielnik-Jurkiewicz, & Stankiewicz-Szymczak, 2016; Earwood, Rogers, & Rathjen, 2018). Several factors that play a role in the pathogenesis of CSOM include anatomical factors, environmental factors, and the body's defense mechanisms (Qureishi, Lee, Belfield, Birchall, & Daniel, 2014).

The body's defense mechanism as an agent of innate immunity in warding off pathogens is related to the production of various cytokines. T-helper cells (TH-17) release pro-inflammatory cytokines such as interleukin-17 (IL-17), where IL-17 has an important role in defense against autoimmune diseases and infections. In addition, IL-17 plays a role in controlling chemokines and other cytokines that cause the mobilization of neutrophils to sites of inflammation (Isailovic, Daigo, Mantovani, & Selmi, 2015; Kuwabara, Ishikawa, Kondo, & Kakiuchi, 2017).

IL-17 also plays a role in initiating the ability of neutrophils to eliminate infections by increasing the production of cytotoxic molecules such as reactive oxygen species (ROS) (Chen, Wang, Huang, Zhou, Zhao, Zhang, & Xia, 2018). ROS is a pathological mechanism of various diseases, occur during the body's metabolic processes and produce various contents that are toxic to body tissues, such as hydroxyl free radicals, hydrogen peroxide, lipid peroxide, hydroxyl peroxide, and other derivatives. The destruction of proteins, nucleic acids, cell membranes and lipids in tissues is the etiology of ROS. In preventing oxygen free radicals and their derivatives (oxygen-derived free radicals), the body activates a body defense complex called the antioxidant defense complex. Oxidation formed by free radicals will be inhibited by antioxidants through mechanisms of stabilization and free radical capture. As a result, the body will initiate an enzyme system as an antioxidant against toxic substances resulting from the oxidation process, including the formation of malondialdehyde (MDA) (Kalyanaraman, 2013).

High levels of polyunsaturated fatty acids and low levels of antioxidants influence the susceptibility of pro-oxidant agents. Lipoperoxidation (LPO) is the main reaction of reactive oxygen species (ROS), where this reaction involves polyunsaturated fatty acids which are then bound to phospholipid membranes where malondialdehyde (MDA) is the end product of lipid peroxidation induced by ROS (Shi, Wu, Li, Cai, Liu, Gao, & Chen, 2017). In line with these two studies, another study concluded that the IL-17 pathway plays an important role in the pathogenesis of acute otitis media through increasing ROS including MDA levels (Abdelhafeez, & Mohamed, 2021).

A study found that increased levels of oxygen free radicals and deficiency of antioxidant defense complexes may be involved in the pathogenesis of chronic otitis media (CSOM) (Zielnik-Jurkiewicz, & Stankiewicz-Szymczak, 2016). Oxidative stress may be a causal factor in the CSOM (Garça, Aslan, Tuna, Kozan, & Cankaya, 2013). Although various studies have mentioned the role of IL-17 in the pathogenesis of CSOM through the ROS mechanism, to date research discussing the correlation of IL-17 and MDA levels in the blood of CSOM patients is still limited.

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RESEARCH METHOD

Analytical observational research with a cross sectional approach was conducted in December 2022 at RSUD Dr. Moewardi, the research sample was patients with chronic suppurative otitis media without active phase cholesteatoma. Sampling used consecutive sampling, namely a non-random sampling method, so that a sample size of 40 participants was obtained, divided into two groups, namely the CSOM group and the healthy group.

The independent variable is interleukin 17 (IL17) levels, while the dependent variable is MDA levels. The inclusion criteria in this study were patients with chronic suppurative otitis media without active phase cholesteatoma, the patient's family had no history of chronic suppurative otitis media, and the patient was willing to be a sample in this study with informed consent. Meanwhile, the exclusion criteria are CSOM patients who received oral antibiotic therapy or topical drops in the last 7 days, have systemic infectious diseases, immunocompromise, external ear infections, and congenital ear abnormalities.

