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# The pH and Bismuth Oxide Particle Size can Affect Diametral Tensile Strength of Mineral Trioxide Aggregate

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## ABSTRACT

**Objective:** The aim of the present study was to evaluate the effect of different pHs (4.4, 5.4, 6.4, 7.4, 8.4, and 9.4) and three different particle sizes of bismuth oxide on diametral tensile strength (DTS) of white Mineral Trioxide Aggregate (WMTA).

**Methods:** Thirty cylindrical moulds were divided into six groups of five; WMTA was mixed, placed inside the moulds, and wrapped in pieces of gauze soaked in synthetic tissue fluid (STF) with pH values of either 4.4, 5.4, 6.4, 7.4, 8.4, 9.4. For bismuth oxide, eighteen similar molds were divided into three groups of six (n=6). Then bismuth oxide with three particle sizes, including fine (120 nm), medium (200 nm), and coarse (10 µm), were provided and added to the Portland cement, which did not have any bismuth oxide to create WMTA. Then WMTA was mixed, placed inside cylindrical molds. After incubation at 95% humidity for 48 hours, samples were subjected to DTS testing by an Instron Universal testing machine with a crosshead speed of 1 mm/min. Then, one sample from each group was subjected to scanning electron microscope (SEM) analysis. Data were analysed by ANOVA and Tukey tests (α=0.05).

**Results:** The comparison of DTS in pH groups were: 8.4>7.4>9.4>6.4>5.4>4.4 ($P<0.05$); and in bismuth oxide groups were: fine particles > medium particles > coarse particles ($P<0.05$). Acidic pH, negatively affected the distribution of $Ca^{2+}$ and $Si^{4+}$ ions, while bismuth oxide with fine particles enhanced it.

**Conclusion:** Acidic pH can decline the DTS of MTA significantly. However, reducing the particle size of bismuth oxide can increase the DTS of MTA significantly.

**Keywords:** Bismuth oxide, calcium silicate cement, diametral tensile strength, mineral trioxide aggregate, pH value

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### HIGHLIGHTS

- Special caution must be regarded during application of calcium silicate-based cements (CSCs) in heavy occlusal load bearing areas, where the cement is exposed to acidic pH because of surrounding tissue inflammation.
- The usage of CSCs with nano-sized particles can be regarded as an effective strategy to fortify the cement structure in these clinical conditions.

## INTRODUCTION

Mineral trioxide aggregate (MTA) is a calcium silicate-based cement (CSC), widely used since 1993 (1) because of its unique properties such as biocompatibility, positive bioactivity, antibacterial effect, sealing ability, and pro-angiogenic characteristics (2, 3). However, MTA drawbacks are poor handling characteristics and prolonged setting time (4, 5). Additionally, the sealing properties of MTA can be negatively affected by surrounding tissues in applied areas (6). In most clinical cases, MTA cement is applied in inflamed areas of acidic pH (7). The changes of pH, specifically toward acidic, are shown to induce a negative influence on the properties of MTA, such as compressive strength (8). However, the effect of acidic, neutral, and alkaline pH on other physical properties such as diametral tensile strength (DTS) remains unaddressed.

Tensile strength is an important measure of the mechanical properties of a dental material (8). Direct measurement of the tensile strength of brittle and friable dental materials such as CSCs is difficult. DTS is an easier form of biomaterial measurement (10). DTS is a widely accepted standard method for the measurement of the tensile strength of materials because of the simplicity and reproducibility of outcomes (11, 12). In general, the strength of dental materials is defined as the amount of stress necessary to cause material fracture because of occlusal forces in stress-bearing areas. CSC such as MTA can be subjected to these forces in clinical situations such as a furcation, where MTA is used for the management of furcation perforations (13).

