#### 204 - EFFECT OF SWIMMING EXERCISE ON SPONGY BONE IN THE OVARIECTOMIZED RATS

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#### INTRODUCTION

The bone is a tissue that serves as a support for the soft tissue and shell of various ions among them calcium (STEVENS AND LOWE, 1997). In aging, the process of resorption exceeds bone formation and with it begins the phase of bone loss is called osteoporosis. The same process occurs when using the technique of ovariectomy to induce menopause (KALU, 1991). Osteoporosis is characterized by asymptomatic and silent loss of bone mass leading to fragile and weakening of bones that become susceptible to fractures (COTRAN et al., 1997). Osteoporosis currently, due to an aging world population, has been a public health problem. Osteoporosis is installed through two main mechanisms: increased activity of osteoclasts (cells that absorb bone), resulting in accelerated bone resorption, or the reduction of osteoblasts (replacement cells) leading to an inadequate bone remodeling. Among these factors bone formation is physical activity, among others, responsible for local and general changes of body tissues that will influence the formation of bones. Thus, its role for both the control and prevention of osteoporosis is advocated by several authors, due to their involvement in the genesis and mineralization of bones and distribution of blood flow and skeletal muscle and its mechanical action caused by the exercise on density and bone formation. It is believed that exercises that generate traction, compression and torsion are the most effective for the prevention of osteoporosis in the elderly, the most recommended for this body building, however little is known about the power of the exercises, after the installation of a picture of osteoporosis, the maintenance of bone integrity. Our objective was to determine whether, after installing the menopause by ovariectomy, an exercise protocol would be sufficient to control or minimize the loss of bone density or the maintenance of microarchitecture of spongy and compact bone that make up a long bone.

### **METHODOLOGY**

This study was approved by the Ethics Committee process 223/2007. We used 20 Wistar rats (Rattus norvegicus Albinus Wistar), aged 60 days. All animals were fed a standard balanced diet (Purina) and water ad libitum, distributed, 5 rats in each cage, at controlled room temperature 25 ° C and a photoperiod of 12 h light / 12h dark. The rats were randomly assigned to four groups of five animals named: 1- Sedentary Control (S): rats not subjected to the training protocol, not ovariectomized; 2- Castrated Sedentary (SC): Ovariectomized rats not subjected to the training protocol; 3 - Trained Control (T): Trained only; 4 - Trained Castrated (TC) held the training protocol and were ovariectomized. The rats were anesthetized with pentobarbital sodium 2% intraperitoneal and ovariectomy was performed. The rats were placed in cage with clean Dipyrone sodium dissolved in water during the first two days after surgery. The training protocol consisted of swimming, five times a week over nine weeks. In the first week the rats went through a period of adaptation to the water for a period of 20 minutes five days without swimming overhead. After this period started using an overhead equal to 5% of their body weight, coupled with his elastic body. In the following weeks will adaptation training time was increased by 5 minutes per week, finishing in ninth week with a time of 60 minutes. The water temperature was maintained between 31 and 32 degrees, considered as thermally neutral body temperature of rats. The animals were weighed for the assessment of body mass, once a week throughout the experiment by a scale semi - analytical. This procedure was performed on the last day of the week. After the period of 9 weeks of training, the animals were sacrificed by guillotine and femur of both was collected for electron microscopy. The middle third of the diaphysis, and this was fixed in a Karnovsky solution for 48 hours and was divided along for the bone marrow could be withdrawn by jets, with a syringe of saline. This procedure allowed the spongy inner wall of the segment was exposed. Later the material was dehydrated, dried in critical point apparatus and coated with gold. The material was observed in Scanning Electron Microscope Philips (Laboratory of the Biology Department, IB-UNESP-RC). Statistical analysis was performed by analysis of variance and t-Student and Bonferroni's test.

#### **RESULTS**

To make sure that the rats were menopausal by ovariectomy, was made the observation of atrophy of the uterus. In some cases the uterus was non-existent or was not found. The control of body weight of animals studied was given weekly, with a weight gain greater in castrated (SC and TC), relating the initial weight groups, and their final weights, and weight gain in percentage, as seen in Table 1.