The instruments in this research were an otoscope, headlamp, PPE equipment, office stationery, 3cc or 5cc syringe, tourniquet, alcohol swab, and vacutainer tube. This research was conducted by collecting research participants based

on inclusion criteria. Continue taking samples of the participant's venous blood and placing the blood sample in an EDTA tube. Then the blood sample is sent to the anatomical pathology laboratory for analysis.

6 The BMI variable is categorized as underweight if <18.5, normal weight between 18.5-24.9, overweight between 25-29.9, and if >30 it means obesity. Social status is categorized as low if the income is ≤minimum wage and high if it is ≥minimum wage. The level of hearing loss is divided into mild if 26-40 dB, moderate if 41-60 dB, severe if 61-90 dB, and most severe if >90 dB. Types of hearing loss are divided into conductive hearing loss (CHL), which is a condition of hearing loss caused by disturbances in the function of the auditory ossicles and mixed hearing loss (MHL), which occurs due to simultaneous sensorineural hearing loss (SNHL) and CHL.

The data analysis used is univariate and bivariate analysis using parametric tests, namely the product moment correlation test and if it is not normally distributed (p-value <0.05) then use the Spearman rank correlation test.

This research has received permission and recommendations from the Health Research Ethics Committee of Dr. Moewardi General Hospital with number 1.611/XII/HREC/2022.

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RESEARCH RESULTS

Table 1. Participant Demographic Data (N=40)

Variables	CSOM Group (n=20)	Healthy Group (n=20)
Age (Mean±SD)(Range)(Years)	(43.00±15.89)(22-70)	(38.35±10.16)(27-61)
Gender (n/%)		
Male	8/40.0	11/55.0
Female	12/60.0	9/45.0
Employment (n/%)		
Unemployment	1/5.0	6/30.0
Housewife	8/40.0	6/30.0
Self-employed	8/40.0	0/0.0
Teacher	1/5.0	0/0.0
Laborer	1/5.0	3/15.0
Employee	1/5.0	0/0.0
Retired	0/0.0	3/15.0
Civil servants	0/0.0	2/10.0
Education (n/%)		
Low	10/50.0	10/50.0
High	10/50.0	10/50.0
Socioeconomic Status (n/%)		
Low	8/40.0	10/50.0
High	12/60.0	10/50.0
BMI (n/%)		
Normal	11/55.0	15/75.0
Underweight	5/25.0	0/0
Overweight	3/15.0	5/25.0
Smoking (n/%)		
Yes	4/20.0	0/0
No	16/80.0	20/100
Level of Hearing Loss (n/%)		
No Distractions	0/0	20/100
Mild	7/35.0	0/0
Moderate	11/55.0	0/0
Severe	2/10.0	0/0
Very Severe	0/0	0/0
Types of Hearing Loss (n/%)		
No Distractions	0/0	20/100
CHL	19/95.0	0/0
MHL	1/5.0	0/0

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Based on Table 1, the average age in the CSOM group was 43 years with a standard deviation of 15.89, while in the healthy group it was 38.35 ± 10.16 . The majority of participants in the CSOM group were 12 female (60.0%), while in the healthy group the majority were male 11 people (55.0%). The most common occupations in the CSOM group are self-employed and housewives, 8 people each (40.0%), while in the healthy group, the majority unemployed and are housewives, namely 6 people each (30.0%).

The education of the participants in the CSOM group was balanced, namely 10 people each had high (High School/College) and low education (Primary School/Junior High School) (50.0%), while in the healthy group there were 10 people (50.0%) and those with low education (50.0%). In the CSOM group, the majority have high socio-economics, namely 12 people (60.0%), the same thing in the

group of healthy people who have high socio-economics, namely 10 people (50.0%). The BMI in the CSOM group was mostly normal, namely 11 people (55.0%), while in the healthy people group the majority was also normal, namely 15 people (75.0%). Most of the participants in the CSOM group did not smoke, namely 16 people (80.0%), while all of the healthy group did not smoke, namely 20 people (100%).

