Naunyn-Schmiedeberg's Archives of Pharmacology
https://doi.org/10.1007/s00210-022-02293-5

RESEARCH

Evaluation of the oxytocin effect in a rat model with experimental periodontitis

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Received: 24 May 2022 / Accepted: 8 September 2022

Abstract

The aim of the present study was to evaluate the inhibitory effects of oxytocin on the development of periodontitis based on its properties against bone loss and resorption. Thirty-two Wistar albino rats were divided into four equal groups: control, periodontitis + saline, periodontitis + 0.5 mg/kg/day oxytocin, and periodontitis + 1 mg/kg/day oxytocin. Periodontitis groups received 4.0 silk ligatures around their cervixes of the right and left mandibular incisors in an "8" shape, kept for 14 days. Animals in oxytocin groups were injected once every day during 14 days with oxytocin. The mandibles were fixed and scanned using microcomputed tomography to quantify bone resorption and volumetric measurements. Blood samples were collected to analyze the concentrations of macrophage colony-stimulating factor (M-CSF), receptor activator of nuclear factor-κB ligand (RANKL), osteoprotegerin (OPG), matrix metalloproteinase-8 (MMP-8), tumor necrosis factor-alpha (TNF-α), interleukin (IL)-6, glutathione peroxidase (GPx), superoxide dismutase (SOD), and malondialdehyde (MDA). Histopathological evaluations were conducted to examine the gingiva and alveolar bone. Oxytocin prevented the development of periodontitis by decreasing ligament deteriorations and leukocytes in the gingival connective tissue and promoting reintegration with the alveolar bone. Bone resorption in all regions was less in the periodontitis + 1 mg/kg/day oxytocin group than in the periodontitis + saline group. Although TNF-α, IL-6, and RANKL values were lower in the periodontitis + 1 mg/kg/day oxytocin group, OPG was higher than that in the periodontitis + saline group. M-CSF, MMP-8, and MDA were lower in the oxytocin groups than in the periodontitis + saline group. Oxytocin may be an effective agent for periodontal diseases because it decreased bone resorption, oxidative stress, and inflammation in an experimental periodontitis.

Keywords Alveolar bone loss · Antioxidant · Oxytocin · Periodontal diseases · Rats

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Published online: 17 September 2022

Introduction

Oxytocin is an anabolic hormone produced in the hypothalamus and carried into the bloodstream where it plays a role in lactation in mammals (Amar and Weiss 2003). In addition to its physical and mental effects such as behavior regulation, social memory, stress management, learning, sexual behavior, initiation of labor, and promoting breastfeeding (Lee et al. 2019), it has recently been identify as having protective effects on bone metabolism (Elabd et al. 2007, 2008; Tamma et al. 2009). Systemic administration of oxytocin stimulates bone formation by regulating osteoblast differentiation and osteoclast functions (Colaianni et al. 2011; Tamma et al. 2009). The absence of oxytocin or the oxytocin receptor causes a decrease in bone formation, and its progression can result in osteoporosis (Colaianni et al. 2014). In addition to being an antioxidant, its immunomodulatory and anti-inflammatory effects have been identified (Düşünceli et al. 2008). These effects can also be altered by suppressing proinflammatory cytokines and increasing antioxidant enzyme levels. When evaluated within this context, oxytocin has been shown to have a protective effect on inflammation in models of sepsis, ischemia/reperfusion, pyelonephritis, and colitis (Allameh and Salamzadeh 2016; Dou et al. 2021; Düşünceli et al. 2008; Franco et al. 2021).

Periodontitis is a chronic inflammatory disease that is characterized by the progressive destruction of the periodontium around the teeth. Although microorganisms play a role in its primary etiology, the pattern of destruction from the disease is shaped by the host's response. In a healthy adult, there is a balance between bone formation by osteoblasts and bone resorption by osteoclasts (Hienz et al. 2015). Osteoclasts are the cells responsible for the destruction of mineralized bone and, as they increase and mineralized bone is destroyed, that balance is disturbed and alveolar bone is resorbed.

To our knowledge, there are no published studies on the effect of oxytocin on periodontal disease; therefore, the aim of the present study was to evaluate how oxytocin prevents the development of periodontitis by protecting bone deterioration and resorption.

Materials and methods

Study design

The present study comprised 32 Wistar albino rats weighing 220–240 g each. The rats were separated into groups as follows: (a) control, (b) periodontitis + saline (injected subcutaneously with saline, (c) periodontitis + 0.5 mg/kg/day oxytocin (subcutaneous injection), and (d) periodontitis + 1 mg/kg/day oxytocin (subcutaneous injection).

Animals in oxytocin groups were injected once every day during 14 days with oxytocin, and in periodontitis + saline groups, animals were injected subcutaneously with saline once a day for 14 days.

A power calculation was carried out (Mester et al. 2019), and the sample size was calculated to be at least three rats for identification of a substantial difference in terms of BV/TV% between groups at a power level of 95% and an alpha error of 5%, and the effect size is 6.1176 (G*Power 3.1 software; Heinrich Heine University, Dusseldorf, Germany). Among these patients, eight rats were collected for each group as a precaution in case of loss due to any reason and insufficient sample size.

The rats were housed two per cage in a quiet room under controlled temperatures (21–22 °C) and a reversed light–dark cycle (12 h/12 h) and had free access to standard food and water. Experiments carried out in accordance with the guidelines laid down by the National Institute of Health (NIH) in the USA regarding the care and use of animals for experimental procedures. The experimental protocol was examined and approved by the Animal Experiment Ethics Committee of Near East University (number: 2019/05–77).

Ligature placement around the cervical region of the mandibular incisors

A combination of ketamine (Ketalar; Pfizer, New York, NY, USA) (90 mg/kg i.p.) and xylazine (Rompun; Bayer, Istanbul, Turkey) (10 mg/kg i.p.) was used to anesthetize the animals after an overnight fast (Da Silva Freitas Ribeiro et al. 2018). In the periodontitis groups, we induced alveolar bone loss by placing 4.0 silk ligatures around the cervix of the right and left mandibular incisors in an "8" shape and buccally knotting (Mester et al. 2019). Any loose or missing ligatures found during a daily inspection were immediately replaced.

Euthanasia and collection of specimens

The rats were euthanized 2 weeks after the study was initiated. Animal euthanasia was performed by decapitation method. The mandibles were excised and 3 mm of gingival tissue from the area surrounding the lower incisors affected by induced periodontitis was obtained for micro-computed tomography (micro-CT) and histopathology analyses. Blood was taken for biochemical analysis.

