

Effects of the systemic administration of omega-3 polyunsaturated fatty acid on experimental periodontitis

Aveen Ageel Jalal,¹ Zewar A. AL-Qassab,¹ and Rafel A. AL-Rawi²

¹College of Dentistry, Hawler Medical University, Erbil, Kurdistan Region, Iraq.

²College of Pharmacology, Hawler Medical University, Erbil, Kurdistan Region, Iraq.

Correspondence to Aveen Ageel Jalal (email: aveenyawar445@gmail.com).

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Objective The present study was aimed to investigate and evaluate the effects of eicosapentaenoic acid (EPA) on ligature induced periodontitis in rats through the biochemical analysis for rat's serum level of CRP, alkaline phosphatase (ALP) and MDA.

Methods Periodontitis was induced for the studied animals by ligation around the upper central incisor. Twenty five animals were used as a control and gavaged by water only, and 100 animals with the induced periodontitis were divided into four equal groups according to the treatment used which was started at time of ligation removal: Water (P/W), scaling/root planing (P/SRP), 60 mg/kg EPA (P/EPA), and SRP together with EPA (P/SRP + EPA). Blood was taken by cardiac puncture, after 3, 24 h, 3 days, 1 week and 2 weeks for biochemical analysis.

Results The P/SRP + EPA treatment group showed significant decrease ($p < 0.05$) in serum CRP, significant increase in serum ALP, significant decrease in serum MDA after 1 week, 3 days, and 3 h respectively in comparison with the P/W treatment group.

Conclusion The treatment by SRP with 60 mg/kg EPA is better than each one alone and can be used for treatment of periodontitis.

Keywords periodontitis, eicosapentaenoic acid, alveolar bone resorption, omega-3

Introduction

Chronic periodontitis is an inflammatory disease affecting the supporting tissues of teeth, and its expression results from the interaction of host defense mechanisms, microbial agents, environmental, and genetic factors.¹ A biomarker is a substance used to indicate a biologic state and is an objective measure to evaluate the disease activity and considered as an indicator of normal biologic processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention.² Several biomarkers are associated with periodontal disease like the systemic inflammatory marker such as CRP, bone markers such as alkaline phosphatase (ALP), and oxidative stress markers such as MDA.³⁻⁵

Ide et al.⁶ found that CRP has been associated with the presence of various bacterial infections, including periodontitis. Perumal et al.⁵ found that there was a decrease in serum bone-specific ALP level in severe periodontitis patients as compared with moderate chronic periodontitis group level. Other study found that patients with periodontitis had higher values of MDA whereas healthy patients had lower levels of MDA which is commonly known as a marker of oxidative stress.⁷

Polyunsaturated fatty acids are fatty acids with more than one carbon-carbon double bond. Omega-3 are the bioactive lipids which consist of three major omega-3 fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) from marine sources, and alpha-linolenic acid (ALA) which is from plant sources.⁸ Periodontal disease affects almost 90% of the population,⁹ and the incidence increase with the growing ageing population.^{10,11} The most common periodontal treatment is still mechanical removal of plaque and calculus deposit and local antibiotic application. Thus, a therapy of natural origin, if effective, might be safer method for treatment of periodontitis. For this reason, the present study was aimed to investigate and evaluate the effects of 60 mg/kg EPA¹² on ligature-induced periodontitis in rats through the biochemical analysis for serum CRP, ALP, and MDA.

Materials and Methods

Rats and Housing

All the Wister-albino rats used in the study was aged about 8–10 weeks, weighing 200–300 g and cared in the animal house of College of Medicine, Hawler Medical University, Erbil, Iraq. They were allowed to adapt to the housing conditions for 1 week prior to the commencement of the study. Five rats were housed in each wire cage and maintained on a 12-h light/dark cycle at $20 \pm 5^\circ\text{C}$ and 20–30% humidity. The animals were kept in standard room conditions and fed with a standard rat chow and allowed to drink water ad libitum. The research project was approved by the Research Ethics Committee at College of Dentistry, Hawler Medical University under protocol.

