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# Tissue Dissolution Ability of Sodium Hypochlorite, EDTA, Ambroxol Hydrochloride and Triphala Evaluated and Compared when used as an Irrigant on Bovine Pulp Tissue

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**Abstract:-**

- ***Aim:***

**The aim of the study is to evaluate pulp tissue dissolution ability of Sodium hypochlorite (NaOCl), Ethylenediaminetetraacetate (EDTA), Ambroxol hydrochloride and Triphala.**

- ***Methods:***

**Pulp tissue of *BOS Taurus primigenius* was extracted from 20 mandibular incisors of 4 bovine jaws .Decoronation was done at the cementum–enamel junction and roots were resected at a length of 3 mm from the apex with a diamond disc connected to a electric micromotor. Pulp tissue was then taken out of the pulp chamber of the bovine tooth with a periodontal probe and cotton pliers. Specimens were weighed using a hermetic precision electronic balance (AND Company LTD) before the test to analyze the initial weight (T0) of the samples. Extracted bovine pulp tissue specimens were weighed and randomly distributed in to 4 experimental groups of 3% sodium hypochlorite, 17% EDTA, Ambroxol hydrochloride(35mg/ml), Triphala(100mg/ml) that were taken in 4 centrifugation tubes. Contents are filtered and undissolved tissue remnants collected and reweighed.**

- ***Results:***

**Results obtained indicate that highest pulp dissolution is for NaOCl, followed by EDTA, Ambroxol Hydrochloride and least for Triphala.**

- ***Conclusion:***

**Highest pulp dissolution is for NaOCl, followed by EDTA, Ambroxol Hydrochloride and least for Triphala.**

***Keywords:-*** *Sodium Hypochlorite, EDTA-Ethylenediaminetetraacetate, AMB-Ambroxol Hydrochloride, Triphala.*

## I. INTRODUCTION

For a successful endodontic therapy cleaning, shaping and chemical disinfection of the root canals are mandatory requisites. Mechanical preparation when done solely cannot end up at disinfecting the complex internal anatomical spaces of the pulp space. Weighing this in mind, manual or rotary root canal preparations should always be aided with irrigating solutions. Irrigation augments bacterial elimination and aids in the debridement of necrotic tissue and dentine shavings from the root canal. Irrigants can avert buildup of the infected debris apically in the root canal and into the periapical area[1].

Sodium hypochlorite (NaOCl) is the irrigant with the maximum pulp dissolution ability and aids in the elimination of elimination of bacteria biofilm. But it is not able to eliminate the inorganic matter. It is used in concentrations varying from 0.5% to 5.25%; it kills bacteria very effectively even at low concentrations[1].

Ethylenediaminetetraacetate is a polyaminocarboxylic acid and is a colorless, water-soluble solid. During the shaping and cleaning of root canals, it contributes to the eradication of the smear layer and thus facilitates the infiltration of irrigants into the dentinal tubules, thus adequate disinfection of the root canals is achieved. Smear layer removal could also improve the permeability of dentinal tubules, thus intensifying the efficacy of intracanal medication, and aids better bonding of the obturating material.[2]

Ambroxol hydrochloride (AMB) is used in pulmonology and interferes directly in the composition of mucus by reducing its viscosity and aiding in its elimination. Until now there are limited studies regarding the pulp tissue dissolution ability of ambroxol hydrochloride.[4]

Triphala, an Ayurvedic meicinal plant comprises dried, powdered fruits of three medicinal plants *Terminalia bellerica, Terminalia chebula,* and *Emblica officinalis* has tannic acid as its principal constituent.[6]

There are limited studies regarding the comparative assessment of tissue dissolution property of sodium hypochlorite (NaOCl), Ethylenediaminetetraacetate (EDTA), Ambroxol hydrochloride and Triphala when used alone as a root canal irrigant.

