Bone repair after osteotomy with diamond burs and CVD ultrasonic tips – histological study in rats

Reparação óssea após osteotomia com broca esférica diamantada e pontas de diamante CVD – estudo histológico em ratos

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Abstract

This study histologically evaluated the behavior of bone tissue of rats submitted to osteotomy with conventional diamond burs in high speed and a new ultrasonic diamond tips system (CVD – Chemical Vapor Deposition), at different study periods. The study was conducted on 24 Wistar rats. Osteotomy was performed on the posterior paws of each rat, with utilization of diamond burs in high speed under thorough water cooling at the right paw, and CVD tips at the left paw. Animals were killed at 0, 7, 14 and 21 days after treatment, followed by removal of connective tissue and bone at the area of osteotomy. Specimens were fixated, demineralized, dehydrated and sectioned for histological analysis by polarized light microscopy (25, 100 and 200x magnification) by two examiners. The results demonstrated that there were no differences in the repair of bone defects between the two different materials employed for osteotomy.

Uniterms

Crown lengthening; diamond bur; osteotomy.

Introduction

The restoration of shape, function and recovery of esthetic characteristics of teeth should be the main goals of Restorative Dentistry, yet they should be in harmony with the periodontium to allow adequate oral health (Venturini et al.[24], 2002). These goals may only be met if restorations are fabricated within proper biological principles to allow effective dental plaque control, maintaining the biological width (Stoll & Novaes[22], 1997; Melo Filho et al.[15], 2000).

The difficulty to achieve a correct relationship between the gingival margin and the periodontal tissue has been addressed by several authors for years. Invasion of these tissues may be very harmful, especially if the prosthetic-restorative treatment contacts the marginal periodontal tissues (Reeves[19], 1991). Subgingival preparations with cervical extension to the junctional epithelium or underlying tissues are potentially harmful to the periodontium (Sivers & Johnson[21], 1995; Melo Filho et al.[15], 2000; Venturini et al.[24], 2002), leading to alveolar bone loss for achievement of the physiological space required for new attachment of dentogingival joining components (Allen[2], 1993). The biological width, from the alveolar bone crest to the most coronal portion of the junctional epithelium, should be respected (Gargiulo et al.[9], 1961), at a minimum distance of 3.0mm above the alveolar bone crest (Kaldahl et al.[12], 1984; Bragger et al.[6], 1992; Hagge & Rector[10], 1993).

When the biological width is invaded by deep caries and/or fractures, subgingival margins, or in patients presenting clinical crowns with poor retention, crown lengthening procedures may be required to adequate the biological width, allowing correct positioning of the margin (Waal & Castelucci[26], 1993; Rockenbach et al.[20], 1995; Wilson & Kornman[28], 2001). This procedure, isolated or combined with endodontic, orthodontic or periodontal treatment, often assures successful tooth recovery (Baima[4], 1986).

Recovery of biological width by osteotomy and osteoplasty with apical repositioning flap requires removal of supporting bone for achievement of space for dentogingival connection and correction of bone architecture; osteotomy is often extended to adjacent tissues to avoid discrepancies in the natural parabolic curve of the bone margin (Lacaz Netto et al.[13], 1992). Surgical or orthodontic procedures or association of both may be used for biological width recovery. The surgical technique may be performed with burs in high or low speed and chisels applied manually or with a hammer (Abouzgia & James[1], 1997; Marzola et al.[14], 2000). Selection of the technique depends on the surgeon's manual dexterity and preferences (Abouzgia & James[1], 1997).

Among the high speed burs employed for osteotomy, conventional diamond burs fabricated by galvanic welding of diamond powder on metallic rods have been widely employed for more than 40 years (Hartley & Hudson[11], 1958). However, limitations in their technology led to the development of innovative diamond burs (Vieira & Vieira[25], 2002).

This evolution is represented by the advent of CVD (Chemical Vapor Deposition) diamond tips, which have several advantages in Dentistry compared to conventional burs. The CVD tips are obtained by a process of chemical deposition in vapor state inside a reactor, from a mixture of methane, tetrafluoromethane and hydrogen, which pass through a region of activation with a fiber. Adequate control of the mixture flow, substrate temperature, fiber temperature and internal pressure of the reactor allow the appearance of a polycrystalline diamond film (Valera et al.[23], 1996; Vieira & Vieira[25], 2002). A schematic drawing of the achievement of a CVD diamond is presented in Figure 1.

These diamond burs last longer, are easier to clean, and allow greater contact between diamond and the tooth structure, besides producing better finishing of prepared surfaces compared to conventional diamond burs (Valera et al.[23], 1996; Borges et al.[5], 1999).

