

REVIEWER'S REPORT

Manuscript No.: IJAR-52562

Date: 01/07/2025

Title: COMPARATIVE EVALUATION OF HERBAL IRRIGANTS ON MICROHARDNESS OF DENTIN- AN IN VITRO STUDY.

Recommendation:

Accept as it is

Accept after minor revision.....Yes.....

Accept after major revision

Do not accept (*Reasons below*)

Rating	Excel.	Good	Fair	Poor
Originality	•			
Techn. Quality		•		
Clarity		•		
Significance	•			

Reviewer Name: Dr. Sireesha Kuruganti

Date: 01/07/2025

Reviewer's Comment for Publication.

(To be published with the manuscript in the journal)

The reviewer is requested to provide a brief comment (3-4 lines) highlighting the significance, strengths, or key insights of the manuscript. This comment will be Displayed in the journal publication alongside with the reviewers name.

The manuscript, "COMPARATIVE EVALUATION OF HERBAL IRRIGANTS ON MICROHARDNESS OF DENTIN- AN IN VITRO STUDY," presents an interesting and relevant topic in endodontics, exploring natural alternatives to conventional chemical irrigants. The study design is generally clear, and the methodology is adequately described.

Detailed Reviewer's Report

Here's an in-depth review of the manuscript, with specific line numbers mentioned where applicable:

General Comments:

The manuscript, "COMPARATIVE EVALUATION OF HERBAL IRRIGANTS ON MICROHARDNESS OF DENTIN- AN IN VITRO STUDY," presents an interesting and relevant topic in endodontics, exploring natural alternatives to conventional chemical irrigants. The study design is generally clear, and the methodology is adequately described. However, there are several areas that require clarification, more detailed explanation, and improvement in presentation to enhance the scientific rigor and readability of the manuscript.

Specific Comments and Suggestions:

Title:

* Line 1-2: The title is clear and concise.

Abstract:

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* Line 4: "Common chemicals, such as 17% EDTA and sodium hypochlorite, are used to remove the smear layer." While true, it's important to clarify that NaOCl primarily dissolves organic tissue and CHX is an antimicrobial, while EDTA is the chelating agent specifically for the smear layer's inorganic components. This distinction is crucial for understanding their individual roles.

* Line 14: "Chlorhexidine showed the highest reduction in dentin microhardness, followed by Green Tea, Triphala, Neem, Tulsi, and Distilled water." This sentence is a direct result. The abstract should ideally present the statistical significance of these findings or at least indicate if the differences were significant.

* Line 15: "Within limitation of this study it can be concluded that herbal showed less effect on microhardness of dentin." This conclusion is a bit vague. "Less effect" compared to what? It would be stronger to state "less reduction" or "maintained microhardness better than chlorhexidine."

Introduction:

* Line 17-18: "Complete eradication of bacteria and their byproducts is essential for successful endodontic therapy. This is achieved through mechanical instrumentation and primarily with antimicrobial irrigants." Clear and well-stated.

* Line 19: "Commonly used irrigants include sodium hypochlorite (NaOCl), chlorhexidine gluconate (CHX) and EDTA." Good to cite common irrigants.

* Line 20: "Chemical irrigants can alter dentin's composition, affecting microhardness and the adhesion of dental materials." This is a key point and well-introduced.

* Line 21-23: "Triphala derived from Terminalia chebula, Terminalia bellerica and Emblica officinalis, contains tannic acid with antibacterial and anti-inflammatory properties." This should be moved to the "Preparation of the Test Solutions" section or the discussion, as it's a detail about the specific irrigant rather than a general introduction to herbal alternatives. The citation here is to the beginning of the line itself which is incorrect based on the guidelines.

* Line 46-54: The descriptions of Triphala, Green Tea, Neem, and Tulsi and their properties are good, but their placement in the Introduction (specifically on page 2) feels slightly out of place. This information might be better suited for the "Preparation of the Test Solutions" section or a dedicated "Background on Herbal Irrigants" sub-section if expanded upon.

* Line 55-58: "Despite their efficacy, chemical irrigants may negatively affect dentin microhardness, increasing fracture risk. Thus, safer alternatives like herbal irrigants Triphala, Green Tea, Neem and Tulsi are being explored for their low toxicity and high antimicrobial potential." This effectively sets the rationale for the study.

Material and Methods:

* Sample Preparation (Lines 61-67):

* Line 63-64: "Freshly extracted 30 permanent mandibular premolars extracted tooth with completely formed roots were collected." "Extracted tooth" is redundant. Just "Freshly extracted 30 permanent mandibular premolars with completely formed roots were collected."

