Ghizlane Chafai

The predicting factors of bisphosphonate-related osteonecrosis of the jaw

Universidade Fernando Pessoa

Faculdade de Ciências da Saúde

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Candidate signature

Abstract

Osteonecrosis of the jaw (ONJ) is a main adverse effect related to Bisphosphonates (BPs). Bisphosphonates are potent inhibitors of bone resorption and are most widely prescribed drugs for the treatment of osteoporosis, and are also used in malignant bone metastases, multiple myeloma, and Paget's disease. With increasing use, evidence is emerging that patients taking BPs are at risk of developing bisphosphonate related-osteonecrosis of the jaw (BRONJ), sometimes occurring spontaneously, but more commonly following dental extractions or oral bone surgery. However, the pathogenesis of BRONJ remains unknown. Attempts have been made to predict the development of BRONJ. The aim of this study is to review and to discuss if the biomarkers and the systematic markers of bone turnover can be used to predict the risk of BRONJ.

The methodology used in the accomplishment of this work was based on a bibliographic review for the subject under study.

Key-words: bisphosphonates, osteonecrosis of the jaw, CTX, systematic markers of bone turnover.

Resumo

Osteonecrose da mandíbula (ONJ) é um dos principais efeitos adversos relacionados com a utilização dos bifosfonatos (BPs). Os bifosfonatos são inibidores potentes da reabsorção óssea e são drogas amplamente prescritas para o tratamento da osteoporose, sendo também usadas nas metástases ósseas, mieloma múltiplo, e doença de Paget. Com o seu uso crescente, há uma crescente evidência de que os doentes que tomam BPs estão associados a um maior risco de desenvolver osteonecrose da mandíbula relacionada com bifosfonatos (BRONJ). Esta lesão surge, por vezes, de forma espontânea, ou, mais vulgarmente após extracção dentária ou cirurgia óssea oral. Apesar destas evidências, a patogénese da BRONJ permanece desconhecida. Diversos estudos têm tentado prever o desenvolvimento desta patologia.

O objetivo deste estudo é analisar e discutir se os biomarcadores e os marcadores sistémicos de renovação óssea podem ser usados para prever o risco de BRONJ.

A metodologia utilizada na realização deste trabalho foi baseada numa revisão bibliográfica sobre o tema em estudo.

Palavras-chave: bifosfonatos, osteonecrose da mandíbula, CTX, marcadores sistémicos de renovação óssea.

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Index of Abbreviations:

AAOMS	The American Association of Oral and Maxillofacial Surgeons
ALP	alkaline phosphatase
BAP	bone specific alkaline phosphatase
BPs	bisphosphonates
BRONJ	bisphosphonate-related osteonecrosis of the jaw
CI	confidence intervals
CTX	C-terminal telopeptide
DPD	deoxypyridinoline
ivBPs	intravenous bisphosphonates
LD	linkage disequilibrium
MMP2	matrix metalloproteinase 2
NTX	N-terminal telopeptide
ONJ	osteonecrosis of the jaw
OC	osteocalcin
OPG	osteoprotegerin
OPN	osteopontin
SNPs	single nucleotide polymorphisms
VEGF	vascular endothelial growth factor

I. Introduction

Bisphosphonate-related osteonecrosis of the jaw (BRONJ) has become one of the most prominent enigmas not only in the dental community but in the field of skeletal as a whole. Confusion surrounding BRONJ exists for several reasons, including a lack of understanding about how and why this condition manifests. (Allen *and* Burr, 2009).

The incidence of BRONJ in the USA report was 0.8 - 12% with intravenous BPs and 0.7/100.000 person-years of exposure with oral BPs. In Europe, the incidence was 95/100.000 person-years of exposure with intravenous administration and 1/100.000 person-years of exposure with oral administration. BRONJ occurred in 1.15% of intravenously administered patients and in 0.04% of the orally administered patients in Australia. Another report from the USA indicated that the incidence of BRONJ was 0.1% with oral BPs administration (Ikebe, 2013).

Bisphosphonates (BPs) are antiresorptive agents that have been used for more than a decade, mainly for the treatment of metabolic bone diseases, such as osteoporosis and osteopenia, and to control the skeletal complications associated with multiple myeloma and metastases of solid tumors to the bone (Glover *et al.*, 1994; Boissier *et al.*, 2000; Lazarovici *et al.*, 2010).

Intravenous BPs are primarily used and effective in the treatment and management of cancer-related conditions, including hypercalcemia of malignancy, skeletal-related events associated with bone metastases in the context of solid tumors such as breast cancer, prostate cancer, and lung cancer, and management of lytic lesions in the setting of multiple myeloma (Berenson *et al.*, 2002). Although BPs have not been shown to improve cancer-specific survival, they have had a significant positive effect on the quality of life for patients with advanced cancer involving the skeleton. Before 2001, pamidronate was the only drug approved in the United States for the treatment of metastatic bone disease. In 2002, zoledronic acid was approved for this indication by the US Food and Drug Administration (FDA). More recently, a once-yearly infusion of zoledronate and a parenteral formulation of ibandronate administered every 3 months have been approved by FDA for management of osteoporosis (Ruggiero *et al.*, 2009). Oral BPs are approved to treat osteoporosis and frequently used to treat osteopenia as well. They are also used for a variety of less common conditions such as Paget's disease of bone

and osteogenesis imperfecta of childhood. By far the most prevalent and common indication, however, is osteoporosis (Ruggeiro *et al.*, 2009).

BPs, which are potent inhibitors of osteoclast-mediated bone resorption, are associated with bisphosphonate-related osteonecrosis of the jaw (BRONJ).To date, the etiology and pathogenesis of BRONJ development remain unknown. It is hypothesized that long-term therapy may impair bone turnover and angiogenesis processes, predisposing to BRONJ development (Thumbigere-Math *et al.*, 2016).