The CVD tips are connected to an ultrasound machine, which is very favorable, since the rod performs vibrating instead of rotating motion. This allows angulation of the active rod and better access to regions where intact structure must be removed compared to conventional diamond burs in high speed (Vieira & Vieira[25], 2002). Moreover, many authors demonstrated that the excessive heat generated by high speed burs produces deep bone changes (Eriksson & Albrektsson[8], 1984; Okamoto[17], 1994). In the high speed system, the water spray may be obstructed by an adjacent tooth or by a wall of the same tooth, whereas water cooling in the ultrasound flows throughout the active point (Vieira & Vieira[25], 2002).

This new technology in CVD diamond tips suggests promising outcomes, especially for accomplishment of osteotomy, because of favorable characteristics as less trauma, better cooling and consequently less heat generation, thus favoring tissue repair. Therefore, this study evaluated the bone repair after osteotomy with CVD diamond tips compared to conventional diamond burs in high speed.

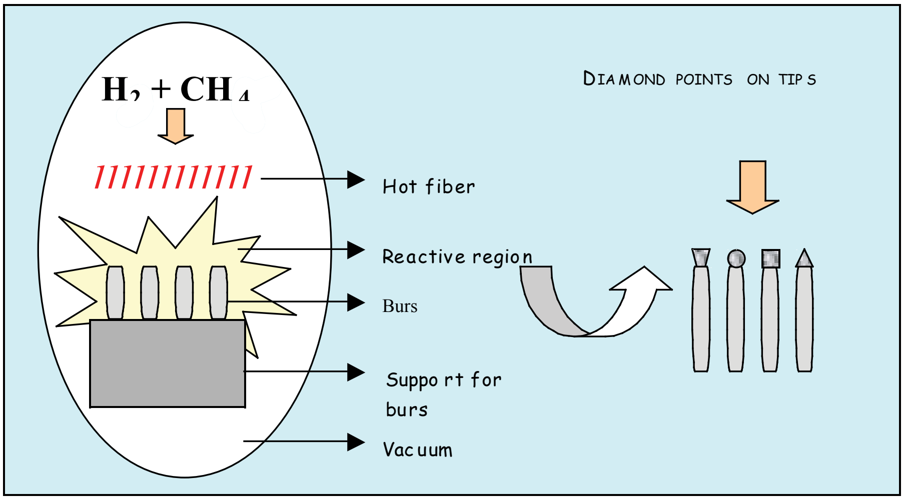

FIGURE 1 – Schematic drawing for achievement of a CVD diamond

Material and Methods

Division of groups

The sample was composed of 24 Wistar rats weighing 300 to 400g, obtained from the Animal Laboratory of São José dos Campos Dental School – UNESP. Osteotomy was performed on the posterior paws of each rat, with utilization of diamond burs in high speed under thorough water cooling at the right paw, and CVD tips at the left paw, thus constituting two study groups.

Anesthetic procedure

Before surgery, the rats were weighed for calculation of the correct anesthetic dose. General anesthesia was employed by intramuscular injection of xylazine hydrochloride 0.2g (Virbaxyl, Virbac do Brasil Ind. e Com. Ltda., São Paulo, Brazil) and ketamine 1.0g (Francotar, Virbac do Brasil Ind. e Com. Ltda., São Paulo, Brazil), at a ratio of 1:0.5 ml and dose of 0.1/100g body weight.

Accomplishment of osteotomy

After trichotomy and antisepsis with iodine solution, two lateral incisions measuring approximately 1.0cm in craniocaudal dimension were performed on the animals' paws. The underlying connective tissue was dissected with curved scissors to allow accomplishment of osteotomy. After exposure of bone tissue, osteotomy was performed with 2.0-mm diameter, as follows:

- Right paws – osteotomy with round diamond burs n. 1016 HL (ISO 018) connected to a high speed turbine under constant irrigation (Kavo).
- Left paws – osteotomy with CVD diamond tips connected to an ultrasound machine vibrating at 20,000 cycles per second under thorough irrigation (Gnatus).

The diameter of perforations was standardized with aid of a pachymeter with pre-established 2.0-mm diameter.

Incisions were closed with mononylon 4-0 simple suture (Ethicon, Johnson & Johnson) to allow healing by first intention. The animals received solid food and water *ad libitum* throughout the study.

Sacrifice of animals

Animals were killed by anesthetic overdose at 0, 7, 14 and 21 days after treatment. Three animals were killed at each period, adding up to 12 animals per group and 24 animals in the sample.

