

Histologic Study of Use of Microfibrillar Collagen Hemostat in Rat Dental Sockets

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The aim of this paper was to evaluate if the placement of microfibrillar collagen hemostat (MCH) into a dental socket interfered with healing. General anesthesia was administered to 30 adult male *Albinus* Wistar rats and the maxillary right central incisor was extracted. In the control group after each tooth was extracted, the socket was sutured. In the MCH group after each tooth was extracted, MCH was placed into the socket before suturing. Postoperatively, 5 animals were sacrificed from each group at 7, 21 and 28 days. The right maxilla was removed from each animal and histologic slides were stained with Masson's trichromic and hematoxylin and eosin. Quantitative and qualitative analyses were done. The percentage of bone area in the dental socket was quantified using the Image Lab 98 image analysis system. The bone area formation for the control and MCH groups was: 8.1% and 3.3% at 7 days, 34.4% and 33% at 21 days and 41% and 41.3% at 28 days, respectively. We concluded that MCH interferes with the beginning of dental socket healing but does not interfere with the final healing of the dental socket.

Key Words: tooth extraction, hemostat, bleeding, dental socket.

INTRODUCTION

Microfibrillar collagen hemostat (MCH) is a topical agent composed of resorbable microfibrillar collagen prepared to preserve the native bovine collagen molecules. The MCH attracts platelets and allows for the formation of a blood clot when it comes into contact with blood (1).

The manufacturer (MedChem Products Inc., Woburn, MS) suggests the use of MCH Avitene to achieve hemostasis with or without conventional methods or in situations in which conventional methods are not practical. This hemostatic agent is contraindicated as the sole hemostatic control in patients who have systemic problems causing hemorrhage.

To date, there is no experimental research in the literature relating the wound healing of a dental socket to MCH. The aim of this study was to determine whether or not the placement of MCH into a dental socket interferes with socket healing.

MATERIAL AND METHODS

Thirty male albino rats (*Ratus albinus norvegicus*, Wistar) weighing 160-220 g were used in this study. After general anesthesia (30 mg/kg thionembutal; Abbott do Brasil, Sao Paulo, SP, Brazil), the maxillary right central incisor was extracted using specially designed instruments (2). The control group consisted of 15 animals that did not receive any treatment to their dental sockets. The MCH group consisted of 15 animals that had MCH (Avitene; MedChem Products Inc., Woburn, MS) placed in the apical and middle thirds of their dental sockets, using a small adapted instrument. Each dental socket of both groups was sutured with 5-0 vicryl (Ethicon; Johnson & Johnson do Brasil, São Jose dos Campos, SP, Brazil). Five animals from each group were sacrificed at postoperative days 7, 21 and 28. The right maxilla was separated using a median sagittal incision. The specimen containing the right socket was removed with straight scissors, cutting tan-

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gentially to the distal surface of the last molar.

The specimens were fixed in 10% formalin for 24 h, decalcified in formic acid and sodium citrate solution, clarified and embedded in paraffin so the sockets could be cut in a longitudinal plane (3). The blocks were semi-serially sectioned at 6 μ m thickness and the sections were stained with Masson's trichromic and hematoxylin and eosin.

Qualitative and quantitative analyses of bone formation were done. The percentage of bone area in each dental socket was quantified using the IMAGE LAB 98 image analysis system. Data were analyzed with the Tukey test and analysis of variance.

RESULTS

Qualitative Analysis

Control Group: After 7 days, the alveolus was partially filled with newly formed connective tissue, blood clot and showed thin osseous trabeculae principally in the apical third (Figure 1). On day 21, the alveolus exhibited thick osseous trabeculae and some areas of mature connective tissue (Figure 2). On day 28, the alveolus was filled with mature osseous trabeculae (Figure 3).

MCH Group: At day 7, osteoid tissue and thin osseous trabeculae were present at the apical third. There was part of the MCH material in some samples (Figure 4) and polymorphonuclear neutrophil infiltration. Thick osseous trabeculae were present at the apical and middle thirds at 21 days (Figure 5). The mature osseous trabeculae present on the 28th day filled almost all alveoli (Figure 6).

Quantitative Analysis

The quantification of the trabecular bone area in the dental sockets was done by carefully following all of the dental sockets linearly. The mean percentile values of trabecular bone area in the dental socket at 7, 21 and 28 days were: control group: 8.1%, 34.4% and 41.0%, respectively; MCH group: 3.3%, 33.0% and 41.3%, respectively.

The Tukey test was applied and the interaction values were then subjected to analysis of variance. There was a significant statistical difference between the experimental and the control groups only at postop-

erative day 7 and no significant statistical difference between the groups at postoperative days 21 and 28.

DISCUSSION

It is well-documented that MCH has local hemostatic action. Kubo et al. (4) placed it in contact with bleeding surfaces after great vessel hemostasis during urologic surgery. They concluded the material quickly controlled hemorrhage in all of the patients studied. Low et al. (5) used MCH in laparoscopy because of its effectiveness as a hemostatic agent. Nakamura et al. (6) used MCH on the brain cortex and reported that the hemostatic action was 95% effective without any side effects in all of their experimental surgeries. Low et al. (5) and Nakamura et al. (6) concluded that MCH is biocompatible in their areas of research. The dental socket, however, presents its own peculiarities when compared with other areas of the body (7).

