

Studies Results Regarding Bisphosphonate Therapy in Dental Medicine

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Osteoporosis is a health problem reported worldwide, which predominantly affects women in menopause. Since the percentage level in this population category is quite high, a treatment plan was elaborated to minimize the negative effects of this bone disease, to restore the general state of comfort and allow women to reintegrate in society.

Keywords: bisphosphonates, osteonecrosis of the jaw, osteoporosis, creatinine, alkaline phosphatase

Osteoporosis is defined as a bone cell dysfunction, characterized by a reduction of bone mass and bone tissue integrity, leading to bone fragility and decreased bone hardness [1]. This bone cell dysfunction results in an increase of fracture risk. Estrogen deficiency is the main contributing factor in the formation of osteoporosis in women. The treatment of osteoporosis is bisphosphonate drugs, which by their action at the cellular level stops the action of osteoclasts and even improves the bone architecture [2]. Bisphosphonates represent the category of drugs most commonly used in the treatment of osteoporosis in postmenopausal women. The primary function of bisphosphonates is the direct inhibition of mineralization, thus inhibiting the action and activity of osteoclasts and stimulating the emergence of mature osteoclast cells [3-5]. All these processes prevent bone resorption. Bisphosphonate therapy for osteoporosis, osteoarthritis, Paget's disease, etc. have a number of benefits to the quality of life of the patient, stopping the evolution of localized or generalized bone degradation. The side effects of bisphosphonates with mouth-to-mouth localization are associated with loss of dental and even bony units. It occurs most often after a dental invasive (surgical) procedure such as dental extraction. Thus, in a routine check, the dentist must detect even dental bone changes in this group of patients and indicate, prior to the start of the treatment itself, the recovery of event dental focuses. The pathology of osteoporosis in postmenopausal women is at the intersection of various and diverse specialties such as medical oncology, hematology, oromaxillo-facial surgery, radiotherapy and general dentistry. Bisphosphonate therapy in this patient segment is a challenge for the general dentist, requiring interdisciplinary collaboration with results and benefits for the patient being treated.

Experimental part

Specialty literature abounds in information about generic bisphosphonates and how they are administered. However, as any drug obtained synthetically in pharmaceutical laboratories, presents both advantages and disadvantages. In an effort to bring novelty to this segment of pharmaceuticals, we conducted an animal model

experiment that yielded satisfactory results in terms of therapeutic indications and adverse effects of these drugs.

The present paper aims to expose an experimental comparison made on the animal model - females wistar mice - between two types of bisphosphonates (ibandronic acid and denosumab), in the conditions of osteoporosis induced by ovariectomy, extractions at the superior central incula and the occurrence of osteonecrosis of the jaw bone. The main goal in this experiment was the observation of the occurrence of maxillary bone necrosis as a secondary effect of the administration of bisphosphonates under conditions of dental extractions, and, also a retrospective study on patients with bisphosphonate therapy and the imaging results of mandibular osteonecrosis associated with bisphosphonate treatment.

The first experiment described in this article was carried out at the Biobase of UMPH Cluj-Napoca and in the experimental laboratories of USAMV Cluj-Napoca, according to the standards and directives of the Ethics Commission, in compliance with Law 43/2014 (published in Monitorul Oficial, Part I, no. 326, of 6 May 2014) regarding the protection of animals used for scientific purposes.

Study 1

Clinical implications of treatment with two types of bisphosphonates in dental medicine - an animal model experiment

The objectives of the study was an experimental comparison in an animal model between two types of bisphosphonates (ibandronic acid and denosumab), under the conditions of osteoporosis induced by ovariectomy, extraction of upper central incisors and development of osteonecrosis of the jaw. The aim of this experiment was to observe the development of necrosis of the jaw as a side effect of bisphosphonate administration under the conditions of tooth extractions.

This study was aimed at observing the development of osteonecrosis of the jaw under the conditions of tooth extractions in an animal model, on the background of treatment with two types of bisphosphonates. The first part of the study included 30 female Wistar rats, in which osteoporosis was induced by ovariectomy.

