

## **Basic Research - Technology**

### **Surface Changes of Mineral Trioxide Aggregate after the Application of Bleaching Agents: Electron Microscopy and an Energy Dispersive X-ray Microanalysis**

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## **Abstract**

**Introduction:** The aim of this study was to investigate the changes in the surface structure and chemical composition after applying bleaching agents to completely hardened mineral trioxide aggregate. **Methods:** A total of 12 samples of MTA blocks were divided into 3 groups, two different bleaching agents, and a control group. The surface structure was observed using a scanning electron microscope (SEM). The changes in elemental composition were analyzed by an energy dispersive X-ray microanalysis (EDX) system. **Results:** The surface of the MTA covered with each bleaching agent changed in terms of both color and structure compared to the control. EDX showed that both bleaching agents affected the elemental distribution. A decrease in Ca and an increase in Si were demonstrated, and this tendency was especially pronounced in the higher hydrogen peroxide concentration group. **Conclusions:** The acidic conditions induced by bleaching agents brought about the deterioration of the MTA surface. These findings suggest that MTA is an insufficient barrier against tooth bleaching.

## **Key Words**

MTA, bleaching agent, acidic condition, SEM, EDX

Mineral trioxide aggregate (MTA) has been used as root-end filling material (1-3), a root or furcal perforation repair material (4, 5), and a pulp-capping material (6-10).

The bleaching of non-vital, discolored teeth is a low-risk popular procedure. The placement of a cervical base before internal bleaching treatment is recommended for preventing cervical resorption after bleaching (11-13). In the walking bleaching technique, after an access cavity is prepared, the root-canal filling material is shortened 2-3 mm subgingivally. An impermeable base, such as resin-modified glass-ionomer cement, is laid on the root-canal filling material to prevent cervical root resorption. Previous study of the apical sealing ability of MTA after root resection has indicated that root resection does not significantly affect the sealing ability of MTA when at least 3 mm of the MTA remained (14). Furthermore, the sealing ability of MTA used as a cervical barrier in the intracoronal bleaching was demonstrated *in vitro* to have a non-statistically significant difference compared to the cyanoacrylate-sealed control (15). Based on these findings, there appears to be a possibility that a bleaching agent may be applied on the MTA in the discolored non-vital tooth after a cervical perforation site has been filled with MTA.

The surface changes of MTA after the application of various chemicals (16) and a physiological solution pH (17) have been examined together with physical properties. Furthermore, the structural changes of MTA after the application of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) solution as acidic conditions have been investigated using SEM and the EDX system (18-23). Each report

presents different data and results, which is thought to be due to the differences in the concentrations of the H<sub>2</sub>O<sub>2</sub> solution, the reaction time, and the time lapse after MTA setting. However, little is known about the structural and chemical changes of the MTA under clinically-relevant conditions. The interaction between MTA and other dental materials is important to understand the indications of using the MTA application as a filling material. Therefore, the aim of the present study was to investigate the changes in the surface structure and elemental distribution after applying commercially available bleaching agents on completely hardened MTA. The test hypothesis was that structural deterioration, together with change in the elemental level, occurs after the application of bleaching gel to MTA.

## **Materials and Methods**

The standard powder/liquid ratio (1 g of white MTA powder to 0.33 mL purified water), according to the manufacturer's instructions, was used for this study. The mixed MTA was placed in silicon tubes (4 mm in length×4 mm in diameter) and maintained in a container for 21 days at room temperature in which the relative humidity was 100%. After a complete setting, the cylindrical-shaped MTA was cut horizontally into halves with a disposable scalpel (Feather Co., Tokyo, Japan). The divided plane was provided to the present experiment. A total of 12 samples of MTA blocks were divided into 3 groups, two groups exposed to bleaching agents (each: 5 samples) which consisted of a SHOFU HiLite<sup>®</sup> (Shofu Co., Kyoto, Japan) group and a PYRENEES<sup>®</sup> (Mitsubishi Gas Chemical Com. Inc., Tokyo, Japan) group (Table 1), and a control group (2 samples). The following clinical standard bleaching methods recommended by manufacturers were used in this study.