Based on the data presented, the CSOM group sample had normal hearing thresholds with moderate hearing loss as many as 11 people (27.5%), however the type of hearing loss most experienced by participants was conductive hearing loss (CHL) as many as 19 people (47.5%). Whereas in the healthy group all of them were normal and had no distraction of hearing.

Table 2. Description of Research Subject

Variables	IL-17	MDA
CSOM Group	(78.76±28.01)	(138.37±123.96)
Healthy Group	(47.68±21.75)	(55.84±47.36)

Based on the table above, the IL- 17 level in the CSOM group obtained an average of 78.76 ± 28.01 . As for the MDA, the average value was 138.37 ± 123.96 . In the group of healthy people, the average value was 47.68 ± 21.75 , while the MDA obtained an average value of 55.84 ± 47.36 .

Table 3. Normality Test

Variables	Shapiro-Wilk	
	Statistic	Sig.
IL-17		
CSOM Group	0.968	0.702
Healthy Group	0.951	0.380
MDA		
CSOM Group	0.948	0.332
Healthy Group	0.952	0.394

Based on the normality test with the Shapiro-Wilk test on IL-17 and MDA levels in the CSOM group and the group of healthy people, the p value was >0.05 . Because the p value <0.05 , the data for IL-17 and MDA levels were normally distributed, so a parametric test was used.

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Table 4. Comparison Test

Variables	CSOM Group	Healthy Group	p-value
IL-17	78.76±28.01	47.68±21.75	0.000
MDA	138.37±123.96	55.84±47.37	0.008

Based on the table, it was found that the mean IL-17 level in the CSOM group was 78.76 ± 28.01 and the healthy group was 47.68 ± 21.75 with a p = 0.000 < 0.05. The MDA mean value for the CSOM group was 138.37 ± 123.96 and for the healthy group it was 55.84 ± 47.37 with a value of p = 0.008 < 0.05. Because the p value < 0.05 on the IL-17 and MDA variables, there is a significant difference between CSOM and the group of healthy people.

Table 5. Correlation Test

Variables	Coefficient Correlation	p-value
IL-17 MDA	0.623	0.000

5 Based on the product moment test correlation test table, the correlation coefficient value was 0.623 with a value of p = 0.000. Because the p value < 0.05, there is a significant positive relationship between IL-17 levels and MDA in the blood of patients with chronic suppurative otitis media (CSOM).

DISCUSSION

Chronic otitis media occurs due to inflammation of the mucous lining of the air space of the middle ear and temporal bone, accompanied by perforation of the tympanic membrane with a duration of more than 3 months. The prevalence of CSOM is between 65 and 330 million people, and 60% have significant hearing loss. The incidence of cholesteatoma is estimated at 9 per 100,000 people. Many factors are involved in the pathophysiology, such as the presence of osteoclastic activity, cytokines, chronic inflammation, endotoxins and lipid peroxidation products, where these factors also lead to the production of free oxygen radicals (Serban, Badescu, Palade, Badescu, Filip, Diaconu, & Cobzeanu, 2019).

The existence of a balance between oxidative free radicals and antioxidant levels indicates the body's resistance or resistance. In many pathological conditions, there is accelerated formation of reactive oxygen species (ROS), resulting in an imbalance between oxidative factors and protective antioxidant systems. Reactive oxygen species (ROS) are important pathological mediators of many diseases under normal physiological conditions (Serban et al., 2019; Harabagiu, Cosgarea, Mogoantă, Leucuța, & Maniu, 2017).

A decrease in the activity of the antioxidant system and an increase in ROS production are known to play a role in the pathogenesis of ROS. Ultimately, ROS can directly cause oxidative injury to cells by damaging nucleic acids, proteins, lipids, and cell membranes in tissues. When oxygen free radicals interact with polyunsaturated fatty acids in the membrane or lipoproteins, the process of lipid peroxidation is initiated, and lipid peroxide (LPO) can be measured. Through a lack of antioxidant defense systems, increased oxygen free radicals may contribute to the formation of effusion otitis media. Lipid peroxidation is a reaction in which membrane proteins and receptors and the enzymes they are bound to are deactivated through the disintegration of the integrity of the cell membrane. In addition, lipid peroxidation reduces and affects membrane permeability. MDA is formed from peroxidation of membrane lipids and is the most commonly used indicator for free radicals (Sagiroglu, Ates, Tolun, & Oztarakci, 2019; Abraham, Ntunaguzi, Kahinga, Mapondella, Massawe, Nkuwi, & Nkya, 2019; Garça et al., 2013).