Induction of Experimental Periodontitis

The upper incisor was chosen because the induced periodontal disease occurs more rapidly in that location due to the porosity of the spongy bone in the maxilla. The rats were anesthetized by intraperitoneal administration of ketamine (0.5 ml/kg b.w.) and the animals were placed on a proper operating table, which allowed open-mouth maintenance of the rats to facilitate access to the teeth. After that a 3.0 sterile black braided silk threads was placed around the cervix of maxillary right incisor for each animal and kept for 2 weeks. The ligatures are knotted on the labial side of the tooth, resulting in subgingival positioning on the palatal side and supragingival position on the labial side. Daily we perform ligatures control and checking, and if any had been lost or become loose, it was replaced. This ligature acts as a gingival irritant for 14 days and promoted the accumulation of plaque and subsequently development of periodontal disease.¹³

Experimental Design for Biochemical Analysis

One hundred and twenty five animals were used in the study. Twenty five animals (normal control), and 100 animals with the induced periodontitis were randomly assigned into four

experimental groups (25 animals each) according to the treatment used.

NC/W

Normal control + distilled water treatment group.

P/W

Ligature-induced periodontitis + distilled water treatment group.

P/SRP

Ligature-induced periodontitis + scaling and root planing treatment group.

The right incisors were subjected to SRP with manual #1–2 mini-five curettes through 10 distal–mesial traction movements in the labial and palatal aspects. The interproximal areas were scaled with the same curettes using cervico-incisal traction movements.¹⁴

P/EPA

Ligature-induced periodontitis + 60 mg /kg EPA treatment group.

P/SRP+EPA

Ligature-induced periodontitis + SRP + EPA treatment group.

The ligatures were removed at day 14 (day 0) and the different types of treatment was started directly. The treatment by intragastric gavage of distilled water or EPA were done daily once for 2 weeks.

Blood Sampling for Biochemical Assays

Blood was taken by cardiac puncture, 3 h after ligation removal (day 0), 24 h, 3 days, 1 week and 2 weeks for immunological analysis. First by anesthetizing the rat, placed on its back, the left index finger was placed at the level of the lowest ribs without applying any pressure, and the heart is located 1 cm above this point, slightly to the right.¹⁵ The 5 ml syringe was hold at a 45° angle and the needle was inserted between two ribs, the plunger was pulled on slowly to fill the syringe. Then the samples were placed in non-heparinized tubes, allowed to clot for 2 h at room temperature and centrifuged at 3000 rpm for 10 min at 4°C, and then the supernatant was collected. The serum was frozen at –20°C and used within 1 month to avoid loss of bioactivity and contamination.

Chemiluminescence immunoassay detection kit was used for determination of high sensitivity CRP (23961, Diagnostic

Automation Inc., Calabasas, CA, USA), colorimetric determination was used of the ALP (02160, Biolabo, France), and enzyme-linked immunosorbent assay kit was used for MDA determination (MBS764641, USA).

Statistical Analysis

Data were analyzed using SPSS software version 23, and were summarized using means and standard deviations. Statistical analysis with one-way analysis of variance was performed to compare the differences in the means among groups, and when it revealed that there was a statistically significant difference; Mann–Whitney *U*-test was performed to assess individual pair of groups for statistically significant finding. *p*-Value ≤0.05 was considered statistically significant.

Results

Serum CRP

The result showed a significant increase ($p < 0.05$) in serum CRP in P/W, P/SRP, P/EPA, and P/SRP + EPA treatment groups in the first 3 days in comparison with the NC/W treatment group. The P/EPA and P/SRP + EPA treatment groups showed a non-significant difference in serum CRP level from that of the NC/W after 7 days treatment, but not lower level to that of the healthy control group. The P/SRP treatment group showed a non-significant difference from that of NC/W treatment group after 2 weeks (Table 1).

Statistical analysis showed non-significant differences ($p > 0.05$) in serum CRP present between all treatment groups in the first 3 days. After 1 week the P/SRP + EPA treatment group showed significant decrease ($p < 0.05$) in serum CRP in comparison with the P/W, P/SRP, and P/EPA treatment groups. After 2 weeks a non-significant differences ($p > 0.05$) were seen between P/SRP and P/EPA, P/SRP and P/SRP + EPA, P/EPA and P/SRP + EPA treatment groups (Table 2).

