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PREVENTIVE EFFICIENCY OF CURCUMIN ON ALVEOLAR BONE OF HYPOTHYROIDISM RAT MODELS WITH INDUCED PERIODONTITIS (HISTOLOGICAL, ULTRASTRUCTURE AND MICRO-ELEMENTAL STUDY)

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ABSTRACT

INTRODUCTION: Experimental animals used as periodontitis models have been used to investigate the sequence of the disease with the related alveolar bone resorption and periodontal tissue destruction. Hypothyroidism oral manifestations show variations including periodontal tissues inflammation associated with resorption of alveolar bone. Curcumin is a yellow polyphenol that is used frequently in a variety of medical applications due to its various protective effects.
AIM: Investigating Curcumin efficiency on the histological, ultrastructure and micro-elemental composition of alveolar bone with induced periodontitis in hypothyroidism rat models.
MATERIALS AND METHODS: 30 adult male rats were divided into 3 groups. Group I: Control, Group II: Periodontitis + hypothyroidism, Group III: Periodontitis + hypothyroidism + Curcumin. In groups (II) and (III), hypothyroidism was induced using daily oral dosage (10 mg / kg body weight propylthiouracil), together with periodontitis induction. Rats in group (III), Curcumin was treated with oral gavage with a dosage (75 mg/kg body weight) along with propylthiouracil. After 30 days, mandibles were dissected and prepared for histological examination, ultrastructure and elemental microanalysis.
RESULTS: Histological evaluation revealed increased osteoclasts and bone resorption in group II compared to the control group. However, significant improvement of the alveolar bone histology could be seen in the group III. Ultrastructural and micro-elemental results confirmed the histological findings indication smoother bone surface and increased calcium and phosphorus levels in the treated group III than group II.
CONCLUSION: Curcumin is considered an effective prophylactic agent, with great antioxidant and anti-inflammatory properties, that preserve alveolar bone in periodontitis in hypothyroid cases.
KEYWORDS: Hypothyroidism, Alveolar bone, Curcumin.
RUNNING TITLE: Efficiency of curcumin on hypothyroidism rats with induced periodontitis.

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INTRODUCTION

Periodontitis prevalence is high worldwide where it affects 50% of adults and is the sixth most widespread disease. Together with dental caries, periodontitis shows high prevalence among dental diseases. It is characterized by periodontal tissue destruction due to the adherence of dental plaque or dental biofilm to the dento-gingival junction [(1)].

As an inflammatory disease, periodontitis is prompted by periodontopathogenic bacteria in the oral cavity. Persistent inflammation causes destructive effects on all tissues of periodontium involving gingiva, periodontal ligament, cementum and alveolar bone. Consequently, alveolar bone destruction leads to damage of tooth support system, eventually leading to teeth loss [(2)].

As all bones in the body, alveolar bone undergoes repeated resorption by osteoclasts and deposition by osteogenic cells in a continuous remodelling process. However, when an imbalance occurs, bone resorption proceeds, while the compensatory effect of new bone formation cannot cope with the resorption, resulting in bone loss. The inflammatory nature of periodontal diseases causes cytokines production which in turn promote the function of osteoclasts [(3,4)].

Alveolar bone proper represents the inner cortical wall of the alveolar bone that surrounds roots of teeth and contains Sharpey's fibers reflecting its importance in the attachment of teeth in their sockets. On the other hand, the supporting alveolar bone supports the alveolar bone proper which is structurally formed of cortical and trabecular bone [(5)].

Mimicking human periodontitis, experimental animal models with ligature-induced periodontitis are used as a successful attempt to stimulate inflammation and subsequent bone loss describing the disease pathogenesis [6]. Pathogenic bacteria take the leading role in the sequence of periodontitis, causing the attraction of immune cells along with inducing the release of signaling molecules as cytokines and growth factors resulting in periodontitis progression and accelerating its severity [7].

Diversity of systemic conditions has strong connections with periodontal diseases. Some of which includes cardiovascular and cerebrovascular diseases, diabetes mellitus, osteoporosis, obesity and stress. Moreover, it has been confirmed that the aggressiveness and incidence of periodontitis is much higher in autoimmune-diseased individuals such as those suffering from rheumatoid arthritis. Common immunoinflammatory pathways in diseases pathogenesis, might explain the correlation between those medical conditions and periodontal disease. Some inflammatory mediators have been hypothesized to be locally released in cases of periodontitis resulting in distortion in bone hemostasis and at the same time play an important risk role in other systemic diseases. Some of these mediators include prostaglandin E2, interleukin-1β, tumor necrosis factor-α, and matrix metalloproteinases have been. [8,9].