### **SHOFU HiLite<sup>®</sup> (SHilite) group**

The bleaching agent was mixed according to the manufacturer's instructions and placed at a 2 mm in thickness on the MTA. It was reacted with the MTA for 5 minutes and activated with an LED light source for 3 minutes (JET LIGHT 3000; Morita Co., Kyoto, Japan). This procedure was repeated three times. After that, the surface of the MTA was cleaned with tap water for 1 minute, dried with air, and kept in a container at room temperature for 1 week.

### **PYRENEES<sup>®</sup> (Pyrenees) group**

Two bleaching solutions were mixed for 1 minute according to the manufacturer's instructions and placed at a 2 mm in thickness on the MTA. The solutions were activated with an LED light source for 3 minutes (JET LIGHTE 3000). This procedure was repeated three times. After that, the surface of the MTA was cleaned with tap water for 1 minute, dried with air, and kept in a container at room temperature for 1 week.

### **Control group**

The surface of the MTA in the control group did not receive any treatment.

### **SEM and EDX**

The samples were mounted on aluminum holders with adhesive carbon tape, and observed using an operating microscope. Thereafter, the same samples were coated with carbon using a vacuum evaporator (JEE-400; JEOL Ltd., Tokyo, Japan). They were then examined in a SEM (S-3500N; Hitachi Ltd., Tokyo, Japan) fitted with an EDX (EMAX-7000 Type S; Horiba Ltd., Kyoto, Japan) system. SEM observation (magnification:  $\times 50$ ,  $\times 250$ ,  $\times 500$ ) was conducted using a secondary electron imaging technique operating at 15 kV and a working distance of 15 mm. In this study, EDX by area mode (analytic area:  $0.01 \text{ mm}^2$ ) was done on 3 parts of the MTA surface selected at random. EDX was performed using the following parameters: an X-ray take-off angle of 35 degrees, a probe current of 0.3 nA, and a counting time of 100 seconds. Weight percentages of Mg, Si, P, Ca, and bismuth (Bi) were

calculated using an AUTO-ZAF program. Furthermore, a high-speed additional element mapping was performed at 20 kV for 30 seconds  $\times$  10 cycles.

## **Results**

### **Surface color**

The color of the MTA was white or cream-color in the control group. However, after interaction with the bleaching agents, the surface color was changed from white to gray in both bleaching agent groups.

### **SEM and EDX**

Plate-like, globular, and rod-like structures were also observed together with air bubbles on the surface of the control intact MTA (Fig. 1A). In the SHilite group (Fig. 1B), globular structures and numerous woodpecker holes (around 5  $\mu\text{m}$  in diameter) were observed on the uneven surface. In the Pyrenees group (Fig. 1C), there were numerous creases and/or folds throughout the surface, and globular structures (5-15  $\mu\text{m}$  in diameter) were also observed in some places.

Table 2 shows the elements detected in the 3 groups. The Si values of the SHilite and Pyrenees groups were approximately 6-fold and 3-fold, respectively, compared to those of the control group. The Ca values of the SH and P groups were about one third and one half, respectively, compared to the control group. Elemental mapping revealed the detected elements to be distributed throughout the surface in the 3 groups. Ca in the control and Si in the SH group were strongly detected by such mapping.

## Discussion

The data obtained using SEM and EDX clearly demonstrated the present hypothesis to be validated, because the discoloration of MTA was confirmed compared to the control after the application of bleaching agent. This was brought about by the low pH of the H<sub>2</sub>O<sub>2</sub> in the bleaching gel. Then, an etchant and self-etching primer, in combination with the resin bonding system, could also cause the discoloration of the MTA.