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After 3 months, of the 30 rats, only 24 were left, which were divided into three groups and were administered ibandronic acid (group 1), denosumab (group 2) and placebo (group 3). During the experiment, blood tests (creatinine and alkaline phosphatase) and radiological analyses were performed in order to monitor osteoporosis by stage, as well as the effects of the two types of bisphosphonates administered under the given conditions. Tooth extractions (extraction of the upper central incisor) were performed in each rat and the development of osteonecrosis of the jaw was observed based on the mentioned paraclinical examinations.

The histopathological study of the collected samples demonstrated the degree of osteonecrosis, bone resorption, the presence of mononuclear cell and neutrophil inflammatory infiltrate, the number of osteoclasts, the degree of fibrous connective tissue proliferation and local neoangiogenesis, which were very conclusive for the study concerned.

Study 2

Imaging results of mandibular osteonecrosis associated with bisphosphonate treatment

The objectives of the study was to present three cases with different stages of mandibular osteonecrosis developed on the background of bisphosphonate therapy in patients with cancer pathologies. Also a retrospective study was performed during the period 2010-2012 in patients with osteonecrosis of the jaw on the background of bisphosphonate therapy; obtaining statistical results regarding these data.

This was a retrospective clinical study performed at the Clinic of Oromaxillofacial Surgery II Cluj Napoca, in 22 patients (8 men and 14 women) with various stages of osteonecrosis of the jaw, in the period 2010-2012. Clinical signs showed necrotic bone exposure associated with pain and infection, extra-oral fistulas and extensive osteolysis up to the lower cortex. Imaging results evidenced thickening of the lamina dura, osteolysis, thickening of the mandibular canal, sequestered bone.

Results and discussions

Study 1

The use of bisphosphonates in the treatment of osteoporosis had a profound beneficial effect on the management of this pathological entity in postmenopausal women, and implicitly in the reduction of bone mass loss, minimizing the risk of fractures in this category of patients [6,7]. In the treatment of osteoporosis in postmenopausal women, bisphosphonates are elective in stopping osteoclast activity and implicitly in reducing bone turnover translated by rarely bone formation. Their administration i.v has both strengths and disadvantages [8-11]. According to our experiment, administration of bisphosphonates has antibacterial effects, decreased bone resorption and, implicitly, decreased osteoclast activity. This is beneficial to postmenopausal osteoporosis patients who have received ibandronic acid or denosumab under dental surgeries (dental extractions). It has been shown that denosumab with ibandronic acid has stronger antiseptic effects, and dental extractions in these situations can be successfully performed without the occurrence of septic complications or local bone-necrosis. Thus, denosumab (PROLIA) is a new generation bisphosphonate which, compared to ibandronic acid, has beneficial local and general effects, the risk of secondary effects and complications being less pronounced.

In our experiment on females rats induced osteoporosis by ovariectomy, they were divided into three groups and

then two types of bisphosphonates (ibandronic acid, denosumab) were administered, then dental extractions (extraction of the upper central incisor for each rat in each batch) thus wishing to observe the occurrence of bone necrosis at the level of the post-operative alveoli.

During the experiment, blood samples (PAL, Creatinine) and rat exposures were made. Thus, 3 months after the ovariectomy procedure of each rat, blood sampling and radiological sampling was performed for each rat in each batch. Following these preliminary tests, ibandronic acid (group 1), denosumab (group 2) and saline (control group 3) were administered. We waited for 7 days, and a set of blood tests was performed. At 20 days, a new set of blood and radiological tests was performed. At this stage, dental extractions were also carried out. 14 days after the extractions were performed, a new set of blood tests was performed, aiming at euthanasia of the rats for histopathological sampling