Thyroid gland secretes two main hormones; Thyroxine (T4) and Triiodothyronine (T3) which are essential for mostly all metabolically active cells. As a result, a lack of or reduced activity of these hormones can cause hypothyroidism [10]. Hypothyroidism has an extensive array of manifestations, ranging from symptomless individuals to patients with multisystem failure owing to decreased production of T3, T4, and calcitonin. Essentially, deficiency in these hormones leads to decreased bone metabolism, development, and remodeling, besides, destructively affecting bone homeostasis [11].

Affecting the healing capacity of tissues is one of the main negative manifestations of thyroid disease, which can be attributed to the imbalance in body homeostasis and metabolic slowdown. Bone healing capacity is thus, consequently affected in cases of hypothyroidism. Inhibition of the activity of bone cells including differentiation and maturation, could be clearly observed in such cases leading to reduction of bone resorption and formation [12].

However, according to a recent study done in 2023, the potential link between periodontitis and hypothyroidism has only been investigated in a limited number of high-quality studies. The metabolic activity of fibroblasts decreases due to hypothyroidism, delaying the healing of wounds. Due to the long-term exposure of the injured tissue to pathogenic organisms, a delay in the healing process may be associated with a greater risk for infection [10].

Curcumin (CMN) is a natural compound that can be obtained from the rhizomes of *Curcuma longa L.*, that belongs to the Zingiberaceae family. It is categorized as one of the phenolic acids with a variety of biological activities attaining great curing potential for disease treatment. Among these important biological properties are anti-inflammatory, anti-oxidant and anti-tumor. Its anti-inflammatory effect is exerted by controlling the inflammatory signaling pathways along with decreasing the production of inflammatory mediators [13,14]. Moreover, CMN has been reported as a free radical scavenger where it has the ability to get rid of reactive oxygen species (ROS) such as hydroxyl radical and amine superoxide. CMN has been speculated to have exceptionally good tolerance and thus to be safe for humans and animals. [15].

It has been further shown that CMN has beneficial influence on pain, metabolic syndromes, degenerative eye conditions and kidney diseases. The previously mentioned biological properties along with anti-proliferative, apoptosis and angiogenesis, renders CMN an effective agent in thyroid gland disorders [16].

Curcumin can be afforded in different ways as in the form capsules, tablets, ointments, energy drinks, soaps, and cosmetics, where it has been approved by the US Food and Drug Administration (FDA) as "Generally Recognized As Safe" [17]. Recently, it was mentioned that curcumin has the ability to relieve some symptoms in some autoimmune diseases such as rheumatoid arthritis and inflammatory bowel disease. Moreover, its effect in obesity therapy was studied due to its lipophilic molecule that facilitate its cellular penetration, besides its association with lipid metabolism [18].

The effect of CMN on various oral diseases associated with chemotherapy. has been previously studied, where it has been proven to have a positive influence on leukoplakia, oral submucosal fibrosis, and other oral mucosal lesions. Moreover, CMN has antifungal effect which can prevent candidiasis, representing a common side effect of corticosteroids [19]. The antibacterial effect of CMN has been attributed to its action against periodontopathic bacteria as P gingivalis and Provotella intermedia which thus can be considered an important agent for preventing periodontal diseases. Moreover, its anti-inflammatory potential acts via downregulating some inflammatory cytokines synthesis as tumor necrosis factor-alpha (TNF-a) interleukin (IL-1, -2, -6, -8, and -12) and matrix metalloproteinase (MMP), resulting in preventing disease progression. Besides, it regulates the activation of certain transcription factors like activating protein-1 (AP-1) and NF-κB and by

inhibiting intercellular signaling proteins such as protein kinase C, thus blocking cytokine gene expression [20, 21].

Little studies have focused on CMN effect on the alveolar bone of periodontitis in hypothyroidism. So, the current study aimed to investigate the effect of CMN on periodontitis related alveolar bone histology and ultrastructure in hypothyroidism induced animal models.