The SHilite group developed more obvious surface changes compared to the Pyrenees group. SHilite contains 35% H<sub>2</sub>O<sub>2</sub> (pH 3.19), while Pyrenees contains 3.5% H<sub>2</sub>O<sub>2</sub> (pH 6.13) (24). These findings suggest that the lower pH derived from the higher concentration of H<sub>2</sub>O<sub>2</sub> affected the surface structure of MTA more strongly. The previous studies concerning the structural changes of MTA by acid attack were designed for different setting times after mixture. If the test was performed after insufficient setting time, it is easier to dissolve the components from the surface. Although the compressive strength of MTA at 24 hours after mixture was inferior to that of Super EBA cement, there is no statistical difference between two materials after 21 days (25). This data means that the MTA hardens completely at 21 days after mixture. Based on this finding, a 21 day-period after mixture was used in this study to clarify the exact structural changes in fully-hardened MTA. The present results clearly demonstrate that bleaching agents may disturb even the surface of completely hardened MTA. A previous study showed that the interaction between MTA and H<sub>2</sub>O<sub>2</sub> produced bubbling, and the soluble oxygen concentration increased in the reaction solution (23).

Woodpecker holes formed as a surface defect, and were especially notable in the SHilite group, are likely a by-product of this oxygen-bubbling. Microleakage is suspected to occur through the cracks around woodpecker holes in SHilite group, and through the fissures at the creases or folds in Pyrenees group.

White MTA contains about 44.23 weight% of CaO, 21.20 weight% of SiO<sub>2</sub>, and 16.13 weight% of Bi<sub>2</sub>O<sub>3</sub> (26). A 22.45 weight% of Bi in the present study was consistent with this reference value. However, EDX showed that the concentration of Bi was 3.85 weight% after the reaction of the MTA surface with 3% H<sub>2</sub>O<sub>2</sub> (18, 19). The Bi concentration was 2.2-fold higher in the Pyrenees group, which contained a similar percentage of H<sub>2</sub>O<sub>2</sub>. The discrepancy results from the differences in the semi-quantitatively analyzed area in the MTA. The central part of hardened MTA was analyzed using the EDX system in the present study (see the Materials and Methods), which means that Bi distributes more to the central part of hardened MTA.

The concentration of Si in the SHilite and Pyrenees groups was 6 times and 3 times higher than that in the control group, respectively. The elemental mapping of Si showed the specific pattern of distribution consistent with the beehive-like structures. These findings indicate that Si precipitates deposit at the uneven surface through oxygen-bubbling produced by the H<sub>2</sub>O<sub>2</sub> in the bleaching gel. The general deterioration of industrial concrete blocks is caused by 8 factors; dryness, release of Ca ion by water, erosion by sulfate, expansion by delayed ettringite  $\{(CaO)_6(Al_2O_3)(SO_3)_3 \cdot 32H_2O$ , in hydrated Portland cement system} formation, freezing and fusion, thaumasite

{Ca<sub>6</sub>Si<sub>2</sub>(CO<sub>3</sub>)<sub>2</sub>(SO<sub>4</sub>)<sub>2</sub>(OH)<sub>12</sub>·24H<sub>2</sub>O, in hydrated Portland cement system} formation, neutralization, and chemical degeneration (27). Among these processes, the release of Ca ion by water and chemical degeneration of hydrates are closely related to the changes after the application of bleaching agents. The decrease of Ca and the increase of Si were demonstrated at pH 3.5-4.0 in Portland cement, which is basically similar in composition to MTA (28). The acidic condition of the bleaching agent attacked the surface of the MTA, which induced the release of Ca ion through these phenomena, and subsequently the relative concentration of Si increased.

In conclusion, the present findings suggest that acidic attack of bleaching agents makes it prudent to protect the MTA with an intermediate resin before a bleaching procedure.