The American Association of Oral and Maxillofacial Surgeons (AAOMS) published a position paper in 2007, with revision in 2009 (Ruggiero *et al.*, 2009) and 2014 (Ruggiero *et al.*, 2014), to address the need for clarity to diagnosis the BRONJ. The AAOMS suggest the following criteria to establish the diagnosis of BRONJ: 1) current or previous treatment with BPs; 2) exposed necrotic bone in the maxillofacial region that has persisted for more than 8 weeks; 3) and no history of radiation to the jaws. The 2014 AAOMS position paper update recommended a change of the term BRONJ to medication-related osteonecrosis of the jaw (MRONJ) to reflect the growing number of osteonecrosis cases associated with non-BP antiresorptive drugs and antiangiogenic therapies (Enciso *et al.*, 2016; Ruggiero *et al.*, 2014; Frietas *et al.*, 2016).

However, in 2015 the International Task Force on Osteonecrosis of the Jaw (Gavalda and Bagan, 2016) defined ONJ as follows:

- Exposed bone in the maxillofacial region that fails to heal in 8 weeks after identification by a health professional.
- Exposure to an antiresorptive agent.
- No history of craniofacial radiotherapy.

Patients taking BPs have risk factors associated with dental surgery involving the jaw, including tooth extraction, dental implant, and other alveolar surgeries, all of which can initiate osteonecrosis (Dal Pra *et al.*, 2017; Yamashita *et al.*, 2010).

Several cross-sectional studies have attempted to investigate that predictive value of bone turnover markers (eg, C-terminal telopeptide (CTX), N-terminal telopeptide (NTX),

osteocalcin (OC), and alkaline phosphatase (ALP) and angiogenesis markers (eg, VEGF) in BRONJ development (Thumbigere-Math *et al.*, 2016).

This study aims to review the available literature about the possible predictive factors and markers such as markers of bone turnover, angiogenesis, endocrine function, and inflammation for the development of bisphosphonate-related osteonecrosis of the jaw.

1. Materials and methods

Search Strategies: The Pub Med, Medline and B-on databases was electronically researched for articles published to February 2017 without date restrictions. The following keywords was using jointly or individually: Bisphosphonates, osteonecrosis, bisphosphonate-related osteonecrosis of the jaw, CTX, biomarkers of bone turnover. In additional, manual search was made by each one of the researchers.

Exclusion criteria: articles published in another language other than English, studies that the main topic was not the relation between osteonecrosis of the jaw and the bisphosphonates, full text articles were not available on the database, single case report, duplicated articles, and commentaries.

The initial search resulted in a list of 293 articles. In turn, titles were analyzed and based on exclusion criteria only 87 abstracts were included. After reading of the available abstracts, 45 articles were included in the study.

II. Development

1. Chemistry and Mechanism of Action of the Bisphosphonates

BPs are synthetic compounds with a chemical structure similar in that of inorganic pyrophosphate, and endogenous regulator of bone mineralization. While pyrophosphate is comprised of two phosphate groups linked by phosphoanhydride bonds (P-O-P structure), BPs are compromise of two phosphonate groups linked by phosphoether bonds (P-C-P structure). There are two main types of BPs: Nitrogen containing (aledronate, risedronate, pamidronate) and non-nitrogen containing (etidronate, tiludronate). The BPs act almost exclusively on bone when administered at physiological doses because of specific affinity to bone, where they deposit both in newly formed bone and in proximity to the osteoclasts. BPs act on bone through several mechanisms simultaneously. The first is exemplified by internalization by osteoclasts, causing disruption of osteoclast-mediated bone resorption, the second by inhibiting osteoclasts recruitment and accelerating programmed cell death (apoptosis) of osteoclasts, thus reducing osteoclast numbers. Both of mechanisms lead to reduction of bone and to a decrease in bone turnover (Gupta *et al.*, 2013). BPs are hydrophilic molecules thereby not passing directly through biological membranes. Evidence suggests that BPs are held in bone several years even after the drug therapy is discontinued. BPs bind tightly to hydroxiapatite crystals situated in bone surfaces and there remain for long time, since they are not susceptible to enzymatic degradation by osseous pyrophosphatase. BPs are constantly and gradually released from bone tissue into haematic circulation, entering in osteoclasts by endocytosis (Izzotti *et al.*, 2013).

2. Pathogenesis

There have been many pre-clinical and clinical studies on the pathophysiology of BRONJ, but the exact mechanism of why osteonecrosis occurs is under investigation. In particular, there are many theories being presented on why this type of osteonecrosis only occurs on the jaw and not in other areas. Several review articles propose a relationship to excessive of the jaw bone turnover, infection/inflammation, angiogenesis inhibition, soft tissue toxicity, the immune system, and accumulation of micro-fractures (Kim *et al.*, 2015; Allen *and* Burr, 2009).

2.1 Suppression of bone turnover

BPs inhibits the differentiation and promotes apoptosis of osteoclasts, so that the resorption and formation of bone is decreased (Russell *et al.*, 2008). Based on the action mechanisms of these medications, it had been reported that bone turnover plays an important role in osteonecrosis. The reason why osteonecrosis occurs in the jaw rather than in other long bones is explained by the strong suppression of the bone turnover in jaw after experimental BPs administration in preclinical study (Allen,. 2011), and more rapid cortical bone turnover in the human alveolar bone than in the long bones (Allen *et al*,. 2010). However, there is contradictory opinion based on the facts that bone turnover is not decreased in the ONJ lesions (Lesclous *et al.*, 2009), osteoclasts exist in the osteonecrotic areas, and that active bone resorption is occurring in these areas (Kim *et al.*, 2015).

2.2 Soft tissues toxicity

BPs act primarily on osteoclasts, they also have direct toxicity towards soft tissue such as oral epithelial cells (Kim *et al.*, 2015). In vitro, BPs were indeed shown to inhibit proliferation of endothelial cells and angiogenesis (Wood *et al.*, 2002), and furthermore, reduce the viability of periodontal ligament fibroblasts and macrophages (Vemeer *et al.*, 2016). BPs suppresses the proliferation and transportation of oral keratinocytes, which can increase the chances of latent bone exposure and subsequent infection (Yamashita and McCauley, 2012).