From the point of view of other similar experiments in the literature, speculations have also been made about the pathogenic role of certain strains of microorganisms such as Actinomycetes and the occurrence of bone osteonecrosis [12]. Another study on the pathogenesis of bone osteonecrosis associated with bisphosphonate treatment suggested that semiologically, soft bone tissue is affected. On this basis, the authors concluded that any bone injury caused by local surgical procedures or other local trauma results in the release of a certain amount of bisphosphonates in the periodontal tissues. This release of bisphosphonates is toxic to the local epithelium and ultimately results in inhibition of local cell proliferation and implicitly results in poor postoperative healing. Thus, these lesions provide an entry gate for oral microflora, which by local pathological action leads to bone infections and bone necrosis [13]. New generation bisphosphonates by the experiment on rats revealed that denosumab (Prolia) has local antibacterial effects over other bisphosphonates (ibandronic acid). Thus, the administration of the two types of bisphosphonates not only resulted in a decrease in bone marrow extraction, but also the thickening of the hard lamina with the formation of bone callus, a reduction in bone resorption, antibacterial effects in the dental extraction conditions and implicitly a reduction of the purulent material at the level postextractional alveoli, decreased osteoclast number. All these aspects compared to the control group that received only saline.

The two types of bisphosphonates (ibandronic acid, denosumab) administered under the conditions of induced osteoporosis cause changes in blood alkaline phosphatase and creatinine levels. From a radiological point of view, bone callus formation was observed particularly in the humerus, and bone necrosis in postextraction alveoli was also detected.

From a histopathological point of view, the occurrence of: bone necrosis (group 1,2) (fig. 1), large purulent material in group 3, versus 1 and 2, higher bone resorption gradient

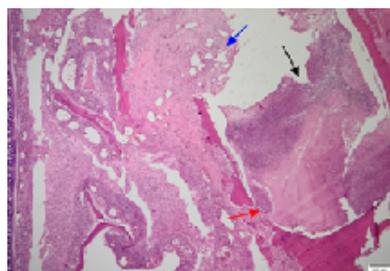


Fig. 1. Post-operative cavity - severe lesion with osteonecrosis Red arrow - severe bone resorption. The black arrow - purulent intraalveolar material. The blue arrow - moderate grain weave

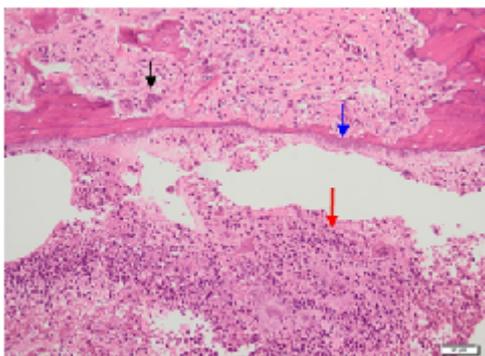


Fig. 2. Post-surgical cavity; Blue arrow - Severe bone resorption with the formation of Howship loopholes Black arrow - Increase in osteoclast number Red arrow - Increased number of degenerated neutrophils and serum exudate

at group 3 versus 1 and 2, proliferation of fibrous connective tissue more at lot 3 than 1 and 2, local neoangiogenesis (fig. 2) [14].

Thus, the administration of bisphosphonates not only caused the decrease of bone marrow extraction, but also the thickening of the hard lamina with the formation of bone callus, a reduction in bone resorption, antibacterial effects in the conditions of dental extraction and implicitly a reduction of the purulent material in the postextraction alveoli, decreasing the number of osteoclasts. All these aspects compared with the control group (group 3) who received only saline.

At the end of the period of development of osteoporosis, we divided the rats into three groups and we analyzed the blood collected from the infra-orbital fossa in order to monitor the fluctuating values of alkaline phosphatase and creatinine. The results obtained in this experiment clearly show that alkaline phosphatase and creatinine values play an exceptional role in the induction of osteoporosis by ovariectomy in female Wistar rats. The data obtained based on the blood tests for alkaline phosphatase (U/L) and creatinine (mg/dL) at 7, 14, 30 days in the three groups were centralized in tables (fig. 3,4). The ANOVA test was performed to compare values between the groups and see whether significant differences were present or not, and REGRESSION correlations were established, where R^2 showed correlations between the values obtained in each diagram.

