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Effects of Ovariectomy and Bilateral Molar Teetless on Mandibular Condyle of Rat

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Abstract: This study, researchers examined the ovariectomy and bilateral molar teetless of mandibular condyle in rat by histologically and histomorphometrically. In this part of the study, 12 Wistar Albino rats, 120 days old and weighing 200-250 g were used. The rats were divided into three groups, a control and two experimental group. The experimental groups of rats were subjected to ovariectomy in order to create oestrogen deficiency and bilateral molars extraction. After 35 days of ovariectomy, control and experimental group rats were killed and their mandible condyle removed. The speciemens were stained with Hematoxylin and Eosin (HE) and examined for structure of histologically and histomorphometrically. Histomorphometric analysis; mandibular condyle of control and experimental groups were statistically significant results of the histological layers ($p<0.05$). It was concluded that ovariectomy application and molar teetless can induce histopathological and histomorphometrically changes in the structure of the condyle.

Key words: Mandible condyle, rat, bilateral molar teetless, ovariectomy, Turkey

INTRODUCTION

Osteoporosis is a major public health concern, its effect on oral bone has not been clarified. More important may be the effect of estrogen depletion on the response of oral bone to dental treatments such astoothextraction or pathologic processes such as prosthodontic disease.

Osteoporosis will be one of the most common health problems with respect to geriatric health condition. This systemic disease is more common in geriatric women. Occurrence of spontaneous fracture is known especially in the advenced stage of systemic disease in geriatric patients particulary fort he postmenoposal women who was rapidly decreased eastrogen hormonal level.

Temporomandibular Joint (TMJ) also known as jaw joint or mandibular joint is an ellipsoid variety of synovial joints, right and left joints forming a bicondylar articulation. The articular disk is a thin, oval plate, placed between the condyle of the mandible and the mandibular fossa. Its upper surface is concavo-convex from before backward, to accommodate itself to the form of the mandibular fossa and the articular tubercle. It's under surface, in contact with the condyle is concave. Its circumference is connected to the articular capsule and in front to the tendon of the Pterygoideus externus (Herring, 2003).

The precise relationship between osteoporosis, periodontal mandibular bone and tooth loss is currently not well understood (Bollen *et al*., 2004; Hildebolt *et al*., 1997; Mavropoulos *et al*., 2007).

Despite the growing evidence that Ovariectomy (OVX) could be associated with tooth loss rather conflicting results have been observed in different animal studies on the effect of Ovariectomy (OVX) on the mandibular bone (Jiang *et al*., 2003; Johnson *et al*., 2002; Tanaka *et al*., 2003).

Numerous epidemiological studies have pointed out a higher frequency of Temporomandibular Disorder (TMD) in women than in men which indicates the involvement of a sex hormone such as estrogen in the pathogenesis of TMD. Although, estrogen is known to play pivotal roles in osteoarthrosis or rheumatoid arthritis in systemic joints, there have been few reports about the role of estrogen in the temporomandibular joint (Yamada *et al*., 2003). The effect of postmenopausal osteoporosis on the mandible is also attracting the

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attention of dental specialists in recent years. It has been reported that in ovariectomized rats boneloss may occur in the mandibular body and the mandibular condyle (Tanaka *et al*., 2000, 2002).

Despite the growing evidence that Ovariectomy (OVX) could be associated with tooth loss rather conflicting results have been observed in different animal studies on the effect of OVX on the mandibular bone (Johnson *et al*., 2002; Moriya *et al*., 1998).

The main purpose of this study is to present histomorphologic influence on mandibular condyle in the rats with loss of mastication function and decreased hormonal level of eostrogen in partiallly edentulus posterior regions.

MATERIALS AND METHODS

Animals: In this part of the study, 12 Wistar Albino rats, 120 days old and weighing 200-250 g were used. The rats were obtained from the Department of Medical Science Application and Research Centre of Dicle University (DUSAM). They were housed in invidual cages in temperature-controlled environment (22°C) with a 12:12 h light-dark cycle. All animals were fed standard pellet food and adlibitum tap water which were performed according to the Declaration of Helsinki with the permission of the Governmental Animal protection committee. The animals were divided to randomly three groups.