* Line 64-65: "Teeth were cleaned with the help of ultrasonic scaler followed by storage in saline solution until the start of the experiment." Specify the type of saline solution (e.g., physiological saline).

* Line 65-66: "Teeth were decoronated at the CEJ with diamond disc." The type/size of the diamond disc should be specified for reproducibility.

* Line 67: "The surface of the specimens were smoothened using a fine gritted silicon carbide abrasive papers 180,320,600,800,1000 grit followed by polishing of the specimens were done with alumina." This is very detailed and good for reproducibility.

* Preparation of the Test Solutions (Lines 71-84):

* Line 71-73 (Triphala): "500 mg of Triphala powder was dissolved in 10 ml of 10% DMSO. Sterile water was added to make up a final volume of 100 ml." Specify if the Triphala powder was standardized or a commercial product. If commercial, provide the manufacturer. "Sterile water was added to make up a

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final volume of 100 ml" implies a 0.5% w/v solution. It's better to explicitly state the final concentration (e.g., "resulting in a 0.5% w/v Triphala solution").

* Line 75-77 (Green Tea Extract): "5 grams of green tea extract was added to 100 ml of boiling sterile water, steeped for 5 minutes, and then filtered using filter paper." Specify the type of green tea extract (e.g., brand, form). Was it pure extract or just green tea leaves? If leaves, indicate the type of leaves. If a commercial extract, state the concentration or purity. "Boiling sterile water" is good.

* Line 79-82 (Neem and Tulsi): "100 grams each of dried neem and tulsi leaves were tied in a cloth pouch and soaked in 800 ml of distilled water. The pouch was boiled gently over a low flame. After cooling, the pouch was removed, and the solution was filtered to obtain a clear liquid." This is a good description of the extraction process.

* Line 83: "A 25% concentration was achieved by measuring the final volume post-filtration." How was the 25% concentration verified? Was it based on initial weight of leaves to final volume, or was a specific constituent analyzed for concentration? This needs clarification for scientific accuracy.

* Grouping of Samples (Lines 85-92):

* Line 85: "Based on the irrigating solutions, specimens were randomly divided into 6 groups of 10 samples each, namely-" Good to state the sample size per group (n=10).

* Line 87: "Group I-2% Chlorhexidine [Postive control]" - Typo: "Positive control."

* Line 92: "Group VI - Distilled water [Negative control]" - Good to specify controls.

* Line 93: "Each sample was immersed in respective irrigating solution for 15 minutes." This immersion time is consistent with the discussion (Line 150).

* Microhardness Testing (Lines 96-101):

* Line 97-98: "Specimens were tested for microhardness using Vickers Microhardness test with a 200g load with a dwell time of 20 seconds." Good to specify the load and dwell time.

* Line 98-100: "Specimen were horizontally mounted on Vickers testing machine and three indentations for each specimen were made on the coronal, middle and apical thirds of the root." This ensures comprehensive evaluation across the root.

* Line 101: "Mean length of the diagonals of indentations were recorded and computed." Standard procedure.

* Statistical Analysis (Lines 111-115):

* Line 111: "The data were analysed using statistical package for social sciences version (SPSS) 20.0." Good to state the software and version.

* Line 112: "The level of statistical significance was set at 95% (P=0.05)." Standard.

* Line 112-113: "P-value > 0.05 was non-significant." Clear.

* Line 113-114: "The data of the present study were subjected to statistical analysis to interpret the differences and significance among groups." Good general statement.

* Line 114-115: "One-way ANOVA test was applied to compare measurements of microhardness values among six study groups at each root level." Appropriate test.

* Line 115: "Post hoc tukey test analyses multiple pair wise individual group comparisons." Appropriate post-hoc test.

Results:

* Line 117-123: "The microhardness values across all groups were highest at the apical region, followed by the middle, and lowest at the cervical region. Group I showed the highest value at the apical (57.90), with the lowest at the cervical (54.90). Group II also had the highest at the apical (58.20), but the lowest at the middle region (54.90). Groups III, IV, and V followed a similar trend, with peak values at the apical (61.10, 60.70, and 61.00 respectively) and the lowest at the cervical. Group VI showed the highest at the apical (61.30), followed by cervical, and lowest at the middle (57.30)." This descriptive summary of the results is clear and easy to follow.

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* Line 124-125: "Overall, the apical region recorded the highest mean microhardness (60.03), with the middle (57.23) and cervical (56.82) showing progressively lower values as seen in TABLE 1" Good summary.

* Table 1 (Line 126-127):

* The table is well-formatted and presents the data clearly.