The main ingredients of MTA include Portland cement (a mixture of dicalcium silicate, tricalcium silicate, tricalcium aluminate, and tetra calcium aluminoferrite), bismuth oxide, and gypsum (14). The bismuth oxide is added to MTA as a radiopacifier agent. However, it increases the porosity of the cement, which can negatively affect the longevity of the material (15). Bismuth oxide also reduces the release of calcium hydroxide from MTA and affects the dimensional stability of the set cement (16). One possible solution for minimizing the negative effect of bismuth oxide on physical properties is the reduction of the bismuth oxide particle size. As shown previously, reduction of particle size to nanoscale improved the physicochemical properties of CSCs such as the setting time, microhardness (17), push-out bond strength (18, 19), compressive strength (8), osseous reaction (20), and resistance in acidic environments (6, 21). However, the effect of reducing the particle size on the DTS of CSC materials was not well investigated.

According to this information, this study aimed to evaluate the effect of six different pH values, including 4.4, 5.4, 6.4, 7.4, 8.4, 9.4, and three different particle sizes of bismuth oxide, including fine, medium, and coarse particles on DTS of MTA cement. The first null hypothesis noted that the acidic pH of the storage medium did not have a negative effect on the DTS of MTA. The second null hypothesis noted that the reduction of particle size of bismuth oxide did not increase the DTS of MTA.

## MATERIALS AND METHODS

The present study was performed in two parts to evaluate the effect of pH value and bismuth oxide particle size on the DTS of WMTA cement.

### pH Value

Thirty cylindrical moulds (Scientific Labware Glass, China) with a 4mm diameter and 6 mm height were provided and divided into six groups of five in each (n=5). Pro Root WMTA (Tooth-colored Formula, ProRoot MTA; Dentsply, Tulsa, OK, USA) was provided and mixed with distilled water according to manufacturer instruction, placed inside cylindrical moulds by hand condenser and hand pressure. Then samples were wrapped in pieces of gauze soaked in STF with pH of either 4.4, 5.4, 6.4, 7.4, 8.4, 9.4. The preparation of STF was performed according to a method previously described (7). At first, we prepared STF by using 1.7 g of $KH_2PO_4$, 11.8 g of $Na_2HPO_4$, 80.0 g of NaCl, and 2.0 g of KCl (pH=7.4). This STF solution was divided into six parts, and for acidic pH groups (4.4, 5.4, and 6.4), the pH of STF was adjusted by using butyric acid (pH=4.4), and for alkaline pH groups (8.4 and 9.4), STF was adjusted by using potassium hydroxide (pH=10.4). For the control group, normal saline (pH=7.4) was used. After wrapping with soaked gauze, samples were incubated at 95% humidity for 48 hours before subjecting to DTS measurement. The soaked gauze was refreshed each day during incubation.

### Bismuth oxide particle size

Eighteen cylindrical molds, similar to moulds that were used for the first part, were provided and divided into three groups of six (n=6). Bismuth oxide with a mean particle size of 10 µm (Sigma-Aldrich, 223891 CAS Number: 1304-76-3), 200 nm (Inframat Advanced Materials. Stock 83N-0801), and 120 nm (US Research Nanomaterials, Stock US3788) was added to ProRoot WMTA [Special edition, without radiopacifier], (DENTSPLY Tulsa Dental Specialties, Tulsa, OK) to create a CSC with 20% (wt%) nano-size bismuth oxide in each of the groups. After the addition of bismuth oxide to each group, WMTA was mixed with distilled water according to manufacturer instruction, placed inside cylindrical moulds by hand condenser and hand pressure, and set at 95% humidity for 48 hours.

### DTS measurement

After setting, samples were subjected to DTS measurement by an Instron 85215 (Instron Corp, Canton, MA) testing machine. Before removing the samples from the moulds, they were probed with an explorer to verify the complete setting and solidity of the cement. A compression rate of 1 mm/min was applied, and the load at fracture (MPa) was recorded.

### Scanning electron microscopy (SEM) analysis

After DTS measurement, one specimen from each of pH groups (4.4, 7.4, and 9.4) and the bismuth oxide groups was examined by SEM electron energy-dispersive X-ray spectroscopy mode to analyze elemental distribution. Specimen surfaces were polished and sputter-coated with 10 nm of gold and observed under an SEM (VEGA; TESCAN, Brno, Czech Republic). Energy-dispersive X-ray spectroscopy colour dot map analysis was performed for each sample at X1000 for the evaluation of Ca and Si dispersion as the main constitutions of WMTA.