Achievement of histological slides

After sacrifice, the connective tissue and bone at the area of osteotomy were removed. Specimens were immediately fixated in buffered 10% formalin solution at neutral pH for a minimum period of 72 hours.

After fixation, the specimens were placed in a demineralizing sodium citrate and formic acid solution until complete demineralization, and then rinsed in tap water for 24 hours.

Following the routine histotechnical procedures, specimens were dehydrated in increasing alcohol concentrations, diaphanized in xylol and embedded in paraffin.

Semi-serial sections were obtained from the paraffin blocks in a microtome with 5-μm thickness. Sectioning was performed in axio-mesio-distal and axio-bucco-lingual planes. Hematoxylin-eosin staining was performed to allow microscopic analysis of specimens.

Histological analysis

The histological slides were analyzed by polarized light microscopy at 25x, 100x and 200x magnifications by two examiners. The inflammatory response was analyzed by histomorphometry, by assessment of the amount of bone tissue in the defect.

Results

0-day group

Right paw (round bur at high speed)

Histological sections of specimens in this group presented the following characteristics:

Presence of hemorrhagic area inside the medullary canal and at the area of osteotomy, as demonstrated by the large amount of erythrocytes at the surgical site (Figure 2a). Bone necrosis was not observed at this period, i.e. there were no signs of bone resorption at the periphery of the defect nor giant cells as osteoclasts. Bone spicules were observed in the medullary canal (Figure 2b), which had been originated by osteotomy.

Left paw (CVD ultrasonic tips)

The histological characteristics in this group were similar to the group submitted to osteotomy with high-speed burs. The following characteristics were noticed (Figure 3a).

The defects created with CVD tips originated an hemorrhagic area in the bone marrow, with large amount of erythrocytes and a fibrin network (Figure 3b). Bone spicules formed during osteotomy were also observed in the medullary canal. Some histological specimens exhibited an aspect of basophilic and disorganized tissue on the section surface, which may indicate surface necrosis, yet this was not observed in all specimens in this group (Figure 3c).

7-day group

Right paw (round bur at high speed)

This group presented the following histological characteristics:

Presence of immature, newly formed bone trabeculae originated from the endosteum throughout the defect, not only in width but also in the medullary canal, with wide spaces containing richly vascularized loose connective tissue (Figure 4a). Osteoblasts were observed around the newly formed bone trabeculae, which contained large osteocytes, indicating onset of bone matrix formation (Figure 4b). The defect surface exhibited onset of fiber deposition. Histological

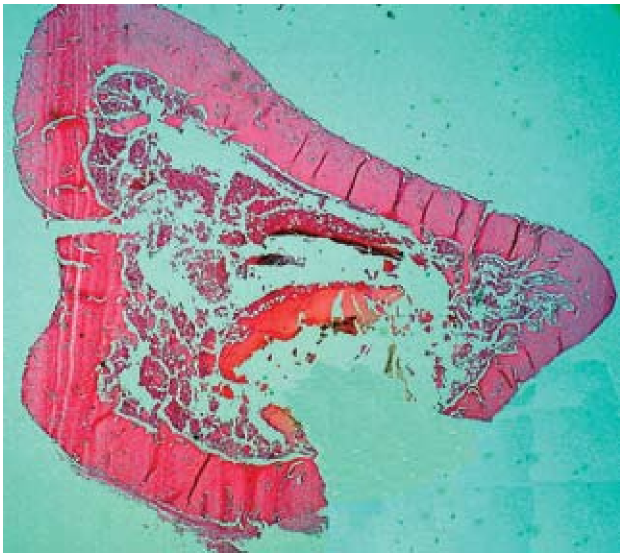

Figure 2a- Hemorrhagic area in the medullary canal caused by osteotomy with high-speed bur.

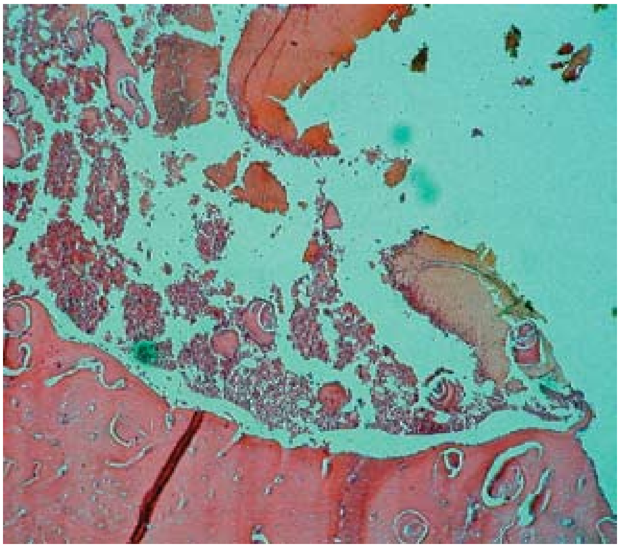

Figure 2b- Bone spicules inside the medullary canal.

