Supplementation of omega-3 fatty acids can reduce tumor necrosis alpha (TNF-α) levels and pain intensity in osteoarthritis patients

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ABSTRAK

Latar Belakang : Osteoarthritis (OA) merupakan penyakit sendi yang paling umum di seluruh dunia, dengan dampak yang kuat pada kesehatan individu dan populasi. Secara global prevalensi OA meningkat sebesar 113,2% dari 247,5 juta pada tahun 1990 menjadi 527,8 juta pada tahun 2019. Prevalensi OA di Indonesia meningkat seiring bertambahnya usia yaitu sebesar 5% pada individu berusia kurang dari 40 tahun, 30% pada usia 40-60 tahun, dan 65% pada usia di atas 60 tahun. Angka kejadian OA lutut relatif tinggi, yaitu 15,5% pada pria dan 12,7% pada wanita. Penyakit ini dapat menyebabkan nyeri kronis, kecacatan, dan menurunkan kualitas hidup penderita. Tingginya kadar sitokin pro inflamasi seperti TNF-α pada penderita OA dapat berperan sebagai faktor utama yang menginduksi kematian krondosit dan menghambat diferensiasi serta proliferasi kondrosit. Penelitian sebelumnya mengatakan bahwa suplementasi omega 3 dapat menurunkan kadar sitokin proinflamasi, memperbaiki nyeri, dan kekakuan fungsi fisik pada individu dengan OA lutut, namun belum ada yang meneliti pengaruh suplementasi omega 3 ini dengan penurunan kadar Tumor Necrosis Factor α (TNF-α).
Tujuan : Menganalisis pengaruh suplementasi asam lemak omega 3 terhadap kadar *TNF-α* dan intensitas nyeri pada penderita osteoarthritis.
Metode : Studi ini merupakan *quasi-experimental* desain kelompok tunggal dengan rancangan penelitian *one group pre-post test without control group design* pada 31 pasien. Pengumpulan data dilakukan dengan pengukuran antropometri, pengambilan sampel darah dan pengukuran skor nyeri dilakukan sebanyak dua kali, yaitu sebelum dan sesudah dilakukannya intervensi. analisa *TNF-α* menggunakan *Enzym Linked Immunosorbent Assay* (*Elisa*), intensitas nyeri menggunakan *numeric rating scale* (NRS). Analisis uji statistik pengaruh suplementasi asam lemak omega 3 terhadap kadar *TNF-α* dan intensitas nyeri menggunakan uji *Wilcoxon*.
Hasil : Hasil yang diperoleh pada *TNF-α* dengan nilai *P*=0,007 dan intensitas nyeri dengan nilai *P*=0,001 yang menunjukkan hasil yang signifikan.
Kesimpulan : Suplementasi asam lemak omega 3 pada penderita OA dapat mengasilkan beberapa mediator anti inflamasi yang mampu menurunkan kadar sitokin pro inflamasi *TNF-α* dan intensitas nyeri secara siginifikan.

KATA KUNCI*: asam lemak omega 3 ; tumor necrosis alpha (TNF-α); intensitas nyeri; inflamasi; osteoarthritis*

ABSTRACT

Background: Osteoarthritis (OA) is the most common joint disease worldwide, strongly impacting individual and population health. Globally, the prevalence of OA increased by 113.2% from 247.5 million in 1990 to 527.8 million in 2019. The prevalence of OA in Indonesia increases with age, namely by 5% in individuals under 40 years, 30% in those aged 40-60 years, and 65% in those over 60 years. The incidence of knee OA is relatively high, namely 15.5% in men and 12.7% in women. This disease can cause chronic pain and disability and reduce the quality of life of sufferers. High levels of pro-inflammatory cytokines such as TNF-α in OA sufferers can act as a major factor in inducing chondrocyte death and inhibiting chondrocyte differentiation and proliferation. Previous studies have shown that omega-3 supplementation can reduce proinflammatory cytokine levels and improve pain and stiffness of physical function in individuals with knee OA. Still, no one has studied the effect of omega-3 supplementation on reducing TNF-α levels.
Objective: To analyze the effect of omega-3 fatty acid supplementation on TNF-α levels and pain intensity in patients with osteoarthritis.
Method: This study is a quasi-experimental single-group design with a one-group pre-post test without a control group design in 31 patients. Data collection was carried out by anthropometric measurements, blood sampling, and pain score measurements twice, namely before and after the intervention. TNF-α analysis using Enzyme-Linked Immunosorbent Assay (Elisa), pain intensity using a numeric rating scale (NRS). Statistical analysis of the effect of omega 3 fatty acid supplementation on TNF-α levels and pain intensity using the Wilcoxon test.
Results: The results obtained on TNF-α with a P value = 0.007 and pain intensity with a P value = 0.001 showed significant results.
Conclusion: Supplementation of omega-3 fatty acids in OA patients can produce several anti-inflammatory mediators that significantly reduce the levels of pro-inflammatory cytokines TNF-α and pain intensity.

