Evidence for feasibility of aluminum potassium sulfate (alum) solution as a root canal irrigant


ABSTRACT

Background: The purpose of this study was to evaluate, in vitro, the antimicrobial activity and cleaning efficiency of the aluminum potassium sulfate (alum) solution.

Materials and methods: The antibacterial action of alum solution (1 mg/mL at pH 3.6) against bacterial isolates found in infected root canals, including facultative anaerobic microorganisms (Escherichia coli, Staphylococcus aureus and Klebsiella sp.), and aerobic species (Pseudomonas aerogena ) using agar well diffusion test. The investigation of the debridement and smear layer removing efficiency, on the cervical, middle and apical thirds of root canals of freshly extracted human single-rooted teeth were done by a scanning electron microscopy study.

Results: Alum solution were able to demonstrate antibacterial activity against all the bacteria tested, and produced inhibitions zones of 27, 25, 24 and 22 mm against Staphylococcus aureus, Pseudomonas aerogena , Escherichia coli, and Klebsiella sp. respectively. Alum solution was effective in removing the debris and smear layer. Removal of the smear layer and other debris was more effective in the coronal and middle third than in the apical third.

Conclusion: The findings of this study suggested that alum solution has potential for use as an endodontic irrigant, during chemomechanical root canal preparation.

Key words: antimicrobial, chemomechanical debridement, smear layer removing activity. (J Bagh Coll Dentistry 2012; 24(sp. Issue 1):1-5).

INTRODUCTION

It is known that removal of vital and necrotic remnants of pulp tissues, microorganisms, and microbial toxins from the root canal system is essential for endodontic success. (1, 2) Although this might be achieved through chemomechanical debridement. (3) An uninstrumented area with organic and inorganic debris still present and it is impossible to shape and clean the root canal completely. (4, 5) Therefore, irrigation is an essential part of root canal debridement because it allows for cleaning beyond what might be achieved by root canal instrumentation alone. (6, 7)

During mechanical root canal instrumentation, formation of a smear layer occurs which consists primarily of fine inorganic particles, along with some organic material from necrotic and/or viable pulp tissue, odontoblastic processes, bacteria and blood cells. (8, 9) Despite controversy over maintaining the smear layer, it has been shown that the smear layer itself may contain bacteria and protect the bacteria within the dentinal tubules. The smear layer has also been shown to hinder the penetration of intracanal disinfectants and sealers into dentinal tubules and can potentially compromise the seal of the root canal filling. (10, 11)

Natural products have been used for centuries in treating human diseases and they contain components of therapeutic value. Natural products are environmentally safer, easily available, and cheap. (12) Alum (Aluminum potassium sulfate), the crystallized double sulphates with the formula KAl(SO$_4$)$_2$·12H$_2$O, are generally odourless, colourless crystalline solids that turn white in air, which is used as an astringent and antiseptic in various food preparation processes such as pickling and fermentation and as a flocculants for water purification among other things. (12) Moreover the FDAs over the –Counter Advisory Panel has recommended alum as category I active ingredient in mouthwashes. (13)

Potassium aluminum sulfate (Alum) in concentrations of 10% and 100% is used in mechanico-chemical gingival displacement. (14, 15)

The purpose of this study was to evaluate, in vitro, the aluminum potassium sulfate (alum) solution, in respect to their antibacterial action against common bacterial isolates found in infected root canals, including facultative anaerobic microorganisms (Staphylococcus aureus, Escherichia coli and klebsiella sp), and aerobic bacteria (Pseudomonas aeruginosa), using agar well diffusion test, and to investigate their debridement and smear layer removing efficiency from the prepared root canals on freshly extracted human teeth, by a scanning electron microscopy study.

MATERIALS AND METHODS

This study received approval from the ethics committee of the University of Sulaimani, Sulaimani, Iraq.

Preparation of alum solutions

Seven hundred grams of alum material were purchased from the local botanical market of Sulaimani, and were identified in the College of Science, Department of Chemistry, Sulaimani University. Crystals of alum KAl(SO$_4$)$_2$·12(H$_2$O) dissolved completely in hot (distilled) water at 92°C, to obtain a final concentration of 1 gm/mL, at pH 3.6
Bacterial samples
The bacterial isolates used in this experiment were obtained from the College of Science (Department of Biology), University of Sulaimani. These bacteria were: *Staphylococcus aureus*, *Klebsiella sp.*, *Psedomonas aerogenosa* and *Escherichia coli*. All the isolates were collected from infected root canals and cultivated in suitable culture medium; *Staphylococcus aureus* in Muller-Hinton agar and the other isolates were cultivated in Mackoncky agar.

