Evaluation of Rat Alveolar Bone Response to Angelus MTA or Experimental Light-cured Mineral Trioxide Aggregate Using Fluorochromes

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Abstract

Introduction: The aim of this study was to evaluate the rat alveolar bone response after the implantation of experimental light-cured mineral trioxide aggregate (MTA) or Angelus MTA (Angelus, Londrina, Paraná, Brazil) by histological and fluorescence analysis. **Methods:** Thirty Wistar Albino rats were divided into three groups. In the control group, empty polyethylene tubes were inserted into the rat alveolar sockets immediately after extraction. In the other groups, the tubes were filled with light-cured MTA or Angelus MTA. Five animals from each group were injected with calcein on day 7, alizarin on day 14, and oxytetracycline on day 21. On day 30, these animals were killed, and the right hemimaxillas were removed and histologically processed. Half of the maxillas were processed and stained with hematoxylin and eosin. The remaining maxillas were processed for fluorescence analysis and stained with Stevenel blue and alizarin red. New bone was histomorphometrically evaluated using a Merz grid. **Results:** The light-cured MTA presented a similar response when compared with Angelus MTA; it was characterized by a mild inflammatory response and complete bone healing. In the light-cured MTA group, the fluorescence areas were more evident at 21 days, showing an increase in bone formation. However, dystrophic mineralization was observed only with Angelus MTA. **Conclusions:** It was concluded that both materials present a similar inflammatory response and bone healing, but dystrophic mineralization was observed only with Angelus MTA. *(J Endod 2011;37:250–254)*

Key Words

Biocompatibility, fluorochromes, inflammation, mineral trioxide aggregate

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Mineral trioxide aggregate (MTA) was introduced in 1993 (1). It may be a viable material in capping of the dental pulp, root-end closure, repairing root perforation, and apical surgery (2). In addition, *in vivo* studies showed that MTA induces mineralized tissue formation such as dentin and cementum-like tissue (3–11). Its biomineralization process leads to the formation of an interfacial layer with tag-like structures at the cement-dentin interface and positively influenced the push-out bond strength (12). MTA also has insignificant arsenic amounts in its composition (13).

Despite its favorable characteristics, MTA has a short working time, has difficulty in handling and insertion, and requires additional moisture to activate its setting (14). An experimental light-cure MTA has been developed to have similar properties to MTA and also better working properties. Although this experimental material apparently presents positive characteristics, there is only one study that evaluates the subcutaneous tissue reaction after its implantation (15), showing its similarity with Angelus MTA. Moreover, this material is lacking in information regarding the bone healing.

Thus, the aim of this study was to evaluate by histological and fluorescence analysis the rat alveolar tissue response to implanted polyethylene tubes filled with a light-cure MTA or Angelus MTA.

Materials and Methods

Thirty-three–month-old male Wistar Albino rats, weighing between 300 and350 g, were used. The animals were housed in temperature-controlled rooms and received water and food ad libitum. The care of the animals was performed according to the Araçatuba School of Dentistry-UNESP Ethical Committee, which approved the project before the beginning of the experiments.

The rats were anesthetized with ketamine (87 mg/kg Francotar; Virbac do Brasil Ind e Com Ltda, Roseira, Brasil) and xylazine (13 mg/kg Rompum; Bayer S A, São Paulo, Brazil) administered intramuscularly and were divided into three equal groups: light-cure MTA (Bisco, IL), Angelus MTA, and control.

Thirty polyethylene tubes (Abbott Laboratories of Brazil, Sao Paulo, Brazil) with a 1.0-mm internal diameter, 1.6-mm external diameter, and 3.0-mm length had one of their openings sealed with 1 mm of gutta-percha (Hygenic-DFL, Akron, OH) before sterilization in ethylene oxide. Two groups of 10 tubes each were filled with the tested materials, and 10 remained empty to be used as controls.

The materials were handled according to the manufacturer's recommendations and inserted into 10 polyethylene tubes. The light-cure MTA was cured for 60 seconds with a light-cure unit (Ultralux-Dabi Atlante, Ribeirão Preto, Brazil). Each animal had its right upper incisor extracted by using special instruments (16), and after hemostasis the implants were inserted into the depth of the socket. The gingival tissue was sutured with nonresorbable silk 4-0 sutures (Ethicon, Somerville, NJ).

Fluorescent bone markers were injected in each animal for the study of the bone remodeling surround the tube opening. Calcein green, alizarin red, and oxytetracycline hydrochloride were injected after 7 days, 14 days, and 21 days after implantation,