An *in vitro* coagulation mechanism of commercialized products was evaluated by Wagner et al. (8). They classified from better to worse the following hemostatic agents: microfibrillar collagen (Avitene®), collagen sponges (Helistat®, Instat® and Actifoam®), gelatin sponge (Gelfoam®) and oxidized regenerated cellulose (Surgicel®). As a result, the use of MCH for postoperative bleeding in dental sockets appears to be an interesting option. The aim of this paper was not to evaluate hemostasis; however, we did notice that the MCH group did not show the same amount of bleeding as the control group.

Barbolt et al. (9) carried out a comparative study with gelatin sponge, oxidized regenerated cellulose and MCH implanted in the subdural space after craniotomy in rabbits. They reported that the tissue reaction to MCH was characterized by moderate to marked granulomatous inflammation with an acute inflammatory component indicating a greater degree of tissue irritation. Sequelae of this reaction were still observed at 92 days post-implantation.

A question that still remains is: can MCH be used in the dental socket after tooth extraction? It is believed that the majority of materials implanted in the dental socket after dental extraction to treat alveolitis, to avoid alveolus atrophy or to act as a hemostatic agent all promote considerable healing delay (10). We agree with Perri de Carvalho and

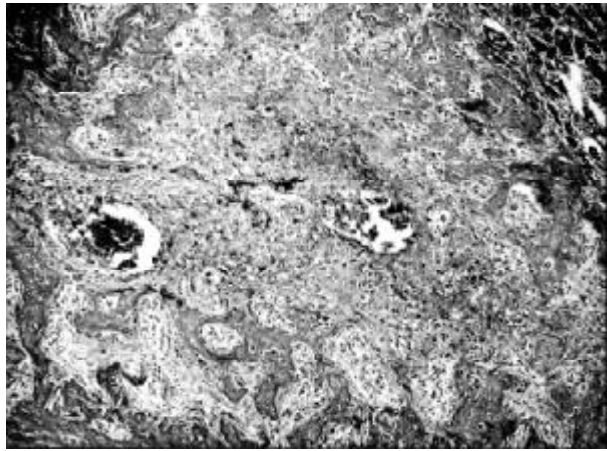


Figure 1. Control Group. 7 days postoperative. Newly formed connective tissue, blood clot and thin osseous trabeculae in the apical third. H&E. Original magnification, 63X.

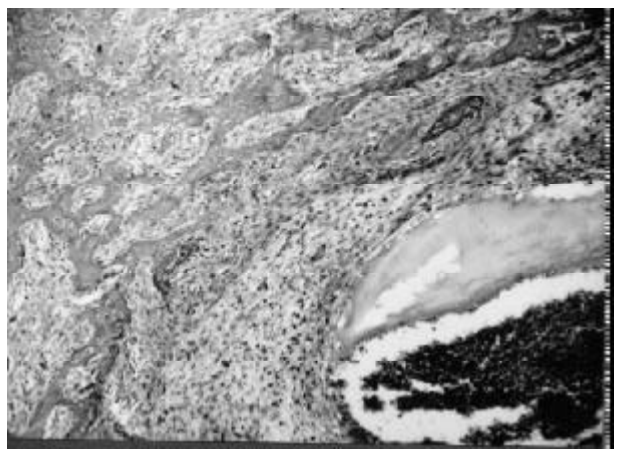


Figure 4. MCH Group. 7 days postoperative. Connective tissue, osteoid tissue and thin osseous trabeculae in the apical third. MCH material is still present. H&E. Original magnification, 63X.

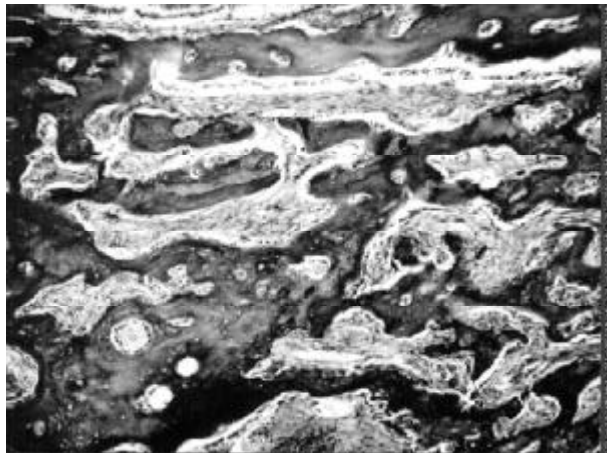


Figure 2. Control Group. 21 days postoperative. Presence of thick osseous trabeculae and some areas of mature connective tissue. H&E. Original magnification, 63X.