TABLE 1 - Body weight initial (PI), final body weight (PF), and weight gain (GP). Measured by the percentage of GP = PF - PI, in (g).

Groups Experimental	Initial Weight (g)	Final weight (g)	Weight Gain (%)
Sedentary Control (n=5)	221 ± 9.6	282.8±16,6	28.1
Castrated sedentary (n=5)	216 ± 6.5	351.8 ± 14.6	63.0
Trained Control (n=5)	227 ± 4.4	281.6 ± 7.7	24.2
Trained Castrated (n=5)	231 ± 12.4	323.6 ± 30,5	40.3

Results in Middle standard deviation for P> 0.05.

The weight gain of groups presented in Table 1 is shown in graphic 1.

Graphic 1 - Initial body weight-matched to the final body weight of experimental groups in (g).

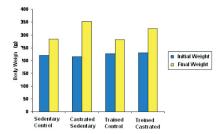


Figure 1 shows a significant increase in final weight of the castrated groups (SC and TC), and an expected increase of the groups (S and T).

Qualitatively, the bones of the sedentary rats did not show the spongy bone that fills the bone cavity (Fig. 1). The inside of these bones were not occupied by any structure that resembles the existence of a trabecular bone that form spongy bone, and that should fill this region. Since the bones of rats submitted to swimming exercise, there was the preservation of spongy bone that fills the bone cavity (Fig. 2). It is evident that the trabecular bone formed within the femurs of the animals in group T (Fig. 3) is more robust and therefore stronger than trabecular bone found in animals from other groups. A more detailed analysis of bone tissue shows that these spikes, as well as throughout the endosteal surface of bones of animals of the sedentary group (S and SC), osteoclasts were present in phagocytic activity evident on the surface of both bone fragments and on the surface besides some endosteal osteoblasts (Fig. 4). This phagocytic activity revealed in the endosteal excavations produced by osteoclasts. In a smaller increase was also observed channels Volkman, since these animals in group S and SC the absence of spongy bone in the spinal canal allowed this observation. In the group trained can be observed that the spikes that form the spongy bone were more numerous and robust in group T than in the CT (Fig. 5). In these animals, despite the presence of spines, these were thinner compared to the group T. Discussion The bone tissue is reabsorbed and deposited continuously, however, with age, menopause, ovarian follicles are no longer active, just as occurs in ovariectomy. The parade in follicular development results in reduced production of the hormone estrogen, reaching almost zero (DELLING; GLUECKSELING 1971, HARRISON, 1996; SIMINOSKI AND JOSS, 1996) and thus the size of the uterus begins to decline even started to disappear as found in this study. According to Yamasaki and Yamaguchi (1989) in both species the loss of bone is greater in spongy bone than in cortical bone which corroborate with the present research. With respect to trabecular bone on this work have also been demonstrated by Castro Netto et al. (2006) and Reddy and Lakshmana (2003). Authors Tervo et al. (2009) observed that young men studying the reduction of exercise leads to reduction of spongy bone and to a lesser extent in the cortical bone. By studying the proximal tibia of rats, the authors Waarsing et al. (2006), noted that changes in the organization of bone architecture occurred after ovariectomy are similar to changes that occur in normal aging of animals. The authors suggest that estrogen upon depletion results in acceleration of normal bone adaptation. Bone loss in young ovariectomized rats according O'Louchin and Howard (1994) is due to reduced intestinal calcium absorption and increased excretion of calcium through the gastrointestinal tract. Yamazaki and Yamaguchi (1989); Arisawa et al. (2000) found that the administration of the hormone calcitonin in ovariectomized rats, after watching osteoporosis can promote more rapid regeneration of bone defect.