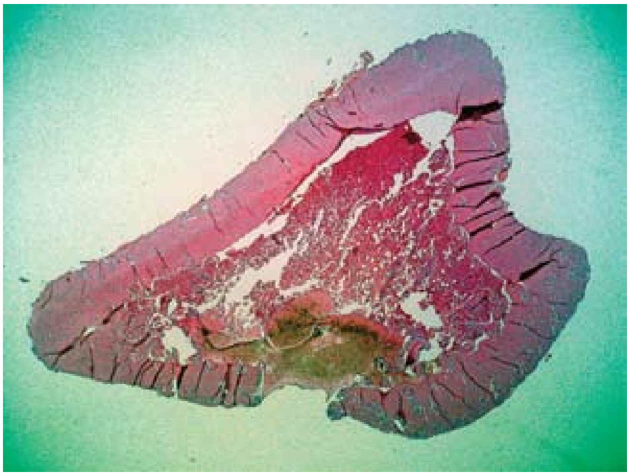

Figure 3a- Presence of hemorrhagic area, fibrin network and bone spicules in the medullary canal.

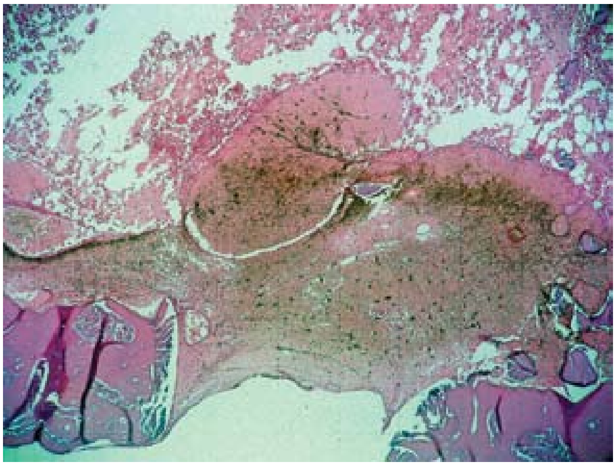

Figure 3b- Detail of fibrin network.

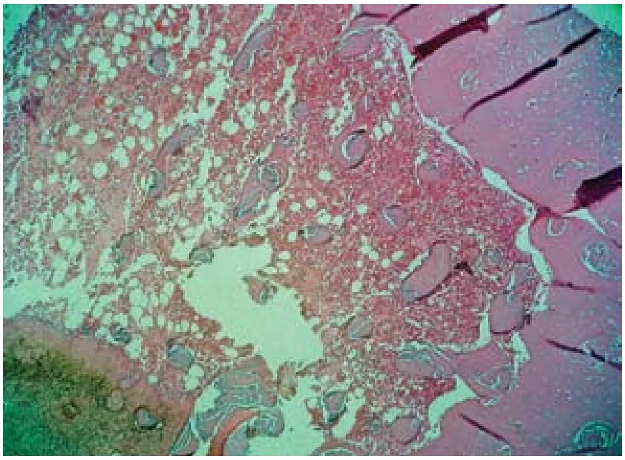

Figure 3c- Basophilic aspect observed only in some specimens.

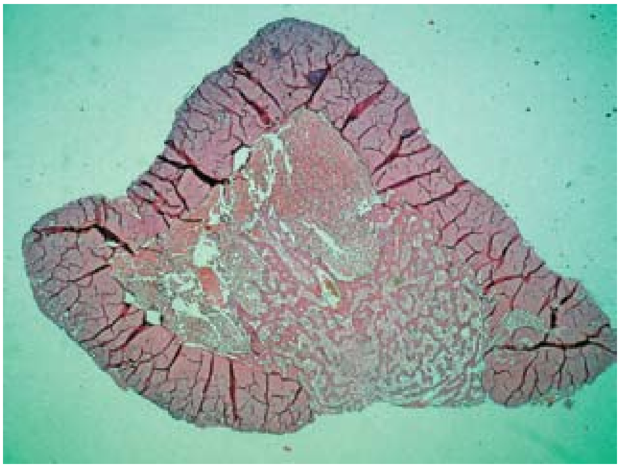

Figure 4a- Image of histological section with presence of bone trabeculae originated from the endosteum.