MATERIALS AND METHODS

Animal housing

Thirty adult male albino rats aging 4-6 months with 200-250 g body weight were involved in the current study. Throughout the experimental period, rats were kept in clean and well aerated plastic cages with constant regular climate. Rats were supplied filtered tap water and laboratory food in accordance with the guidelines of scientific research ethics recommendation of the Research Ethics Committee at Faculty of Dentistry, October 6 University, Giza, Egypt.

Sample size calculation

Calculation of sample size was done by using Power Analysis and Sample Size Software (PASS 2020) "NCSS, LLC. Kaysville, Utah, USA, ncss.com/software/pass". A minimal total hypothesized sample size of 30 adult male albino rats aging from 4-6 months and weighing from 200-250 g body weight, 10 animals per group, was needed to study the preventive effect of CMN on alveolar bone with induced periodontitis and Propylthiouracil induced hypothyroidism rats with an assumption of obtaining an effect size of 20%, with 0.05 probability of type I error and power of 80% using Chi-square test [22].

Chemicals

1. Propylthiouracil (PTU)

PTU was supplied in the powder form which was dissolved in saline. It was purchased from Sigma Aldrich company.

2. Curcumin

CMN was purchased from Sigma Aldrich company.

Experimental design

Rats were randomly divided into 3 equal groups, 10 rats each, as follows:

Group I [Controls]: Animals received daily distilled water by oral gavage (10 ml/kg) for thirty days.

Group II [Periodontitis + hypothyroidism]: Animals received 10 mg / kg body weight PTU dissolved in 0.3 ml saline by oral gavage daily by for thirty days for hypothyroidism induction [23]. Meanwhile, induced periodontitis was established by ligation insertion around the right mandibular first molars till the end of the experiment [24].

Group III [Periodontitis + hypothyroidism+ CMN]: Induction of hypothyroidism was accomplished as mentioned in group II, together with 75 mg/kg body weight CMN daily by oral gavage for thirty days, together with periodontitis induction as mentioned in group II with the same duration [24].

Serological tests

At the end of the experiment, blood samples were assembled from the para-orbital sinus puncture to determine T3 and T4 levels in the sera of rats to ensure hypothyroidism induction.

Periodontitis induction Ligation

Rats in groups II and III were anesthetized with sodium pentobarbital by intraperitoneal injection (6mg/100g). 6-0 silk ligatures were fastened around the right mandibular first molars where they were positioned apical to the interproximal region and pressed into the gingival sulcus and left for 30 days. Ligature were checked every 2 days to ensure their intact placement [25].

Euthanization

Euthanization occurred after the experimental period for all rats of the 3 groups. Mandibles were dissected and cut into halves. The right halves were prepared for histological examination by light microscope, ultrastructural examinations and elemental microanalysis by electron microscope and energy dispersive x-ray respectively.

Histological evaluation

For histological examination, sections from the right halves of the mandibles were fixed in 10% neutral–buffered formalin. After fixation, the mandibles were washed and decalcified with 5% trichloroacetic acid then dehydrated with ascending concentrations (50%, 70%, 90%, and 95%) of ethanol, cleared with xylene, and embedded in paraffin wax blocks. The blocks were cut with an average thickness of 5 µm then stained with Haematoxylin and Eosin stain. Histological features of alveolar bone in different groups were assessed [23].

Scanning electron microscope examination (SEM) and Energy dispersive X-ray microanalysis (EDX)

Right halves of the mandibles were prepared for ultrastructure evaluation and elemental microanalysis. They were preserved in 11% gluteraldehyde and prepared for SEM to observe the surface topography of alveolar bone the different groups. The 3 group specimens were subjected to EDX analysis to assess and measure Ca and P percentages [26].

Statistical analysis

Statistical analysis was done to evaluate the measurements of T3 and T4 in the different groups to detect hypothyroidism induction. It has also been done for elemental microanalysis results detecting calcium and phosphorus levels in the 3 different groups.

Data were fed to the computer and analyzed using IBM SPSS software package version 20.0. (Armonk, NY: IBM Corp). continuous data, they were tested for normality by the Shapiro-Wilk test. Quantitative data were expressed as range (minimum and maximum),

mean, standard deviation and median One way ANOVA test was used for comparing the three studied groups and followed by Post Hoc test (Tukey) for pairwise comparison. Significance of the obtained results was judged at the 5% level [27].