Hence this study aims to evaluate pulp tissue dissolution ability of sodium hypochlorite (NaOCl), Ethylenediaminetetraacetate (EDTA), Ambroxol

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hydrochloride and Triphala when used as an irrigant on bovine pulp tissue.

## II. MATERIALS AND METHODS

➢ *Pulp Specimen:*

Bovine pulp tissue was extirpated from 20 mandibular incisors of bovine jaws (30– 70 months old) removed from the animal after being slaughtered for the purpose of food production. After slaughtering of the aniaml anterior mandibular teeth were extracted within 48 hours. Those teeth were stored in glass vials with 0.1% thymol solution. Decoronation was done at the cementum–enamel junction and roots resected at 3 mm from the apex with the help of an electric micromotor. Periodontal probe and cotton pliers were used to remove the pulp tissue. The debris and excess of blood were removed by rinsing pulp tissue in distilled water. Specimens were blotted dry for 30 seconds. Hermetic precision electronic balance (AND Company LTD) was used to measure the weight of dried specimens before the test to determine the initial weight (T0) of the samples.[1]

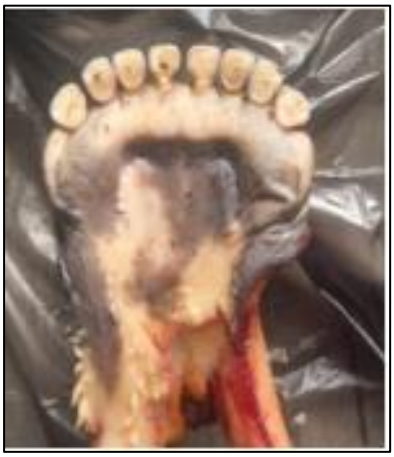

Fig 1 Bovine Mandible

Fig 2 Hermetic Precision Balance

➢ *Groups and Solutions*

The pulp specimens derived from the anterior teeth of the bovine jaw were randomly distributed into 4 experimental groups

- G1—3% NaOCl
- G2— 17% EDTA
- G3- Ambroxol hydrochloride(35 mg/ml)
- G4- Triphala(100mg/ml)


Fig 3 Groups and Solutions

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➢ *Preparation of Ambroxol Hydrochloride Solution*

0.35mg/ml Ambroxol hydrochloride solution is prepared by mixing 0.35 g of ambroxol hydrochloride obtained by crushing contents of 2 capsules with 10ml distilled water.

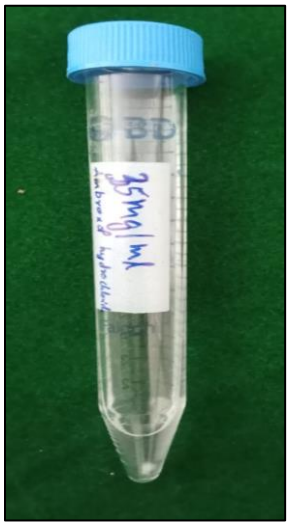


Fig 4 Preparation of Ambroxol Hydrochloride Solution

➢ *Preparation of Triphala*

100 mg/ml solution of Triphala is prepared by mixing commercially available triphala powder (varachoornam)-1g with 10ml distilled water.


Fig 5 Preparation of Triphala

➢ *Tissue Dissolution Method*

Each group of solutions were taken in 4 centrifugation tubes

- G1—3% NaOCl
- G2— 17% EDTA
- G3- Ambroxol hydrochloride (35 mg/ml)
- G4- Triphala (100mg/ml)

Tissue specimens are added into each of the four groups for 10 minutes. After 10 minutes the contents are filtered by using a filter paper. The undissolved tissue remains are collected.2 mL of distilled water was used for washing the samples for 30 seconds to neutralize the actions of the different solutions. Again the samples were blotted dried and reweighed to evaluate remnant tissue.