2.3 Infection/ Inflammation

Apparently, soft tissue toxicity and infection are two closely related phenomena possibly explaining the pathophysiology of ONJ. Unlike long bones, the jaw is constantly exposed to pathogens present in dental plaque, particularly the actinomyces species commonly found in ONJ (Hansen *et al.*, 2007). According to the study of (Silveira *et al.*, 2016) showed that zoledronic acid was associated with non vital bone and that there was no statistically significant association between the presence of microbial colonies and the presence of non-vital bone, they suggested that the presence of microbial colonies is not involved with osteonecrosis (Silveira *et al.*, 2016). Barros *and colleagues*, (2016) conclude that BRONJ is characterized by increases in empty osteocyte lacunae, osteoclast numbers, polymorphonuclear neutrophils, CD68 mononuclear-positive cells, immunostaining for TNF-

α, IL-1β, iNOS, and NF-Kb but not mast cells. BRONJ is also inversely associated with cells exhibiting IL-18 bp.

2.4 Angiogenesis inhibition

BPs have an angiogenic effect (Santini *et al.*, 2003). There are several reports about osteonecrosis of the jaw which happened after administration of anti-angiogenic agents; moreover, it is difficult to explain why the osteonecrosis develops in circulation-rich upper jaw rather other long bones (Kim *et al.*, 2015).

2.5 Immune-related or fracture-related theories

BPs controls the activity of various cells which involve in the immune response (Oizumi *et al.*, 2010). The risk of osteonecrosis after tooth extraction becomes significantly higher if steroids or chemotherapeutic agents, which may influence the innate/acquired immune system, are given during bisphosphonates administration (Thumbierger-Math *et al.*, 2012). Bone tissue is constantly undergoing the repetitive micro-fractures and healing process throughout the life, and such micro-trauma is slowly accumulated by age, and due to the suppressive effect of bisphosphonates on osteoclasts or osteoblasts, resulting in latent osteonecrosis lesions (Kim *et al.*, 2015). Where high concentrations of BPs are present, the remodelling is inhibited; with a consequent increase is bone density (Rosini *et al.*, 2015). Therefore, BRONJ is probably caused by multiple, combined factors that cannot be explained by single path physiologic mechanism.

3. Biomarkers for predicting BRONJ

3.1 Bone turnover markers (CTX, NTX, bone-specific alkaline phosphatase (BAP))

Marx *and colleagues,* (2007) took 30 consecutive cases with presence of no healing expose bone in maxilla or mandible that has persisted for more than 8 weeks in a patient who has received a systemic bisphosphonates but has not received local radiation therapy. Each included patient was asked for detailed history concerning the BPs: dose, the frequency, the duration of the BPs therapy, co morbidities and indication for the BPs. Results in part noted a higher incidence related to alendronate, 94.7% predilection for the posterior mandible and a 50% occurrence spontaneously, with the remaining 50% resulting from an oral surgical

procedure, mostly tooth removals. Just over 53% of patients were taking their oral BPs for osteopenia, 33.3% for documented osteoporosis, and 13.4% for steroid-induced osteoporosis related to 4 or more years of prednisone therapy for an autoimmune condition. There was a direct exponential relationship between the size of the exposed bone and the duration of oral BPs use. There was also a direct correlation between reports of pain and clinical evidence of infection. The morning fasting serum C-terminal telopeptide (CTX) test results were observed to correlate to the duration of oral BPs use and could indicate recovery of bone remodelling with increased values if the oral BPs was discontinued. A stratification of relative risk was seen as CTX values less than 100 pg/ml representing high risk, CTX values between 100 pg/ml and 150 pg/ml representing moderate risk, and CTX values above 150 pg/ml representing minimal risk. The CTX values were noted to increase between 25.9 pg/ml and 26.4 pg/ml for each month of a drug holiday indicating a recovery of bone remodelling and a guideline as to when oral surgical procedures can be accomplished with the least risk. In addition, drug holidays associated with CTX values rising above the 150 pg/ml threshold were observed to correlate to either spontaneous bone healing or a complete healing response after an office-based debridement procedure.

Kunchur *and colleagues*,(2009) reported that of 348 patients, 215 were taking oral BPs alendronate (63.5%) and taking long-term oral BPs were older (71 ± 11.6 years), were predominantly women with osteoporosis, and were medically compromised. The average CTX value was 238 ± 144 pg/ml, with 98 having a value less than 200 pg/ml. One patient with a CTX value of 126 pg/ml developed ONJ after an extraction. Seven intravenous BPs patients underwent extractions with no cases of ONJ developing. The CTX value was 329 ±354, with 4 less than 200 pg/ml. Fifteen patients developed ONJ, 12 after extractions and 3 spontaneously. Of these, 7, who were still taking a BPs at presentation, had a CTX value of 116 pg/ml. A CTX value of less than 150 pg/ml did not correlate with the clinical risk factors of age, gender, comorbidities, bone disease, or BPs duration. A statistically significant difference in the CTX value was found for those taking alendronate compared with those taking risedronate ($p < 0.0001$). If the BPs was ceased, the CTX value increased at approximately 25 pg/ml per month. The CTX test is not predictive of the development of ONJ for an individual patient but does identify those in the "risk zone," which is a value of less than 150 pg/ml to 200 pg/ml. If medically appropriate, the BPs can be ceased so that the CTX value increases to bring the patient out of the "risk zone."

Lazarovici *and colleagues*, (2010) investigated the predictive value of serum levels of CTX, bone specific alkaline phosphatase (BAP), and parathyroid hormone (PTH) for the development of BRONJ. The Data on the demographics, comorbidities, and BPs treatment were collected from 78 patients scheduled for dentoalveolar surgery. Of the 78 patients, 51 had been treated with oral BPs and 27 had been treated with frequent intravenous infusions of BPs. Blood samples for CTX, BAP, and PTH measurements were taken preoperatively. Surgery was performed conservatively, and antibiotic medications were prescribed for 7 days. Of the 78 patients, 4 patients taking oral BPs (7.8%) and 14 receiving intravenous BPs (51.8%) developed BRONJ. A CTX levels less than 150 pg/ml was significantly associated with BRONJ development, with an increased odds ratio of 5.268 (p = 0.004). The BAP levels were significantly lower in patients taking oral BPs who developed BRONJ. The PTH levels were similar in patients who did and did not develop BRONJ. They found that the CTX serum level is not a definitive predictor factor for the development of BRONJ; it might have an important role in the risk assessment before oral surgery. Serum BAP levels might beneficial, but their true value awaits additional investigation. PTH does not appear to have any predictive value for the development of BRONJ.