KEYWORDS: omega 3 fatty acids; tumor necrosis alpha (TNF-α); pain intensity; inflammation; osteoarthritis

INTRODUCTION

Osteoarthritis (OA) is a chronic joint disease characterized by degeneration and destruction of the joint cartilage (1). OA is the most common joint disease worldwide, strongly impacting individual and population health (2). Globally, the prevalence of OA increased by 113.2% from 247.5 million in 1990 to 527.8 million in 2019. The prevalence of OA in Indonesia increases with age, with 5% in individuals aged less than 40 years, 30% in those aged 40-60 years, and 65% in those aged over 60 years. The incidence of knee OA is relatively high, at 15.5% in men and 12.7% in women (3).

OA is a leading cause of disability in the elderly, affecting more than 50 million people in the United States. The disease can cause chronic pain and disability and reduce patients' quality of life (4,5). The pathological process of osteoarthritis involves complex interactions between immune, metabolic, hormonal, and genetic factors.

These factors damage and destroy articular cartilage, bone, synovium, and other tissues, causing joint pain and dysfunction. However, the exact mechanism causing this disease remains unclear (6,7).

Many risk factors influence the pathogenesis of OA, including age, gender, genetic factors, obesity, and physical factors, including trauma and joint damage. Aging can be characterized by progressive tissue and organ function loss, which is the greatest risk factor for OA (8). Hormonal differences between men and women play a role in the development of osteoarthritis. Most cases of osteoarthritis generally occur in postmenopausal women because the hormones estrogen and progesterone, which were originally balanced, are reduced, resulting in an increased risk of developing arthritis (9). Estrogen deficiency causes an unbalanced acceleration of bone remodeling where osteoclast activity is much higher than osteoblast activity. This causes thinning of the bone cortex, which results in thinning of trabecular bone and loss of trabecular elements (10).

Patients with OA often experience pain, stiffness, swelling, and limited movement in the affected joint. Articular cartilage is composed of chondrocytes and extracellular matrix. Normally, chondrocytes play an important role in synthesizing extracellular matrix and degrading it with the help of *proteolytic* enzymes. In patients with OA, there is an imbalance of these two processes. The degradation process increases, possibly triggered by trauma, mechanical stress, and inflammatory processes that produce cytokines Interleukin-1 (IL-1), Interleukin-6 (IL-6), and Tumor Necrosis Factor-alpha (TNF-α) (7,11).

Omega-3 fatty acids (EPA and DHA) produce several anti-inflammatory mediators, such as resolvins, protectins, and maresins, which are released in large amounts during inflammation (12, 13). Omega-3 fatty acid supplementation can significantly relieve arthritis pain and improve joint function in OA patients. EPA reduces apoptosis caused by oxidative stress and chondrocyte matrix loss by inhibiting metalloproteinase13 expression and chondrocyte apoptosis (14).

No studies have assessed the effect of omega-3 supplementation on *TNF-α* levels and pain intensity in Indonesia until now. Therefore, this study is proposed to determine and evaluate the effect of omega-3 supplementation on reducing *TNF-α* proinflammatory cytokine levels as a new nutritional perspective on reducing pain in patients with OA.

MATERIALS AND METHODS

This study is a *Pre-experimental* single-*group* design with a research design of *one group pre-post-test without a control group* design. It only looks at the treatment results in one group of objects without any comparison group or control group. The independent variable in this study is *omega-3* fatty acid supplementation, while the dependent variable is the *Tumor Necrosis Alpha (TNF-α*) level and pain intensity. This study was conducted at Diponegoro National Hospital (RSND) in Semarang City from May to July 2024.

We evaluated 31 individuals with Osteoarthritis aged 50-70 years old who were given omega-3 fatty acid supplementation obtained commercially from PT. Vita Shopindo Jakarta. The capsule contains 1000 mg fish oil with EPA 180 mg and DHA 120 mg as a soft gel with BPOM SI164307531. The dose is administered per day 3 times with a total dose of 540 mg EPA and 360 DHA. Treatment time is 4 weeks. The exclusion criteria of this study are having a history of trauma, congenital abnormalities, infections, tumors, and autoimmune diseases in the knee joint, as well as performing knee joint replacement surgery. Having an allergy to seafood and taking anticoagulant drugs. This study has received approval from the Ethics Committee of the Faculty of Medicine, Sebelas Maret University Surakarta, with Number 47/UN27.06.11/KEP/EC/2024.