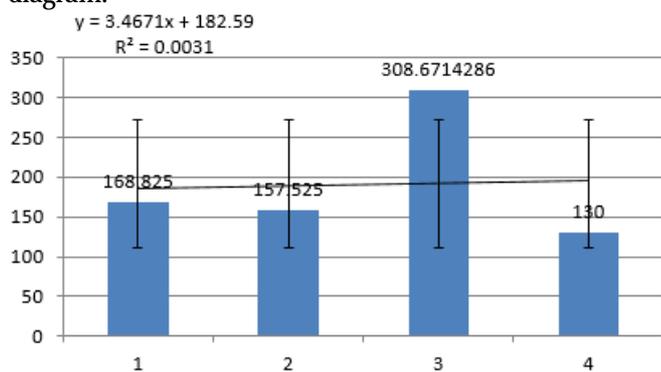


Fig. 3. Alkaline Phosphatase (PAL U / L) Comparison between the three batches with normal values over 30 days

This study provides preliminary observations of bone osteonecrosis in rats with induced osteoporosis receiving the two types of bisphosphonates and following the biological and blood parameters in these circumstances. Thus, rats have more accelerated metabolism than humans, and although we have noticed both beneficial and harmful action of these types of drugs, future studies should be done to determine their antibacterial effect. This

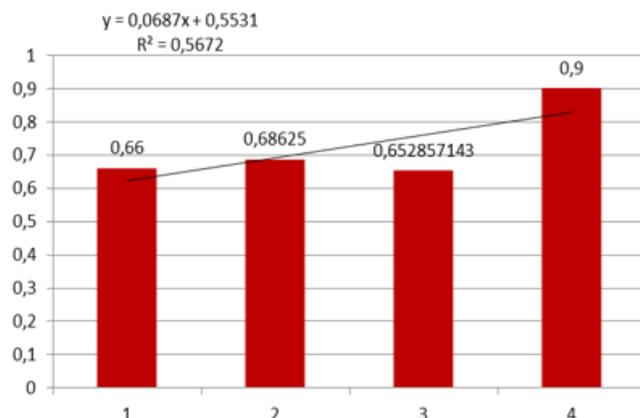


Fig. 4. Creatinine (mg / dL) Comparison of the three batches with normal values over 30 days

model could serve as a starting point for studies with a similar theme for accurately defining the mechanism of action of the two types of bisphosphonates and their antibacterial effects as well as potential pathways in therapeutic interventions.

Bisphosphonate treatment is the first-line and most frequently indicated treatment for osteoporosis in postmenopausal women. In addition to the major benefits of these pharmacological preparations, there is a possibility of development of osteonecrosis of the jaw under the conditions of tooth extractions.

Study 2

Radiological examination is the first choice for the early detection and diagnosis of osteonecrosis of the jaw in patients receiving or having completed bisphosphonate treatment, who require periodic monitoring. In spite of efforts to minimize the risk of occurrence and prevent, BONJ will continue to be diagnosed and will continue to be a challenge faced by topologists, oncologists and their patients. Desiring to investigate the underlying pathology and BONJ biology, based on epidemiological and interventional studies, a basic characterization of this pathology will be done through basic clinical, biochemical, and radiographic aspects. This being said, the incidence of BONJ in cancer patients is approximately 5% [15,16], precise documentation and retrospective reports data will continue to provide important data on this bone pathology. Mostly associated with oral infections and other complications, treatment with bisphosphonates requires an early and specialist therapeutic approach. Radiological examination is a first choice in the early detection and diagnosis of osteonecrosis of the jaw in patients requiring permanent supervision by the physician and dentist.

This retrospective study showed that the prevalence of mandibular osteonecrosis is higher in women than men; this is due to the hormonal factor, women being more affected in this regard. Also, we found in our study that the prevalence of osteonecrosis of the jaw depends on the age factor, the disease being more frequent between 52-59 and 73-80 years of age (fig. 5).