## III. STATISTICAL ANALYSIS

➢ *Data Profile:*

Table 1 Data Profile

| | GROUP 1 BEFORE | Mean | GROUP 1 AFTER | Mean | GROUP 1 CHANGE PERCENTAGE | Mean |
|---|---|---|---|---|---|---|
| 1 | 0.008 | **0.007 ± 0.0008** | 0.001 | **0.0012 ± 0.0004** | 87.5 | **83.1 ± 4.80** |
| 2 | 0.007 | | 0.001 | | 85 | |
| 3 | 0.008 | | 0.002 | | 75 | |
| 4 | 0.006 | | 0.001 | | 83 | |
| 5 | 0.007 | | 0.001 | | 85 | |
| | **GROUP 2 BEFORE** | | **GROUP 2 AFTER** | | **GROUP 2 CHANGE PERCENTAGE** | |
| 1 | 0.011 | **0.010 ± 0.0016** | 0.005 | **0.0048 ± 0.0004** | 54 | **52 ± 5.29** |
| 2 | 0.012 | | 0.005 | | 58 | |
| 3 | 0.009 | | 0.005 | | 44 | |
| 4 | 0.011 | | 0.005 | | 54 | |
| 5 | 0.008 | | 0.004 | | 50 | |
| | **GROUP 3 BEFORE** | | **GROUP 3 AFTER** | | **GROUP 3 CHANGE PERCENTAGE** | |
| 1 | 0.009 | **0.008 ± 0.0014** | 0.005 | **0.0056 ± 0.0013** | 44 | **36.10 ± 8.29** |
| 2 | 0.008 | | 0.005 | | 37.5 | |
| 3 | 0.011 | | 0.008 | | 27 | |
| 4 | 0.007 | | 0.005 | | 28 | |
| 5 | 0.009 | | 0.005 | | 44 | |
| | **GROUP 4 BEFORE** | | **GROUP 4 AFTER** | | **GROUP 4 CHANGE PERCENTAGE** | |
| 1 | 0.01 | **0.008 ± 0.013** | 0.006 | **0.0052 ± 0.0008** | 40 | **40.70 ± 2.43** |
| 2 | 0.009 | | 0.005 | | 44 | |
| 3 | 0.008 | | 0.005 | | 37.5 | |
| 4 | 0.01 | | 0.006 | | 40 | |
| 5 | 0.007 | | 0.004 | | 42 | |

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➢ *Anova Table*

Table 2 Anova Table

| Groups | Mean ± SD | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|---|
| **Group 1** | 83.1 ± 4.80 | **Between Groups** | 6719.53 | 3 | 2239.846 | 71.219 | **< 0.001** |
| **Group 2** | 52 ± 5.29 | **Within Groups** | 503.2 | 16 | 31.45 | | |
| **Group 3** | 36.10 ± 8.29 | **Total** | 7222.73 | 19 | | | |
| **Group 4** | 40.70 ± 2.43 | | | | | | |

An ANOVA analysis indicated a statistically significant variation in the average percentage change in weight among the four groups ($F(3, 16) = 71.21$, $P < 0.001$). There were significant differences between each group, with only the exception of groups three and four, according to the pairwise comparison Bonferroni test.

Table 3 Bonferroni Pairwise Comparison

| (I) Group | (J) Group | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 31.10 | 3.5468 | **< 0.001** | 20.43 | 41.77 |
| | 3 | 47.00 | 3.5468 | **< 0.001** | 36.33 | 57.67 |
| | 4 | 42.40 | 3.5468 | **< 0.001** | 31.73 | 53.07 |
| 2 | 3 | 15.90 | 3.5468 | **0.002** | 5.23 | 26.57 |
| | 4 | 11.30 | 3.5468 | **0.034** | 0.63 | 21.97 |
| 3 | 4 | -4.6 | 3.5468 | 1 | -15.27 | 6.07 |

