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COMPARATIVE EVALUATION OF THE TISSUE DISSOLUTION CAPABILITY OF 3% OF SODIUM HYPOCHLORITE, 5%POMEGRANATE, AND 5%MISWAK

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ABSTRACT

Objective: This study aimed to evaluate of the tissue dissolution capability of 3% of sodium hypochlorite, 5%pomegranate, and 5% miswak.

Methods: 45 freshly extracted premolar teeth were used**.** The teeth were grouped into three groups (n=15) ***Group I:*** The pulp tissue was immersed in 3% sodium hypochlorite irrigating solution. ***Group II:*** The pulp tissue were immersed in Punicagranatum (5%pomegranate) Nano particles irrigating solution. ***Group III:*** The pulp tissue were immersed in Salvadorapersica (5%miswak) Nano particles irrigating solution.

The pulp tissue obtained was inserted into a small plastic test tube and 10ml of the irrigating solution were added to it and stored for 15 min. The ultrasonic tip were inserted inside the tube with irrigating solution and allowed to work for 1 minute to achieve proper agitation. After 15 min the solution was filtered through absorbing filter paper for 3 min to remove any excess moisture and left the sample to dry. Then the weight of the residual pulp tissue was measured and the difference between the initial and final weight was used to calculate the weight loss that were represent the dissolving power of each solution.

Results: The highest value of dissolution percentage was found in Sodium hypochlorite group (98%), followed by Pomegranate group (78.3%). while the least value was found in Miswak group (66.8%). p= ≤0.001**.**

Conclusion: On contrary of miswak, Pomegranate gives promising results in relation to NaOCl in terms of tissue dissolving capacity.

INTRODUCTION

Cleaning, shaping for three dimensional filling of the root canal are an obligatory procedures for achieving a good results [(1)]. Although clinical preparation is done using clinical preparation of the root canal, complete cleaning of such complex morphology is not achievable due to bacterial contamination [(2)]. Furthermore, bacteria and their byproducts play an essential role in the progress of pulpal and periapical pathologies [(3)].

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One of common species of bacteria that are predominating in endodontic infection is *Enterococcus faecalis* [4]. It is an anaerobic bacteria that can invade the dentinal tubules and it is resistant to irrigating materials and medications [5].

To overcome bacterial contamination, NaOCl solution is considered the irrigant solution of choice due to its anti-bacterial activity and pulp dissolution power [6]. But, it has cytotoxicity effect on the living tissues. So, the need for finding a safe irrigating solution to NaOCl is raised in various studies [7]. Now, herbal products were used as an irrigating solution due to their antibacterial activity, and biocompatibility [1].

Miswak, was used as a chewing stick. Extracts of miswakcontains saponins, flavonoids, salvadorine, traces of tannins, chloride, and trimethylamine, sterol and fluoride [8].

Various herbal extracts like Punica granatum (pomegranate), Chamomile and Echinacea used to treat gingival and periodontal disease due to minimal side effect with high safety [9].

Thus, this study aimed to evaluate of the tissue dissolving capability of 3% of sodium hypochlorite, 5%pomegranate, and 5%miswak.

MATERIALS AND METHODS

Selection and Preparation of Samples

45freshly extracted premolar teeth were used in this study. They were collected from oral and maxillofacial surgery department, Al Azhar University. The selected teeth were extracted for either orthodontic or periodontal reasons after confirming the pulp vitality using thermal test. Before extraction periapical radiographs were taken to exclude any tooth with periapical radiolucency, internal or external root resorption, internal calcifications, root fracture and the teeth that have endodontic thereby. After extraction, the teeth were cleaned from any soft or hard debris on its surface with curette and ultrasonic scaler (Woodpecker WAP-UDSE China) before they used in the study.

Preparation of different irrigant solutions

1. Sodium hypochlorite:

The sodium hypochlorite solution was purchased as readymade solution from the market with concentration of 3%. (Calix. Dharma Research, made in USA. Vendor: Acrostone).

2. Salvadora persica (miswak):

800 g of miswak chewing sticks were cut. The resulting pieces of miswak were ground to powder with food blender. 120mL of 70% ethanol was added to 40 g of powder in a sterile well capped flask, left for 3 days at room temperature and then filtered using number 1 filter paper. The extract then evaporated in a rotary evaporator at 40°C until ethanol removing. The extract stored in sterile screw-capped vials in the refrigerator until needed.

3. Pomegranate peel:

The peel of Punicagranatum fruits was separated manually and extracted with a 90% hydro alcoholic solution, by maceration at room temperature for 5 days in a dark room. The hydro alcoholic extracts were filtered, evaporated under vacuum at 40°C, lyophilized, and kept in a freezer at 10°C.