Evaluation of antibacterial activity of the alum solution
Agar well diffusion method was used to evaluate, in vitro, the antibacterial effect of the alum against the common bacterial isolates found in infected root canals, by means of agar-well diffusion assay. Fifteen milliliters of the molten agar (45°C) were poured into sterile petri dishes (Ø 90 mm), 50 µl from each bacterial isolate (Cell suspensions containing 10⁸ CFU/ml cells), were taken separately and evenly spread onto the surface of the agar plates of Mueller-Hinton agar using a micropipette as described by Bauer et al. Wells (6 mm diameter, and 4 mm height) were bored using a sterile cork borer. Different concentrations of the test solution were placed into the wells and the plates were incubated aerobically and under CO2 incubation at 37°C for 24 h. After 24 hours of incubation, the plates were removed from the incubator and are examined for the inhibition zone around each well (if present), by using the ruler (minimum calibration: 1 mm).

Scanning electron microscopy study
The goal of this part of the study is to assess by mean of scanning electron microscopy, the debridement and smear layer removing ability of alum solution tested as an irrigants, on the cervical, middle and apical thirds of root canals of human extracted teeth. Six freshly extracted human single-rooted teeth with straight roots, mature apex and less than 5 degree curvature, were selected for this part of the study. The crown of each tooth was removed at the cemento-enamel junction using a diamond disk. The working length of each canal was determined by placing and moving a #15 K file apically in the canal until it exited from the apical foramen.

Root canals were manually instrumented according to a step-back type of instrumentation using sequential K-type files up to size #40. First, the root canals were filled with the tested endodontic irrigants through the pulp chamber using 3 ml disposable syringes and 30-gauge needles, which were placed to approximately 3–4 mm from the working length without binding as described by Monika and Izabel. Each time the files were substituted, the canals were thoroughly rinsed with alum solution, aspirated and refilled with a new quantity of this solution.

After final irrigation with 5.0 ml of distilled water to terminate the action and eliminate any precipitates from the irrigants according to methodology described by Manuele et al. The root canals were carefully dried with paper points. Then, by using a diamond disk mounted on a low-speed handpiece, with a constant water spray, longitudinal and transversal grooves, which did not penetrate into the canal, were prepared along the buccal and lingual surfaces of each root. Afterwards, the roots were carefully fractured with the aid of a chisel and a surgical mallet. The cervical, middle and apical thirds were divided, thereby providing three sections from each portion. The roots were mounted on stubs, put in a vacuum chamber, sputter coated with gold-palladium ~35 nm thick with a sputter coater for SEM evaluation. After that an observation with a scanning electron microscope is given.

Specimen grading
Randomly assessment in each third of each half-root at a magnification of 1000x was done. One photomicrograph for each specimen was taken to visualize the coronal, middle, and apical portion of the root canal system. The areas examined for each sample were standardized using parameters similar to those proposed by AL-Hadlaq et al. and Soares et al. with some modifications.

A total of 36 images were analyzed by a calibrated, blinded evaluator using the following scoring system: Score 1, clean surface with very little to no debris, presenting open dentinal tubules throughout the canal wall (figure 1A); Score 2, clean surface with some scattered debris and/or thin homogenous smear layer with some open or partially open dentinal tubules (figure 1B); Score 3, mostly unclean surface containing debris and smear layer with few visible open or partially open dentinal tubules (figure 1C); Score 4, unclean surface with large amount of debris and smear layer with no visible dentinal tubules (figure 1D).
Figure 1: (A) This sample received a score of 1. It shows most of the dentinal tubules are open with a clean surface and very little debris. (B) This sample received a score of 2. It shows a clean surface with very little debris, a thin homogenous smear layer, and some partially open dentinal tubules. (C) This sample received a score of 3. It shows a mostly unclean surface containing debris and smear layer and few open dentinal tubules. (D) This received a score of 4. It shows an unclean surface with large amounts of debris and smear layer with no open dentinal tubules.