O'connell *and colleagues*, (2012) evaluated 23 patients underwent a morning fasting CTX test. 19 patients were taking alendronic acid orally, 2 risedronate sodium orally and 2 zoledronic acid intravenously. The indications for BPs were as follow: osteoporosis, multiple myeloma, osteopenia, breast cancer. The mean duration of BPs treatment was 30 months. The mean CTX value at or less than 180 pg/ml, with 11 patients having a value at or less than150 pg/ml. The mean follow-up period was 5 months. None of the patients, who underwent removal of one or more teeth, subsequently developed BRONJ. They concluded that the CTX test was not predictive for the development of BRONJ following oral surgery.

Hutcheson *and colleagues*, (2014) reported the value of CTX test at the moment of the extraction and after extraction in cases of BRONJ in a large prospective cohort. All patients took an oral BPs for osteoporosis and had extractions from January 2007 to June 2013. 950 patients on oral BPs for osteoporosis had 2,461 extractions performed under local anesthesia.181 patients had a CTX level lower than 150pg/ml. Four patients developed BRONJ, all had a CTX level lower than 150pg/ml. All were older than 70 years, and were on alendronate, and had medical comorbidities, but were not immunocompromised. The case – control comparison approached significance (< 150 pg/ml; p= 0.073). Alendronate was

associated with a low CTX level ($p< 0.05$). A CTX levels lower than 150pg/ml had a sensitivity of 100% and specificity of 81%. They conclude that the risk of BRONJ for patients with osteoporosis on BPs having extractions is approximately 0.2%. A CTX level lower than 150 pg/ml, is sensitive and is associated with an approximately 3-fold greater risk of BRONJ.

As a biomarker of bone resorption, N-telopeptide of type I collagen (NTX) is responsible for cross-linking of the bone matrix and bone-specific alkaline phosphatase (BAP) is a highly specific isoenzyme in bone and reflects ostoeblastic cellular activity.

Morris *and colleagues*, (2012) investigated N-telopeptide of type I collagen (NTX) and bone-specific alkaline phosphatase (BAP) as potential predictors of ONJ onset. The Patients with ONJ were identified from a prospectively maintained database from the Dental Service at Memorial Sloan-Kettering Cancer Centre. Therefore, the medical records were used to identify possible stored serum from up to 4 approximate points: 12 months before ONJ, 6 months before ONJ, 1 month before ONJ, and at the ONJ diagnosis. Using commercially available enzyme-linked immunosorbent assays, the available serum was used to measure both NTX (Osteomark; normal range 5.5 to 19.5 nmol/L BCE) and BAP (Quidel; normal range 14.2 to 42.7U/L). The absolute values and trends in NTX and BAP over time were examined as possible predictors for ONJ. From March 1998 to September 2009, they identified 122 patients with ONJ. Of these, 56 (46%) had one or more serum samples available. Overall, 55 patients (98%) received BPs. Using the exact dates; no obvious patterns in either NTX or BAP were noted. Similarly, using the ordinal points, no evidence of suppression of NTX or BAP over time was seen. The consecutive median values were as follows: The median NTX values were 8.0 nmol/L (range, 3.8 - 32.9) at 12 months before ONJ; 9.5 nmol/L (range, 4.7 - 42.7) at 6 months; 9.5 nmol/L (range, 4.5 - 24.6) at 1 month, and 10.4 nmol/L (range, 4.4 - 32.5) at the ONJ diagnosis. The median BAP values were 18.0 U/L (range, 7.0 - 74) at 12 months before ONJ; 18.0 U/L (range, 4.0 - 134) at 6 months; 14.0 U/L (range, 4.0 - 132) at 1 month, and 18.0 U/L (range, 0.7 - 375) at the ONJ diagnosis. Only 2 patients (4%) had NTX and 17 (30%) had BAP below the normal range at the ONJ diagnosis. They had seen no trend in the NTX and the BAP levels before the ONJ diagnosis.

Kim *and colleagues*, (2013) investigate in the possible associations of the bone biomarkers osteocalcin (OC), deoxypyridinoline (DPD), CTX, NTX, BAP, PTH and BRONJ development. 37 patients had at least 1 sample available at the time of BRONJ diagnosis and

were included in the present study (age, 73.6 ± 11.2 years, 3 men and 34 women). Then, 37 age- and gender-matched patients composed the control group. The patient's baseline characteristics Patient's personal information and type of BPs taken, dose, dosage instructions, duration of medication use, and indication were recorded. Through an examination, the location and size of the exposed necrotic bone, the presence of infection and pain, and the extension of lesions were recorded. Possible comorbidities, including patient-related factors (Diabetes, Obesity, and renal failure) and iatrogenic factors (steroid use, chemotherapy), were recorded. Of the 37 patients in the BRONJ group, 35 were taking BPs for osteoporosis and 2 patients for bone metastasis. Two patients had a history of chemotherapy use, 8 patients had been using steroid, and 6 patients had a diagnosis of diabetes. The median duration of BP exposure was 73 months (range, 21 – 87 months) for the BRONJ group and 67 months (range, 27 – 94 months) for the control group ($p > 0.05$). The levels of the bone formation markers serum OC and serum BAP, and those of the bone degradation markers urine DPD, urine NTX, and serum CTX were not significantly different between the 2 groups; only levels of serum PTH showed a significant difference ($p < 0.05$). The OC, DPD, CTX, NTX and BAP levels were not significant between the 2 groups ($p > 0.05$). The serum CTX level in reference to a 150 pg/ml cut-off was also not significant for the development of BRONJ ($p > 0.05$). They did not find evidence for the risk prediction of BRONJ of those current bone biomarkers.