Micro-CT analysis

Micro-CT scanning

After air-calibrating the detector to minimize ring artifacts, the specimens were scanned using a high-resolution micro-CT device (Bruker SkyScan 1172, Kontich, Belgium) (voxel size, 13.73 µm, 100 kV, 100 µA, rotation 360° within an integration time of 5 min, Al filter 0.5 mm, a 0.7° rotation step, 250 ms exposure, total time 1 h each). Beam hardening was corrected and optimal contrast limits were used according to the manufacturer's instructions using initial scanning and reconstruction tests (Huang et al. 2017).

Micro-CT imaging analysis

NRecon ver. 1.6.10.4 (SkyScan, Kontich, Belgium) and CTAn ver. 1.17.7.2 (SkyScan) were used to visualize and quantitatively measure the samples by obtaining axial two-dimensional (2D) 1000 × 1000-pixel images. During reconstruction, the ring artifact was corrected and smoothed by setting to 0 and the beam artifact was corrected by setting to 50%. CTAn ver.1.17.7.2 (Bruker micro-CT, Kontich, Belgium) and DataViewer ver. 1.5.6.2 (Bruker Micro-CT) were used to analyze the scans of the sections.

Linear measurements

Multiplanar reconstruction (MPR) images were obtained using DataViewer ver. 1.5.6.2 in the coronal and sagittal directions through the center of the region of interest (*x*–*z* axis and *z*–*y* axis). The software allowed the user to designate a region of interest (ROI) and specify the number of slices within the ROI; therefore, a standardized number of slices was acquired for all specimens in all directions. All measuring points were standardized and measurements were taken manually by a single examiner using the software to minimize errors. Alveolar bone loss was measured linearly from the cementoenamel junction to the alveolar bone crest at the buccal, mesial, and distal surface roots of the incisors, and the mean was calculated (Fig. 1a).

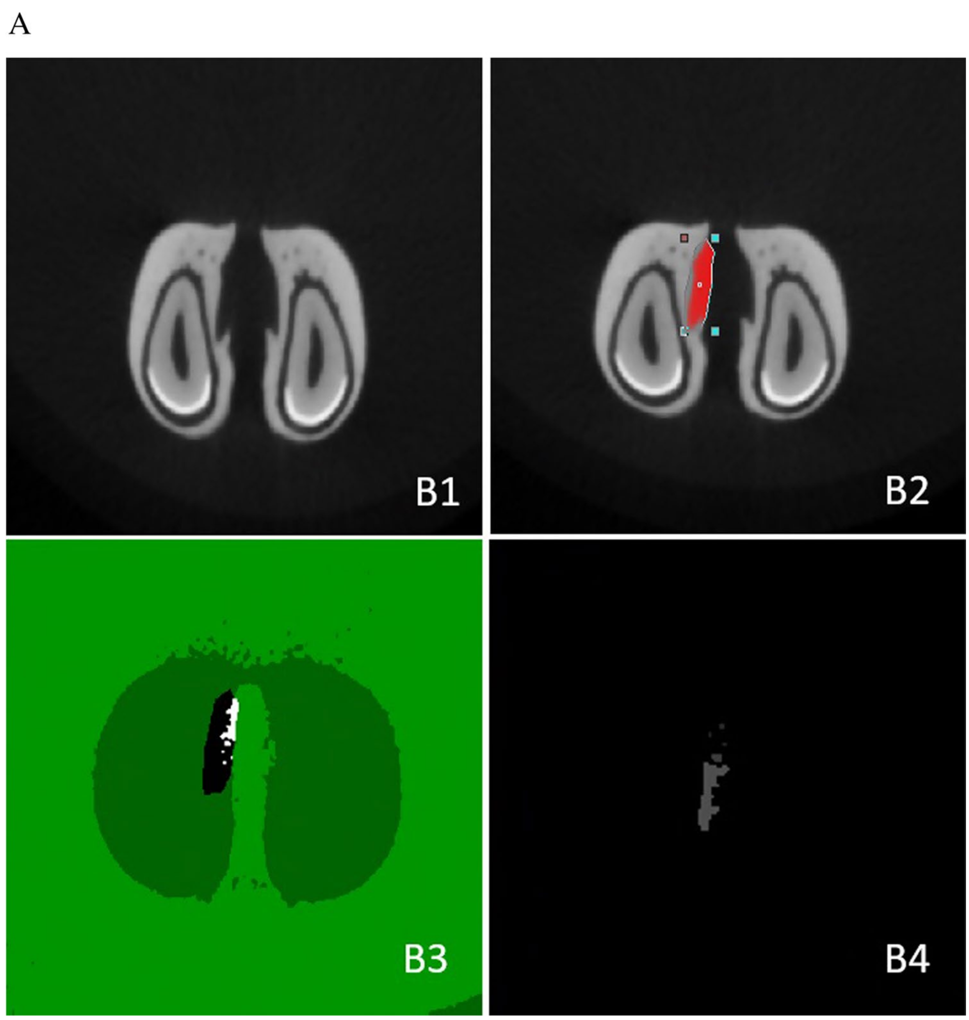

Fig. 1 Three-dimensional micro-CT images. **A** Linear measurements of the alveolar bone loss. The measurements were done linearly from the cementoenamel junction to the alveolar bone crest at the buccal, mesial, and distal surface roots of the incisors. **B** Volumetric measurements of alveolar bone loss. Region of interest was highlighted and draw in all slices for volumetric measurement of alveolar bone loss and used for the calculation of total alveolar bone loss. The images were binarized to distinguish the bone and dental structures according to differences in density using a grey-scale (limits 50–140; threshold 0–140) transformation