* "STD. DEVIATION" is good.

* The " 0.039^{**} ", " 0.041^{**} ", and " 0.004^{**} " indicate statistical significance, which is crucial. It would be beneficial to add a footnote or a legend below the table explaining what the asterisk denotes (e.g., " $P < 0.05$ ").

* Graph 1 (Line 129):

* The bar graph provides a good visual representation of the data presented in Table 1.

* The axes are clearly labeled.

Discussion:

* Line 135-139: "The microhardness of the dentin is influenced by many factors such as mineral content as the quantity of hydroxyapatite in the intertubular substance, tubular density along with diameter of tubule." Good explanation of factors influencing microhardness.

* Line 140-142: "Dentin being a biological construction is not homogeneous the density of dentin tubules increases as one moves from the dentino-enamel junction in coronal part of dentin towards the pulp chamber and in radicular part of dentin, it induces from the cervical to apical area." This explanation of dentin homogeneity and tubular density is relevant. "induces" should be "increases" or "decreases" - it's a typo/grammatical error. Based on line 212, it increases from cervical to apical. So, "it increases from the cervical to apical area."

* Line 143: "Hardness correlates with resistance to fracture, modulus of elasticity, yield and bond strength." Important clinical implication.

* Line 144-146: "Although a change in composition of dentin makes the instrumentation process easier during the root canal treatment, it also compromises the root construction by decreasing the microhardness." This highlights the trade-off.

* Line 147-148: "As a result, teeth that have undergone root canal therapy are more likely to break. 9" The "9" at the end is likely a stray character or an incorrect citation format.

* Line 150-151: "All samples were submerged in their designated irrigating solutions for 15 mins prior to undergoing microhardness testing. Goldberg et al. recommended an application duration of 10-15 mins to achieve optimal outcomes that aligns better with practical clinical settings.8" Good justification for the immersion time. The "8" at the end is likely a stray character or an incorrect citation format.

* Line 152-154: "The choice of the Vickers microhardness tester over the Knoop hardness tester was based on the Vickers test's greater suitability and practicality for assessing surface alterations in deeper hard tissues of teeth. In contrast the Knoop hardness tester is typically utilized for evaluating superficial dentin at a depth of 0.1 mm rather than for deeper dentin.2" Good justification for the choice of microhardness tester. The "2" at the end is likely a stray character or an incorrect citation format.

* Line 155-156: "Distilled water was utilized as a control because it does not make any chemical alteration on dentin.11" The "11" at the end is likely a stray character or an incorrect citation format.

* Line 162-163: "All the irrigants used that had not observed any significant impact on microhardness reduction of radicular dentin." This statement seems to contradict the conclusion in the abstract (Line 14), which states Chlorhexidine showed the highest reduction. If the reduction was not statistically significant for all irrigants, this needs to be clearly stated and reconciled with the abstract and results. Looking at Table 1, the asterisk indicates significance for all three regions (cervical, middle, apical). This sentence requires significant revision or removal as it seems to be contradictory to the results and other statements. The phrasing "had not observed any significant impact" is also a bit awkward. It should be "did not show a significant impact."

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* Line 164-165: "In this study, it was also obtained that CHX decreased the dentin microhardness. The result was in accordance with Oliveira et al., the microhardness of the dentin treated with CHX (2.0%) was reduced. CHX being a cationic chemical, which has the capability to bind to anionic molecules viz., the phosphates in the hydroxyl apatite structure, which may change the Ca/P ratio and help to explain the decreased microhardness of the dentin when exposed to CHX. 12" The "12" at the end is likely a stray character or an incorrect citation format. This explanation for CHX's effect is good.

* Line 170-171: "Smear layer may play a role as barrier, which allows for only minimal alterations in microhardness by limiting irrigant interaction with the dentin and it reduced the Ca and P levels along with the microhardness of root dentin." This sentence is a bit convoluted. Consider rephrasing for clarity, e.g., "The smear layer may act as a barrier, limiting irrigant interaction with dentin and thus allowing for only minimal alterations in microhardness, which also correlated with reduced Ca and P levels and root dentin microhardness."

* Line 173: "18 Amin et al. reported reduction of microhardness with the use of CHX. 13" The "18" and "13" are likely stray characters or incorrect citation format.

* Line 174-175: "The current study also found that the green tea extract and neem did not significantly reduce the microhardness of dentine was similar to the study of Durgavandi et al. and they also reported that green tea and neem had no significant impact on microhardness of the dentin..." This contradicts the statement on Line 14 in the abstract and the overall trend of results, where Green Tea did show a reduction, albeit less than CHX. This needs clarification and reconciliation. If the difference between them and distilled water was not significant, that's one thing, but saying they "did not significantly reduce" needs context.