Nano particles preparation from these extracts by Green synthesis method:

Synthesis process: is initiated by addition of extracts obtained from plant parts into the aqueous solution of metal ions. With the materials present in the plant extract, such as sugar, flavonoid, protein, enzyme, polymer and organic acid, acting as a reducing agent, takes charge in bio induction of metal ions into nanoparticles.The achieved precipitation was placed in a water bath of the temperature of 35ºC [10,11]. within 30 min the color of the solution of samples changed from light yellow to dark brown. Then the sample was washed with 70 % ethanol solution and ultra-pure water respectively and then dried at 150˚C for 8 hrs. Thus nano particles were obtained.

Fig. (1): A photograph showing the irrigating material used in this study: 1) 3% sodium hypochlorite. 2) 5%pomegranate. 3) 5% miswak.

Grouping of the Samples:

Computer software (IBM SPSS Statistics Software) were used to randomly divide the samples according to the used irrigating solution into three groups (n=15) fig. (1).

Group I:

The pulp tissue was immersed in 3% sodium hypochlorite irrigating solution.

Group II:

The pulp tissue were immersed in Punicagranatum (5%pomegranate) Nano particles irrigating solution.

Group III:

The pulp tissue were immersed in Salvadoraper-sica (5%miswak) Nano particles irrigating solution.

Each tooth was notched longitudinally from the mesial and distal sides using Low speed diamond covered disc that was mounted on straight hand piece (Dental NSK Style Low Speed Hand piece 2 Hole Straight. Made in: China SKU: 200444) with coolant to the point just before reaching the pulp tissue. Final separation of the tooth into two longitudinal parts was done using chisel instrument (Dental chisel. RAI-3250. Pakistan) **Fig. (2)**.

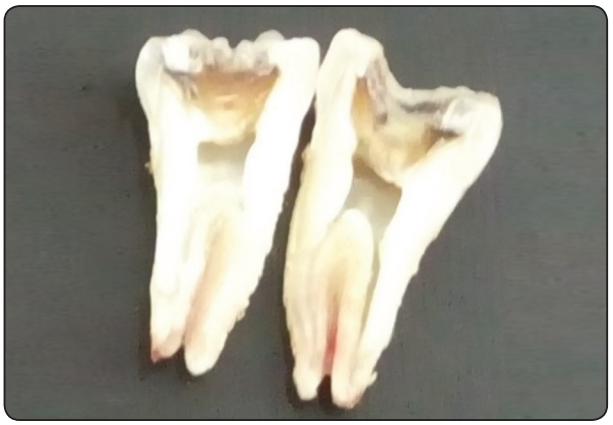

Fig. (2): A photograph showing tooth after splitting with chisel.

Final pickup of the pulp tissue of each tooth was done using tissue forceps (Model Number: OEM. Made in China (Mainland)). The picked pulp tissue was dipped into saline to wash it from blood and debris.

Furthermore, dryness of the pulp tissue from the remaining saline was done using absorbing filter paper. Using the analytical balance ((Sartorius AG WeenderLandstr. 94-10837075, Made in Germany)), the samples of Pulp tissue were standardized to a weight from 0.030 to 0.040 without the weight of filter paper **Fig. (3)**.

Moreover, the pulp tissue were Inserted into a small plastic test tube and 10ml of the irrigating solution were added to it and stored for 15 min. the ultrasonic tip were inserted inside the tube with irrigating solution and allowed to work for 1 minute to achieve proper agitation. After 15 min the

solution was filtered through absorbing filter paper for 3 min to remove any excess moisture and left the sample to dry. Then the weight of the residual pulp tissue was measured and the difference between the initial and final weight was used to calculate the weight loss that were represent the dissolving power of each solution **Fig. (4)**.

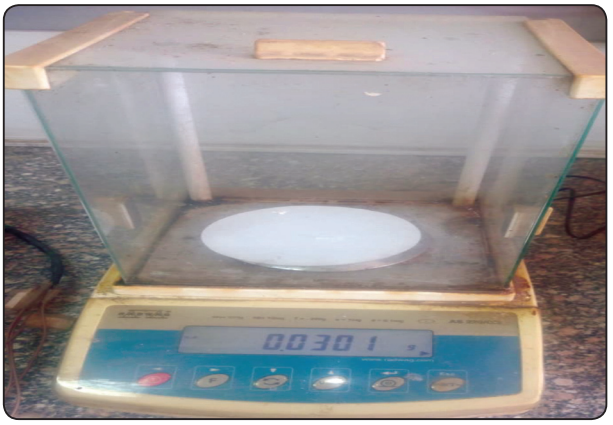

Fig. (3): A photograph showing the analytical balance and pulp tissue on filter paper on it.