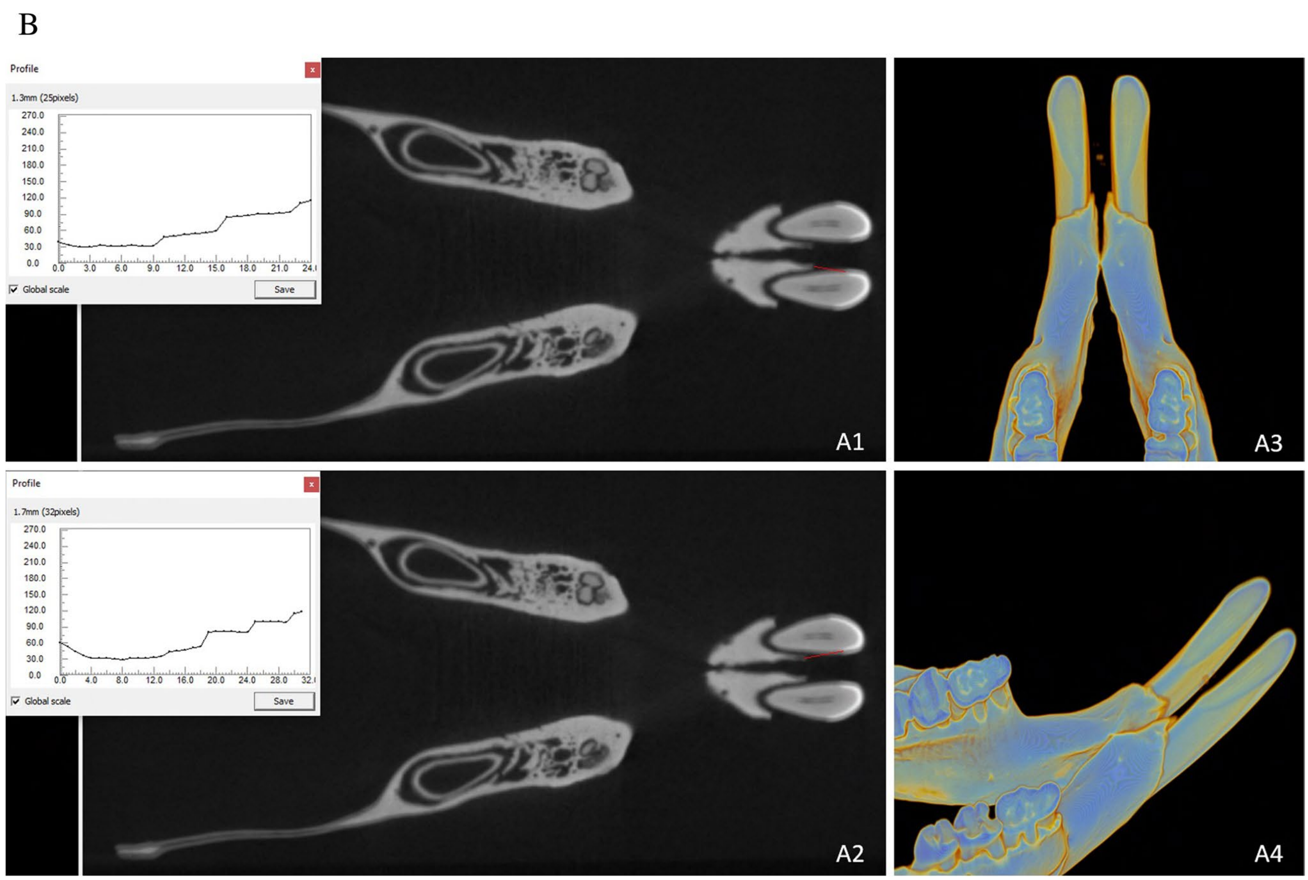

Fig. 1 (continued)

Volumetric measurements

Connectivity, bone volume/tissue volume (bone volume fraction, BV/TV), and bone surface/bone volume (BS/BV) were analyzed using CTAn ver. 1.17.7.2 (SkyScan). An interpolated ROI using that software included the region from the incisor root apex to the incisal edge roof. Binarizing the images using grayscale (limits 50–140; threshold 0–140) enabled the differentiation of bone and dental structures according to differences in density (Fig. 1b). Skyscan CTVox ver. 3.3.0 (SkyScan) was used to further process the reconstructed images for visualization. An experienced dentomaxillofacial radiologist conducted all image reconstructions and measurements.

Biochemical analyses

Blood samples were collected into serum separator tubes and sera were separated by centrifugation at 1500 × *g* for 10 min after coagulation (K241, BRK5324; Centurion Scientific, West Sussex, UK). Sera were stored at – 80 °C until analyses.

Concentrations of macrophage colony-stimulating factor (M-CSF), receptor activator of nuclear factor κB ligand (RANKL), osteoprotegerin (OPG), and matrix metalloproteinase-8 (MMP-8), which are indicators of alveolar bone remodeling, and the development and function of osteoclasts and tumor necrosis factor (TNF)-α and interleukin (IL)-6, which are proinflammatory cytokines, were determined from the serum samples using commercially available rat-specific enzyme-linked immunosorbent assay kits. Assays were conducted, and the results were calculated following the manufacturers' instructions.

Ready-to-use commercial test kits (RANSEL and RANSOD; Randox, County Antrim, UK) and an automatic analyzer (BS120; Mindray, Shenzhen, China) were used to evaluate glutathione peroxidase (GPx) and superoxide dismutase (SOD) activity to assess oxidative–antioxidative processes.

Malondialdehyde (MDA), the most stable lipid peroxidation product, was measured from sera. The test was conducted using the spectrophotometry method based on a reaction with thiobarbituric acid at 100 °C in an acidic environment; the absorbance of the reaction mixture was measured at 530–540 nm (Ohkawa et al. 1979).

Histopathological analyses

The samples were fixed in 10% neutral buffered formalin solution for 24–72 h. The bone tissue was decalcified in decalcification solution (Shandon TBD-2 decalcifier, Thermo Scientific, Ref.6764004) until the bone tissue was soft enough to examine. The tissues were processed routinely and embedded in paraffin, cut into 4-μm-thick sections using a microtome (Leica RM 2125, Wetzlar, Germany) in the mesiodistal direction on the sagittal axis, and stained for histopathology with hematoxylin and eosin. The Olympus BX light microscope was used to examine the stained sections (Tokyo, Japan) for histopathological evaluation.

Statistical analyses

GraphPad Prism 5.0 (GraphPad Software, Inc., San Diego, CA, USA) was used to conduct the statistical analyses. The Shapiro–Wilk test was used to check normal distribution. One-way analysis of variance was used to analyze the normally distributed data and Tukey's test was used to assess paired comparisons. The Kruskal–Wallis test followed by Dunn's test was used to assess the non-normally distributed data. A P value < 0.05 was considered statistically significant.

Results

Histopathological results

Figure 2 shows representative images of the histological sections. The dentin, gingiva, alveolar bone, and periodontal ligament were observed to have a regular structure in the control group (Fig. 2a). Congestion and increased leukocytes in the gingival connective tissue, enlargement of capillaries and congestion in alveolar bone, and deterioration and congestion in the periodontal ligament were observed in the periodontitis + saline group (Fig. 2b), while decreased ligament deterioration and reintegration with the alveolar bone, and decreased leukocytes in the gingival connective tissue were evident in the periodontitis + 0.5 mg/kg/day oxytocin group (Fig. 2c). The periodontal ligament, dentin, and alveolar integration were improved, and decreased leukocyte density was observed in the periodontitis + 1 mg/kg/day oxytocin group (Fig. 2d).