Under normal physiological conditions, the antioxidant defense system controls tissue damage

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caused by oxidants. This defense consists of enzymatic and non-enzymatic systems. The most important antioxidant enzymes are superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and catalase (CAT); and the most important non-enzymatic antioxidants are glutathione, tocopherol (vitamin E), ascorbic acid (vitamin C), carotene (vitamin A), albumin, bilirubin and uric acid. Oxidant-induced DNA damage in chronic diseases can prevent the synthesis of antioxidant enzymes and glutathione, thus, the damage can be exacerbated as the process continues. In chronic infections, antioxidants slowly decrease when oxidative stress reaches neutralizing antioxidant levels. Thus, oxidative stress can be both a cause and a result of chronic infections (Garça et al., 2013).

Based on descriptive data regarding the frequency of sex in the sample, this study showed that CSOM patients were dominated by female, namely 12 people (60%), while male participant in this study totaled 8 people (40%). This is slightly different from previous literature which states that the percentage of CSOM patients is dominated by males, although there is no significant difference between male and female patients. This difference can be influenced by several factors, such as taking sample patients in an area where the proportion of males and females is quite different which will affect the percentage of CSOM patients seeking treatment at the health center where the study was conducted (Abraham et al., 2019; Abdelhafeez, & Mohamed, 2021). The type of hearing loss that is often experienced is CHL. This is in accordance with previous research which showed that in CSOM, more CHL were found compared to patients who experienced MHL (Subramaniam, Ashkar, & Rai, 2020). Participants' education was balanced between high and low categories. Apart from that, socioeconomic status is in the high category. This shows that a person's high level of education is directly proportional to their high awareness of health and desire to receive treatment.

Most of the research subjects did not smoke. Other studies showed a prevalence of otitis media of 36.7% when there were family members who smoked. Otitis media was found to have a statistically significant association with passive smoking among parents and family members (Paneru, Shah, & Chettri, 2021). The effect of cigarette smoke is

damage to the cilia and airway epithelium, making a person more susceptible to bacterial infections. This will cause otitis media and can progress to CSOM. Although there are many anti-tobacco campaigns and advertisements highlighting the carcinogenic effects of smoking, there is little awareness that passive smoking can have negative impacts on respiratory and otological health.

In resisting oxygen free radicals and their derivatives (oxygen-derived free radicals), the body activates a body defense complex called the antioxidant defense complex and oxidation formed by free radicals will be inhibited by antioxidants through a mechanism of stabilization and capture of free radicals. As a result, the body will initiate an enzyme system as an antioxidant against toxic substances resulting from the oxidation process, including the formation of malondialdehyde (MDA) (Kalyanaraman, 2013).

Based on the results of data analysis, it was found that there was a positive and significant correlation between Interleukin 17 (IL-17) levels and malondialdehyde (MDA) levels in the blood of patients with chronic suppurative otitis media (CSOM) with a high coefficient value of 0.623. This study is in line with the results concluding that the IL-17 pathway plays an important role in the pathogenesis of OME by increasing ROS, including MDA levels (Abdelhafeez, & Mohamed, 2021).

CONCLUSION

There is a positive and significant correlation between IL-17 levels and MDA in the blood of patients with chronic suppurative otitis media (CSOM).

SUGGESTION

The results of this research can be used as reference material for future researchers to carry out similar and more in-depth research and for further research it is hoped that they can be added to the examination of the enzymes nitric oxide (NO), catalase (CT), myeloperoxidase (MPO), superoxide dismutase (SOD), glutathione peroxidase (GPX) which can attack patients with chronic suppurative otitis media (CSOM) and samples of middle ear secretions were examined in CSOM patients.

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