### Statistical analysis

The data were analyzed by Kolmogorov-Smirnov, one-way ANOVA, and Post Hoc Tukey tests at a level of significance P<0.05.

## RESULTS

### pH value results

The mean±standard deviation of DTS values of pH groups were as follows: 6.10±0.18 (pH=4.4), 7.14±0.61 (pH=5.4), 8.98±0.40 (pH=6.4), 10.36±0.49 (pH=7.4), 11.12±0.63 (pH=8.4), and 10.00±0.60 (pH=9.4). The statistical analysis showed significant differences between experimental groups (P<0.001). The highest DTS was seen in 8.4, and the lowest was seen in 4.4 groups. The values of DTS in 4.4 and 5.4 groups were significantly lower than other groups (P<0.001), while significant differences were not noticed between these two groups (P=0.054). Results of the 6.4 group, was significantly different from all groups (P<0.001), except for the 9.4 group (P=0.061). Results of 7.4 were significantly different from all groups (P<0.001), except for 8.4 (P=0.259) and 9.4 group (P=0.893), respectively. DTS values of the 8.4 group were significantly different from all groups (P<0.001), except for 7.4 (P=0.259). Finally, the results of the 9.4 group were significantly different from all groups (P<0.001), except for 6.4 (P=0.61) and 7.4 (P=0.893) (Fig. 1).

### Bismush oxide particle size results

The mean±standard deviation of DTS values of bismuth oxide groups was as follows: 13.28±0.85 (fine particles), 9.35±0.80 (medium particles), 6.90±0.44 (coarse particles). The DTS value in the fine particles group was significantly higher than medium and coarse particles (P<0.001), and the medium par-