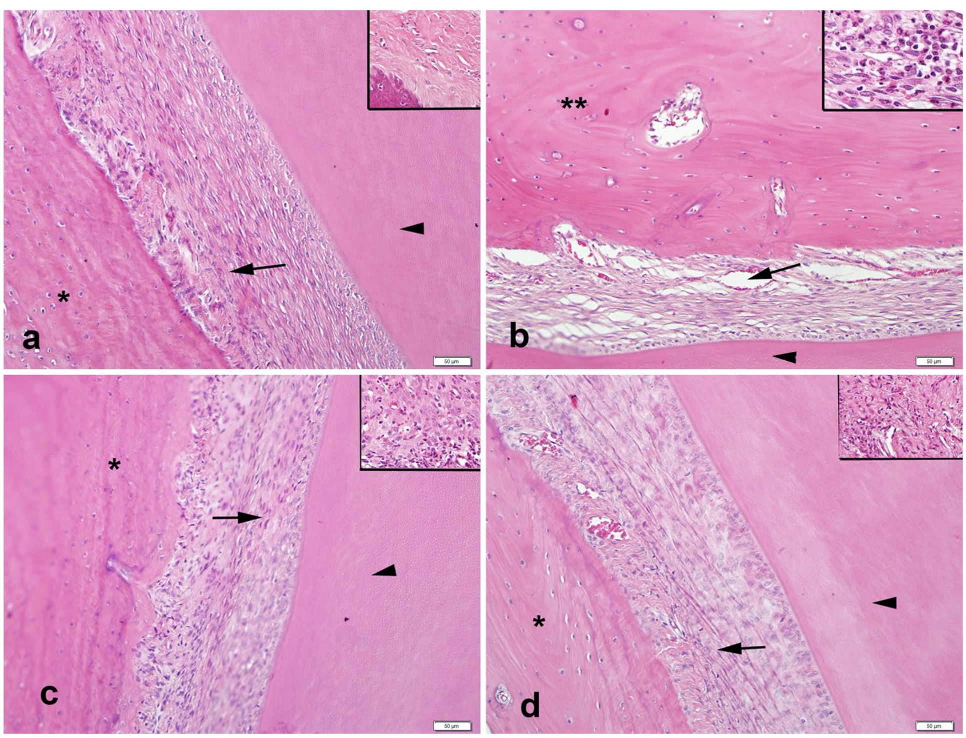

Fig. 2 Representative histological images of the study groups. **a** Control: normal histopathological findings, intact alveolar bone (*), smooth dentin (arrowhead), periodontal ligament (arrow), and gingival mucosa (small image—inset). **b** Periodontitis + saline: separation and deterioration in the periodontal ligament (arrow), dentin (arrowhead), intense congestion in alveolar bone (**), and leukocyte increase in the gingival mucosa (small image—inset). **c** Periodontitis + 0.5 mg/kg/day oxytocin: alveolar bone (*), periodontal ligament (arrow), and dentin (arrowhead) reintegration; decreased leukocytes in gingival mucosa (small image—inset). **d** Periodontitis + 1 mg/kg/day oxytocin: integration of alveolar bone and periodontal ligament (arrow) dentin (arrowhead), new ossification lines in alveolar bone (*), decreased leukocytes in the gingival mucosa (small image—inset)

Microtomography results

Results of the microtomography parameters are shown in Fig. 3. Bone resorption in the lingual, mesial, and distal regions was significantly higher in the periodontitis + saline group ($P < 0.001$) and in the periodontitis + 0.5 mg/kg/day oxytocin group ($P < 0.01$) compared to that in the control group, while bone resorption in the lingual, mesial, and distal regions was statistically significantly lower in the periodontitis + 1 mg/kg/day oxytocin group ($P < 0.01$) than in the periodontitis + saline group (Fig. 3a).

The difference in BV/TV values between the control and periodontitis groups was not statistically significant ($P > 0.05$). The BV/TV values in the periodontitis + 0.5 mg/kg/day oxytocin ($P < 0.05$) and periodontitis + 1 mg/kg/day oxytocin groups ($P < 0.0001$) were significantly higher than that in the periodontitis + saline group (Fig. 3b).

The difference between the control and periodontitis + saline groups was statistically significant ($P < 0.001$). The BS/BV values in the periodontitis + 0.5 mg/kg/day oxytocin ($P < 0.01$) and periodontitis + 1 mg/kg/day oxytocin groups ($P < 0.0001$) were significantly higher than that in the periodontitis + saline group. We also observed that the BS/BV values in the periodontitis + 1 mg/kg/day oxytocin group were statistically significantly higher than those in the periodontitis + 0.5 mg/kg/day oxytocin group ($P < 0.05$) (Fig. 3b).

The connectivity in the control group was higher than that in the periodontitis + saline group ($P < 0.0001$), and that in the periodontitis + 0.5 mg/kg/day oxytocin and periodontitis + 1 mg/kg/day oxytocin groups was significantly higher than that in the periodontitis + saline group ($P < 0.0001$) (Fig. 3b).