Data analysis was carried out using descriptive analysis and hypothesis testing. Descriptive analysis of dependent variables will be presented in tabular form by presenting the mean and standard deviation. The data normality test was carried out using the *Saphiro-Wilk* test, and then the hypothesis test was used using the *Wilcoxon* non-parametric test. Data analysis was carried out using SPSS Ver. 23 for windows.

RESULTS AND DISCUSSIONS

Table 1. Characteristics of Research Respondents

Characteristics	n(%)	mean±SD
Gender		
Male	6 (19.4%)	-
Female	25 (80.6%)	-
Age	-	61.26±5.86
IMT		27.10±4.01
TNF-α		
Pre	-	62.60±77.22
Post	-	52.03±53.71
Pain intensity		

Pre	-	4.85±2.09
Post	-	3.44±2.17

In this study, it can be seen in **Table 1** that the majority of subjects were (80.6%) female and (19.4%) male. Their average age was 61.26±5.86 years, while the average BMI of the subjects was 61.26±5.86, which is included in the Obesity I category.

The average *TNF-α* level before omega 3 fatty acid supplementation was 62.60 ± 77.22, and after omega 3 supplementation with a dose of 540 EPA and 360 DHA per day for 4 weeks to 52.03 ± 53.71 while the average pain intensity before omega 3 supplementation was 4.85 ± 2.09 to 3.44 ± 2.17 after omega 3 fatty acid supplementation.

Table 2. Effect of Omega 3 Fatty Acid Supplementation with *TNF-α* and Pain Intensity

Variables	**Median (Min-Max)**	***p***
TNF-α		
Pre	39.4 (7.10-392.70)	0.007
Post	36.5 (6.00-272.70)	
Intensity Nyeri		
Pre	4.7 (1.50-9.25)	0.001
Post	3.0 (0.50-9.25)	

Table 2 shows that the median *TNF-α* level before supplementation was 39.4 with a range of 7.10 to 392.70, while the median *TNF- α* level after supplementation was 36.5 with a range of 6.00 to 272.70. This significant decrease in *TNF-α* levels occurred after patients were given omega-3 fatty acid supplementation over 28 days. Furthermore, the statistical test results using the Wilcoxon method showed a significant *p-value* ($p<0.05$), indicating that the observed changes in *TNF-α* levels were statistically significant. Thus, these results indicate a positive effect of omega-3 fatty acid supplementation on reducing *TNF-α* levels in patients suffering from osteoarthritis (OA).

Table 2 shows that the median pain intensity score before supplementation was 4.75, with a range of 1.50 to 9.26, while the median pain intensity score after supplementation was 3.00, with a range of 0.50 to 9.25. A significant decrease in pain intensity scores occurred after the subjects were given omega-3 fatty acid supplementation over 28 days. Furthermore, the statistical test results using the

Wilcoxon method showed a significant p-value ($p<0.05$), indicating that the observed changes in *TNF-α* levels were statistically significant. These results indicate a positive effect of omega-3 fatty acid supplementation on reducing *TNF-α* levels in patients suffering from osteoarthritis (OA).

Omega-3 fatty acids (EPA and DHA) produce several anti-inflammatory mediators, such as resolvins, protectins, and maresins, which are released in large amounts during inflammation (12). Resolvins, protectins, and maresins are all oxylipin metabolites classified as lipid modulators with specific abilities to reduce inflammation (15).

Resolvins, which include the E series of EPA (such as RvE1, RvE4, and 18S-RvE1) and the D series of DHA (such as RvD1 to RvD6), function as compounds that trigger or activate specific biological responses to regulate or control the peak phase of inflammation (4). Resolvins are metabolically processed by several enzymes, including lipoxygenase (LOX), cytochrome P450, and cyclooxygenase (COX). Resolvins bind to G- protein receptors found on various cell types, such as monocyte-macrophages, lymphocytes, endothelial cells, vascular smooth muscle cells, and neutrophils, each cell- and organ-specific. The cellular targets of resolvins depend on the specific receptors, but several resolvins subtypes may have the same cell targets but with different effects. RvD2 inhibits the movement of neutrophils, which helps reduce inflammation. Resolvins also affect macrophage responses to inflammation by converting macrophages from pro-inflammatory type (M1) to anti-inflammatory type (M2), decreasing monocyte conversion, modulating macrophage polarization, and affecting macrophage-associated IL-10 and 5- LOX. In addition, resolvin regulates leukocytes, which play a major role in tissue damage due to inflammation, by reducing CD4+ T cell expression, facilitating NK cell migration to eliminate eosinophilic granulocytes, and reducing TNF-α and IL-6 secretion (16).