Experimental protocol: Eight rats underwent bilateral Ovariectomy (OVX) after intramuscular injection of ketamine hydrochloride 90 mg kg^{-1} (Ketalar-Eczacibasi, Turkey). Four rats were subjected to bilateral Ovariectomy (OVX). The other four rats were subjected to bilateral ovariectomy and bilateral molar teetless (n:4).

After 35 days, control and experimantal group rats were sacrificiezed under over dose sodium pentobarbital and their mandible condyle removed.

Histologic evaluation: The mandible condyles were fixed in 10% neutral formalin solution for 24 h, decalcified in 5% formic acide and embedded into paraffin. The block was cut in to 5 μm. They were stained with Hematoxylin and Eosin (HE) and examined for structure of mandible condyle under a light microscope. The microphotographs were taken by Nikon 400 Eclipse microscope.

Histomorphometrical analysis: The sections were used to measure the thickness of the condylar cartilage layer. The articaular cartilage layer was divided into fibrous (articular), proliferative (chondrogenic) and maturative/hypertrophic (cartilaginous) zones. The histomorpometric analyses were performed by image analyses program Nikon New Version.

Statistical analysis: The Kruskall-Wallis and Mann-Whitney U-tests were used to determine the differences between the groups in relation to the thickness of the layers in the mandibular condyle. The Statistical Package Social Science (SPSS), Version 15.0 (SPSS Inc., Chicago, IL, USA) was used for data analyses. For all statistical tests, the significance level was $p<0.05$.

RESULTS

Histologic findings

Group-I (control): Histological examination of the rat mandibular condyle of the control subjects; layers followed the normal structures (Fig. 1).

Group-II (Ovariectomy-OVX): The rats were ovariectomized compared to control mandibular condyle; fibrous layer showed degenerative changes. Proliferative layer expands, the contraction suffered by hyphertropic layer, a layer of erosion was significantly less activation is selected and the reduction in osteoblastic lesions. Thin bone trabecula and bone marrow areas of ossification observed that there is a significant increase in the amount as shown in Fig. 2.

Group-III (Ovariectomy-OVX and bilateral molar teetless): Mandibular condyle of rats after ovariectomy and ovariectomy created molar edentulous mandibular condyle of rats generated very clear that the differences