3.2 Angiogenesis Markers

Vincenzi *and colleagues*, (2012) evaluated the role of vascular endothelial growth factor (VEGF) as predictive marker of BRONJ. Of 81 patients, 6 (7.4%) developed BRONJ following intravenous BPs administration (1 under treatment with pamidronate, 3 under treatment with zoledronic acid on standard schedule and 2 under treatment with zoledronic acid on metronomic administration). These 6 patients were affected by breast cancer (n = 3), prostate cancer (n = 2) and renal cancer (n = 1). The median number of BPs administrations was 9 in BRONJ group vs. 11 in non-BRONJ group ($p = 0.196$) and the median time of BRONJ development was 9 months (range 6 - 22). Compared to the patients who didn't develop BRONJ, these 6 patients showed the strongest decrease in VEGF circulating levels at day 7 (median = 540.81, confidence intervals (CI) 95% 129.35-634.64 vs. median = 788.55, CI 95% 728.96-870.44, $p < 0.0001$) and at day 21 (median = 458.00, CI 95% 369.05 - 601.29 vs. median = 710.81, CI 95% 638.66 - 955.53, $P < 0.0001$) after the first administration of

either zoledronic acid or pamidronate. The detection limit of the VEGF was 62.5pg/ml. If confirmed, serum VEGF levels at day 7 and 21 after the first administration of intravenous BPs could represent an effective early predictive marker of BRONJ.

3.3 Other markers

Mücke *and colleagues*, (2016) in their study examined the value of preoperative measurements of inflammatory mediators in blood in 212 patients with BRONJ who were studied prospectively. Multiple logistic regression analysis was used to assess the importance of the amounts of substance in the blood that are independently associated with the dependent variable recurrence of BRONJ. The only factor that significantly influenced the development of recurrence BRONJ was reduction in the white cell count ($p < 0.0001$, hazard ration 5.324; 95% CI 2.373 to 11.945). Neither white cell counts nor C-reactive protein concentration within or above the reference ranges were significant associated with recurrence BRONJ. Patients whose white cell counts were lower than the reference range were at increased risk of recurrence BRONJ.

Thumbigere-Math *and colleagues*, (2016) analyzed serum markers of bone turnover, angiogenesis, endocrine function, and inflammation in patients with BRONJ who discontinued long-term intravenous BPs therapy. Serum samples were obtained from 25 BRONJ patients who had discontinued long-term ivBPs for an average of 11.4 ± 8.7 months and 48 non-BRONJ controls who continued receiving ivBPs therapy. Samples were analyzed for total alkaline phosphatase, bone-specific alkaline phosphatase, osteocalcin, C-telopeptide, vascular endothelial growth factor, triiodothyronine, thyroxine, thyroid-stimulating hormone, 25-hydroxuvitamin D, and C-reactive protein. They compared serum markers between a subgroup of BRONJ patients (n= 10) and controls (n= 48) whom they closely matched for mean age (64 years) and number of BPs infusions (mean, 19 infusions). Within this well-matched subgroup, and consistent with the unmatched results, they found no significant differences in levels of bone turnover or endocrine markers between BRONJ patients and controls (table I). However, $\log_{10}$ VEGF (2.92 ± 0.38 pg/ml vs. 2.41 ± 0.35 pg/ml, $p< 0.001$) and CRP (34 ± 26 mg/l vs. 13 ± 8 mg/l, $p <0.01$) levels were significantly higher in BRONJ patients compared with controls. Of the 10 BRONJ patients in the subgroup analysis, 5 had discontinued BPs for 6 months or less (range, 2 to 6 months) and 5 had discontinued BPs for more than 6 months (range, 7 to 22 months). Both in this subset of BRONJ patients and the

entire BRONJ group, none of the serum markers were correlated with the duration of intravenous BP discontinuation.

Table I. Serum biochemical markers matched according to mean age and number of bisphosphonate, (Adapted from Thumbigere-Math *et al*., 2016).

Mean or Mean ±SD

Characteristics	BRONJ (n=10)	Non BRONJ(n=48)	Reference Range	P Value*
Age, yr	64.4	64.4	—	>.99
No. of BP infusions	19 ± 8	19 ± 7	—	0.92
BP discontinuation, mo	9	—	—	—
Biochemical markers				
Bone formation				
Total alkaline phosphatase ,U/L	101 ± 52	85 ± 76	40-150	0.54
Bone-specific alkaline phosphatase, mg/L	12 ± 6	14 ± 8	6.5-22	0.47
Osteocalcin, ng/mL	13 ± 4	14 ± 10	11-50	0.56
Bone resorption: C-telopeptide, pg/mL	271 ± 167	222 ± 119	50-580	*0.27*
Angiogenesis: log10 vascular endothelial growth factor, pg/mL	2.92 ± 0.38	2.41 ± 0.35	1.78-2.85	<0.001
Endocrine				
Triiodothyronine, ng/dL	120 ± 26	110 ± 30	60 ± 180	0.33
Thyroxine, mg/dL	10 ± 2	9 ± 2	5 ± 11	0.40
Thyroid-stimulating hormone, mIU/mL	2 ± 1	2 ± 2	0.4-3.0	0.29
25-hydroxyvitamin D, mg/L	33 ± 14	31 ± 10	30-75	0.81
Inflammation: C-reactive protein, mg/L	34 ± 26	13 ± 8	0-10	<0.01

* P values were calculated from 2-sample *t* tests.