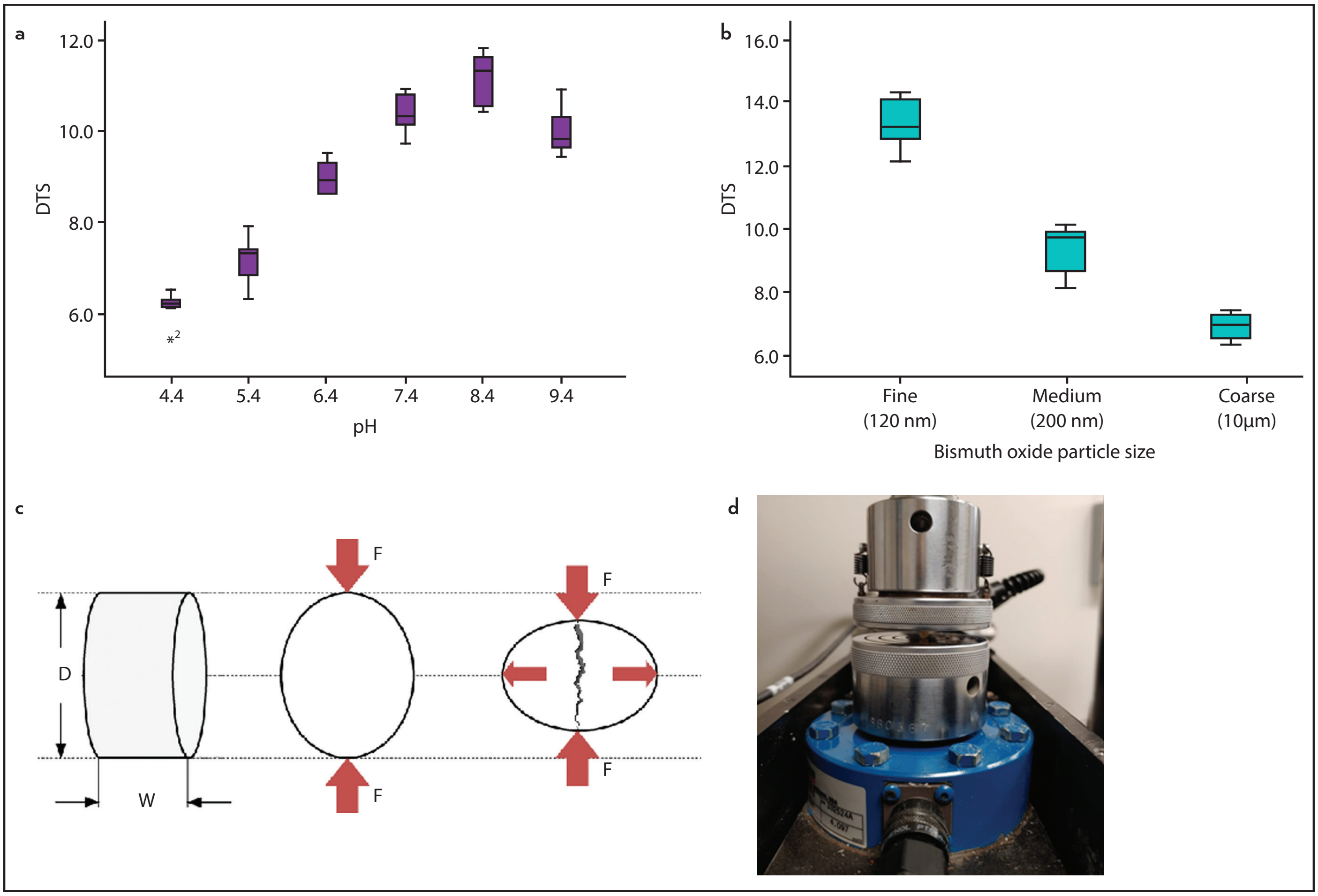


**Figure 1.** (a) The box plots of the means and standard deviations of DTS values of pH groups. (b) The box plots of the means and standard deviations of DTS values of bismuth oxide groups. (c) Schematic of diametral tensile test. (d) Method of placement of Samples for DTS experiment

ticle group's DTS was significantly higher than the coarse particles group (P<0.001) (Fig. 2). Lowest values were noticed in coarse particles group.

### SEM results

The SEM analysis of pH groups samples showed that the reduction of pH value could affect the distribution of Ca and Si in a sample that was exposed to 4.4 pH (Fig. 2a-c). However, these changes were not noted in the other two groups (7.7 and 9.4) (Fig. 2). In addition, regarding bismuth oxide group samples, the SEM colour dot map confirmed the absence of agglomeration of bismuth oxide even at lower particle size (120 nm) (Fig. 2d-f).

## DISCUSSION

The present study intended to evaluate the effect of different pH and bismuth oxide particle size on the DTS of MTA. In the study, both null hypotheses were rejected. As previously mentioned, DTS is the indirect method for the evaluation of the tensile strength of brittle and friable dental materials such as MTA. In DTS measurement, a cylindrical sample is subjected to compressive force in the diametral plane, which is perpendicular to the longitudinal axis. It is shown that most of the mastication forces are compressive forces, and these forces can result in the breakdown of brittle materials such as CSC in occlusal load-bearing areas such as in furcations (12).

MTA is a widely used dental cement (22, 23), which is exposed to different clinical conditions such as different pH values, including acidic or alkaline pH. It has been shown that the pH of tissues could be reduced and become acidic because of infection and inflammation (24). In addition, most local anaesthetics have low pH (3.5-4) to make the anaesthetic solutions more stable and increase shelf life (6). According to this information, butyric acid was used to adjust the pH of the storage medium to become acidic to simulate the clinical conditions where periapical or periodontal tissues have an acidic pH.