RESULTS

1. Serological results

Serological results showed a significant decrease in T3 and T4 levels in hypothyroidism group with induced periodontitis when compared with the control and hypothyroidism induced group with CMN. However, no significant difference was detected between the control group and the hypothyroidism induced group with CMN (Table 1)

2. Histological results

Group I (Control)

Light microscopic examination of the alveolar bone of the control group revealed smooth and regular bone surface with continuous lining of osteoblast cells. normal osteocytes in their lacunae appeared embedded within the thick trabeculae of the alveolar bone. Regular parallel resting lines were clearly seen demarcating regular bone formation. Normal orientation and thickness of periodontal ligament fibers could be detected with regular insertion of the Sharpey's fibers. (Fig 1, a &b)

Group II [Periodontitis + hypothyroidism]:

Regarding the hypothyroidism in periodontitis related alveolar bone, resorption of the alveolar bone surface was clearly seen as irregular bone surface with discontinuation in the lining of the osteoblasts. Multiple osteoclasts in their Howship's lacunae were seen demarcating bone resorption. Absence of regular resting line within the bone trabeculae. Irregular orientation and widening in the thickness of the periodontal ligament fibers were clearly detected. (Fig 1, c & d)

Group III [Periodontitis + hypothyroidism + CMN]:

In comparison to group II, relative regaining of the regular alveolar bone surface could be obviously noticed. Continuous regular osteoblasts were seen lining the alveolar bone surface. Less number of osteoclasts could be still noted on the surface of alveolar bone. Regular regaining of the normal arrangement and architecture of periodontal ligament fibers could be detected. (Fig 1, e & f)

3. Ultrastructural results

Scanning electron microscopic results revealed variations in the topography of the buccal cortical plate of bone among the different groups. However, all specimens of the same group showed similar results.

Group I (Control)

Scanning electron microscopic picture revealed intact normal level of bone around mandibular first molar with generalized smooth bone surface topography showing multiple nutritive canals with regular intact borders. (Fig 2: A,D)

Group II [Periodontitis + hypothyroidism]:

Dramatic resorption in the level of the alveolar bone around mandibular first molar with irregular resorbed bone surface topography could be clearly detected with multiples craters with Il-defined nutritive canals interpreting active bone resorption. (Fig 2: B,E).

Group III [Periodontitis + hypothyroidism + CMN]:

Partial preservation in the level of alveolar bone around mandibular first molar with relative preservation of the smooth surface topography. Significant increase the various sized nutritive canals were detected (Fig 2: C,F)

4. Energy dispersive microanalysis results

Levels of calcium and phosphorus were detected in the 3 different groups. Statistical significant decrease in calcium and phosphorus levels was observed in group II when compared to the control and the treated group III. However, these levels revealed no significant difference between the control group I and the treated group III. (Table 2)

Table (1): Comparison between the three studied groups according to T3 and T4

	Control (n = 10)	**Periodontitis + hypothyroidism (n = 10)**	**Periodontitis + hypothyroidism + CMN (n = 10)**	**p**
T3				
Mean ± SD.	78.5 ± 2.6	57.3 ± 5.7	74.9 ± 4.6	
Median (Min. – Max.)	78.5 (74 – 83)	58.5 (46 – 65)	75.5 (68 – 81)	<0.001*
Sig. bet. grps.	p_1<0.001*, p_2=0.188, p_3<0.001*			
T4				
Mean ± SD.	6 ± 0.51	3.1 ± 0.40	6.4 ± 0.45	
Median (Min. – Max.)	5.9 (5.2 – 6.8)	3.1 (2.5 – 3.7)	6.4 (5.8 – 7.1)	<0.001*
Sig. bet. grps.	p_1<0.001*, p_2=0.143, p_3<0.001*			

SD: **Standard deviation**

F: **F for One way ANOVA test**, Pairwise comparison bet. each 2 groups was done using **Post Hoc Test (Tukey)**

p: p value for comparing between the studied groups

p_1: p value for comparing between **Control** and **Periodontitis + hypothyroidism**

p_2: p value for comparing between **Control** and **Periodontitis + hypothyroidism + Curcumin**

p_3: p value for comparing between **Periodontitis + hypothyroidism** and **Periodontitis + hypothyroidism + Curcumin**