Choi *and colleagues*, (2015) recruited 45 individuals (2 men, 43 women; age: 68.7± 12.3). All visited the Yonsei University Dental Hospital for surgical intervention from January 2012 to January 2013 and had a history of BP prescription (oral and/or intravenous). The following inclusion criteria were employed: Case group: received BPs; none healing, exposed bone in the maxilla or mandible that had persisted for more than 8 weeks; no history of local radiation therapy. Control group: received BPs; history of surgical dental intervention including extraction, implant placement, and periodontal surgery involving osseous injury; no none-healing exposed bone in the maxilla or mandible that had persisted for more than 8 weeks. Exclusion criteria for their study were as follows: uncontrolled systemic disease; uncontrolled hemorrhagic disease; psychiatric disorder; pregnancy; and any factor that compromised the

appropriateness of a patient for the study, according to the medical judgment of the investigators. As a result, 26 individuals diagnosed with BRONJ were designated cases, and 19 without BRONJ were designated controls. They analyzed three VEGF single nucleotide polymorphisms (SNPs) rs699947 (in promoter region), rs2010963 (in the 5' -UTR), and *rs3025039* (in the 3' UTR) to investigate the association between VEGF polymorphisms and BRONJ. The genotype permutation test showed all genotypes were within the Hardy −Weinberg equilibrium (P>0.05). Of all three SNPs, rs2010963 and rs3025039 differed significantly between cases and controls. The frequency of genotype CC in rs2010963 and rs3025039 was significantly different between cases and controls (P= 0.04, P= 0.03, respectively) when multiple logistic regression analysis was applied. The rs699947 and rs2010963 polymorphisms were in tight linkage disequilibrium (LD), while the rs3025039 polymorphism was only weakly associated with the other polymorphisms. Haplotypes were reconstructed using LD coefficient values, and their frequencies in cases and controls were compared. The frequency of haplotype C-C (-2578/ 634) and C-G (-2578/-634) was found to show no statistical difference between case and control. They also selected extended haplotypes, predicted using genotype data, for analysis they excluded haplotype C-C-T (-2578/-634/+936) due to a frequency <3%. Extended VEGF haplotype C-G-T (-2578/-634/ + 936) was inversely associated with BRONJ, more common among controls than cases (P=0.039). By contrast, haplotype C-C-C (-2578/-634/ + 936) was more common among cases (P=0.177), though the difference was not statistically significant. The CC homozygotes of rs2010963 and rs3025039 polymorphisms in the VEGF gene were associated with an increased risk of BRONJ in the Korean population.

III. Discussion

The morbidity of BRONJ, its high incidence among patients receiving frequent intravenous infusions of BPs, and the large number of patients using oral BPs has spurred a vigorous search for predictive and prognostic factors for the development of BRONJ (Marx *et al*,. 2007; Kunchur *et al*., 2009). This bibliographic review evaluated the efficacy of the CTX test, biomarkers of bone suppression and formation, angiogenesis markers, inflammation markers, endocrine hormones, as predictors of the development of BRONJ. Despite the initial work of Marx and colleagues, (2007) the cut-off values for the CTX is not established till today, no consensus in the literature was been shown. Some works confirmed that the serum fasting value below 150 pg/ml was associated to high risk of BRONJ (Hutcheson *et al*., 2014;

Lazarovici *et al.*, 2010; Kunchur *et al.*, 2009), other studies was controversial and reported that CTX test did not predict the development of BRONJ (Bagan. 2008; Baim *and* Miller. 2009; O'connell *et al.*, 2012; Thumbigere-Math *et al.*, 2016; Dal Pra *et al.*, 2017). For those reasons the American Society for Bone and Mineral Research Task Force on BRONJ and American Dental Association, have not endorsed CTX test findings as a predictor for BRONJ development (Schwartz. 2008; Thumbigere-Math *et al.*, 2016). Finally, the validity of CTX test as a predictor of BRONJ must be explored in future studies. Due of the relatively low incidence of BRONJ among patients taking oral BPs, more cohort study should be conduct.

For the other biomarkers NTX, BAP, OC, DPD, PTH, no consensus in the literature was found regarding on the use of biomarkers levels in clinical practice, with conflicting and controversial findings reported. The small number of cases studied in the published series and the dispersion of the results do not make possible to establish a relation between articles, to date. No trends were seen in the NTX and BAP levels before the ONJ diagnosis (Morris *et al.*, 2012). Kim *and colleagues*, (2013) investigated in the possible associations of the bone biomarkers OC, DPD, NTX, BAP, PTH and BRONJ development, they did not find evidence for the risk prediction of BRONJ of those current bone biomarkers. Thumbigere-Math and colleagues, 2016 do not find significant difference in mean BAP, OC levels between BRONJ patients and controls.

The role of VEGF as predictive angiogenic marker in BRONJ development was not concluding, Vincenzi *and colleagues*, (2012) in their study showed a significant decrease in serum VEGF levels at day 7 and day 21 after the first BPs administration, but they didn't have any control group. Thumbigere-Math *and colleagues*, (2016) didn't find significant difference in the serum VEGF levels between BRONJ patients who discontinued long-term ivBPs therapy and controls. More prospective and extensive studies should be done.

The endocrine markers like PTH, triiodothyronine, thyroxine, and thyroid - stimulating hormone didn't have a significant association with the BRONJ development. Thumbigere-Math *and colleagues*, (2016) related in their study that the levels of bone turnover and endocrine markers in BRONJ patients who discontinue long-term intravenous BP therapy are similar to those in non-BRONJ controls receiving intravenous BP therapy. Further studies containing a greater number of patients are needed to prove the utility of those markers in the prediction of the BRONJ.

Recently there have been several cases reports indicating that genetic variation is strongly associated with BRONJ risk. Genetic variation between individuals may increase or decrease the BRONJ susceptibility. Sarasquete *and colleagues*, (2008) were the first to report an association between cytochrome P450-2C (CYP2C8) polymorphisms and ONJ. Katz *and colleagues*, (2011) identified a potential association between a combined COL1A1, RANK, matrix metalloproteinase 2 (MMP2), osteoprotegerin (OPG), and osteopontin (OPN) genotype score and BRONJ risk in multiple myeloma patients undergoing ivBPs therapy. Choi *and colleagues*, (2015) found that the CC homozygote of rs2010963 and rs3025039 polymorphism in the VEGF gene were associated with an increased risk of BRONJ in the Korean population. However, as genetic variation among different ethnic groups may have different effects, it is worthwhile conducting genetic association studies on the different ethnic groups to which future genetic diagnostic methods will be applied. Therefore, additional epidemiological cohort studies with larger sample size and functional biological research would be required. The genetic test might be helpful to predict the BRONJ but more investigations should be done.