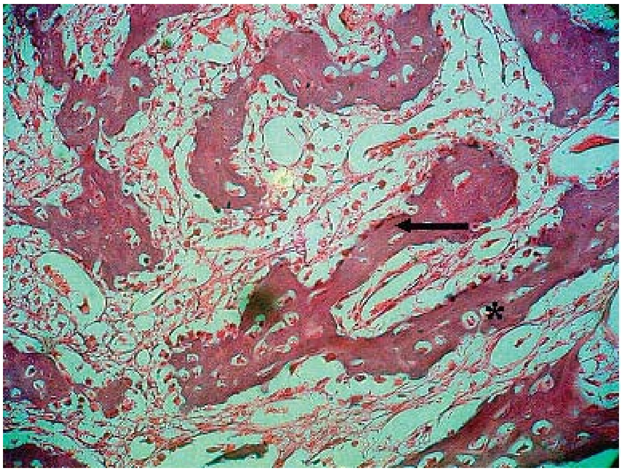

Figure 4b- Presence of osteoblasts (arrow) and osteocytes (mark).

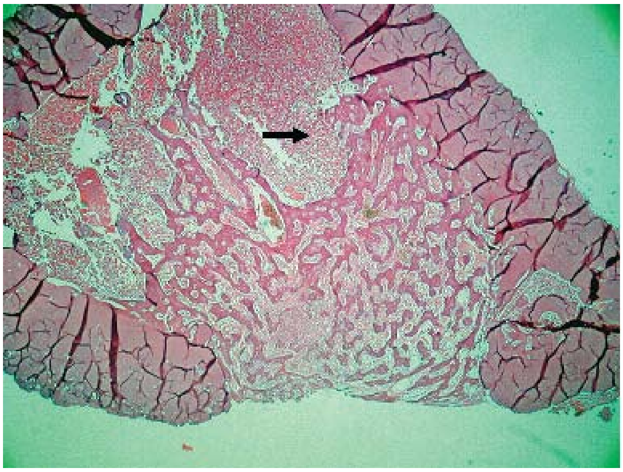

Figure 4c- Granulation tissue and blood clot at the central region of the defect (arrow).

analysis of some specimens revealed granulation tissue, with presence of the blood clot at the central and most superficial region of the defect. There was also new bone formation around the bone chips originated by osteotomy.

Left paw (CVD ultrasonic tips)

There were also similarities between specimens in the groups submitted to osteotomy with high-speed burs or CVD tips, as follows:

Many newly formed bone trabeculae were present and interrelated with the endosteum on the remaining bone tissue (Figure 5a), as well as many osteoblasts, osteocytes in the newly formed bone matrix and remarkable presence of osteoclasts, suggesting onset of bone remodeling (Figure 5b). Also, granulation tissue was observed on the defect surface.

14-day group

Right paw (round bur at high speed)

Specimens in this group exhibited the following histological characteristics:

Bone trabeculae were more organized than in the 7-day group, with many newly formed bone trabeculae joining the edges of the bone defect produced during osteotomy, thus constituting a bone bridge with well-organized periosteum, whose more internal layer presented many cells and a fibrous external layer (Figure 6a). The amount of loose connective tissue was reduced in spaces between bone trabeculae, as observed in the 7-day group, besides an increase in the amount of medullary bone tissue (Figure 6b).

Left paw (CVD ultrasonic tips)

Many similarities were observed in the histological characteristics of specimens in the two groups:

Many bone trabeculae inside the medullary space, presence of bone bridge joining the edges of the bone defect invested by a well organized periosteum, and clear replacement of loose connective tissue amongst the trabeculae by medullary bone tissue (Figures 7a and b).

21-day group

Right paw (round bur at high speed)

This group clearly exhibited the bone maturation process; the histological characteristics observed were as follows;

Formation of thicker bone bridge joining the edges of the defect with better organized bone trabeculae, bone matrix tissue with medullary characteristics amongst e trabeculae, since they represented a lamellar aspect indicating a more organized bone bridge. Osteocytes in bone trabeculae in the medullary canal were

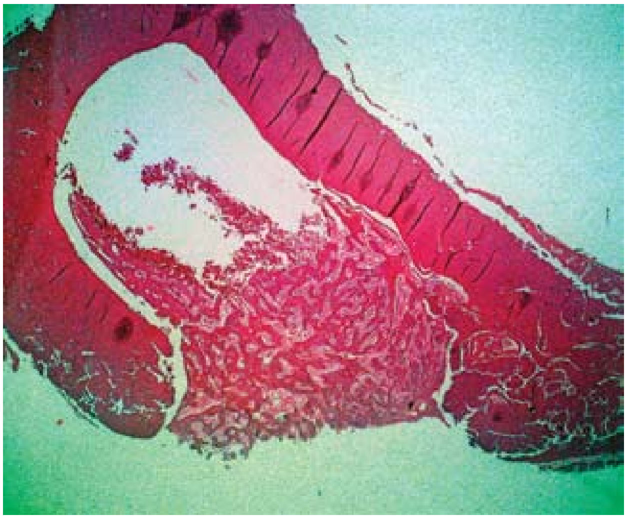

Figure 5a- Presence of bone bridge and granulation tissue on the defect surface.