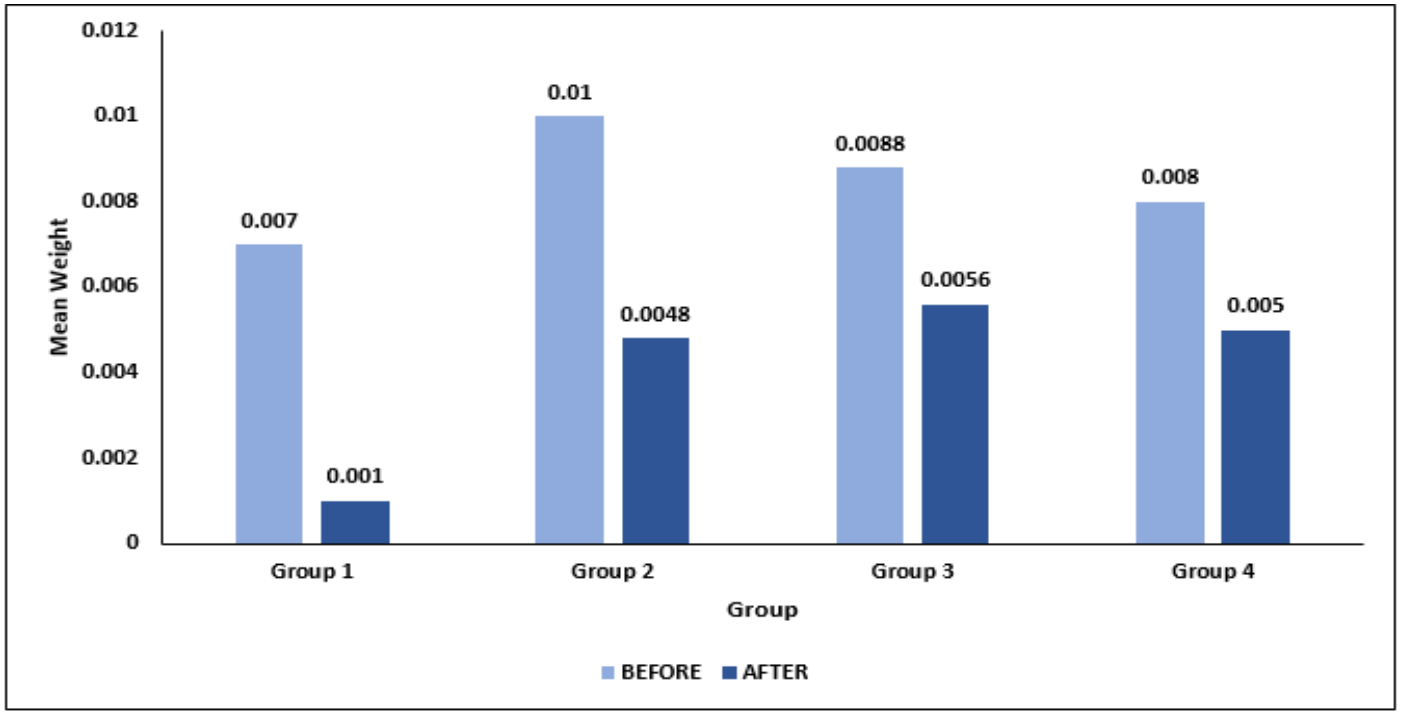


Fig 6 Bar Chart Representing the before and after mean Weight of the 4 Groups

➢ *Sample Size Calculation*

This short study used a convenience sample of 20 samples divided into four groups, each with five participants.

## IV. RESULTS

The mean percentages of remnant tissue over time for each group are shown in table.

When comparing the four groups regarding the average percentage change in weight, there was a statistically significant difference ( $P < 0.001$). There were significant differences within each group, with only the exception of third and fourth groups. Results obtained indicate that highest pulp dissolution is for NaOCl, followed by EDTA, Ambroxol hydrochloride and least for Triphala.

## V. DISCUSSION

Irrigation is considered as an integral part of endodontic treatment, mainly for the purpose of removal of root canal flora. During and after instrumentation, irrigating solutions help in the killing and eradication of microorganisms, inflamed tissue, necrotic and dentine debris.