IV. Conclusions

This bibliographic review finds the follow conclusions:

- The CTX test has no predictive ability to detect the risk of BRONJ.
- The bone turnover biomarkers (BAP, OC, NTX, DPD), endocrine markers (thyroid hormones and PTH), inflammation marker (C- reactive protein) were unable to establish an evident association with BRONJ.
- The angiogenesis marker (VEGF) could represent an effective early predictive marker of BRONJ but more studies should be done.
- The genetic tests may be useful as a diagnostic tool to predict the BRONJ; additional investigations are needed to explore the real potential of those markers and their impact for the clinical practice.

V. References

Allen, M.R. (2011). The Effects of Bisphosphonates on Jaw Bone Remodeling, Tissue Properties, and Extraction Healing. *Odontology*, 99 (1), pp. 8–17.

Allen, M.R. e Burr, D.B. (2009). The Pathogenesis of Bisphosphonate-Related Osteonecrosis of the Jaw: So Many Hypotheses, so Few Data. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 6, 5 Suppl, pp. 61–70.

Allen, M.R. *et al*. (2010). Cancer Treatment Dosing Regimens of Zoledronic Acid Result in Near-Complete Suppression of Mandible Intracortical Bone Remodeling in Beagle Dogs. *Journal of Bone and Mineral Research: The Official Journal of the American Society for Bone and Mineral Research* 25 (1), pp. 98–105.

Bagan, J.V. (2008). Collagen Telopeptide (Serum CTX) and Its Relationship with the Size and Number of Lesions in Osteonecrosis of the Jaws in Cancer Patients on Intravenous Bisphosphonates. *Oral Oncology*, 44 (11), pp. 1088–1089.

Baim, S. e Miller, P. (2009). Assessing the Clinical Utility of Serum CTX in Postmenopausal Osteoporosis and Its Use in Predicting Risk of Osteonecrosis of the Jaw. *Journal of Bone and Mineral Research: The Official Journal of the American Society for Bone and Mineral Research*, 24 (4), pp. 561–574.

Barros Silva, P.g. *et al*. (2016). Immune Cellular Profile of Bisphosphonate-Related Osteonecrosis of the Jaw. *Oral Diseases*, 22 (7), pp. 649–657.

Berenson, J.R. *et al*. (2002). American Society of Clinical Oncology Clinical Practice Guidelines: The Role of Bisphosphonates in Multiple Myeloma. *Journal of Clinical Oncology: Official Journal of the American Society of Clinical Oncology*, 20 (17), pp. 3719–3736.

Boissier, S. *et al*. (2000). Bisphosphonates Inhibit Breast and Prostate Carcinoma Cell Invasion, an Early Event in the Formation of Bone Metastases. *Cancer Research* 60 (11), pp.2949–2954.

Choi, H. *et al*. (2015). Genetic Association between VEGF Polymorphisms and BRONJ in the Korean Population. *Oral Diseases*, 21 (7), pp. 866–871.

Dal Prá, K.j. *et al*. (2017). Systematic Review: Efficacy of the C-Terminal Telopeptide Test in Predicting the Development of Bisphosphonate-Related Osteonecrosis of the Jaw: A Systematic Review. *International Journal of Oral & Maxillofacial Surgery* , 46 (February), pp.151–156.

Enciso, R. *et al*. (2016). Assessing the Utility of Serum C-Telopeptide Cross-Link of Type 1 Collagen as a Predictor of Bisphosphonate-Related Osteonecrosis of the Jaw: A Systematic Review and Meta-Analysis. *Journal of the American Dental Association*, 147 (7), pp. 551–560.

Freitas, N.R. *et al*. (2016). Bisphosphonate Treatment and Dental Implants: A Systematic Review. *Medicina Oral, Patologia Oral Y Cirugia Bucal* , 21 (5), pp.644-651.

Gavaldá, C. e Bagan, J. (2016). Concept, Diagnosis and Classification of Bisphosphonate-Associated Osteonecrosis of the Jaws. A Review of the Literature. *Medicina Oral, Patología Oral y Cirugía Bucal*, 21 (3), pp. 260–270.

Glover, D. *et al*. (1994). Intravenous Pamidronate Disodium Treatment of Bone Metastases in Patients with Breast Cancer. A Dose-Seeking Study. *Cancer*, 74 (11), pp. 2949–2955.

Gupta , *et al*. (2013). Bisphophonates Related Osteonecrosis of the Jaw. *National Journal of Maxillofacial Surgery*, 4 (2), pp. 151

Hansen, *et al*. (2007). Actinomycosis of the Jaws--Histopathological Study of 45 Patients Shows Significant Involvement in Bisphosphonate-Associated Osteonecrosis and Infected Osteoradionecrosis." *Virchows Archiv: An International Journal of Pathology*, 451 (6), pp.1009–1017.

Hutcheson, *et al*. (2014). A C-Terminal Crosslinking Telopeptide Test-Based Protocol for Patients on Oral Bisphosphonates Requiring Extraction: A Prospective Single-Center Controlled Study. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 72 (8), pp. 1456–1462.

Ikebe,T. (2013). Pathophysiology of BRONJ: Drug-Related Osteoclastic Disease of the Jaw. *Oral Science International*, 10 (1), pp. 1–8.

Izzotti, A. *et al*. (2013). Biphosphonates-Associated Osteonecrosis of the Jaw: The Role of Gene-Environment Interaction. *Journal of Preventive Medicine and Hygiene*, 54 (3), pp. 138–145.

Katz, J. *et al*. (2011). Genetic Polymorphisms and Other Risk Factors Associated with Bisphosphonate Induced Osteonecrosis of the Jaw.*International Journal of Oral and Maxillofacial Surgery*, 40 (6), pp. 605–611.

Kim, Jin-Woo. *et al*. (2013). Prospective Biomarker Evaluation in Patients with Osteonecrosis of the Jaw Who Received Bisphosphonates. *Bone*, 57 (1), pp. 201–205.

Kim, Kyoung Min. *et al*. (2015). Medication Related Osteonecrosis of the Jaw: 2015 Position Statement of the Korean Society for Bone and Mineral Research and the Korean Association of Oral and Maxillofacial Surgeons. *Journal of Bone Metabolism*, 22 (4), pp. 151–165.