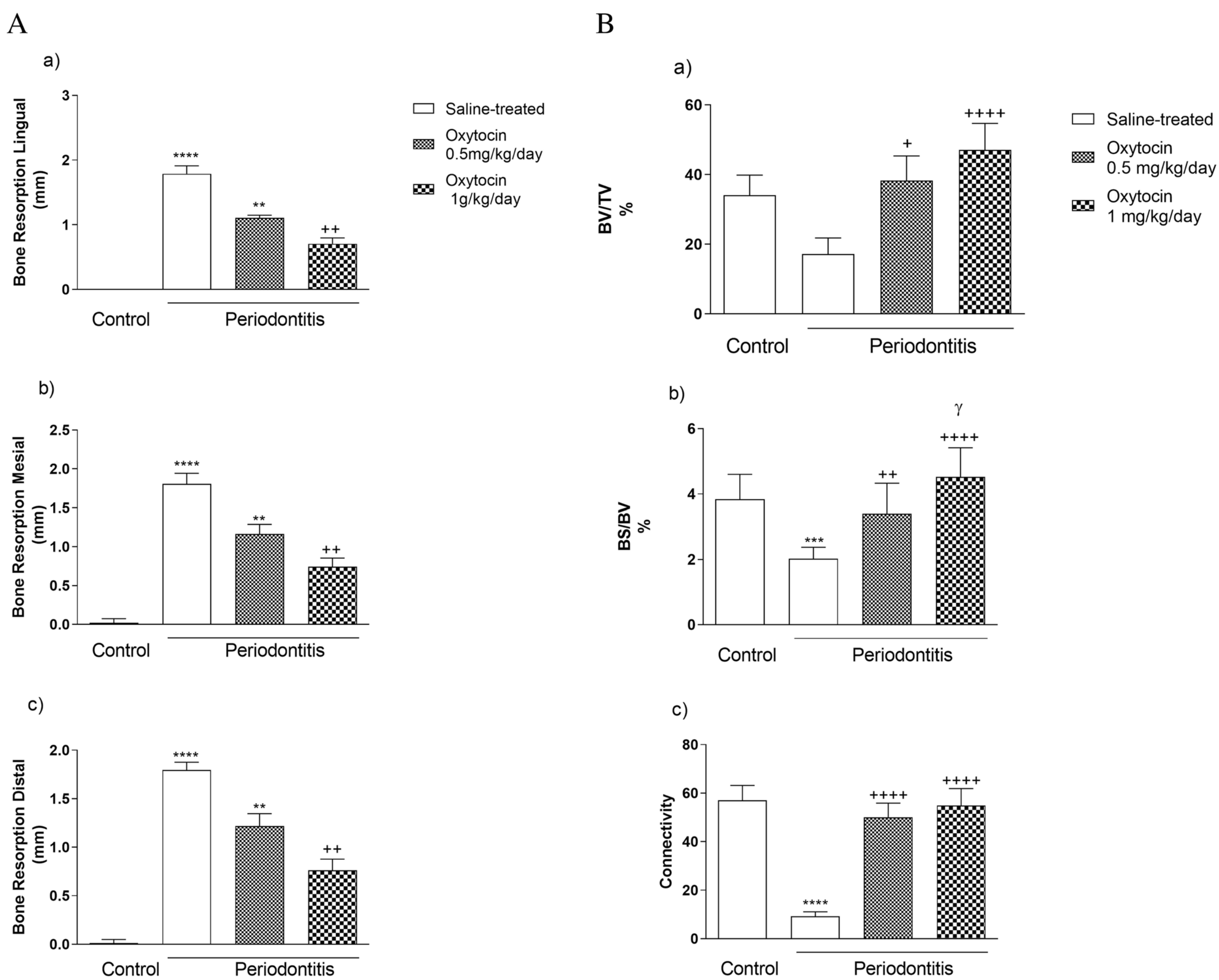

Fig. 3 Microcomputed tomography assessments. **a** Bone resorption in the study groups. **b** Bone volume fraction (BV/TV), bone surface/bone volume (BS/BV), and connectivity of the study groups

Biochemical results

Table 1 presents the results of the biochemical parameters. The TNF-α value was significantly higher in the periodontitis + saline group ($P<0.0001$) and periodontitis + 0.5 mg/kg/day oxytocin group ($P<0.05$) than in the control group, but lower in periodontitis + 0.5 mg/kg/day oxytocin ($P<0.0001$) and periodontitis + 1 mg/kg/day oxytocin groups ($P<0.01$) than in the periodontitis + saline group.

Although the IL-6 value was significantly higher in the periodontitis + saline and periodontitis + 0.5 mg/kg/day oxytocin groups ($P<0.001$) than that in the control group, it was statistically lower in the periodontitis + 1 mg/kg/day oxytocin group than in the periodontitis + saline group ($P<0.05$).

The RANKL value was significantly higher in the periodontitis + saline group than in the control group ($P<0.001$), and it was statistically lower in the periodontitis + 1 mg/kg/day oxytocin group than in the periodontitis + saline group ($P<0.01$). The OPG value was significantly lower in the periodontitis + saline group ($P<0.05$) than in the control group and higher in the periodontitis + 1 mg/kg/day oxytocin group than in the periodontitis + saline group ($P<0.05$).

Both GPx ($P<0.0001$) and SOD ($P<0.01$) values were significantly lower in the periodontitis + saline group than in the control group; both were statistically higher in the periodontitis + 0.5 mg/kg/day oxytocin and periodontitis + 1 mg/kg/day oxytocin groups than in the periodontitis + saline group.

The M-CSF value was significantly higher in the periodontitis + saline group than in the control group ($P<0.0001$). The MMP-8 values were higher in the periodontitis + saline ($P<0.0001$) and periodontitis + 0.5 mg/kg/day oxytocin ($P<0.01$) groups than in the control group. The MDA values were higher in the periodontitis + saline ($P<0.0001$), periodontitis + 0.5 mg/kg/day oxytocin ($P<0.05$), and periodontitis + 1 mg/kg/day oxytocin groups ($P<0.05$).

M-CSF, MMP-8, and MDA were lower in the periodontitis + 0.5 mg/kg/day oxytocin and periodontitis + 1 mg/kg/day oxytocin groups than those in the periodontitis + saline group.

Discussion

The effect of oxytocin on local bone regeneration has been studied by several researchers (Dawood 1995; Park et al. 2014); however, the success of these studies was limited because of the short half-life and insufficient stability of oxytocin against hydrolytic enzymes. In the present study, we administered oxytocin subcutaneously since this protocol reversed bone loss and allowed restoration of bone in mice (Elabd et al. 2008). To the best of our knowledge, there has been no study that has investigated the effects of oxytocin on induced periodontitis. The results of the present study indicated that oxytocin may reduce alveolar bone loss by increasing osteoblastic activity and decreasing osteoclastic activity, oxidative stress, and inflammation.

After recognizing that oxytocin has an anabolic effect on bone metabolism, several studies were conducted on its use in osteoporosis treatment (Elabd et al. 2007, 2008; Tamma et al. 2009). These studies have shown that oxytocin reduces the effects of osteoporosis by acting as an anabolic regulator on bone mass (Elabd et al. 2007, 2008; Tamma et al. 2009). Other studies using ovariectomized rats have found that oxytocin reduces the negative effects of osteoporosis and increases alveolar bone healing and osseointegration of titanium implants (Colli et al. 2012; Wang et al 2016).