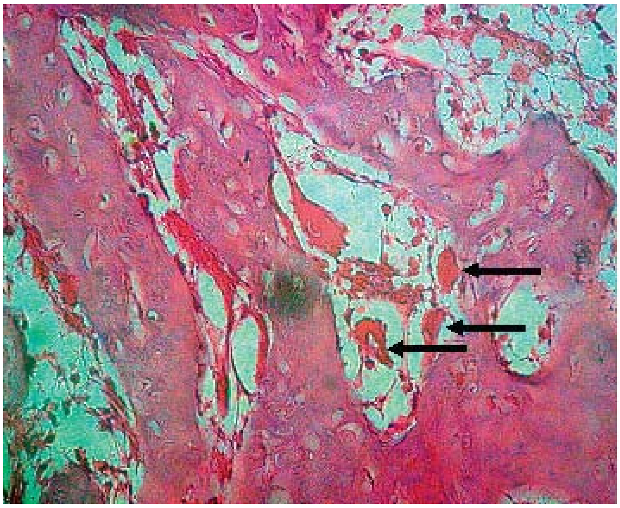

Figure 5b- Osteoclasts (arrows).

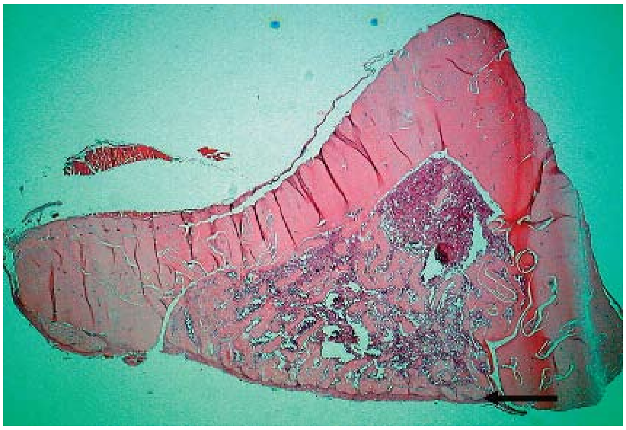

Figure 6a- Be bridge joining the edges of defect covered by periosteum (arrow).

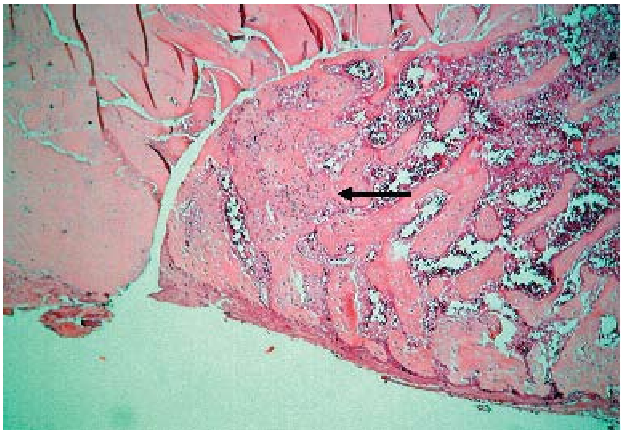

Figure 6b- Aspect exhibiting larger amount of medullary bone tissue (arrow).

smaller, and bone lacunae were increased with a small amount of osteoclasts. Some histological specimens exhibited well-defined areas of cortical and medullary bone tissue.

Left paw (CVD ultrasonic tips)

The specimens in this group presented the same histological aspects as the specimens in the high-speed bur:

The medullary canal was well delimited in the cortical bone, thicker bone bridge joining the edges of the defect, and more mature bone trabeculae than observed in the 14-day group, invested by a well-defined periosteum (Figures 9a and b). A more basophilic aspect was observed compared to the 21-day group submitted to osteotomy with high-speed burs.

Discussion

In 1961, Gargiulo et al.[9] defined biological width as the dimensions present in all human beings, occupied by connective attachment and epithelial attachment of the dentogingival junction. Diffusion of this new discovery changed and somehow guided restorative treatments with a view to maintain the periodontal integrity and increase the longevity of teeth.