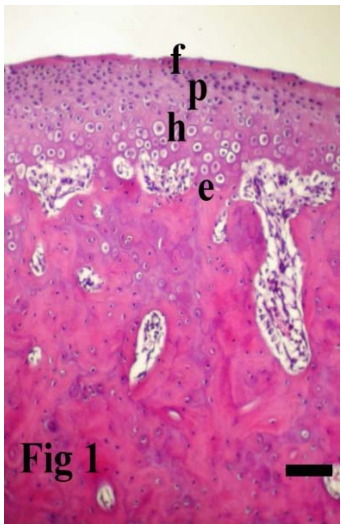

Fig. 1: Control group of rat mandible condyle

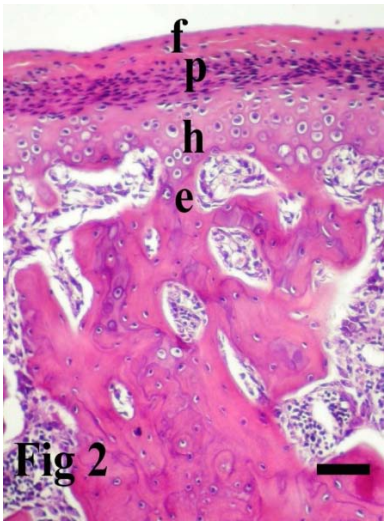

Fig. 2: Ovariectomy group of rat mandible condyle

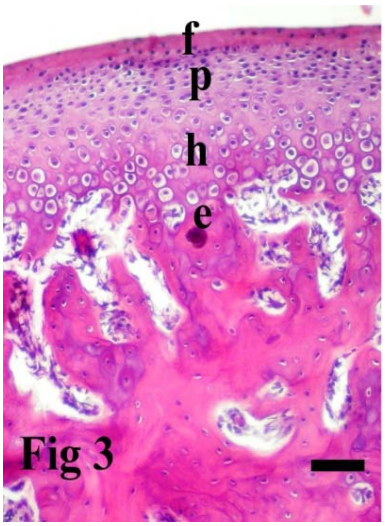

Fig. 3: Ovariectomy and bilateral molar teetless group of rat mandible condyle; f: fibrous (articular); p: proliferative (chondrogenic); h: maturative/ hypertrophic (cartilaginous) and e: eresion zones. Stain: Hematoxylen-Eosin (H&E), scale bar: 50 μm

observed histologically compared. Layer of fibrous degeneration of the mandibular condyle in this group is higher than the proliferative layer, expansion, contraction hypertropic layer, a layer of erosion rather decreased significantly and osteoblastic activity was observed. This group is too weak areas of the condyle ossification in the rest of irregular and fine bone trabecula watched. Increasing in the bone marrow but were markedly reduced ossification (Fig. 3).

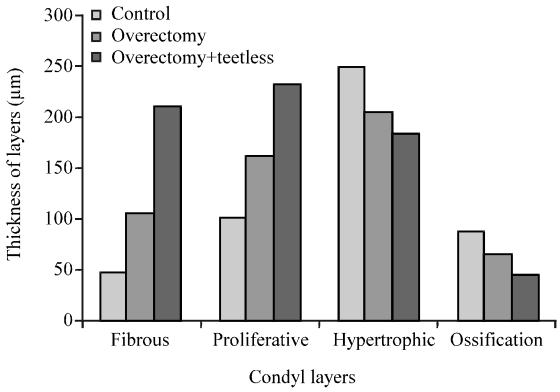

Fig. 4: Histomorphometric evaluation of the control and experimental groups and layers of condylar

Histomorphometric measurement results: Nikon New Version computer program, the control and experimental groups, the histological layers of the mandibular condyle micrometer (μm) statistical evaluation of the measurement was performed (Fig. 4).

Histomorphometric analysis between the control and experimental groups, statistical evaluation of mandibular kondilerinin the following results were obtained from histological layers. Control groups were ovariectomized and ovariectomized + bilateral molar toothless fibrous mean values ($p = 0.07$), proliferative ($p<0.01$), hypertrophic ($p<0.01$), ossification ($p<0.05$) was significantly higher than parameter varies.

Control, ovariectomized and ovariectomized + bilateral molar edentulous groups fibrous of average values ($p = 0.07$), proliferative ($p<0.01$) and hypertrophic ($p<0.01$), ossification ($p<0.05$) between the layers of the important parameters degree varies.

Ovariectomy compared with the control group, fibrous ($p<0.05$), proliferative ($p<0.05$) and hypertrophic ($p = 0.021$) and statistically significant parameters, ossification ($p = 0.110$), a statistically insignificant parameter.

Ovariectomy + molar edentulous compared with the control group, fibrous ($p<0.05$), proliferative ($p<0.05$) and hypertrophic ($p<0.05$) and ossification ($p<0.05$), the parameters are statistically significant.

Ovariectomy and ovariectomy + bilateral molar edentulous comparison of the groups; fibrous ($p<0.05$), proliferative ($p<0.05$) and hypertrophic ($p<0.05$) and ossification ($p<0.05$), the parameters are statistically significant.

Histomorphometric analysis; mandibular condyle of control and experimental groups were statistically significant results of the histological layers ($p<0.05$).

These data showed that ovariectomy and bilateral molar teetless leads to an increase in mandible condyle disorder as well as a unique ability to alter cell and morphology.