Table 1 Mean and standard error of the mean of blood biomarkers

Biomarkers	Control	Periodontitis	Periodontitis + 0.5 mg/kg/day oxytocin	Periodontitis + 1 mg/kg/day oxytocin
TNF-α (pg/mL)	31.81 ± 2.39	59.63 ± 3.15****	38.63 ± 1.57^{++++}	42.77 ± 3.12*$^{, ++}$
IL-6 (pg/mL)	33.18 ± 3.88	81.52 ± 8.56***	76.52 ± 5.19***	55.50 ± 6.77^{+}
RANKL (pg/mL)	51.66 ± 3.50	97.1 ± 10.46***	74.84 ± 5.75	62.09 ± 3.03^{++}
OPG (pg/mL)	215.5 ± 23.7	141.7 ± 7.9*	197.4 ± 12.4	207.8 ± 15.4^{+}
M-CSF (pg/mL)	35.18 ± 1.80	99.24 ± 13.27****	53.67 ± 4.78^{++}	54.31 ± 7.49^{++}
MMP-8 (pg/mL)	55.02 ± 2.45	130.7 ± 7.2****	90.1 ± 5.5**$^{, ++}$	80.1 ± 9.5^{+++}
MDA (µmol/L)	5.96 ± 0.36	10.51 ± 0.51****	7.81 ± 0.55*$^{, ++}$	7.97 ± 0.34*$^{, ++}$
GPx (U/L)	2853 ± 58	1936 ± 108****	2696 ± 104^{+++}	2737 ± 121^{++++}
SOD (U/mL)	0.33 ± 0.08	0.17 ± 0.01**	0.28 ± 0.01^{+}	0.30 ± 0.02^{+}

TNF-α tumor necrosis factor-alpha, *IL-6* interleukin-6, *RANKL* receptor activator of nuclear factor kappa-B ligand, *OPG* osteoprotegerin, *M-CSF* macrophage colony-stimulating factor, *MMP-8* matrix metalloproteinase, *MDA* malondialdehyde, *GPx* glutathione peroxidase, *SOD* superoxide dismutase. Mean ± SEM (standard error of mean). *$P<0.05$, ** $P<0.01$, ***$P<0.001$, ****$P<0.0001$ versus the control group. $^{+}P<0.05$, $^{++}P<0.01$, $^{+++}P<0.001$ versus the periodontitis + saline group

In the present study, there was less bone resorption in the periodontitis + 1 mg/kg/day oxytocin group than that in the periodontitis + saline group. In addition, the BV/TV, BS/BV, and connectivity values increased to varying degrees in the groups with oxytocin compared with those in the periodontitis + saline group. Similar to our results, the results of one study that investigated the effects of oxytocin on distraction osteogenesis have shown that BV/TV and connectivity values were higher in the groups administered oxytocin than in those not administered oxytocin (Altay et al. 2020). The results of the present study on bone volumetric changes using oxytocin are also consistent with the findings of this study. In addition, the histopathologic evaluation in the present study showed congestion and increased leukocytes in the gingival connective tissue, deterioration, and congestion in the periodontal ligament in the periodontitis + saline group but not in the oxytocin groups, and decreased ligament deteriorations, reintegration with the alveolar bone, and a decrease in leukocytes in the gingival connective tissue. In a recent study (Ge et al. 2019), it was shown that oxytocin receptor was expressed on periodontal ligament stem cells, and oxytocin promoted proliferation, migration, and osteogenic differentiation of periodontal ligament stem cells in vitro. In our study, it is thought that the positive changes in the periodontium that occurred in the oxytocin administered groups may be due to the effects and properties of oxytocin described in that study. The results of the present study provided evidence that oxytocin is crucial in the prevention of the development of the periodontitis process. Exogenous oxytocin administration has been shown to alleviate tissue damage in a variety of animal models (Akdemir et al. 2014; Karelina et al. 2011; Yuan et al. 2016). However, no information was available on the effects of oxytocin on periodontitis and its underlying molecular mechanisms. Additional studies are necessary to clarify the exact mechanisms by which oxytocin affects histological changes in the rat model of ligature-induced periodontitis.

Periodontitis is a multifactorial disease in that periodontopathogens induce inflammatory infiltration with the recruitment of polymorphonuclear neutrophils, leukocytes, and macrophages (Hajishengallis et al. 2015). Neutrophils degranulate large quantities of tissue-destroying enzymes (e.g., matrix metalloproteinases) and generate reactive oxygen species; these cells release several proinflammatory cytokines, such as TNF-α, IL-6, and M-CSF, which are involved in inflammation responses and bone resorption (Cavalla et al. 2014) and act through the RANK/RANKL/OPG pathway (Barbato et al. 2015). Studies using animal models have reported that oxytocin may help decrease the levels of circulating proinflammatory cytokines (Akman et al. 2015; Tugtepe et al. 2007) and neutrophil infiltration to the injury site (Al-Amran and Shahkolahi 2014; Iseri et al. 2005). One study has shown that oxytocin inhibits lipopolysaccharide-stimulated proinflammatory cytokine secretion from macrophages and endothelial cells (Szto et al. 2008). In the present study, although TNF-α, IL-6, and M-CSF levels increased in the ligature-induced periodontitis, they decreased significantly with oxytocin administration. These results suggest that oxytocin may control the cytokine response by inhibiting the overexpression of TNF-α, IL-6, and M-CSF.

The RANK/RANKL/OPG pathway controls osteoclast development, differentiation, activation, and function and mediates bone loss in periodontitis. RANK and RANKL are involved in bone resorption during periodontitis because of the osteoclast differentiation enhancement and osteoclastogenesis stimulation (Mori et al. 2013). Several studies using rats have demonstrated that intramuscular or intraperitoneal injection of oxytocin decreases serum levels of calcium and the RANKL soluble receptor activator, increases OPG levels, and has a slight effect on bone reformation (Elabd et al. 2007; Moghazy et al. 2020). In the present study, we measured the levels of RANKL, OPG, and MMP-8 in serum to evaluate the effects of oxytocin on alveolar bone turnover. Similar to those from other studies (Elabd et al. 2007; Gonzalez-Reyes et al. 2015), the data from the present study showed that the OPG value was lowest and the RANKL value was highest in the periodontitis + saline group. Although the RANKL level decreased with the higher dose of oxytocin, the OPG level increased. The MMP-8 value also decreased with both the high and low doses of oxytocin. The results of the present study suggested that oxytocin may be an effective agent for preventing alveolar bone loss.

Although the data in the literature have presented convincing evidence that oxytocin decreases the levels of oxidative stress markers in different tissues and cells, there are no data on periodontal changes from oxytocin (Iseri et al. 2005; Gonzalez-Reyes et al. 2015; Houshmand et al. 2015); however, several studies have demonstrated that oxytocin decreases the concentration of MDA in plasma and tissues (Imani et al. 2015; Iseri et al. 2008). Stanić et al. (2016) have reported that oxytocin decreases total lipid hydroperoxide and increases the activity of SOD in the plasma of rats chronically treated with corticosterone. No difference in MDA concentration among the experimental groups was observed. In the present study, the MDA levels were lower in the oxytocin groups than in the periodontitis + saline group; however, the GPx and SOD levels were statistically higher in the oxytocin groups than in the periodontitis + saline group.