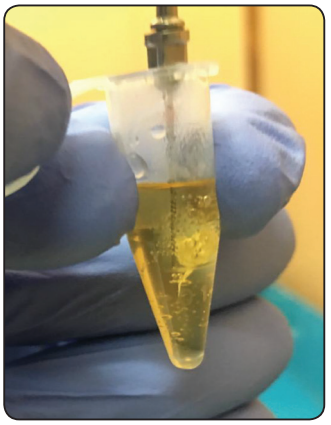

Fig. (4): A photograph showing ultrasonic u file for activation.

RESULTS

The results of this study were listed in table 1 and illustrated in figures (5,6). The results were analyzed by One-way ANOVA test.

The pre-dissolution weight results showed that there was no significant difference among sodium hypochlorite (27 ±19 x 10-3), Pomegranate (27 x 10-3±19 x 10-3) and Miswak (25 x 10-3±14 x 10-3) groups where p=1.000.

The Post dissolution weight results showed that there was a statistically significant difference in dissolution percentage among sodium hypochlorite (0.53 x 10-3 ±0.44 x 10-3), Pomegranate (5.8 x 10-3±3.4 x10-3) and Miswak (8.3 x 10-3±6 x 10-3) groups where p= ≤0.001.

Comparison between tissue dissolution percentages of tested groups. The results in this section were statistically analyzed using One-way ANOVA test.

The highest mean value of dissolution percentage was found in Sodium hypochlorite group (98%), followed by Pomegranate group (78.3%). while the least mean value was found in Miswak group (66.8%). p= ≤0.001 figure (6).

TABLE (1): The mean log10± standard deviation (SD) value and percentage results of comparisons weight reduction in the three groups.

	Sodium hypochlorite	Pomegranate	Miswak	P value
Before	$27 \times 10^{-3} \pm 19 \times 10^{-3}$	$27 \times 10^{-3} \pm 19 \times 10^{-3}$	$25 \times 10^{-3} \pm 14 \times 10^{-3}$	1.000
After	$0.53 \times 10^{-3} \pm 0.44 \times 10^{-3}$	$5.8 \times 10^{-3} \pm 3.4 \times 10^{-3}$	$8.3 \times 10^{-3} \pm 6 \times 10^{-3}$	≤0.001*
Dissolution percentage	[a] 98%	[b] 78.3%	[c] 66.8%	*0.001≥

; significant (p<0.05).

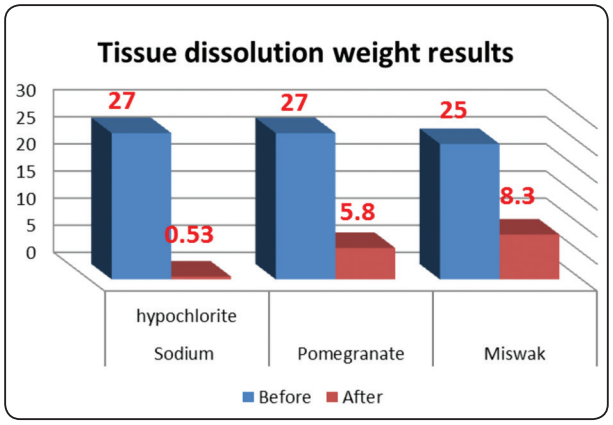

Fig. (5): A column chart of Mean log10± (SD) value of weight reduction for all groups.

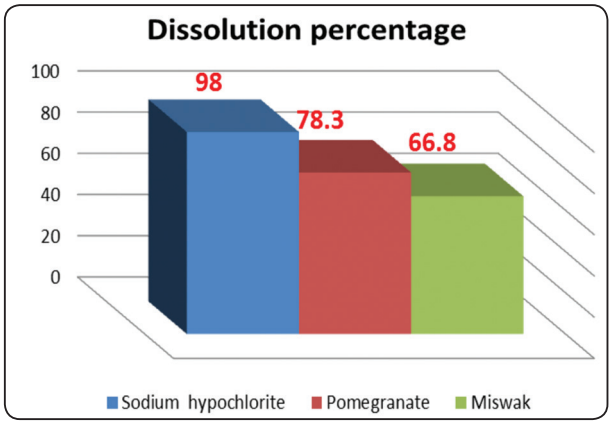

Fig. (6): A column chart of dissolution percentages of the three groups.

DISCUSSION

Elimination the debris from the root canal system is the key to long term success of endodontic therapy. Clinical preparation is one of the most important phases of endodontic treatment[12]. The use of irrigants for ensuring bacterial elimination and eradication of organic tissue remnants[13]. Maximum tissue dissolving effects on the peripheral tissues are some important features of an ideal root canal irrigants. The irregularity of the root canals of teeth negatively affect the success of chemo mechanical endodontic treatment[14].