*: Statistically significant at $p \leq 0.05$

Table (2): Comparison between the three studied groups according to Calcium and Phosphorus Mass%

Mass%	Control (n = 10)	Periodontitis + hypothyroidism (n = 10)	Periodontitis + hypothyroidism + CMN (n = 10)	p
Phosphorus (p)				
Mean ± SD.	14.1 ± 0.73	11 ± 0.65	13.4 ± 1	
Median (Min. – Max.)	13.9 (13.1 – 15.5)	11.2 (10.2 – 12.4)	13.3 (12.2 – 15.5)	<0.001*
Sig. bet. grps.	p_1<0.001*,p_2=0.176,p_3<0.001*			
Calcium (Ca)				
Mean ± SD.	28.7 ± 1.3	17.9 ± 1.3	24.3 ± 0.96	
Median (Min. – Max.)	28.6(27 – 31.2)	17.5 (16.7 – 20.3)	24.4 (22.8 – 25.5)	<0.001*
Sig. bet. grps.	p_1<0.001*,p_2<0.001*,p_3<0.001*			

SD: **Standard deviation**

F: **F for One way ANOVA test**, Pairwise comparison bet. each 2 groups was done using **Post Hoc Test (Tukey)**

p: p value for comparing between the studied groups

p_1: p value for comparing between **Control** and **Periodontitis + hypothyroidism**

p_2: p value for comparing between **Control** and **Periodontitis + hypothyroidism + Curcumin**

p_3: p value for comparing between **Periodontitis + PTU** and **Periodontitis + hypothyroidism + Curcumin**

*: Statistically significant at $p \leq 0.05$

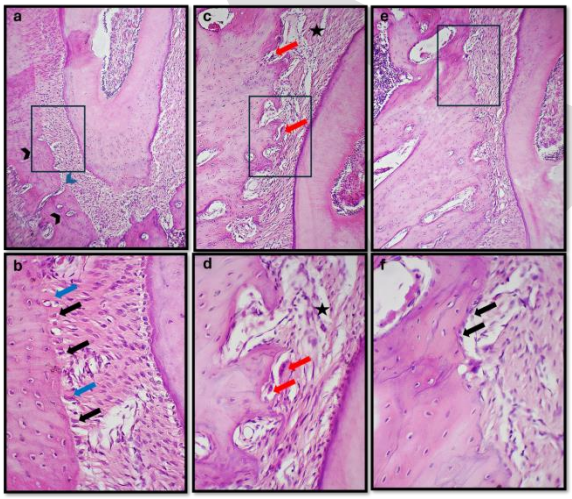

Figure (1): Light micrograph (LM) showing (a) (Control group) regular alveolar bone surface along the root length with normal parallel resting lines (black arrowhead) and nutritive canal (blue arrowhead). Note normal thickness and direction of periodontal ligament (H&E X100). (b) (inset) showing continuous osteoblasts lining (black arrows) along the smooth bone surface. Normal osteocytes within their lacunae. Note normal insertion of PDL fibres (Sharpey's fibres) (blue arrows) (H&E X 400). (c) (Periodontitis + hypothyroidism group) showing irregular alveolar bone surface with multiple resorption lacunae some of which contain osteoclasts (red arrows). Note widening and disorganization of PDL (black star). (H&E X100). (d) (inset) showing resorbed alveolar bone surface with osteoclasts and discontinuity of osteoblasts lining. Note extensive disorganization in the PDL fibres (black star). (H&E X 400). (e) (Periodontitis + hypothyroidism+ CMN group) showing relatively regular alveolar bone surface with relative regaining of the regular thickness and orientation of the. (H&E X100). (f) (inset) showing osteoblasts lining (black arrow) and an osteoclast demarcating active bone remodelling (H&E X 400)