Serum ALP

The P/W treatment group showed a significant decrease ($p < 0.05$) in serum ALP in comparison with the NC/W. Statistical analysis showed a non-significant differences ($p > 0.05$) present between P/EPA and P/SRP + EPA treatment groups in relation with the NC/W treatment groups only after 1 week treatment, but it was still less than the NC/W treatment group. A significant difference ($p < 0.05$) was seen between all groups in all other durations studied (Table 3).

Table 1. Serum CRP (mean ± standard deviation) in normal control/water treatment group in relation with all studied groups

Groups	CRP (ng/ml)									
	3 h	<i>p</i> -Value	24 h	<i>p</i> -Value	3 days	<i>p</i> -Value	1 Week	<i>p</i> -Value	2 Weeks	<i>p</i> -Value
NC/W	11.81 ± 1.186	0.012	11.61 ± 1.328	0.012	11.41 ± 1.104	0.012	11.61 ± 1.843	0.012	11.81 ± 1.177	0.012
P/W	14.61 ± 0.354		14.81 ± 1.18		14.56 ± 1.876		13.954 ± 1.45		13.093 ± 0.963	
NC/W	11.81 ± 1.186	0.012	11.61 ± 1.32	0.012	11.41 ± 1.104	0.012	11.61 ± 1.843	0.012	11.81 ± 1.177	0.731
P/SRP	15.22 ± 0.357		15.32 ± 0.451		15.06 ± 1.96		13.569 ± 1.540		12.03 ± 0.75	
NC/W	11.81 ± 1.186	0.012	11.61 ± 1.328	0.012	11.41 ± 1.104	0.012	11.61 ± 1.843	0.144	11.81 ± 1.177	0.756
P/EPA	14.45 ± 0.344		14.075 ± 0.256		13.99 ± 1.83		12.94 ± 1.275		12.312 ± 0.785	
NC/W	11.81 ± 1.186	0.012	11.61 ± 1.328	0.012	11.41 ± 1.104	0.012	11.61 ± 1.843	0.173	11.81 ± 1.177	0.347
P/SRP + EPA	14.057 ± 1.467		14.99 ± 1.985		13.89 ± 0.34		12.01 ± 0.834		12.01 ± 1.157	

Table 2 The relation of serum CRP (mean ± standard deviation) between different studied treatments groups

Groups	CRP (ng/ml)									
	3 h	p-Value	24 h	p-Value	3 days	p-Value	1 Week	p-Value	2 Weeks	p-Value
P/W	14.61 ± 0.354	0.92	14.81 ± 1.1815	0.731	14.56 ± 1.876	0.920	13.954 ± 1.45	0.833	13.093 ± 0.963	0.012
P/SRP	15.22 ± 0.357		15.32 ± 0.451		15.06 ± 1.96		13.569 ± 1.540		12.03 ± 0.75	
P/W	14.61 ± 0.354	1.00	14.81 ± 1.18	0.400	14.56 ± 1.876	0.828	13.954 ± 1.45	0.250	13.093 ± 0.9631	0.012
P/EPA	14.45 ± 0.344		14.075 ± 0.256		13.99 ± 1.83		12.94 ± 1.275		12.312 ± 0.785	
P/W	14.61 ± 0.354	1.00	14.81 ± 1.18	0.417	14.56 ± 1.876	0.528	13.954 ± 1.45	0.012	13.093 ± 0.963	0.012
P/SRP + EPA	14.057 ± 1.467		14.99 ± 1.985		13.89 ± 0.34		12.01 ± 0.834		12.01 ± 1.157	
P/SRP	15.22 ± 0.35711	0.828	15.32 ± 0.451	0.920	15.06 ± 1.96	0.674	13.569 ± 1.540	0.144	12.03 ± 0.75	0.920
P/EPA	14.45 ± 0.34		14.075 ± 0.256		13.99 ± 1.83		12.94 ± 1.275		12.312 ± 0.785	
P/SRP	15.22 ± 0.357	0.400	15.32 ± 0.451	1.00	15.06 ± 1.96	0.313	13.569 ± 1.540	0.012	12.03 ± 0.75	1.00
P/SRP + EPA	14.057 ± 1.467		14.99 ± 1.985		13.89 ± 0.34		12.01 ± 0.834		12.01 ± 1.157	
P/EPA	14.45 ± 0.344	1.00	14.075 ± 0.256	0.920	13.99 ± 1.83	1.00	12.94 ± 1.275	0.012	12.312 ± 0.785	1.00
P/SRA + EPA	14.057 ± 1.467		14.99 ± 1.985		13.89 ± 0.34		12.01 ± 0.834		12.01 ± 1.157	