According to Nevins & Skurrow[16], in 1984, the biological width comprises the gingival sulcus, epithelial attachment and connective attachment. The biological width may be damaged by several factors, including caries, fractures, iatrogenia, or lack of attention to the signs and symptoms when attempting to restore teeth with alterations in biological width.

Figure 7a- Bone bridge joining the edges of the defect invested by periosteum.

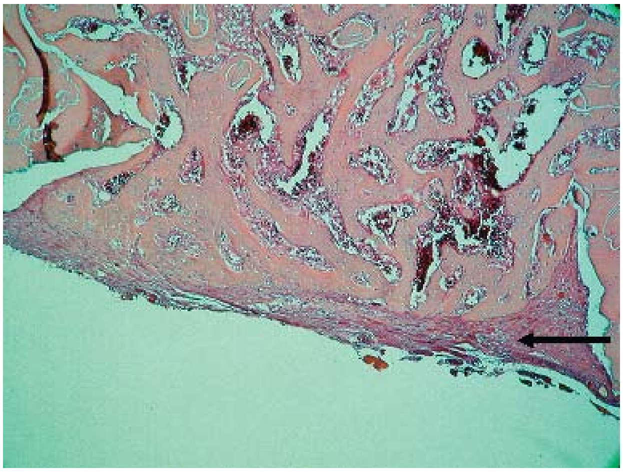

Figure 7b- Detail of periosteum (arrow)

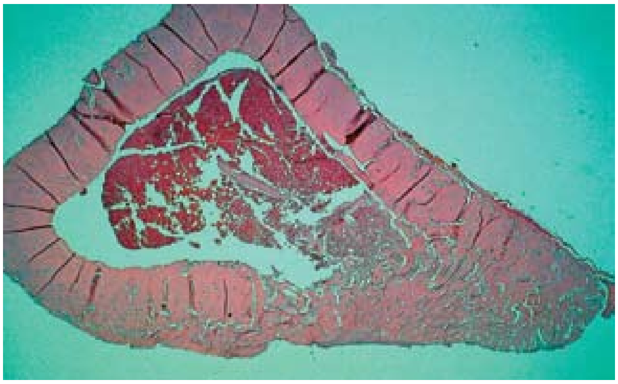

Figure 8- Bone bridge joining the edges of the defect.

When the biological width is invaded, the periodontium may present two responses: marginal recession, when the underlying bone is thin and there is a self-limiting inflammatory response, or when the underlying bone is thick and the inflammatory process leads to formation of a pocket with chronic and progressive characteristics (Wilson & Maynard[27], 1981; Parma-Benfenatti et al.[18], 1985).

Several periodontal surgical techniques as mucoperiosteal flaps either displaced or not, osteotomy procedures and tooth extrusion techniques are currently employed in dental practice to reestablish the biological width and maintain the remaining tooth structure (Lacaz Netto et al.[13], 1992).

According to Abouzgia & James[1], 1997 and Marzola et al.[14], 2000, after accomplishment of mucoperiosteal periodontal flap for achievement of direct access to the bone, the osteotomy procedure may be performed by high-speed burs or chisel; selection of the type of technique depends on the surgeon's manual dexterity and preferences and the access to the bone tissue, which may be influenced by the anatomical conditions or problems originated from limited mouth opening.

High-speed burs have been widely employed in these procedures; according to Amorin-Lopes & Lopes[3], 2001, there are no significant clinical differences as to bone aggression between the final outcomes of osteotomy procedures performed with manual or rotary instruments. We agree with the authors in that utilization of manual instruments is preferred, since they allow better control during removal of bone, limiting possible harms to the adjacent tissues. It should be mentioned that utilization of high-speed burs may cause damage and improper removal of bone generated by the inherent vibration and function of the turbine.

The proven utilization of CVD tips in restorative procedures (Valera et al.[23], 1996; Borges et al.[5], 1999) led us to test their utilization in osteotomy procedures in rats tibiae compared to osteotomy performed with high-speed burs, in order to find means to facilitate surgical procedures without the risks related to high-speed burs.