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## Legends for Figures

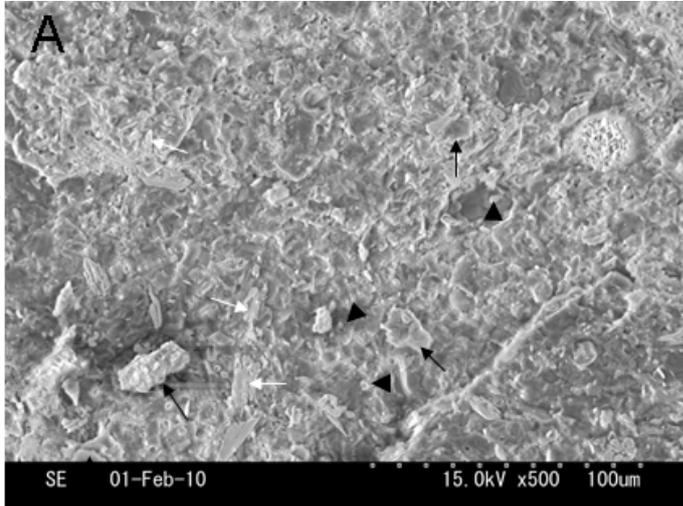


Figure 1A. A scanning electron micrograph of the surface of MTA in the control group. Note plate-like (black arrows), globular (arrow heads), and rot-like (white arrows) structures.

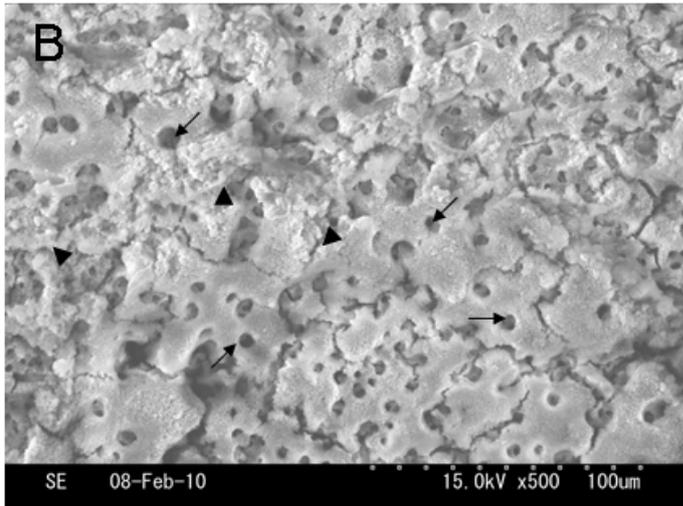


Figure 1B. A scanning electron micrograph of the surface of MTA in the SHilite group. Note globular structures (arrow heads), and woodpecker holes (arrows).

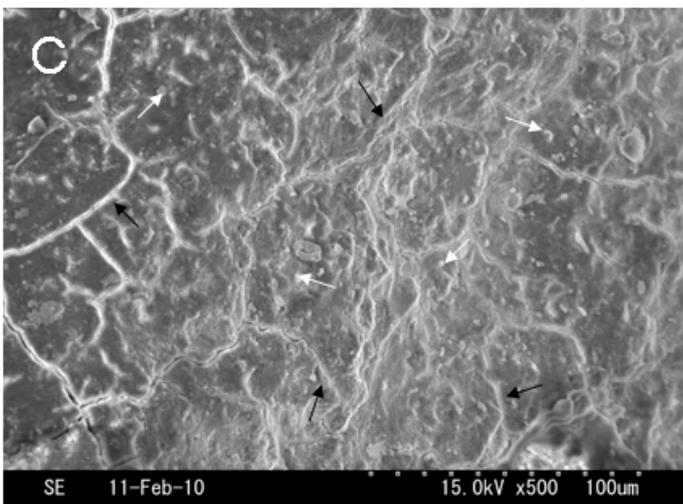


Figure 1C. A scanning electron micrograph of the surface of MTA in the Pyrenees group. Note creases (black arrows), and globular structures (white arrows).