Intra-oral radiography, including retroalveolar, bitewing and occlusal projections, are the main and often the only imaging techniques needed for a precise diagnosis in dental pathology. Normally, they are reviewed and performed by the dentist generalist. The radiologist should be aware that intra-oral radiographs are performed without screen intensification, and this results in higher spatial resolution (20 pairs of lines per millimeter (lp / mm)) than panoramic radiographs (about 5 lp / mm) [17]. Larger spatial resolution allows detection of small carious lesions and periapical lesions, which usually can not always be

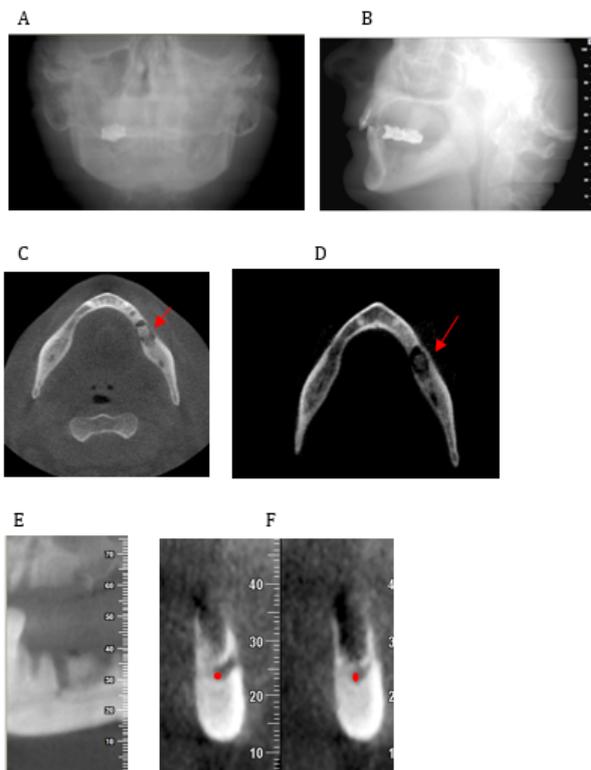


Fig. 5. Radiological results of BONJ located on the left side of the mandible arch. A. Radial skull exposition in frontal view
 B. Radiological exposure of the skull from the side view
 C. Lower mandibular arched axial section with structural modification of the trabecular bone from changes in the thickness and mineral content of the trabeculae until the formation of microlake; D. Localization of the necrotic bone site in the lateral left side of the mandible artery with cortical bone erosion, osteosclerosis and more than 15 mm bone seizure; E. Axial panoramic view of mandibular osteonecrosis
 F. Cross-sectional view, BONJ with mandibular canal (red point is the mandibular canal and the arrow indicates the location of osteonecrosis)

detected with panoramic dental x-rays. CBCT uses a coneshaped X-ray beam (as opposed to the fan-shaped X-ray beam used in conventional CT) to obtain projection data in a flat detector during a single 360° rotation, of which a set volumetric data is reconstructed using similar algorithms to those used in conventional CT. This results in a radiation dose lower than conventional CT, but suffers significant image alterations, and is not suitable for soft tissue assessment. CBCT is capable of higher spatial resolution (with isotropic voxels less than 0.125 mm³) than conventional CT. As with conventional CT, the volume data set can be used to create multiplanar images and three-dimensional reconstructions. CBCT units are generally cheaper and smaller in size than conventional CT scanners, and the patient's position is upright, similar to the position in a dental panoramic radiography unit. Thick multiplanar reconstructions can be used to produce lateral and frontal cephalometric images (without distortion or magnification) for orthodontic evaluation. Some current CBCT units are also capable of performing panoramic tomography and direct cephalometric projections, as used in our study.

A major drawback of conical beam tomography is the lack of standard imaging required for the analysis of the hard lamina [15], in our cases being a very important factor in the diagnosis and treatment of BONJ.