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Irrigation reduces friction between the instrument and dentine, increases the cutting efficiency of the files, dissolves the remnant pulp tissue, and cools the file and tooth mainly during the activation with ultrasonic tips. Use of irrigants can avoid the chance of periapical extrusion of the hard and soft tissue debris, planktonic and biofilm bacteria.[5]

In this study we used bovine pulp tissue was used due to its similarity to human pulp and it is readily available. Inaddition it can be can be cut into standardized specimens in terms of size and maintains the homogeneity among groups.[1]

The present study assessed the efficacy of NaOCl, EDTA, Ambroxol Hydrochloride and Triphala in dissoluting the pulp tissue.

Sodium hypochlorite (NaOCl) has the most effective tissue dissolving capacity and capability of eliminating bacterial biofilm; but it is doesn't have the ability to remove the inorganic matter. NaOCl dissociates in water into sodium (Na+) and the hypochlorite ions, OCl–. It maintains an equilibrium with hypochlorous acid (HOCl). Most of the chlorine exists as HOCl at acidic and neutral pH. But when the pH is nine and above, OCl– is most common ion present. Hypochlorous acid has the strongest antibacterial effect while the OCl– ion is less effective. Hypochloric acid affects directly on the vital functions of the microbial cell, rapidly resulting in cell death.

NaOCl is usually used in the concentrations of range between 1%–6% as an endodontic irrigant. The properties and efficacy of NaOCl depend on its concentration, temperature, pH, frequency and intensity of mechanical agitation, solution refreshment and the surface area of the tissue which is exposed to the irrigant.

EDTA has a chelator kind of action, which is used after irrigation with NaOCl as the final irrigant. EDTA solution has a neutral or slightly alkaline Ph. At acidic pH EDTA has the property of precipitation. EDTA is usually used as a 17% or 15% solution as a root canal irrigant. The recommended time for smear layer removal is around two minutes, but thick layers may require longer times of exposure.

EDTA has the capability to dissolve the inorganic part of dentine and smear layer (hydroxyapatite). But complete removal of the smear layer is achieved only when the canal has been irrigated with NaOCl before the final rinse with EDTA. EDTA has little or no antimicrobial activity. But according to some studies they have got some antifungal activity. However, EDTA has the ability to weaken the bacterial cell membrane but doesn't have the capacity to kill the bacterial cell. However it may work in a synergistic manner when used along with other chemicals.

Ambroxol hydrochloride (AMB) is also used in pulmonology and interferes directly in the composition of mucus by reducing its viscosity and aiding in its elimination. However, no published study on its use in combating endodontic bacterial biofilm exists in the literature. AMB is used in the study as a mucolytic substance that can act directly on the polysaccharide layer of endodontic biofilm, optimising the action of intracanal medication and irrigating solutions.[4]

Triphala fruits is a plant-derived composition developed and most commonly used in India; the powder is a combination of three dried plants naming Terminalia bellerica, Terminalia chebula and Emblica afficinalia. Tanic acid is the main constituent of this composition. It has been used in Indian traditional medicine for treatment of some ailments like headaches, constipation and hepatic disorders etc. Many studies have concluded that it has got bacteriostatic or bactericidal effect on gram-positive as well as gram negative pathogens. It is safe to use and is composed of compounds with proper physiologic effects along with its anti-oxidative and anti-inflammatory properties. The main advantages of Triphala include its easy access, low cost, long-term substantivity, less toxicity and the property tom avoid microbial resistance.[6]

## VI. CONCLUSION

Instrumentation and irrigation are the most vital processes included in root canal treatment procedure. Irrigation has got so many important functions, the most important of which are to dissolve tissue and to have an antimicrobial effect against the microbial endodontic flora. Results obtained from the study indicate that highest pulp dissolution is for NaOCl, followed by EDTA, Ambroxol hydrochloride and least for Triphala.

Antibacterial efficacy of the respective irrigants should also be considered when they are being used in the root canal system as irrigants.

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