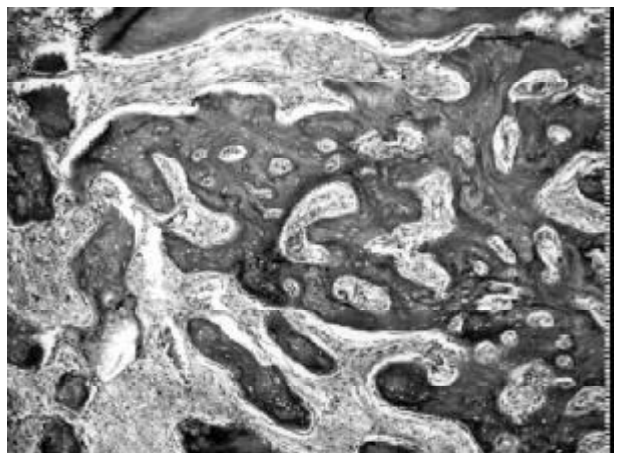


Figure 5. MCH Group. 21 days postoperative. The apical third presents a thick osseous trabeculae. H&E. Original magnification, 63X.

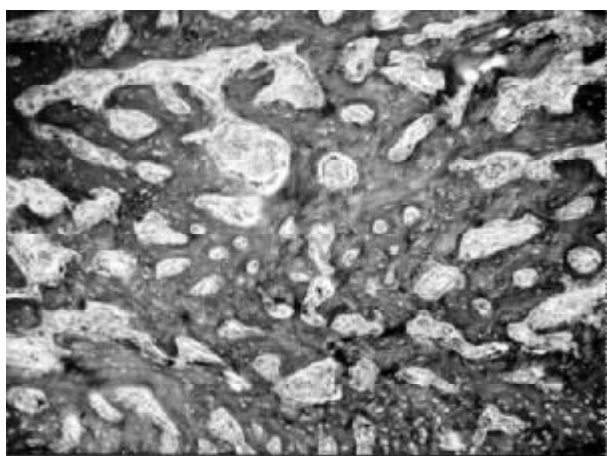


Figure 3. Control Group. 28 days postoperative. Mature osseous trabeculae in the dental alveolus. H&E. Original magnification, 63X.

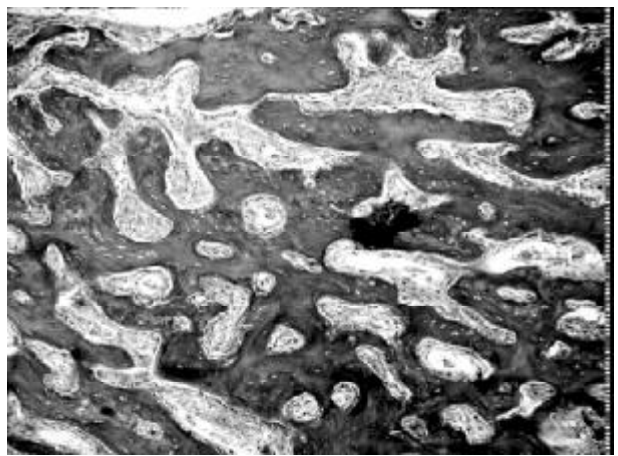


Figure 6. MCH Group. 28 days postoperative. Mature osseous trabeculae fills the alveolus. H&E. Original magnification, 63X.

Okamoto (7) who reported after numerous studies that the use of materials in the dental socket should be restricted to those cases in which the need for hemostasis outweighs the potential morbidity.

In our study, MCH was not totally absorbed at 7 days postoperatively and there was formation of thin trabeculae bone in the apical third of the alveolus; however, the control group showed a greater quantity of new bone at 7 days, most likely due to the presence of MCH in the other group. The final healing quality, however, was similar in both groups. The initial interference of MCH did not influence the quantity of trabecular bone in the dental socket.

We conclude that the hemostatic agent presented here appears to be a viable alternative in controlling intraoperative and postoperative hemorrhage from the dental alveolus because it is both biocompatible and effective.

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RESUMO

O objetivo deste trabalho é avaliar se a colocação do MCH no interior do alvéolo dental interfere ou não no reparo alveolar. Foi feita anestesia geral nos trinta ratos machos adultos *Albinus Wistar* e o incisivo central superior direito foi extraído. No grupo controle, após a exodontia o alvéolo foi suturado. No grupo MCH, após a exodontia introduziu-se MCH dentro do alvéolo que foi suturado. No pós-operatório, cinco animais de cada grupo foram sacrificados aos 7, 21 e 28 dias. Retirou-se a maxila

direita de cada animal e cortes histológicos foram confeccionados. Foi feita análise qualitativa e quantitativa. A porcentagem de área óssea no interior dos alvéolos dentais foi quantificada usando o sistema de análise de imagens ImageLab 98. A área óssea neoformada no grupo controle e no grupo MCH foi: 8.1% e 3.3% aos 7 dias, 34.4% e 33% aos 21 dias e 41% e 41.3% aos 28 dias, respectivamente. Dentro das condições experimentais deste trabalho foi possível concluir que o MCH interfere durante o início do reparo alveolar e não interfere no reparo final do alvéolo dental.

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