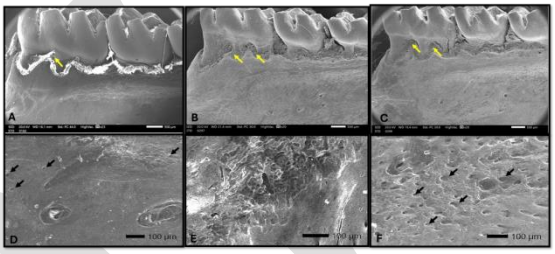

Figure (2): Scanning electron micrograph (SEM) of buccal cortical plate (A, D) (Group I (Control)) showing intact alveolar bone with normal level around mandibular first molar (yellow arrow) (X 20). The cortical plate illustrated a smooth, regular bone surface with multiple nutritive canals (black arrows) (X180). (B,E) (Group II [Periodontitis + hypothyroidism]) showing severe resorption in the level of the alveolar bone around mandibular first molar (yellow arrows) (X 20).The cortical plate revealed rough irregular resorbed bone surface with multiple areas of shallow depressions and craters. No distinct nutritive canals can be seen (X180). (C,F) (Group III [Periodontitis + hypothyroidism + CMN]) showing partial preservation in the level of alveolar bone around mandibular first molar (yellow arrows) (X 20). The surface topography of the buccal cortical plate exhibited a relatively regular bone surface with increased number of well-defined nutritive canals with different sizes (black arrows) (X180).

DISCUSSION

The current work was accomplished to assess the efficacy of CMN on alveolar bone with induced periodontitis in experimentally PTU induced hypothyroidism rats. Hypothyroidism has been induced in the present study by using 6-Propyl-2-thiouracil (PTU) which is considered as an antithyroid drug used in the treatment of hyperthyroidism. PTU has been usually used as an effective mean of inducing hypothyroidism in experimental animals. According to a previous study done on rats to examine the impact of hypothyroidism on pancreatic cells, PTU was used as an induction method which revealed the inhibition in the production of thyroid hormones.

This latter study further demonstrated PTU action in the downregulation of the transformation of T4 to T3 and its effects on the thyroid hormones both, stored in the thyroid gland or circulating in the blood [28]. These results are in accordance with the serological results of the present study which showed significant decrease in T3 and T4 levels in the PTU induced group. Moreover, our results were in agreement with another study that revealed that PTU-induced hypothyroidism cause significant decrease in the thyroid hormones accompanied by morphologic alterations of the gland [13].

However, the usage of CMN as a preventive measure in the third group of periodontitis animal models with PTU induced hypothyroidism, caused significant increase in T3 and T4 levels compared to those of the second group without treatment. These results could be explained by the discussion of a previous study that stated that curcumin caused remarkable improvement in thyroid histology, where the follicular cells appeared healthy. These findings were interpreted in the elevated level of thyroid hormones. Curcumin acts by modulating multiple inflammatory mediators production, and the synthesis of some inflammatory cytokines and inflammatory enzymes such as interleukin-6 and cyclooxygenase-2 respectively, thus demonstrating an effective anti-inflammatory capability. The later study confirmed the antiproliferative action of curcumin [13,29].

Mandibular molars ligation was done in the current study to induce periodontitis in rats. The ligature-induced model is considered the most common method for inducing periodontitis. As mentioned in a recent study, rat models of induced periodontitis represents the best option regarding time, cost, and number of sacrificed animals for short-term studies [30]. Ligature application in the first molar of mice induced several features of periodontitis macro and microscopically, including alveolar bone resorption, gingival recession and inflammatory infiltrate in the studied region, rendering it an applicable process to study this disease [31]. According to a previous study, 30 days duration for periodontitis induction were efficient to induce all signs of periodontitis including clinical signs of gingival inflammation, including color/volume changes and bleeding around the ligated teeth. Besides, morphometric analysis revealed alveolar bone loss around ligated teeth [32].

Histological results of the present study revealed the degenerative effect of hypothyroidism induction in a periodontitis induced animal model, where alveolar bone revealed resorbed surface with multiple osteoclasts and discontinuation of osteoblast lining. Persistant oral manifestations of hypothyroidism showed variations including hypoplastic enamel, postponed wound healing, periodontal tissues inflammation and alveolar bone loss [33].

Previous studies have monitored the relation between periodontitis and hypothyroidism clinically and experimentally using ligature-induced periodontitis model. Periodontitis was evaluated to be more aggressive in rats with thyroid disease. While, clinically, higher pocket depth was found accompanied with clinical attachment loss in hypothyroid patients compared with healthy patients [33,34]. The mechanism of bone loss due to hypothyroidism has been discussed in a previous study revealing increase in the serum and salivary levels of TNF- α and IL-6. Moreover, higher number of osteoclasts was observed demarcating an increase in alveolar bone resorption, thus, influencing both the histological features and density of alveolar bone [35].