Protectin D is a compound formed through the double lipooxygenation process of the omega-3 fatty acid DHA. This process involves the oxidation of DHA by the enzyme lipoxygenase, which produces various metabolites, including Protectin D. This compound plays an important role in modulating inflammatory responses and tissue protection. Protectin DX (PDX) plays an important role in controlling chronic and acute inflammation by affecting the AMPK signaling pathway, which helps regulate the body's response to inflammation (17).

Several studies on cell culture and experimental animals prove resolvin is a good anti-inflammatory and immunomodulator. For example, resolvin D1 can restrain

the expression of TRAP, cathepsin K, TNF-α, IL-1 β, IFN-γ, and PGE2 to inhibit osteoclast differentiation and activation. The mechanism of Resolvin D1 against Rheumatoid Arthritis (RA) has been further elucidated in a mouse model of collagen-induced arthritis (CIA); namely, resolvin D1 alleviates the development of RA by inhibiting CTGF through upregulating miRNA-146a-5p. Furthermore, an arthritic mouse model was used to explore the action of resolving D5, and the results showed that resolving D5 attenuates osteoclast differentiation to interfere with osteoclast genesis involved in RA pathogenesis. Another study showed that GPR101 is a top receptor candidate for resolvin D5 that exhibits antarthritic actions by regulating neutrophil and macrophage responses. Resolvin E1 inhibits bone resorption and osteoclastogenesis by suppressing the expression of the transcription factors nuclear factor of activated T cells c1 (NFATc1) and c-fos (18).

Omega 3 fatty acid supplementation has been shown to have significant effects in relieving arthritis pain and improving joint function in patients with osteoarthritis (OA). One of the main modes of action of omega-3 fatty acids, especially EPA, is reducing the apoptotic process triggered by oxidative stress in the joints. Oxidative stress, which can damage joint cells, often leads to unwanted cell death, including chondrocytes, important cells in joint cartilage tissue. EPA works by inhibiting the expression of metalloproteinase-13, an enzyme involved in extracellular matrix degradation, and reducing chondrocyte apoptosis, thus helping maintain cartilage health and function (14).

In vitro research shows that omega-3 fatty acids lower proinflammatory cytokine levels. Animal experiments show that omega-3 fatty acids can protect cartilage after the transsection of the Anterior Cruciate Ligament (ACL) in mice, which decreases bone loss and angiogenesis so that it can protect against OA pathogenesis. (19) Omega 3 supplementation in rats with Rheumatoid Arthritis (RA) can lower TNF-α levels. (20) Research conducted by Stonehouse et al. on humans evaluated the effect of krill oil with a dose of 4 g/day containing (0.6 g/day EPA, 0.28 g/day DHA, and 0.45 g/day astaxanthin) which can improve pain, stiffness of physical function in individuals with mild to moderate knee OA (21).

In addition, EPA plays an important role in reducing structural damage to the joint by reducing the loss of chondrocyte matrix caused by the inflammatory process. Suppressing metalloproteinase 13 activity with EPA helps protect joint cartilage from further degradation and supports tissue repair, which can reduce pain and improve joint

function so that patients can experience improved quality of life. Thus, omega-3 fatty acid supplementation effectively manages osteoarthritis and improves joint health (14).

CONCLUSIONS AND RECOMMENDATIONS

Omega-3 fatty acid supplementation at 540 mg EPA and 360 mg DHA significantly decreased TNF-α levels and reduced pain intensity in subjects with osteoarthritis. These findings suggest that adding omega-3 fatty acids may benefit the management of pain and inflammation in osteoarthritis, contributing to a comprehensive therapeutic strategy for the condition.