Kunchur,. *et al*. (2009). Clinical Investigation of C-Terminal Cross-Linking Telopeptide Test in Prevention and Management of Bisphosphonate-Associated Osteonecrosis of the Jaws. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 67 (6), pp. 1167–1173.

Lazarovici,. *et al*. (2010). Serologic Bone Markers for Predicting Development of Osteonecrosis of the Jaw in Patients Receiving Bisphosphonates. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 68 (9), pp.2241–2247.

Lesclous, P. *et al*. (2009). Bisphosphonate-Associated Osteonecrosis of the Jaw: A Key Role of Inflammation? *Bone*, 45 (5), pp. 843–852.

Marx, R. E. Cillo,J. e Ulloa, J.J. (2007). Oral Bisphosphonate-Induced Osteonecrosis: Risk Factors, Prediction of Risk Using Serum CTX Testing, Prevention, and Treatment. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 65 (12), pp. 2397–2410

Morris, P.G. *et al*. (2012). Serum N-Telopeptide and Bone-Specific Alkaline Phosphatase Levels in Patients with Osteonecrosis of the Jaw Receiving Bisphosphonates for Bone Metastases. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 70 (12), pp. 2768–2775.

Mücke, T. *et al*. (2016). Do Measurements of Inflammatory Mediators in Blood Predict Recurrence in Patients with Bisphosphonate-Related Osteonecrosis of the Jaws? *British Journal of Oral & Maxillofacial Surgery*, 54 (April), pp. 286–289.

O'Connell, J.e. Kearns, G.j.,e Ikeagwani ,O. (2012). A Role for C-Terminal Cross-Linking Telopeptide (CTX) Level to Predict the Development of Bisphosphonate-Related Osteonecrosis of the Jaws (BRONJ) Following Oral Surgery?. *Irish Journal of Medical Science*, 181 (2), pp. 237–242.

Oizumi. *et al*. (2010). Inhibition of Necrotic Actions of Nitrogen-Containing Bisphosphonates (NBPs) and Their Elimination from Bone by Etidronate (a Non-NBP): A Proposal for Possible Utilization of Etidronate as a Substitution Drug for NBPs. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 68 (5), pp. 1043–1054.

Rosini,S. *et al*. (2015). Understanding Bisphosphonates and Osteonecrosis of the Jaw: Uses and Risks. *European Review For Medical And Pharmacological Sciences*, 19 (17), pp. 3309–3317.

Ruggiero. *et al*. (2009). American Association of Oral and Maxillofacial Surgeons Position Paper on Bisphosphonate-Related Osteonecrosis of the Jaws--2009 Update. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 67, 5 Suppl, pp. 2–1.

Ruggiero. *et al*. (2014). American Association of Oral and Maxillofacial Surgeons Position Paper on Medication-Related Osteonecrosis of the Jaw--2014 Update. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 72 (10), pp. 1938–1956.

Russell, R. G. *et al*. (2008). Mechanisms of Action of Bisphosphonates: Similarities and Differences and Their Potential Influence on Clinical Efficacy. *Osteoporosis International: A Journal Established as Result of Cooperation between the European Foundation for Osteoporosis and the National Osteoporosis Foundation of the USA*, 19 (6), pp. 733–759.

Santini, D. *et al*. (2003). Zoledronic Acid Induces Significant and Long-Lasting Modifications of Circulating Angiogenic Factors in Cancer Patients. *Clinical Cancer Research: An Official Journal of the American Association for Cancer Research*, 9 (8), pp. 2893–2897.

Sarasquete, M.E. et al. (2008). Bisphosphonate-Related Osteonecrosis of the Jaw Is Associated with Polymorphisms of the Cytochrome P450 CYP2C8 in Multiple Myeloma: A Genome-Wide Single Nucleotide Polymorphism Analysis. *Blood*, 112 (7), pp. 2709–2712.

Schwartz, H. C. (2008). Serum CTX Testing. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons* 66 (6), pp. 1319–1320.

Silveira, F.M. *et al*. (2016). Microscopic Evaluation of the Effect of Oral Microbiota on the Development of Bisphosphonate-Related Osteonecrosis of the Jaws in Rats. *Journal of Oral & Maxillofacial Research*, 7 (4), pp. e3.

Thumbigere-Math.. *et al*. (2016). Serum Markers of Bone Turnover and Angiogenesis in Patients With Bisphosphonate-Related Osteonecrosis of the Jaw After Discontinuation of Long-Term Intravenous Bisphosphonate Therapy. *Journal of Oral and Maxillofacial Surgery: Official Journal of the American Association of Oral and Maxillofacial Surgeons*, 74 (4), pp. 738–746.

Thumbigere-Math.. *et al*. (2012). A Retrospective Study Evaluating Frequency and Risk Factors of Osteonecrosis of the Jaw in 576 Cancer Patients Receiving Intravenous Bisphosphonates. *American Journal of Clinical Oncology*, 35 (4), pp. 386–392.

Vermeer, A. F. *et al*. (2016). Osteonecrosis of the Jaw-a Bone Site-Specific Effect of Bisphosphonates. *Current Osteoporosis Reports*, 14 (5), pp. 219–225.

Vincenzi, B. *et al*. (2012). Serum VEGF Levels as Predictive Marker of Bisphosphonate-Related Osteonecrosis of the Jaw. *Journal of Hematology & Oncology*, 5 (September), pp. 56.

Wood, J. *et al*. (2002). Novel Antiangiogenic Effects of the Bisphosphonate Compound Zoledronic Acid. *The Journal of Pharmacology and Experimental Therapeutics*, 302 (3), pp. 1055–1061.

Yamashita, J. e McCauley L.K. (2012). Antiresorptives and Osteonecrosis of the Jaw. *The Journal of Evidence-Based Dental Practice*, 12 ,3 Suppl, pp. 233–247.

Yamashita, J. *et al*. (2010). Updates on Osteonecrosis of the Jaw. *Current Opinion in Supportive and Palliative Care*, 4 (3), pp. 200–206.