However, relative regaining of the normal histological features of alveolar bone has been clearly demonstrated in the histological results of group III. Lesser number of osteoclasts could be detected with relatively smooth bone surface. This was accompanied with relative regaining of the normal thickness and organization of periodontal ligament fibres. A previous study has confirmed the treating effect of CMN on an already established hypothyroid animal model induced also by PTU, where it showed its potential to improve all the histological, immunohistochemical and biochemical markers in pancreatic that were altered by hypothyroidism. This latter study further explained CMN anti-inflammatory potential with its ability to interact with many molecular inflammatory components, which caused inhibition in their activity. Also, some signaling molecules as inflammatory cytokines, TNF α and interleukins were downregulated [36].

Based on the enhanced understanding of function and beneficial effects of CMN on musculoskeletal system as well as osteoporosis, has been studied previously. It has been shown that CMN has the ability to regulate bone remodelling by inhibiting osteoclast bone resorption besides inhibiting osteoclastogenesis by receptor activator of nuclear factor kappa B ligand [37]. Furthermore, other study verified the protective function of CMN in bone health where it elaborated its role in the stimulation of osteoblasts differentiation and inhibiting osteoclasts formation, thus maintain bone health. The proliferative effect of CMN on osteoblast has been explained by its upregulating effect on genes expression related to bone formation, including, alkaline phosphatase (ALP), osteocalcin and RunX2. On the other hand, CMN ability to block osteoclast differentiation from its precursor was attributed to its inhibitory action on the production of chemokine CCL3 and decreasing osteoclast-related gene expression [38]. Another also showed that CMN inhibited osteoclasts differentiation markers as cathepsin K, matrix metalloproteinase (MMP-9 and MMP-13), besides inhibiting iin-α [39].

Ultrastructural findings of the present study further emphasized the histological results where there was a dramatic resorption in the level of the alveolar bone around the mandibular first molar. Moreover, buccal cortical plates of the second group showed resorbed bone surface with multiple craters, however, great preservation in the level of the alveolar bone with smooth surface with increased number of nutrient canals were clearly seen in the third group. These findings are in harmony with another study that was done to reveal the efficacy of CMN nanoparticles on the alveolar bone of rats with induced periodontitis. It showed a smooth surface bone topography with reduced bone loss [(40)]. The EDX microanalysis showed significant decrease in the Ca levels in hypothyroid group with induced periodontitis, however these measures increased relatively returning back to normal by the action of CMN. These results were supported previously by a study that revealed the importance of thyroid hormones in controlling bone mineral homeostasis and density, where, hypothyroidism could be accompanied with reduced bone mineral density (BMD) leading to increased fracture risk [(41)] .Whereas the beneficial effect of CMN regarding bone mineral density was also explained in a previous study that showed that there is a significant increase in BMD accompanied by increasing bone strength, revealing its anti-osteoporotic effect [(37)]. Previously, CMN has been proven to have a protection potential against bone loss associated with ovariectomy animal models elaborated in decreased osteoclastogenesis. Moreover, CMN enhanced microarchitecture of bone and increased mineral density [(42)].

CONCLUSION

Curcumin is a natural anti-inflammatory compound that show multiple biological features allowing it to have a promising role in the prevention of alveolar bone resorption in induced periodontitis in cases of hypothyroidism.

RECOMMENDATIONS:

Curcumin should be widely used to counteract alveolar bone resorption in cases of periodontitis regarded as safe, affordable agent. Further studies should also be conducted to show its effect with different doses and durations and its therapeutic effect rather than preventive.

More studies are required to evaluate its effect on the alveolar bone in other degenerative systemic diseases as diabetes mellitus and autoimmune diseases as rheumatoid arthritis.

DECLARATION SECTION:

Conflict of interest

The authors declare that they have no conflicts of interest neither financial, nor interpersonal.

Funding

No specific funding was received for this work.

Ethical approval

The study was approved by Research Ethics Committee at Faculty of Dentistry, October 6 University, Giza, Egypt, with approval number: RECO6U/37-2023 obtained in its meeting held on December 4, 2023.

ARRIVE guidelines:

The ARRIVE guidelines for the documentation of in-vivo studies in animal research were followed in the conduction of this work.

Data availability:

All data generated and analysed during the current study are available from the corresponding

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