In This study we concerned to compare the tissue dissolution capability of **3% of sodium hypochlorite**, **5% pomegranate,** and **5% miswak**.

In the present study we used:

3% of sodium hypochlorite, chosen as it is popular to use it as irrigant in endodontic treatment, because of its antimicrobial activity, dissolving power, lubricating action, low cost, and its availability[15].

5% pomegranate, it contains substantial amounts of phenolic compounds, including flavonoids (anthocyanins, catechins and other complex flavonoids) and hydrolyzable tannins (punicalin, pedunculagin, punicalagin, gallic and ellagic acid), punicalagin , ellagic acid and Gallic acid, have scavenging properties, Which account for 92% of the antioxidant activity and also possess anti-proliferative activity, antifungal and antibacterial properties[16].

5% miswak, Its chewing sticks contain trimethyl amine, salvadorime chloride and fluoride in large amounts, showed some antimicrobial activity which make it possible to be used as irrigant solution in endodontic treatment against the endodontic pathogens, It can be used as a substitute for sodium hypochlorite and chlorhexidine as root canal irrigant[17]. It has been shown to have various biological properties including significant antibacterial and antifungal effects. The antimicrobial and cleaning effects of miswak have been attributed to various chemicals which can be detected in its extracts which in turn lead to bacterial death[11].

Tissue dissolving depends on 3 factors: frequency of agitation, amount of available tissue surface area, and amount of organic matter in relation to the amount of irrigant in the system [18].

Each tooth sample was notched longitudinally from the mesial and distal sides using Low-speed

diamond-covered disc that was mounted on straight hand piece with coolant to the point just before reaching the pulp tissue. Final separation of the tooth into two longitudinal parts was done using chisel instrument. Final pickup of the pulp tissue of each tooth was done using tissue forceps[6]. The picked pulp tissue was dipped into saline to wash it from blood and debris.

Dryness of the pulp tissue from the remaining saline was done using absorbing filter paper. The samples of Pulp tissue were standardized to a weight range of 0.030: 0.040 g. by cutting them with the help of No. 15 BP blade. The samples were weighed on an analytical balance.

Inserting the pulp tissue into a small plastic tested vial (falcon tube) which contain 10ml from tested irrigating solution were and stored for 15 min during the storage time the ultrasonic tip were inserted inside the tube with irrigating solution and allowed to work for 1 minute to achieve proper immersion and simulate conditions present in the root canal during biomechanical preparation. After 15 min the solution was filtered through absorbing filter paper for 3 min to remove any excess moisture and left the sample to dry. Then the weight of the residual pulp tissue were measured and the difference between the initial and final weight was used to calculate the weight loss that were represent the dissolving power of each solution[19].

In this study there was no statistically significant difference in weight of pulp tissue before immersion in tested irrigated solution between all groups, this confirmed a standardized starting point for evaluation.

When comparing the weight of pulp tissue after immersion in tested irrigated solution between all groups it was founded that the sodium hypochlorite group (group 1) showed the least weight of pulp tissue when compared to other groups.

This difference was statistically significant. This is in agreement with Atul Jain, etal who is comparatively of human pulp tissue dissolution capacities of different irrigating solutions[6]. And in agreement with Almas K. who is comparatively The Effect of Salvadora Persica Extract (Miswak) and Chlorhexidine Gluconate on Human Dentin[20]. And in agreement with Talita TARTAR. et al who is comparatively Tissue dissolution and modifications in dentin composition by different sodium hypochlorite concentrations[21].

In the present study miswak ethanolic extract at 5% concentration was effective in dissolving pulp tissues, but not more than other groups. It is believed that the heat stable polyphenol tannins present in chewing sticks are responsible for this activity28 Benzyl-isothiocyanate (BIT), which is a naturally occurring compound isolated from S persica, acts by inhibiting bacterial growth and its acid production[22].

In our study, the extracts of Punica granatum 5% concentration was effective in dissolving pulp tissues rather than miswak and less than sodium hypochlorite respectively.

Moreover when comparing the tissue dissolution after immersion in tested irrigating solutions, there was a statistically significant difference in tissue dissolution between all groups, with the highest mean of weight was found in (Group3)(miswak) followed by (Group2)(pomegranates), while the least mean weight was found in (Group1)(sodium hypochlorite). This can be attributed to difference in temperature, pH, the amount of protein substances, concentration of each irrigant solution and the inoculation method tend to influence the results which is required for an ideal irrigant solution.

CONCLUSION

On contrary of miswak, Pomegranate gives promising results in relation to NaOCl in terms of tissue dissolving capacity.

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