DISCUSSION

Today, research on mandible condyle is even more important due to the rising number of people suffering from diseases such as chewing problem caused by a breakdown in the structure and functionality of mandible condyle.

Ovariectomized rats have been used by researchers as the model for postmenopausal osteoporosis. Even though limitations exist, certain characteristics in the rat model mimic the bone changes in postmenopausal women and made the study of the human disease possible (Lelovas *et al*., 2008).

The effect of oestrogen on mandibular condylar cartilage has been studied by several researchers. In TMJs from ovariectomized animals the increased condylar cartilage thickness and even degenerative changes were noticed (Okuda *et al*., 1996).

Researchers observed that ovariectomy and molar teetless exposure for 35 days affected the condylar regions histologically in the experimental group, the condylar layer thickness showed significant difference from the control group. In the experimental group of rats, layer of fibrous degeneration of the mandibular condyle in this group is higher than the proliferative layer, expansion, contraction hypertropic layer, a layer of erosion rather decreased significantly and osteoblastic activity was observed.

These findings also indicate that oestrogen may alter condylar remodelling, leading to degenerative changes in the temporomandibular joint. Similar conclusions were drawn by Fujita *et al*. (2001).

Several animal experiments have been done to examine the relationship between the mandibular condyle and mechanical loading by occlusion. One of these studies showed that thickness of cartilage and trabecular density were reduced only in the anterior part of the condyle when rats were subjected to trimming of incisors to lessen the mechanical loading on the condyle (Hinton and Carlson, 1986).

On the other hand, estrogen deficiency had an effect on reducing the bone mass of each region of the condyle. From 30 days onward, both regions of the Ovx condyles showed the tendency to have low bone volume as compared with those of the Sham counterparts. This is probably because the boneresorptive activities immediately elevated and the bone turnover continued to be high in the Ovx group. In contrast, the bone formation rate was also elevated at 60 day postovariectomy, probably due to the coupling phenomena induced by accelerated bone resorption as reported in other parts of skeletons (Wronski *et al*., 1988).

Some experimental studies have indicated that ovariectomy negatively affected bone healing around the implants resulting in reduced bone area and density (Nociti *et al*., 2002; Duarte *et al*., 2005).

In rats, Shimizu *et al*. (2000) observed that the healing process of alveolar bone after molar extraction was delayed after ovariectomy as a result of increased bone resorption. Similarly, Tanaka *et al*. (2001) suggested that ovariectomy might decrease the thickness and mechanical strength of alveolar bone after molar extraction by increased resorption rather than by inhibition of the wound healing process in extracted sockets (Tanaka *et al*., 2001).

Prostodontist should have the responsibility to collobrate with the specialist who teat the systemic disease with respect to define the early diagnosis and treatment. The dentist is in the strategic position with respect to see the patient in initial visit. In the medical phse of the patient consultation, the posibility of being osteoporos would be especially in postmenoposal women.

Ulku *et al*. (2012) observed that histopathologic changes such as: increased thickness of the proliferative layer, hypertrophic layer contraction, inhomogeneous appearance of cells, irregular appearance of bone marrow cavities, irregularities in transition from hypertrophic areas to ossification and irregular appearance in calcification, it could be suggested that hormonal and physiologic changes in ovariectomized rats can affect the bone structure of mandibular condyle.

In rats with unilateral molar edentulism depending on the histopathologic changes in the left mandibular condyle formed. Despite the expansion of the fibrous layer of the left mandibular condyle, hypertrophic layer degradation have been identified (Zengingul *et al*., 2006).

Edentulous very impressed with the lack of estrogen have detected morphological structure of the condyle. The findings are in agreement with earlier studies.

CONCLUSION

The results of the present study showed that oestrogen deficiency, produced by means of ovariectomy and molar teetless application in rats, provoked significant changes in the structural properties of the condylar layers. Researchers observed that the mandibular condylar cartilage is sensitive to changes in estrogen deficiency and bilateral molar teetless.

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