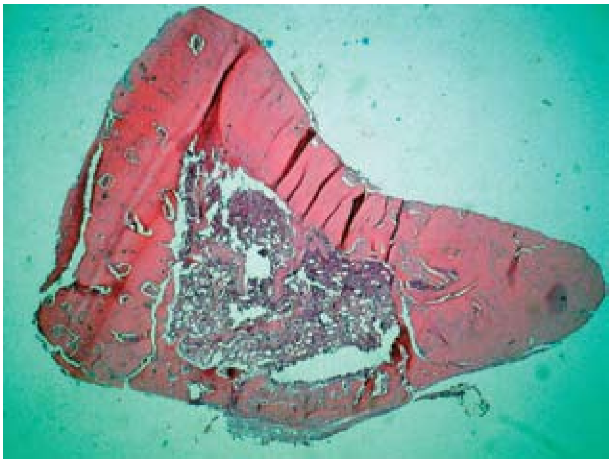

Figure 9a- Mature bone bridge and trabeculae.

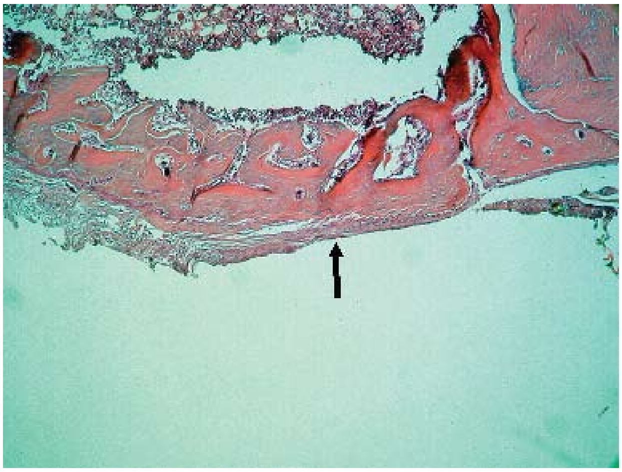

Figure 9b- Detail of periosteum investing the bone bridge

The advantages of CVD tips are less trauma to dental tissues, especially in operative dentistry procedures. According to Vieira & Vieira[25], 2002, this occurs because the ultrasound rods perform a vibrating instead of a rotating motion, thus allowing angulation of the active tip, providing better access to affected areas and keeping the integrity of healthy structures; less heat generation to the structures, since water cooling flows throughout the active point, whereas in high-speed burs it may be blocked by adjacent tooth aspects. According to Erikson & Albrektsson, 1984 and Okamoto[17], 1994, utilization of high speed turbines generates excessive heat and promotes extensive bone necrosis.

This study revealed that, histologically, all groups presented similar results between utilization of high-speed burs and CVD tips in ultrasound, regardless of the study period. With regard to osteotomy procedures, we agree with Amorin-Lopes & Lopes[3], 2001, that high-speed burs are difficult to control, not only due to trepidation but also to their high cutting power, requiring high operator skill.

However, it should be mentioned that utilization of CVD tips requires longer working time, and thus high-speed burs would be more practical if large osteotomies are to be performed, as indicated by Duarte et al.[7], 2002.

Conclusions

It was concluded that CVD tips in ultrasound may be employed for accopmplishment of osteotomy, since they generate repair histological changes in the bone tissues, similar to those observed for conventional high-speed burs. This procedure allowed the following advantages: constant irrigation not impaired by adjacent structures; and possibility to improve dental plaque control during surgery; and the tips' ability to save fost tissue, keeping the integrity of non-involved intact structures.

The results demonstrated that there were no differences in the repair of bone defects between the two different materials employed for osteotomy.

Resumo

O objetivo deste trabalho foi avaliar histologicamente o comportamento do tecido ósseo de ratos submetidos a osteotomia utilizando pontas diamantadas convencionais em alta-rotação e um novo sistema de pontas diamantadas em ultra-som (CVD – Chemical Vapor Deposition), em diferentes tempos de avaliação. Para tanto foram utilizados 24 ratos da raça Wistar. Em cada animal, nas patas posteriores, foram feitas as osteotomias, sendo que na pata direita foi realizada a osteotomia com brocas diamantadas em alta-rotação com refrigeração abundante e na pata esquerda foram utilizadas as brocas CVD para o desgaste ósseo. Os animais foram sacrificados nos dias 0, 7, 14 e 21 após o tratamento e, em seguida, foi realizada a remoção da porção de tecido conjuntivo e osso do local da realização da osteotomia. As peças foram fixadas, desmineralizadas, desidratadas e coradas para obtenção das lâminas histológicas. As lâminas histológicas foram analisadas em Microscópio de Luz Polarizada (25, 100 e 200X de aumento), por dois avaliadores. Os resultados obtidos demonstraram que não houve diferença na reparação óssea dos defeitos entre os dois tipos de materiais empregados na realização das osteotomias.

Unitermos

Aumento de coroa clínica; ponta diamantada; osteotomia.

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