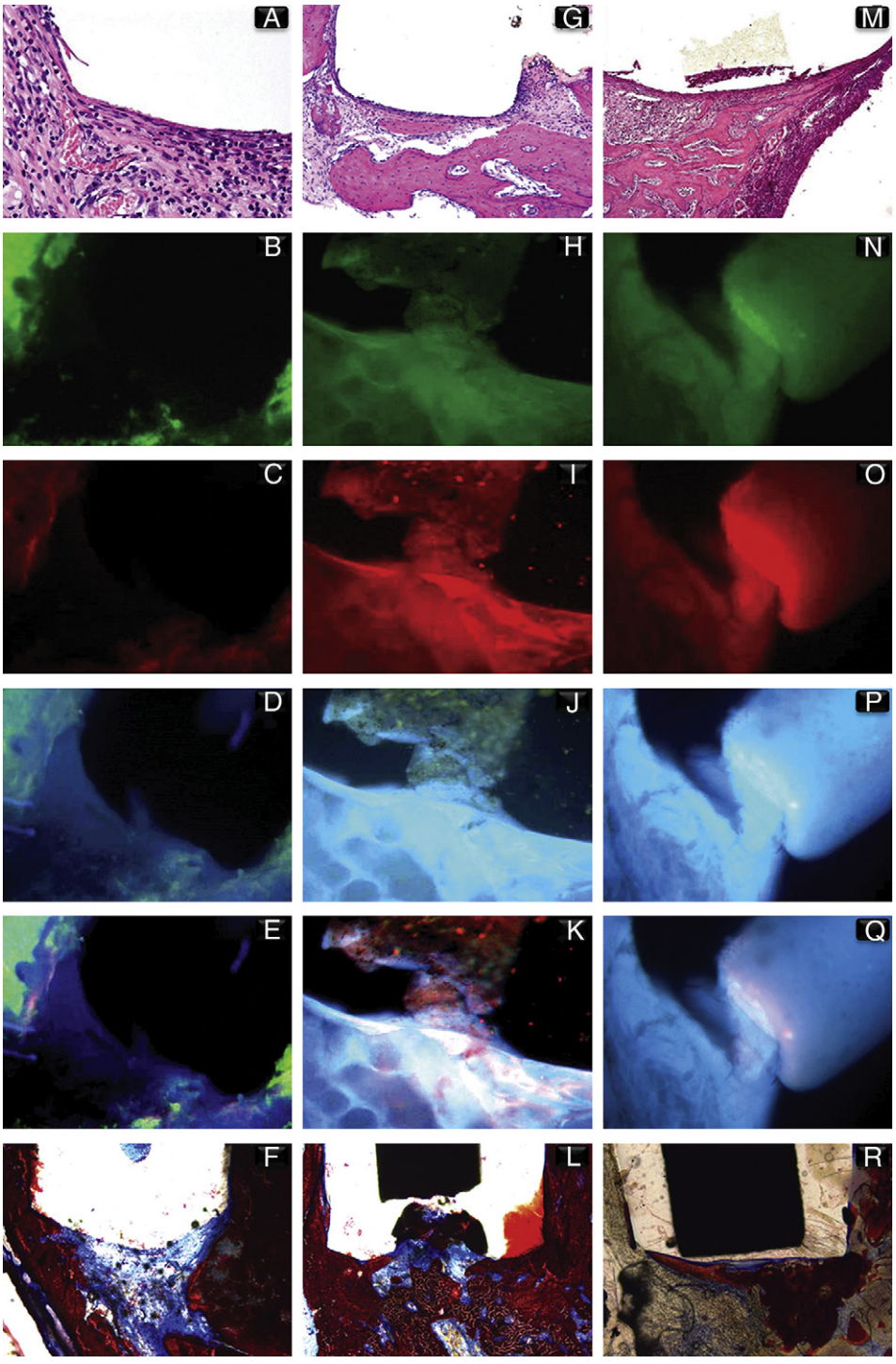

Figure 1. Control group: (*A*) a specimen showing mild chronic inflammatory infiltrate with lymphocytes and giant cells after 30 days (hematoxylin and eosin, ×40); (*B-E*) the absence of calcification areas in close contact with the tube opening (fluorochrome analysis, ×10), and *(F)* the absence of mineralized tissue close to the material (Stevenel blue and alizarin red, ×10). MTA Angelus: *(G)* mild to moderate chronic inflammatory cell infiltration in the fibrous capsule, and the new bone tissue was formed (hematoxylin and eosin, ×10); *(H)* mineralized tissue formed on the seventh day in a region close to the tube end and visualized by the presence of a smooth green marker (calcein green marker, ×10); *(I* and *J)* mineralized tissue present after 14 and 21 days visualized by the distinct presence of red and blue markers, respectively (alizarin red and oxytetracycline hydrochloride, respectively, ×10); *(K)* projected images of the three fluorochromes showing new bone tissue formed along the time and predominant formation on the 21st day (fluorochrome analysis, ×10); and *(L)* the presence of mineralized tissue close to the material (Stevenel blue and alizarin red, ×10). Light-cure MTA: *(M)* mild chronic inflammatory reaction in a thin fibrous capsule between the bone and the material (hematoxylin and eosin, ×10), *(N)* bone tissue was formed on the seventh day in a region close to the tube end (calcein green marker, ×10), *(O* and *P)* the mineralized tissue was more evident and extensive far from the tube end on days 14 and 21 (alizarin red and oxytetracycline hydrochloride, respectively, ×10), *(Q)* the overlaid images showed bone tissue formed predominantly on days 14 and 21, and *(R)* bone tissue close to the material was confirmed by a reddish stain (Stevenel blue and alizarin red, ×10).