The main limitation we had in this study was that the underlying disease, the type of bisphosphonates

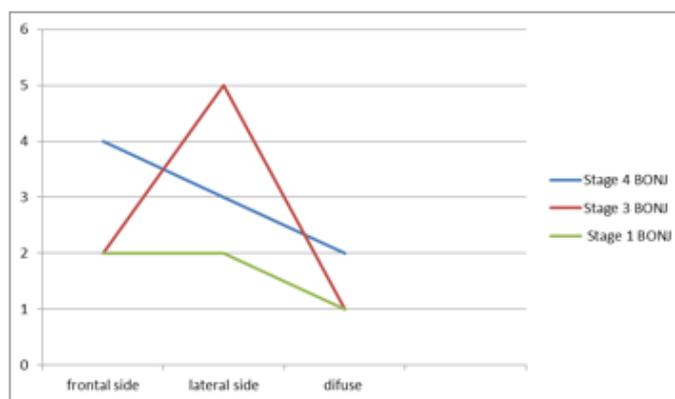


Fig. 6. Radiological findings on mandibular arc related to topographic changes and prevalence of osteonecrosis due to bisphosphonate treatment

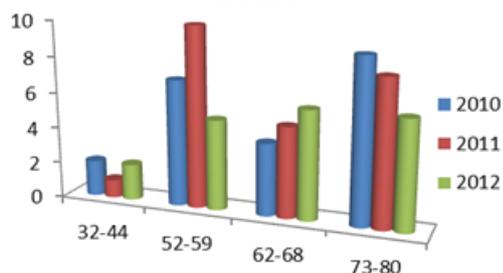


Fig. 7. Distribution by age groups and years of study

administered, the dose and duration of treatment were unknown. The study also lacked the consent of patients hospitalized, so we could not collect data from medical records to obtain such clinical information.

Conclusions

When choosing the type of bisphosphonates depending on the condition, always take into account the dose, the mode of administration and, above all, the potential negative effects. There was a relationship directly proportional to the size of the exposed bone surfaces and duration of treatment with bisphosphonates.

Literature studies have demonstrated both a correlation between treatment duration and bone destruction, and discontinuation of bone remodeling treatment. Even if the benefits of this therapy improve the quality of life of patients, we must always be cautious, because the sequelae of this therapy are serious and lasting. It is of major importance to educate and raise awareness among dentists, maxillofacial surgeons and oncologists about this issue.

The results obtained during these two experiments come to the help of dental practitioners who meet in current practice with patients with osteoporosis and not only who are in treatment or have completed bisphosphonate treatment and the appropriate dental treatment plan in these cases. Osteoporosis is defined as a bone cell dysfunction, characterized by a reduction of bone mass and bone tissue integrity, leading to bone fragility and decreased bone hardness. This bone cell dysfunction results in an increase of fracture risk. Estrogen deficiency is the main causative factor in the development of osteoporosis in women. Treatment of osteoporosis is based on bisphosphonate drugs; through their cellular action, they stop osteoclast activity and even improve bone architecture. Bisphosphonates are the most frequently used drugs in the treatment of osteoporosis in postmenopausal women. The main function of bisphosphonates is direct inhibition of mineralization, and thus, inhibition of osteoclast activity and stimulation of mature osteoclast development. Through all these processes, bone resorption is prevented.

Bisphosphonate therapy administered in the case of osteoporosis, bone metastasis, Paget's disease, etc. has a beneficial effect on the patient's quality of life, stopping localized or generalized bone damage. The side effects of bisphosphonates in the oral cavity are associated with tooth and even bone losses. Routine check-up performed by the dentist detects bone resorption of edentulous alveolar ridges without adequate prosthetic treatment, the presence of generalized periodontal disease and even its aggravation, as well as osteonecrosis of the jaw. This most frequently occurs after an invasive (surgical) dental procedure such as tooth extraction. Thus, during routine check-up, the dentist must detect potential dental bone changes in this group of patients and indicate, before starting treatment proper, the elimination of possible dental foci.

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