#### CONCLUSION

The aspect found in this research, the bones of rats trained to indicate that the resistance to trauma is increased in these bones, as they can sell the forces that the bone is submitted more efficiently than the bones of the sedentary rats. According to the results of the ovariectomy in rats is efficient for the onset of osteoporosis, as evidenced by the observation of a greater number of osteoclasts and the absence of spicules and trabeculae in the sedentary group castrated. Characteristics which mitigated and controlled with the swimming exercise, observed in the group trained castrated. The increase in body weight and destruction of bone micro-architecture has been identified as a result of ovariectomy for the sedentary and trained castrated rats. These more pronounced in the sedentary group, the impression that the swimming exercise, although not the most suitable for the treatment of osteoporosis, was efficient in the case of ovariectomized rats. Therefore, the implementation of a protocol of swimming exercise promotes the maintenance of the microarchitecture of bone, therefore, an exercise that may be recommended for prevention of fractures in osteoporotic individuals.

Figure 1. Photomicrograph electronics endosteal surface of the diaphysis from rat in group (S) in which there is bony wall of the shaft and lack of trabeculae of spongy bone in his spinal canal compared with the control group (T). SEM. Bar = 1 mm.

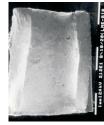


Figure 2. Photomicrograph electronics endosteal surface of the diaphysis from rat in group (TC) where any difference in thickness and content of spongy bone trabeculae when compared to (T). SEM. Bar = 1 mm.

Figure 3. Photomicrograph electronics endosteal surface of the diaphysis from rat in group (T). Notice the difference in

thickness and trabecular pattern of spongy bone compared to control (TC) and the group (S). SEM. Bar = 1 mm

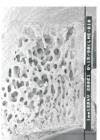


Figure 4. Photomicrograph of bone tissue. Observe osteoclasts in bone fragments adhered to their excavation sites (red arrows) and few osteoblasts produce new bone (yellow arrows).H/E. Bar = 30 micrometers.



Figure 5. Electron photomicrograph showing detail of trabecular spongy bone from animals in group (T). Bar = 1mm.



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### EFFECT OF SWIMMING EXERCISE ON SPONGY BONE IN THE OVARIECTOMIZED RATS ABSTRACT

Introduction The Ovariectomy is characterized by the removal of the ovaries and induces early menopause. Objective evaluate whether the swimming exercise may be indicated for the prevention and control of osteoporosis. Methodology 20 Wistar rats were separated by groups: sedentary control (S), trained control (T), ovariectomized sedentary (OS) and trained ovariectomized (TO). The rats of group T were submitted to swimming exercise, with a load of 5% of body weight for 8 weeks. The induction of menopause, 5 weeks before the start of the experiment was performed by ovariectomy. After the training period the

middle third of the femur was collected, fixed and prepared for ultrastructural analysis. Results morphological analysis of spongy bone shows that when compared to T, the bones of the TO and S groups showed a marked decrease in the thickness and the amount of trabecular, while in group S spongy bone was completely absent. In sedentary animals, the interior of the bone has not spongy bone unlike the trained rats. This found the bones of rats trained us to believe that resistance is increased, since in them, can be disposed of with greater efficiency, the forces of tension, compression and torsion to which the bone is subjected during the daily activities. Conclusion swimming promotes the maintenance of bone architecture and can be recommended for prevention of fractures in individuals with osteoporosis.

KEYWORDS: bone, ovariectomy, osteoporosis

# EFFET DE L'EXERCICE DE NATATION SUR LA OS SPONGIEUX DE RATES OVARIECTOMISÉES RÉSUMÉ

Introduction Ovariectomy est caractérisée par l'ablation des ovaires et induit une ménopause précoce. Objectif évaluer si l'exercice de natation peut être indiqué pour la prévention et le contrôle de l'ostéoporose. Méthodologie 20 rats Wistar ont été séparées par groupes: contrôle sédentaires (S), le contrôle exercitées (T), ovariectomisées OS (sédentaires) et exercitées ovariectomisées (TO). Les rats du groupe T ont été présentées à l'exercice de natation, avec une charge de 5% du poids corporel pendant 8 semaines. L'induction de la ménopause, 5 semaines avant le début de l'expérience a été réalisée par l'ovariectomie. Après la période de exercise, les tiers moyens du fémur ont été recueillis, fixés et préparés pour une analyse en microscopie électronique. Résultats l'analyse morphologique de l'os spongieux montre que, par rapport à T, les os de la TO et des groupes de S ont montre une diminution marquée de l'épaisseur, et le montant des travées, tandis que dans le groupe S os spongieux a été complètement absent. Chez les animaux sédentaires, l'intérieur de l'os n'a pas d'os spongieux, contrairement aux rats formés. Cette enquête nous amène à croire que la résistance des os des rats exercitées est accrue, car en eux, peuvent être éliminées, avec une plus grande efficacité, les forces de traction, compression et torsion à laquelle l'os est soumis à des activités quotidiennes. Conclusion Natation favorise le maintien de l'architecture osseuse et peut être recommandée pour la prévention des fractures chez les individus souffrant d'ostéoporose.