Table 3. Serum ALP (mean ± standard deviation) in normal control/water treatment group in relation with all studied groups

Groups	ALP (U/L)									
	3 h	p-Value	24 h	p-Value	3 days	p-Value	1 Week	p-Value	2 Weeks	p-Value
NC/W	131.07 ± 2.595	0.012	131.27 ± 2.72	0.012	129.87 ± 3.476	0.012	131.47 ± 4.193	0.012	132.27 ± 4.543	0.021
P/W	114 ± 4.149		114.87 ± 3.827		116.076 ± 4.027		116.9 ± 0.761		117.5 ± 1.014	
NC/W	131.07 ± 2.595	0.012	131.27 ± 2.72	0.008	129.87 ± 3.476	0.012	131.47 ± 4.193	0.012	132.27 ± 4.543	0.012
P/SRP	114.47 ± 4.009		113.73 ± 1.896		115.476 ± 3.26		114.4 ± 2.80		118.276 ± 4.246	
NC/W	131.07 ± 2.595	0.012	131.27 ± 2.72	0.012	129.87 ± 3.476	0.012	131.47 ± 4.193	0.060	132.27 ± 4.543	0.094
P/EPA	115.876 ± 4.19		116.45 ± 2.34		120.7 ± 1.256		123.9 ± 2.546		124.5 ± 1.526	
NC/W	131.07 ± 2.595	0.012	131.27 ± 2.72	0.021	129.87 ± 3.476	0.021	131.47 ± 4.193	0.060	132.27 ± 4.543	0.060
P/SRP + EPA	118.5 ± 4.402		120.3 ± 3.389		122.7 ± 1.526		124.1 ± 1.797		124.9 ± 1.968	

The P/EPA and P/SRP + EPA treatment groups showed significant increase ($p < 0.05$) in serum ALP after 3 days in comparison with the P/W treatment group. The P/EPA and P/SRP + EPA treatment groups showed significant increase ($p < 0.05$) in serum ALP after 3 days treatment in comparison with the P/SRP treatment group. The P/SRP + EPA treatment group showed a significant difference from P/EPA treatment group after 3 and 24 h only (Table 4).

Serum MDA

Statistical analysis showed that all types of treatment for periodontitis (water, SRP, EPA, or by SRP + EPA) showed significant increase ($p < 0.05$) in serum MDA in relation with the NC/W treatment group (Table 5).

Statistical analysis showed that the treatment by SRP, EPA, SRP + EPA caused significant decrease ($p < 0.05$) in serum MDA in comparison with the P/W treatment group in all durations studied. No significant differences ($p > 0.05$) were seen between P/SRP and P/EPA, P/SRP and P/SRP + EPA, or P/EPA and P/SRP + EPA treatment groups in all durations studied (Table 6).

Discussion

Serum CRP

CRP is an acute phase protein, synthesized by the liver and circulates in the blood and rise in response to inflammation and used as an inflammatory marker.⁶ The present study showed that P/W treatment group showed significant increase in level of serum CRP. Ekuni et al.¹⁶ found that the serum CRP in normal rats was 1.22 mg/ml, while in the periodontitis group showed a significantly 12% increase compared with that of the control group at time of suture removal. Buhlin et al.,¹⁷ Craig et al.,¹⁸ Thakare et al.,¹⁹ Gupta et al.,²⁰ Masi et al.,²¹ Aziz et al.,²² and Monisha and Savitha⁷ also found that the levels of CRP were significantly higher in periodontitis patients than healthy control. The balance between ROS and antioxidant defense mechanism is important in the pathogenesis of periodontal disease, and those subjects who were infected with periodontal pathogens had clearly higher levels of CRP than those who were not harboring the putative bacteria in their subgingival plaque. These results disagree with that of Buduneli et al.²³ result, they induced experimental periodontitis in rats by

Table 4. The relation of serum ALP (mean ± standard deviation) between different studied treatments groups