Among the limitations of our study are the fact that only serum biomarkers of oxytocin were examined, and for now, it has been studied only in rats. Our study highlights the importance of oxytocin in preventing periodontitis, a chronic inflammation affecting approximately 50% of the global population (Kononen et al. 2019), by evaluating serum biomarkers.

Considering the results of the present study, we suggest that oxytocin may prevent the destruction of periodontal tissue by regulating oxidative stress in induced periodontitis and that there is strong evidence that oxytocin is crucial for preventing the progression of periodontitis. Our findings enhance the understanding of the pathological mechanisms of periodontitis and provide a possible diagnostic and therapeutic target for the disease; therefore, we suggest that additional studies are needed to determine the mechanism by which oxytocin provides a protective and preventive effect on periodontal tissue. In further studies, the effects of oxytocin on the local inflammation process may be evaluated by focusing on biomarkers isolated from the gingival tissues. We believe that in the future, oxytocin or similar modulating anti-inflammatory agents will be able to prevent by host modulation therapy devastating and chronic inflammation such as periodontitis.

Conclusion

The administration of systemic oxytocin exerted positive effects on the progression of induced periodontitis in rats because it increased bone volume/tissue volume, decreased bone resorption, and decreased oxidative stress and inflammation in the experimental model of periodontitis.

Author contribution TP, GU, AÖŞ, RBKÜ, SS, KO, ŞÇ, NBA, and UA conceived and designed the research. TP, GU, AÖŞ, RBKÜ, SS, KO, ŞÇ, NBA, and UA conducted experiments. TP, AÖŞ, SS, KO, and UA contributed new reagents or analytical tools. TP, GU, and AÖŞ analyzed data. TP, GU, AÖŞ, and RBKÜ wrote the first draft of the manuscript. All authors read and approved the final manuscript and all data were generated in-house and that no paper mill was used.

Data availability The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Competing interests The authors declare no competing interests.

Ethics approval Experiments were carried out in accordance with the guidelines laid down by the National Institute of Health (NIH) in the USA regarding the care and use of animals for experimental procedures. The experimental protocol was examined and approved by the Animal Experiment Ethics Committee of Near East University (number: 2019/05–77).

Conflict of interest The authors declare no competing interests.

References

Akdemir A, Erbas O, Gode F, Ergenoglu M, Yeniel O, Oltulu F et al (2014) Protective effect of oxytocin on ovarian ischemia-reperfusion injury in rats. Peptides 55:126–130. https://doi.org/10.1016/j.peptides.2014.02.015

Akman T, Akman L, Erbas O, Terek MC, Taskiran D, Ozsaran A (2015) The preventive effect of oxytocin to cisplatin-induced neurotoxicity: an experimental rat model. Biomed Res Int 2015:167235. https://doi.org/10.1155/2015/167235

Al-Amran FF, Shahkolahi M (2014) Oxytocin ameliorates the immediate myocardial injury in heart transplant through down regulation of the neutrophil dependent myocardial apoptosis. Heart Views 15:37–45. https://doi.org/10.4103/1995-705X.137493

Allameh Z, Salamzadeh J (2016) Use of antioxidants in urinary tract infection. J Res Pharm Pract 5:79–85. https://doi.org/10.4103/2279-042X.179567

Altay B, Dede EÇ, Özgul Ö, Atıl F, Koçyiğit İD, Orhan K et al (2020) Effect of systemic oxytocin administration on new bone formation and distraction rate in rabbit mandible. J Oral Maxillofac Surg 78:1171–1182. https://doi.org/10.1016/j.joms.2020.03.005

Amar AP, Weiss MH (2003) Pituitary anatomy and physiology. Neurosurg Clin North Am 14:11–23. https://doi.org/10.1016/S1042-3680(02)00017-7

Barbato L, Francioni E, Bianchi M, Mascitelli E, Marco LB, Tonelli DP (2015) Periodontitis and bone metabolism. Clin Cases Miner Bone Metab 12:174–177. https://doi.org/10.11138/ccmbm/2015.12.2.174

Cavalla F, Araujo-Pires AC, Biguetti CC, Garlet GP (2014) Cytokine networks regulating inflammation and immune defense in the oral cavity. Curr Oral Health Rep 1:104–113. https://doi.org/10.1007/s40496-014-0016-9

Colaianni G, Di Benedetto A, Zhu LL, Tamma R, Li J, Greco G et al (2011) Regulated production of the pituitary hormone oxytocin from murine and human osteoblasts. Biochem Biophys Res Commun 411:512–515. https://doi.org/10.1016/j.bbrc.2011.06.158

Colaianni G, Tamma R, Di Benedetto A, Yuen T, Sun L, Zaidi M et al (2014) The oxytocin-bone axis. J Neuroendocrinol 26:53–57. https://doi.org/10.1111/jne.12120

Colli VC, Okamoto R, Spritzer PM, Dornelles RCM (2012) Oxytocin promotes bone formation during the alveolar healing process in old acyclic female rats. Arch Oral Biol 57:1290–1297. https://doi.org/10.1016/j.archoralbio.2012.03.011

Da Silva Freitas Ribeiro D, de Oliveira Freire JM, Teixeira AH, do Val DR, da Freitas AR, Gomes FISF et al (2018) Tocoyena sellowiana extract decreases bone loss in an experimental model of periodontitis in rats: putative role for cyclooxygenase-2 and IL-1 beta inhibition. Bomed Pharmacother 98:863–872. https://doi.org/10.1016/j.biopha.2018.01.011

Dawood MY (1995) Novel approach to oxytocin induction-augmentation of labor. Application of oxytocin physiology during pregnancy. Adv Exp Med Biol 395:585–594

Dou D, Liang J, Zhai X, Li G, Wang H, Han L et al (2021) Oxytocin signalling in dendritic cells regulates immune tolerance in the intestine and alleviates DSS-induced colitis. Clin Sci (lond) 135:597–611. https://doi.org/10.1042/CS20201438

Düşünceli F, Işeri SO, Ercan F, Gedik N, Yeğen C, Yeğen BC (2008) Oxytocin alleviates hepatic ischemia-reperfusion injury in rats. Peptides 29:1216–1222. https://doi.org/10.1016/j.peptides.2008.02.010

Elabd SK, Sabry I, Hassan WB, Nour H, Zaky K (2007) Possible neuroendocrine role for oxytocin in bone remodeling. Endocr Regul 41:131–141

Elabd C, Basillais A, Beaupied H, Breuil V, Wagner N, Scheideler M et al (2008) Oxytocin controls differentiation of human mesenchymal stem cells and reverses osteoporosis. Stem Cells 26:2399–2407. https://doi.org/10.1634/stemcells.2008-0127