MOTS-CLÉS: osseuse, l'ovariectomie, l'ostéoporose

# EFECTO DEL EJERCICIO SOBRE NATACION HUESO ESPONJOSO DE RATAS OVARIECTOMIZADAS RESUMEN

Introducción Ovariectomía caracteriza por la extirpación de los ovarios e induce a la menopausia temprana. Objetivo evaluar si el ejercicio de la natación puede ser indicado para la prevención y el control de la osteoporosis. Metodologia 20 ratas Wistar fueron separadas por grupos: control sedentario (S), el control de capacitación (T), ovariectomizadas sedentarias (OS) y entrenadas ovariectomizadas (TO). Las ratas del grupo T fueron sometidas a ejercicio de natación, con una carga de 5% del peso corporal durante 8 semanas. La inducción de la menopausia, 5 semanas antes del inicio del experimento fue realizada por la ovariectomía. Después del período de formación, el tercio medio del fémur fue tomado, fijas y preparadas para el análisis ultraestructural. Resultados el análisis morfológico del hueso esponjoso muestra que en comparación con T, los huesos de los grupos TO y S mostraron una marcada disminución en el espesor y la cantidad de trabéculas, mientras que en el grupo S hueso esponjoso estaba completamente ausente. En los animales sedentarios, el interior del hueso no muestra hueso esponjoso a diferencia de los animales entrenados. Este aspecto de los huesos de las ratas entrenadas sugiere que la resistencia es mayor, ya que en ellos, pueden ser eliminados con mayor eficiencia, las fuerzas de tensión, compresión y torsión a la que el hueso está sometido por las actividades diarias. Conclusión la natación promueve el mantenimiento de la arquitectura ósea y puede ser recomendado para la prevención de fracturas en personas con osteoporosis.

PALABRAS CLAVE: hueso, ovariectomía, la osteoporosis

### EFEITO DO EXERCÍCIO DE NATAÇÃO SOBRE O OSSO ESPONJOSO DE RATAS OVARIECTOMIZADAS RESUMO

Introdução A ovariectomia caracteriza-se pela retirada dos ovários e induz a menopausa precoce. Objetivo avaliar se o exercício de natação pode ser indicado na prevenção e controle da osteoporose. Metodologia 20 ratas Wistar foram separadas por grupos: sedentário controle (S), treinado controle (T), sedentário ovariectomizado (SO) e treinado ovariectomizado (TO). As ratas do grupo T, foram submetidas ao exercício de natação, com carga de 5% do peso corporal, durante 8 semanas. A indução da menopausa, 5 semanas antes do início do experimento, foi realizada através da ovariectomia. Após o período de treinamento o terço médio do fêmur foi coletado, fixado e preparado para análise ultra-estrutural. Resultados A análise morfológica do osso esponjoso mostra que quando comparado ao grupo T, os ossos dos grupos TO e S apresentaram marcante diminuição da espessura e da quantidade de trabéculas, sendo que nos animais do grupo S o osso esponjoso estava totalmente ausente. Nos animais sedentários, o interior do osso não apresenta osso esponjoso ao contrário das ratas treinadas. Este aspecto encontrado nos ossos das ratas treinadas nos leva a acreditar que a resistência está aumentada, já que neles, podem ser escoadas, com maior eficiência, as forças de tração, compressão e torção à qual o osso é submetido durante as atividades diárias. Conclusão A natação promove a manutenção da arquitetura óssea e pode ser preconizado para a prevenção de fraturas em indivíduos com osteoporose.

PALAVRAS CHAVE: osso, ovariectomia, osteoporose

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