The results of the first part of the study showed that samples stored in STF with a pH of 4.4 and 5.4 had the lowest amount of DTS compared with the values of other groups (Fig. 1). These outcomes were consistent with a previously done study. It was indicated that the DTS of white MTA (WMTA) was significantly reduced when it was immersed in a pH 4.0 solution for seven days (25). In addition, similar results were reported by previous studies regarding other physical properties of MTA. By reduction of pH to 4.4, the microhardness of MTA is decreased significantly (26). Also, acidic pH can reduce the push-out bond strength of MTA significantly (27). In the other two similar studies, it is indicated that acidic pH can increase the solubility of WMTA (21) and decrease the compressive strength of WMTA significantly (21). These outcomes can be explained by the fact that acidic pH causes acidic corrosion in the MTA cement

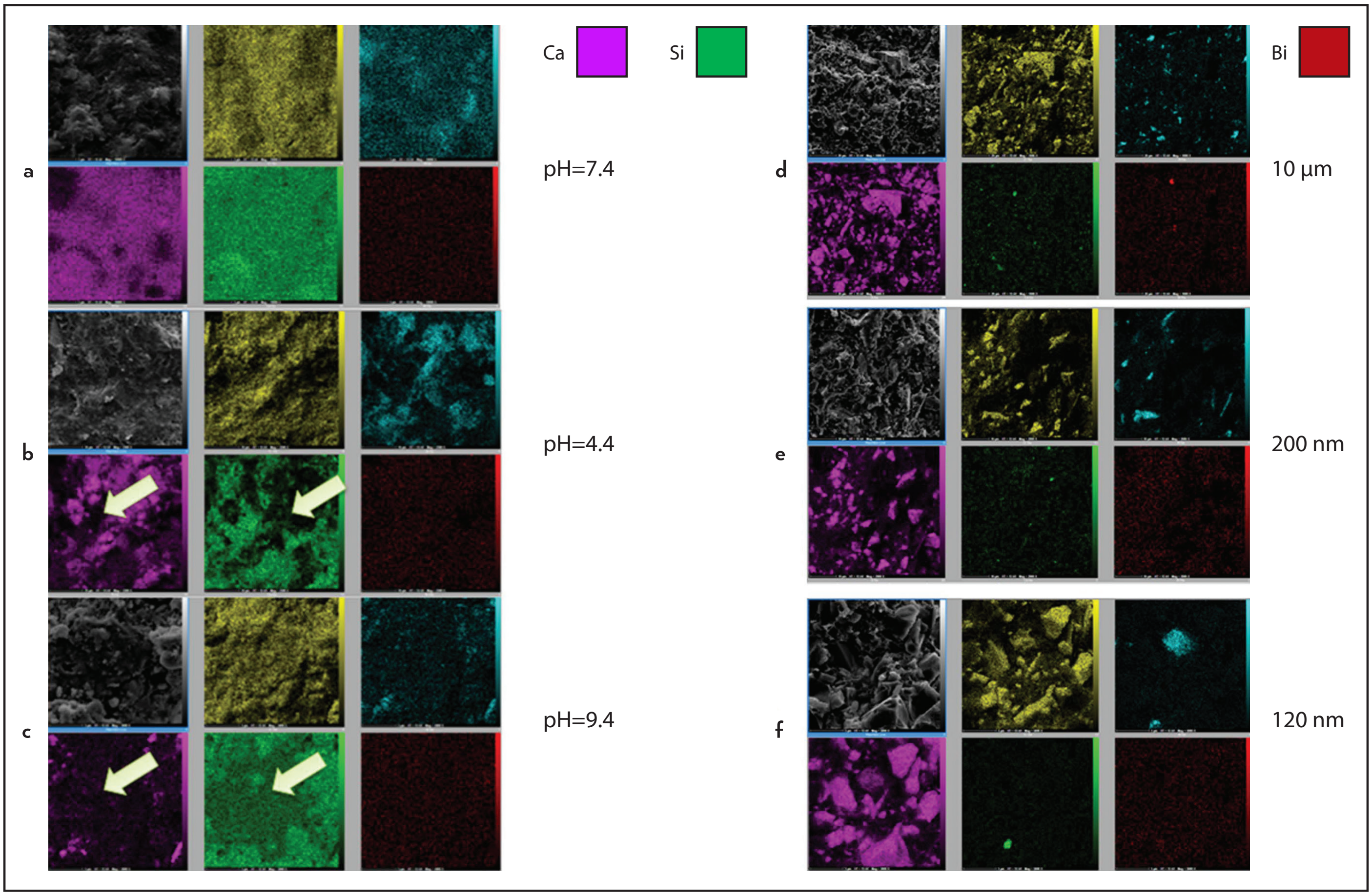


**Figure 2.** Images of SEM analysis of pH groups. (a) sample exposed to STF (pH=7.4) showed normal distribution of Ca and Si, better interlocking, and less porosity over the surface; (b) sample exposed to STF (pH=4.4) showed wide and heterogeneous dispersion of porosity (please note the arrows); (c) sample exposed to pH=9.4 showed normal hydrated products with the normal distribution of Ca and Si, which is similar to sample of 7.4 pH group. (d) Coarse particles (10 µm), (e) Medium particles (200 nm), (f) Fine particles (120 nm). All confirm uniform bismuth oxide dispersion without noticeable agglomeration. It is also showing the number of porosities and poorest integrity of cement particles. Gray (SE), Yellow (O), Blue (Al) Violet (Si), Green (Ca), and Red (Bi)

structure. This corrosion leads to the decomposition of calcium hydroxide, calcium sulfoaluminate, and calcium-silicate-hydrate phases, which increases the porosity of set cement and a decline in the physical properties of MTA (28). In addition to these changes, the SEM analysis of the present study showed that acidic pH could alter the distribution of Ca and Si ions in set cement, which can result in more porosity and a decrease in integration of cement (Fig. 2b). These SEM images can further explain the result of the DTS in acidic pH groups.

The results in alkaline pH groups showed that the DTS amount is increased when MTA is exposed to alkaline pH (8.4) (Fig. 1). These findings are also consistent with previous studies that indicated alkaline pH can increase the microhardness (26) and compressive strength (7) and decrease the solubility of MTA (21). It has been shown in one study that the porosity of MTA cement exposed to alkaline pH is decreased, which