Groups	ALP (U/L)									
	3 h	p-Value	24 h	p-Value	3 days	p-Value	1 Week	p-Value	2 Weeks	p-Value
P/W	114 ± 4.149	0.920	114.87 ± 3.827	0.857	116.076 ± 4.027	0.833	116.9 ± 0.761	0.173	117.5 ± 1.014	0.674
P/SRP	114.47 ± 4.009		113.73 ± 1.896		115.476 ± 3.260		114.4 ± 2.80		118.276 ± 4.246	
P/W	114 ± 4.149	0.920	114.87 ± 3.827	0.603	116.076 ± 4.027	0.0463	116.9 ± 0.761	0.012	117.5 ± 1.014	0.012
P/EPA	115.876 ± 4.19		116.45 ± 2.34		120.7 ± 1.256		123.9 ± 2.546		124.5 ± 1.526	
P/W	114 ± 4.149	0.144	114.87 ± 3.827	0.116	116.076 ± 4.027	0.0164	116.9 ± 0.761	0.012	117.5 ± 1.014	0.012
P/SRP + EPA	118.5 ± 4.402		120.3 ± 3.389		122.7 ± 1.526		124.1 ± 1.797		124.9 ± 1.968	
P/SRP	114.47 ± 4.009	0.756	113.73 ± 1.896	0.412	115.476 ± 3.260	0.036	114.4 ± 2.80	0.012	118.276 ± 4.246	0.021
P/EPA	115.876 ± 4.19		116.45 ± 2.34		120.7 ± 1.256		123.9 ± 2.546		124.5 ± 1.526	
P/SRP	114.47 ± 4.009	0.144	113.73 ± 1.896	0.083	115.476 ± 3.260	0.012	114.4 ± 2.80	0.012	118.276 ± 4.246	0.036
P/SRP + EPA	118.5 ± 4.402		120.3 ± 3.389		122.7 ± 1.526		124.1 ± 1.797		124.9 ± 1.968	
P/EPA	115.876 ± 4.19	0.014	116.45 ± 2.34	0.034	120.7 ± 1.256	0.057	123.9 ± 2.546	0.527	124.5 ± 1.526	1.00
P/SRP + EPA	118.5 ± 4.402		120.3 ± 3.389		122.7 ± 1.526		124.1 ± 1.797		124.9 ± 1.968	

Table 5. Serum MDA (mean ± standard deviation) in normal control/water treatment group in relation with all studied groups

Groups	MDA (ng/ml)									
	3 h	p-Value	24 h	p-Value	3 days	p-Value	One Week	p-Value	Two Weeks	p-Value
NC/W	22.12 ± 0.920	0.012	21.9 ± 0.663	0.012	22.3 ± 1.204	0.012	22.5 ± 0.707	0.012	22.4 ± 0.894	0.012
P/W	32.8 ± 1.303		32.4 ± 1.516		31.6 ± 1.14		31.2 ± 1.303		30.6 ± 0.894	
NC/W	22.12 ± 0.920	0.012	21.9 ± 0.663	0.012	22.3 ± 1.204	0.012	22.5 ± 0.707	0.012	22.4 ± 0.894	0.012
P/SRP	27.54 ± 1.143		26.34 ± 1.275		26.94 ± 1.190		26.74 ± 1.26		26.34 ± 1.768	
NC/W	22.12 ± 0.920	0.012	21.9 ± 0.663	0.012	22.3 ± 1.204	0.012	22.5 ± 0.707	0.012	22.4 ± 0.894	0.028
P/EPA	26.44 ± 1.43		26.54 ± 1.28		26.34 ± 1.275		26.14 ± 1.41		25.74 ± 0.859	
NC/W	22.12 ± 0.920	0.012	21.9 ± 0.663	0.012	22.3 ± 1.204	0.012	22.5 ± 0.707	0.028	22.4 ± 0.894	0.028
P/SRP+EPA	26.5 ± 1.903		25.94 ± 1.663		25.64 ± 1.273		25.34 ± 1.08		25.14 ± 1.431	

Table 6. The relation of serum MDA (mean ± standard deviation) between different studied treatments groups