* Line 175-176: "...both the neem and tulsi have a neutral pH of 6.8 and 6.3, respectively. The dentin which contains 22% organic material have a neutral pH value less than 5.5." The phrasing "have a neutral pH value less than 5.5" is contradictory. A neutral pH is 7.0. A pH less than 5.5 is acidic. This statement needs correction. The pH of dentin should be clarified or removed if not directly relevant to the mechanism of action of the irrigants here.

* Line 177: "Although green tea and neem observed slight variation in pH value, but these were not statistically significant in the study. 14" The "14" is likely a stray character or an incorrect citation format.

* Line 178-180: "Nikhil et al. found green tea decrease the microhardness because of presence of catechins. Catechin a polyphenol with known anti-inflammatory, antioxidant and anticarcinogenic properties. These catechins with an acidic nature may contribute to demineralization of dentin. 15" The "15" is likely a stray character or an incorrect citation format. This provides an alternative perspective on green tea's effect.

* Line 181-183: "The findings of Mirkarimi et al. in contrast observed that green tea extract enhanced the microhardness of eroded dentine and there was an improvement of its texture. Moreover, green tea extract mostly permitted to form a surface deposition by organic components on the dentin, or it could be ascribed to the existence of afresh induced collagen crosslinks. Proanthocyanidin is well-known combination of monomers, oligomers and polymers of flavan-3-ols (catechines), which are extensively exist in extract of green tea and might be interacted with the organic part of the dentin. 58" The "58" is likely a stray character or an incorrect citation format. This section highlights the conflicting findings regarding green tea, which is good for a discussion.

* Line 190-191: "Triphala has demonstrated a lesser decrease in the dentin microhardness when compared to CHX and green tea. The similar result was observed in a comparative study after the usage of NaOCl (5.0%) and EDTA (17.0%)." The comparison to NaOCl and EDTA is relevant.

* Line 192: "The most probable reason for this might be because of the citric acid present in the Triphala fruit, which plays a role on weak chelation. 17" The "17" is likely a stray character or an incorrect citation format. Good explanation for Triphala's effect.

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* Line 193-195: "Tulsi exhibited comparable anti-microbial efficacy due to its active constituents such as Eugenol, Ursolic acid, Carvacrol and Oleanolic acid. Eugenol (1-hydroxy-2-methoxy-4-allylbenzene) the primary potential component in *Ocimum sanctum* L. (Tulsi) were identified as a major contributor to its therapeutic properties. 6" The "6" is likely a stray character or an incorrect citation format.

* Line 207-208: "Maria Philip et al. found the dentin microhardness of induced from the cervical to the apical regions." "induced from" should likely be "increased from" or "varied from." This sentence is incomplete or grammatically incorrect. This point is already covered in the results section (Lines 117-118).

* Line 209-211: "Durgavandi et al. on comparing with different herbal irrigants at cervical, middle and apical third.19 The dentin microhardness decreased as per tubular density increased likely due to a reduction in the quantity of intertubular dentin along with an expansion of tubular diameters individually. 20" The "19" and "20" are likely stray characters or incorrect citation formats. The explanation for the decrease in microhardness with increased tubular density is good.

Conclusion:

* Line 216-218: "The limitation of this study is that the experimental group condition comprised in this research varied considerably as per the clinical conditions. Furthermore, other properties such as biocompatibility, staining and substantively are necessary towards the efficient intracanal irrigation." Well-stated limitations and future research directions.

* Line 220-223: "Many in-vivo and in vitro studies are required to evaluate the efficacy of plant extracts like green tea tea, triphala, neem and tulsi to be used as endodontically irrigation clinically." "green tea tea" is a typo. Should be "green tea". This statement reinforces the need for further research.

References:

* General: The referencing style appears inconsistent with the "" format used in the manuscript text itself. The numerical superscript (e.g., ¹, ², ³, etc.) in the introduction (Line 18, 38, 40) does not match the bracketed format.

* Specific issues with stray numbers at the end of sentences: As noted throughout the review, many sentences in the discussion have a single number (e.g., "9", "8", "2") at the very end, directly following the in-text citation, or sometimes as the only citation. This indicates a formatting issue. These should either be removed if they are stray characters, or correctly integrated into the citation format "" if they are meant to be additional citations.

* Formatting of References: The references themselves (Lines 227-287) appear to be a standard numerical list. Ensure consistency between the in-text citation format and the reference list.