Franco JH, Chen X, Pan ZK (2021) Novel treatments targeting the dysregulated cell signaling pathway during sepsis. J Cell Signal 2:228–234

Ge B, Liu H, Liang Q, Shang L, Wang T, Ge S (2019) Oxytocin facilitates the proliferation, migration and osteogenic differentiation of human periodontal stem cells in vitro. Arch Oral Biol 99:126–133. https://doi.org/10.1016/j.archoralbio.2019.01.007

Gonzalez-Reyes A, Menaouar A, Yip D, Danalache B, Plante E, Noiseux N et al (2015) Molecular mechanisms underlying oxytocin-induced cardiomyocyte protection from simulated ischemia-reperfusion. Moll and Cell Endocrinol 412:170–181. https://doi.org/10.1016/j.mce.2015.04.028

Hajishengallis G, Chavakis T, Hajishengallis E, Lambris JD (2015) Neutrophil homeostasis and inflammation: novel paradigms from studying periodontitis. J Leukoc Biol 98:539–548. https://doi.org/10.1189/jlb.3VMR1014-468R

Hienz SA, Paliwal S, Ivanovski S (2015) Mechanisms of bone resorption in periodontitis. J Immunol Res 2015:615486. https://doi.org/10.1155/2015/615486

Houshmand F, Faghihi M, Zahediasl S (2015) Role of atrial natriuretic peptide in oxytocin induced cardioprotection. Heart Lung Circ 24:86–93. https://doi.org/10.1016/j.hlc.2014.05.023

Huang Y, Celikten B, de Faria VK, Ferreira Pinheiro Nicolielo L, Lippiatt N, Buyuksungur A et al (2017) Micro-CT and nano-CT analysis of filling quality of three different endodontic sealers. Dentomaxillofac Radiol 46:20170223. https://doi.org/10.1259/dmfr.20170223

Imani A, Khansari M, Azizi Y, Rakhshan K, Faghihi M (2015) Stimulation of oxytocin receptor during early reperfusion period protects the heart against ischemia/reperfusion injury: the role of mitochondrial ATP-sensitive potassium channel, nitric oxide, and prostaglandins. Acta Med Iran 53:491–500

Iseri SO, Sener G, Saglam B, Gedik N, Ercan F, Yegen BC (2005) Oxytocin protects against sepsis-induced multiple organ damage: role of neutrophils. J Surg Res 126:73–81. https://doi.org/10.1016/j.jss.2005.01.021

Iseri SO, Gedik IE, Erzik C, Uslu B, Arbak S, Gedik N et al (2008) Oxytocin ameliorates skin damage and oxidant gastric injury in rats with thermal trauma. Burns 34:361–369. https://doi.org/10.1016/j.burns.2007.03.022

Karelina K, Stuller KA, Jarrett B, Zhang N, Wells J, Norman GJ et al (2011) Oxytocin mediates social neuroprotection after cerebral ischemia. Stroke 42:3606–3611. https://doi.org/10.1161/STROKEAHA.111.628008

Kononen E, Gursoy M, Gursoy UK (2019) Periodontitis: a multifaceted disease of tooth-supporting tissues. J Clin Med 8:1135. https://doi.org/10.3390/jcm8081135

Lee HJ, Macbeth AH, Pagani JH, Young WS 3rd (2019) Oxytocin: the great facilitator of life. Prog Neurobiol 88:127–151. https://doi.org/10.1016/j.pneurobio.2009.04.001

Mester A, Ciobanu L, Taulescu M, Apostu D, Lucaciu O, Filip GA et al (2019) Periodontal disease may induce liver fibrosis in an experimental study on Wistar rats. J Periodontol 90:911–919. https://doi.org/10.1002/JPER.18-0585

Moghazy H, Abdeen Mahmoud A, Elbadre H, Abdel Aziz HO (2020) Protective effect of oxytocin against bone loss in a female rat model of osteoporosis. Rep Biochemi Mol Biol 9:147–155. https://doi.org/10.29252/rbmb.9.2.147

Mori G, D'Amelio P, Faccio R, Brunetti G (2013) The interplay between the bone and the immune system. Clin Dev Immunol 2013:720504. https://doi.org/10.1155/2013/720504

Ohkawa H, Ohishi N, Yagi K (1979) Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal Biochem 95:351–358. https://doi.org/10.1016/0003-2697(79)90738-

Park JW, Kim JM, Lee HJ, Jeong SH, Suh JY, Hanawa T (2014) Bone healing with oxytocin-loaded microporous beta-TCP bone substitute in ectopic bone formation model and critical-sized osseous defect of rat. J Clin Periodontol 41:181–190. https://doi.org/10.1111/jcpe.12198

Stanić D, Plećaš-Solarović B, Petrović J, Bogavac-Stanojević N, Sopić M, Kotur-Stevuljević J et al (2016) Hydrogen peroxide-induced oxidative damage in peripheral blood lymphocytes from rats chronically treated with corticosterone: the protective effect of oxytocin treatment. Chem Biol Interact 256:134–141. https://doi.org/10.1016/j.cbi.2016.07.006

Szto A, Nation DA, Mendez AJ, Dominguez-Bendala J, Brooks LG, Schneiderman N et al (2008) Oxytocin attenuates NADPH-dependent superoxide activity and IL-6 secretion in macrophages and vascular cells. Am J Physiol Endocrinol Metab 295:E1495–E1501. https://doi.org/10.1152/ajpendo.90718.2008

Tamma R, Colaianni G, Zhu LL, DiBenedetto A, Greco G, Montemurro G et al (2009) Oxytocin is an anabolic bone hormone. Proc Natl Acad Sci U S A 106:7149–7154. https://doi.org/10.1073/pnas.0901890106

Tugtepe H, Sener G, Biyikli NK, Yuksel M, Cetinel S, Gedik N et al (2007) The protective effect of oxytocin on renal ischemia/reperfusion injury in rats. Regul Pept 140:101–108. https://doi.org/10.1016/j.regpep.2006.11.026

Wang M, Lan L, Li T, Li J, Li Y (2016) The effect of oxytocin on osseointegration of titanium implant in ovariectomized rats. Connect Tissue Res 57:220–225. https://doi.org/10.3109/03008207.2016.1141902

Yuan L, Liu S, Bai X, Gao Y, Liu G, Wang X et al (2016) Oxytocin inhibits lipopolysaccharide-induced inflammation in microglial cells and attenuates microglial activation in lipopolysaccharide-treated mice. J Neuroinflammation 13:77. https://doi.org/10.1186/s12974-016-0541-7