Groups	MDA (ng/ml)									
	3 h	p-Value	24 h	p-Value	3 days	p-Value	1 Week	p-Value	2 Weeks	p-Value
P/W	32.8 ± 1.303	0.012	32.4 ± 1.516	0.012	31.6 ± 1.14	0.012	31.2 ± 1.303	0.012	30.6 ± 0.894	0.012
P/SRP	27.54 ± 1.143		26.34 ± 1.275		26.94 ± 1.190		26.74 ± 1.26		26.34 ± 1.768	
P/W	32.8 ± 1.303	0.012	32.4 ± 1.516	0.012	31.6 ± 1.14	0.012	31.2 ± 1.303	0.012	30.6 ± 0.894	0.012
P/EPA	26.44 ± 1.43		26.54 ± 1.28		26.34 ± 1.275		26.14 ± 1.41		25.74 ± 0.859	
P/W	32.8 ± 1.303	0.012	32.4 ± 1.516	0.012	31.6 ± 1.14	0.012	31.2 ± 1.303	0.012	30.6 ± 0.894	0.012
P/SRP + EPA	26.5 ± 1.903		25.94 ± 1.663		26.34 ± 1.275		25.34 ± 1.08		25.14 ± 1.431	
P/SRP	27.54 ± 1.143	0.920	26.34 ± 1.275	0.920	26.94 ± 1.190	0.920	26.74 ± 1.26	0.920	26.34 ± 1.768	0.347
P/EPA	26.44 ± 1.43		26.54 ± 1.28		26.34 ± 1.275		26.14 ± 1.41		25.74 ± 0.859	
P/SRP	27.54 ± 1.143	0.920	26.34 ± 1.275	0.674	26.94 ± 1.190	0.920	26.74 ± 1.26	0.920	26.34 ± 1.768	0.250
P/SRP + EPA	26.5 ± 1.903		25.94 ± 1.663		26.34 ± 1.275		25.34 ± 1.08		25.14 ± 1.431	
P/EPA	26.44 ± 1.43	0.400	26.54 ± 1.28	0.603	26.34 ± 1.275	0.920	26.14 ± 1.41	0.920	25.54 ± 0.841	1.00
P/SRP + EPA	26.50 ± 1.903		25.94 ± 1.663		26.34 ± 1.275		25.34 ± 1.08		25.74 ± 0.859	

repeated injection of purified lipopolysaccharide (LPS) derived from *Escherichia coli* endotoxin and found no significant differences present between any of the studied groups in serum CRP.

The present study also showed significant increase in serum CRP in P/SRP treatment group in comparison with the NC/W, but this increase was not significant in relation with the and P/W treatment group. The present result agrees with that of Ide et al.'s⁶ study, they found no statistically significant changes in levels of any of CRP after SRP. Tonetti et al.²⁴ was found that within 24 h after periodontal treatment, there were significant increase in CRP indicating an acute systemic inflammatory response, and by day 7, the periodontal treatment group has equal or lower circulating inflammatory biomarkers than the control group. The increase within 24 h following periodontal therapy may be a result of intense transient bacteremia and soft tissue damage resulting from periodontal instrumentation. These two results disagree with that of Mattila et al.²⁵; they reported a reduction of CRP concentrations on 30 patients with chronic periodontitis after non-surgical periodontal treatment. Miyajima et al.²⁶ found that the serum CRP concentrations were not different between the control and the periodontitis rats (control; 10.9 6 0.6 ng/ml, periodontitis; 11.0 6 0.3 ng/ml).

The present study also showed that after 1 week the P/SRP + EPA treatment group showed significant decrease ($p < 0.05$) in serum CRP in comparison with the P/W, P/SRP, and P/EPA treatment groups. This means that the treatment by EPA with SRP is better than each one alone. Phillips et al.²⁷ also found that omega-3 supplementation reduces inflammation as measured by decreased levels of CRP as a marker of inflammation. Naqvi et al.²⁸ found that EPA was associated with lower CRP in linear secondary analyses. This may be due to its anti-inflammatory effects.²⁹ Serhan and Savill³⁰ have been found that omega-3 in animal models of periodontitis can be a substrates for neutrophil production of resolvins and protectins, which appear central to the resolution of inflammation. Other animal studies have suggested omega-3 may have a protective effect on periodontitis by decreasing the host inflammatory responses to common microbial pathogens such as *Porphyromonas gingivalis* and this result in less tissue breakdown.³¹ Vardar-Şengül³² study disagree with the present study, they found that omega-3 cannot be used for treatment of experimental periodontitis induced by repeated injections of *E. coli* LPS in rats, and omega-3 fatty acid administration did not seem to influence the circulating levels of CRP. This may be due to different methods used.