REFERENCES

1. Permadi AW. Fisioterapi Manajemen Komprehensif Praklinik. Penerbit Buku Kedokteran; 2019.
2. Hunter DJ, Bierma-Zeinstra S. Osteoarthritis. Vol. 393, The Lancet. Lancet Publishing Group; 2019. p. 1745–59.
3. Siddik M, Darjanti Haryadi R. The Risk Factors Effect of Knee Osteoarthritis Towards Postural Lateral Sway. Vol. 14, Indian Journal of Forensic Medicine & Toxicology.
4. Liu S, Roemer F, Ge Y, Bedrick EJ, Li ZM, Guermazi A, et al. Comparison of evaluation metrics of deep learning for imbalanced imaging data in osteoarthritis studies. Osteoarthritis and Cartilage. 2023 Sep 1;31(9):1242–8.
5. Sudaryanto WT, Muhlisdo A, Soemanto RB. Biopsychosocial Factors Affecting Functional Disability and Depression in Patients with Osteoarthritis in Surakarta, Central Java. Journal Epidemiology Public Health [Internet]. 2018;3(2):292–306. Available from: https://doi.org/10.26911/jepublichealth.2018.03.02.08
6. Liao Z, Han X, Wang Y, Shi J, Zhang Y, Zhao H, et al. Differential Metabolites in Osteoarthritis: A Systematic Review and Meta-Analysis. Vol. 15, Nutrients. Multidisciplinary Digital Publishing Institute (MDPI); 2023.
7. Coaccioli S, Sarzi-Puttini P, Zis P, Rinonapoli G, Varrassi G. Osteoarthritis: New Insight on Its Pathophysiology. Vol. 11, Journal of Clinical Medicine. MDPI; 2022.
8. He Y, Li Z, Alexander PG, Ocasio-Nieves BD, Yocum L, Lin H, et al. Pathogenesis of osteoarthritis: Risk factors, regulatory pathways in chondrocytes, and experimental models. Vol. 9, Biology. MDPI AG; 2020. p. 1–32.
9. Peshkova M, Lychagin A, Lipina M, Di Matteo B, Anzillotti G, Ronzoni F, et al.

Gender-Related Aspects in Osteoarthritis Development and Progression: A Review. Vol. 23, International Journal of Molecular Sciences. MDPI; 2022.

10. Inayati R, Darma Isasih W. Peran Estrogen Remodelling Tulang Pada Kasus Osteoporosis Post-Menopausal The Role Of Estrogen On Bone Remodelling In Post-Menopausal Osteoporosis. Jurnal Inovasi Pendidikan dan Sains. 4(3):159–66.
11. Pramana Putra B, Putri Sahara M, Istri Agung Asvini C, Gusti Bagus Surya Ari I, Diah Permata K, Pranayoga K, et al. Nutraceutical dari Teripang sebagai Terapi Adjuvan Osteoartritis. Vol. 1, Lombok Medical Journal. 2022.
12. Gropper SS, Smith JL. Advance Nutrition and Human Metabolism. 6th ed. Canada: Wadsworth Cengage Learning; 2013.
13. Oliver L, Dietrich T, Marañón I, Villarán MC, Barrio RJ. Producing omega-3 polyunsaturated fatty acids: A review of sustainable sources and future trends for the EPA and DHA market. Vol. 9, Resources. MDPI AG; 2020. p. 1–15.
14. Deng W, Yi Z, Yin E, Lu R, You H, Yuan X. Effect of omega-3 polyunsaturated fatty acids supplementation for patients with osteoarthritis: a meta-analysis. Journal of Orthopaedic Surgery and Research. 2023 Dec 1;18(1).
15. Mustonen AM, Nieminen P. Fatty Acids and Oxylipins in Osteoarthritis and Rheumatoid Arthritis-a Complex Field with Significant Potential for Future Treatments. 1926; Available from: https://commons.wikimedia.org/wiki/File:Docosahexaenoic-
16. Shawl M, Geetha T, Burnett D, Babu JR. Omega-3 Supplementation and Its Effects on Osteoarthritis. Vol. 16, Nutrients. 2024.
17. Piao S, Du W, Wei Y, Yang Y, Feng X, Bai L. Protectin DX attenuates IL-1β-induced inflammation via the AMPK/NF-κB pathway in chondrocytes and ameliorates osteoarthritis progression in a rat model. International Immunopharmacology. 2020 Jan 1;78.
18. Liu C, Fan D, Lei Q, Lu A, He X. Roles of Resolvins in Chronic Inflammatory Response. Vol. 23, International Journal of Molecular Sciences. MDPI; 2022.
19. Cordingley DM, Cornish SM. Omega-3 Fatty Acids for the Management of Osteoarthritis: A Narrative Review. Vol. 14, Nutrients. MDPI; 2022.
20. Lanchais K, Capel F, Tournadre A. Could omega 3 fatty acids preserve muscle health in rheumatoid arthritis? Vol. 12, Nutrients. MDPI AG; 2020.
21. Stonehouse W, Benassi-Evans B, Bednarz J, Vincent AD, Hall S, Hill CL. Krill oil improved osteoarthritic knee pain in adults with mild to moderate knee

osteoarthritis: a 6-month multicenter, randomized, double-blind, placebo-controlled trial. The American Journal of Clinical Nutrition. 2022 Sep 1;116(3):672–85.