can result in superior physical properties in the set cement (26). However, our results showed that higher alkaline pH such as 9.4, can reduce the value of DTS compared with specimens exposed to pH of 8.4 (Fig. 2). This difference might be explained by the dispersion of Ca and Si ions, which are more affected by higher alkaline (pH=9.4) and high acidic pH (pH=4.4 and 5.4). These results were also consistent with the outcomes of SEM analysis, which showed that the Ca and Si ion dispersion in samples in the 8.4 pH group were less affected than the samples in high alkaline (9.4) or high acidic (4.4 and 5.4) pH (Fig. 2b, c).

The outcomes of DTS in bismuth oxide groups showed that the addition of bismuth oxide with fine particle size could increase the DTS significantly compared with medium and coarse particle sizes (Fig. 2). By reducing the particle size of bismuth oxide to the nanoscale, it was noticed that DTS was increased significantly. Similar results were reported in previously done studies, where authors showed that a new modification of MTA, known as Nano WMTA, had superior physical properties, including compressive strength (7), push-out bond strength (17), dislodgement force (18), and lower solubility (21). These results can be explained by the increased surface area and uniform distribution of ingredients of nano-sized-particle cement materials. The increased surface area and better distribution of cement particles resulted in better reaction and hydration of powder and decrease the porosity by the better interlocking of particles, which enhanced the physical properties of set cement (7, 16, 17, 29). These outcomes were further confirmed by the evaluation of images taken in SEM analysis, which showed more homogenous and better dispersion of Ca and Si ions in samples composing of fine particle size of bismuth oxide (Fig. 2f). However, it should be mentioned that besides bismuth oxide, there are other bioinert radiopacifiers such as zirconimum oxide or tantalum pentoxide, which have showed promising results in previous studies (30, 31). It would be beneficial to perform studies similar to the present study with the inclusion of different radiopacifers with different particle sizes in future studies.

## CONCLUSION

According to the limitations of this study, it can be concluded that changing environmental pH can affect the DTS of MTA. Acidic pH can decrease DTS of MTA significantly, while moderate alkaline pH can reinforce the DTS of MTA. Further, the reduction of bismuth oxide particle size can increase the DTS of MTA, suggesting a possible improvement in clinical performance.

### Disclosures

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