Serum ALP

Alkaline phosphatase is a metalloenzyme anchored to the cell membrane, and it is distributed particularly in the liver, bowel, placenta and bone and its activity and function can be modulated by environmental conditions.³³ The present study showed that the ligation-induced periodontitis caused a significant decrease in ALP serum levels. Kose et al.³⁴ found that the normal serum ALP in rats was 124.86 ± 12.40 U/L, while in periodontitis group was 98.86 ± 9.40 U/L after 28 days from ligation removal. Cetinkaya et al.³⁵ found that the serum alkaline phosphatase levels were 903.90 ± 6.76 U/L in normal control group and 811.0 ± 6.56 U/L in experimental periodontitis group after 2 months ligation around the mandibular first molar. Arabacı et al.³⁶ also found the same results. The significant reduction in ALP levels indicates that osteoblastic activity

decreased in the periodontal area. Contrary to these results, the measurements of periodontal destruction (probing depth, gingival bleeding, and suppuration) are related to higher levels of ALP in saliva.³⁷

The present study also showed that the P/SRP showed no significant difference from that of the P/W treatment group regarding the ALP serum levels. Singh et al.³⁸ also found that the changes in the level of ALP in serum after SRP was not statistically significant in the study groups with gingivitis, aggressive periodontitis and chronic periodontitis patients. This may be probably due to the fact that the local changes in periodontium may not have a direct effect on the levels of this enzyme in serum. The changing concentration in serum depends on function of other organ systems such as bone, kidney, liver etc.³⁹

The P/EPA and P/SRP + EPA treatment groups showed a significant increase ($p < 0.05$) in serum ALP after 3 days in comparison with the P/W treatment group. The statistically significant elevation in ALP levels provided by EPA treatment in rats with periodontitis suggests that this therapeutic agent have a significant effect on osteoblasts in periodontitis tissues. Appleton et al.⁴⁰ found that omega-3 can inhibit the production of inflammatory cytokines such as IL-1, IL-6, and TNF- α , which provide an important stimulus for osteoclastic bone resorption, and thus may inhibit bone resorption and prevent bone loss. Other study found that supplementation of the diets of growing rats with omega-3 fatty acids results in greater bone formation in rats, and also they found an increase in ALP activity in osteoblastic cells in culture.⁴¹ Al-Hashemi et al.⁴² found that the total alkaline phosphatase in rat's serum was significantly elevated after treatment with 400 and 600 mg/kg of omega-3 compared with other studied group. Attia et al.⁴³ found that rats fed with 20 mg/kg b.w. showed significant increase in serum ALP. Abdou et al.⁴⁴ also found the same increase in rat's serum ALP.

Serum MDA

Periodontal disease is one of the most common chronic inflammatory diseases, and high oxidative stress might have important roles in the etiopathogenesis of periodontitis.⁴⁵ The present results showed significantly higher levels of serum MDA in rats in the P/W treatment group compared with the NC/W treatment group. Celec et al.,⁴⁶ Panjamurthy et al.,⁴⁷ Aziz et al.,²² Trivedi et al.⁴⁸ and Ahmadi-Motamaye et al.⁴⁹ also found that serum MDA level was significantly higher in the periodontitis group. Based on the results of these studies, periodontitis can induce systemic oxidative stresses and alter serum MDA levels, and synthesis of MDA might be due to a decrease in antioxidant in destroyed in periodontal tissues.

The present study also showed that treatment by SRP can significantly decrease serum MDA in comparison with the P/W treatment group. Several studies found that successful periodontal therapy increased total antioxidant capacity levels⁵⁰ and decreased MDA levels,⁵⁰ therefore, periodontal therapy can be very useful for the patient.

Conclusion

The treatment of periodontitis by SRP with the EPA is better than each one alone.

Conflict of Interest

None. ■

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