RESULTS
Agar well diffusion assay
The mean diameters of the zones of bacterial inhibition for the tested solution against the bacterial isolates are shown in Table 1. Alum solution were able to demonstrate antibacterial activity against all the bacteria tested, and produced inhibitions zones of 27, 25, 24 and 22 mm against Staphylococcus aureus, Pseudomonus aerogenosa, Eschericia coli, and Klebsiella sp. respectively. The results revealed that the wider inhibition zone was seen against Staphylococcus aureus, and the least inhibition zone was against Klebsiella sp.

Table 1: The mean diameters of the zones of bacterial inhibition for the tested solution against the bacterial isolates

<table>
<thead>
<tr>
<th>Bacterial isolates</th>
<th>Inhibition zones (in mm) produced by test solution</th>
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<tbody>
<tr>
<td>Staphylococcus aureus</td>
<td>27</td>
</tr>
<tr>
<td>Pseudomonus aerogenosa</td>
<td>25</td>
</tr>
<tr>
<td>Eschericia coli.</td>
<td>24</td>
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<tr>
<td>Klebsiella sp</td>
<td>22</td>
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Scanning electron microscopy study
The data is summarized in Table 2. The tested solution removed debris and smear layer better at the coronal and middle level than the apical level.

Table 2: The effect of alum solution on the debris and smear layer at the three locations in the root canals

<table>
<thead>
<tr>
<th></th>
<th>Score 1</th>
<th>Score 2</th>
<th>Score 3</th>
<th>Score 4</th>
</tr>
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<tbody>
<tr>
<td>Coronal third</td>
<td>9</td>
<td>3</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Middle third</td>
<td>7</td>
<td>3</td>
<td>2</td>
<td>-</td>
</tr>
<tr>
<td>Apical third</td>
<td>3</td>
<td>4</td>
<td>3</td>
<td>2</td>
</tr>
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</table>

Figure 2: Representative SEM photomicrographs of samples attributed to different root thirds: A) Score 1-coronal third, B) Score 2-Apical third, C) Score 3-Middle third, D) Score-4 Apical third.
DISSCUSSION

A biocompatible irrigant with antimicrobial activity and which removes smear layer along with necrotic and organic debris is desirable, as long as predictable and complete bacterial elimination does not appear to be possible, either with traditional hand instrumentation or with newer rotary NiTi - systems. With the latter at least 35 per cent of root canal surfaces still remain uninstrumented. (24)

This study is the first to report the feasibility of alum as an endodontic irrigant, based on the most important requirements of an ideal root canal irrigant, which are antimicrobial activity, debridement and smear layer removing activity. Two facultative anaerobic bacterial isolates were tested (S. aureus and E. coli), which are best representing endodontic infections and were good models to be tested for antibacterial sensitivity, because these are present in all phases of the development of an infection in root canals. (25, 26)

Another two bacterial isolates (Pseudomonas aeruginosa and Kiebsiella sp.) also tested; they have been isolated in open necrotic root canal system and after contamination of the root canal during the treatment.

In this study, no attempt was made to test all associated organisms, because endodontic infections are polymicrobial, the antimicrobial sensitivity testing of all associated organisms is difficult and produce a great deal of confusing data, also no absolute correlation has been made between any specific microbial species or combination of species with clinical signs and symptoms. (27, 28)

Analysis of the dentinal walls of all the specimens demonstrated that cleaning have been more effective on the coronal and middle thirds than on the apical third. It is possible that the size of the canals in these thirds, allowed better circulation and action of the irrigating solution, making the complete removal of the smear layer and debris more possible. These results are in agreement with those of various authors who have observed an effective cleaning action on these thirds even when different volumes of solutions and times of irrigation were employed. (29, 30)

Based on the results of this investigation, it seems that alum is an effective solution for the removal of the smear layer when used as a final rinse. It does not significantly change the structure of the dentinal tubules. Studies are in progress to determine the efficacy of alum as a root canal irrigant with and without NaOCl for removing the smear layer and completely disinfecting the root canal system.

CONCLUSIONS

Based on the results of this study, it seems that alum solutions have acceptable antimicrobial effect on tested bacterial isolates, however this finding is promising and warrants further laboratory experiments on different types of bacteria, including strict anaerobic and species has been significantly found to persist after treatment procedures. The degree of cleanliness obtained with alum solution (concerning debris and smear layer), was highly satisfactory, however the cleaning effect was more pronounced in the coronal and middle thirds than in the apical parts of the root canals.

Finally, other properties beyond antimicrobial and debridement activity must also be investigated before the final choice of an irrigant solution for clinical use, such as tissue dissolution capacity, and acceptable biologic compatibility.

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