

Formulation and in-Vitro Characterization of a Novel Herbal Tooth Serum With Activated Charcoal and Cocos Nucifera Extracts for Enamel Remineralization

Saathvika Srinivasan ¹, Nivethigaa Balakrishnan ², Aravind Kumar Subramanian ³

¹ Saveetha Dental College and Hospital, Saveetha Institute of Medical and Technical Sciences, Chennai - 600077, Tamil Nadu, India. 152308004.sdc@saveetha.com

² Saveetha Dental College and Hospital, Saveetha Institute of Medical and Technical Sciences, Chennai - 600077, Tamil Nadu, India. nivethigaab.sdc@saveetha.com

³ Saveetha Dental College and Hospital, Saveetha Institute of Medical and Technical Sciences, Chennai - 600077, Tamil Nadu, India. aravindkumar@saveetha.com

KEYWORDS

activated charcoal, Cocos nucifera, herbal tooth serum, remineralization, physical properties, sustainable resources

ABSTRACT

Background: An ideal novel herbal tooth serum should possess potent antiplaque and remineralizing properties, effectively combating dental plaque buildup while promoting the restoration of tooth enamel. It should be free from powerful chemicals, artificial flavors, or dyes, ensuring a natural and gentle oral care experience. With a balanced abrasive effect, it must efficiently remove surface stains without causing irritation or toxicity, thus safeguarding oral health.

Aim: Combining the efficacy of various natural resources, this study aims to formulate and assess the physical properties of a novel tooth serum incorporating activated charcoal and Cocos nucifera extracts.

Materials and Methodology: The herbal tooth serum was prepared by using the dry gum method. The novel herbal tooth serum incorporates activated charcoal and extracts of *Cocos nucifera* as a remineralizing agent alongside calcium silicate and hydroxyapatite, facilitating the natural mineralization of enamel. Gentle abrasives such as silica and calcium carbonate ensure effective yet mild cleaning action, while limonene, an antibacterial essential oil, enhances oral hygiene. The formulation is fortified with cinnamon and thyme extracts, serving as antioxidants, and sorbitol functions as a natural sweetener. The base ingredients include aqua, glycerin, and cellulose gum, providing a stable and user-friendly activator gel. Other powdered ingredients are shifted together and added gradually to the mucilaginous mixture with continuously gentle stirring in a conical flask. The exact proportion of the tooth serum ingredients for 100 g of tooth serum was measured and taken, and physical properties were evaluated.

Results: The physical properties, such as color, homogeneity, spreadability, foamability and pH, were determined and evaluated. The color was black and stable with no staining; the mixture was smooth with no abrasive particles, and the pH was optimum for intraoral usage.

Conclusion: A herbal tooth serum with activated charcoal and *Cocos nucifera* extracts was formulated with ideal physical properties that could serve as a natural alternative to synthetic tooth serums used in dentistry for enamel remineralization.

1. Introduction

Decalcification of the enamel surface adjacent to fixed orthodontic appliances is an important and prevalent iatrogenic effect of orthodontic therapy. The banding and bonding of orthodontic appliances to teeth increases the plaque retention sites, and as a result, oral hygiene becomes more difficult. The low pH of plaque adjacent to orthodontic brackets hinders the remineralization process, and decalcification of enamel can occur. As enamel translucency is directly related to the degree of mineralization, initial enamel demineralization usually manifests itself as a “white spot lesion” [1].

Testing and diagnosing these lesions involve various methods to assess their presence, severity, and potential for progression. This includes direct visual examination, fiber optic transillumination, laser-induced fluorescence, microhardness testing, and digital x-rays. Previous studies have shown that the extent of WSLs depends on the duration of orthodontic treatment. Increased duration of orthodontic treatment was associated with increased extent of WSLs as assessed on digital photographs using an application [2,21].

Herbal goods are grown and harvested using ecologically friendly practices in developing countries such as India, which have a huge range of cultures, ethnicities, and diverse climatic conditions.

Researchers are making an effort to emphasize more on herbal drugs because of the vile side effects caused by commercial therapies [3].

An ideal tooth serum should be designed to support oral care effectively with good physical properties such as homogeneity, spreadability, antibacterial properties, and optimum pH for clinical usage. An ideal tooth serum would blend these properties to provide comprehensive oral care while being safe and user-friendly [4].

An ideal novel herbal tooth serum should possess potent antiplaque and remineralizing properties, effectively combating dental plaque buildup while promoting the restoration of tooth enamel [5]. It should be free from powerful chemicals, artificial flavors, or dyes, ensuring a natural and gentle oral care experience [6]. With a balanced abrasive effect, it must efficiently remove surface stains without causing irritation or toxicity, thus safeguarding oral health. Moreover, it should leave no unsightly stains on teeth, enhancing the aesthetic appeal of the smile. Accessibility and affordability are key, making it widely available and cost-effective for consumers seeking holistic dental care solutions [7].

The process of eliminating water and other volatile components from carbon-based products yields activated charcoal, a tasteless, odorless, and black powder [8]. This substance has several uses in the medical profession, including the treatment of pain, the alleviation of burning and swelling, and its antibacterial, anti-inflammatory, and remineralizing qualities [3,9]. Charcoal's nanopores increase the surface area and make ion exchange in the mouth easier. The product can remineralize the enamel surface and absorb and remove stains, pigments, and chromophores that cause teeth to discolor.

Cocos nucifera has a wealth of therapeutic properties because of its bioactive components. It has antibacterial qualities linked to lauric acid that help combat against a variety of infections, such as bacteria, viruses, and fungus. Its antioxidant concentration also supports skin health and wound healing by having anti-inflammatory effects. These qualities make coconut a natural medicine that can be used for wound healing, skin care, and general health maintenance. Researchers found that the leaf extract of *Cocos nucifera* had significant antibacterial activity against both Gram-positive and Gram-negative bacteria [10]. This product, commonly used in households, has also been proven to have remineralizing efficacy. Several studies have been conducted on white spot lesions, exploring various treatment strategies using different formulations, including mouthwashes, primers, and toothpastes, with both herbal and synthetic ingredients [11–13].

Combining the efficacy of various natural resources, this study aims to develop and assess the effectiveness of a novel tooth serum based on herbs incorporating activated charcoal on the remineralization of enamel surfaces.

2. Materials and Methodology

Composition of Novel Herbal Tooth Serum

The novel herbal tooth serum incorporates activated charcoal and extracts of *Cocos nucifera* as a remineralizing agent alongside calcium silicate and hydroxyapatite, facilitating the natural

mineralization of enamel. Gentle abrasives such as silica and calcium carbonate ensure effective yet mild cleaning action, while limonene, an antibacterial essential oil, enhances oral hygiene. The formulation is fortified with cinnamon and thyme extracts, serving as antioxidants, and sorbitol functions as a natural sweetener. The base ingredients include aqua, glycerin, and cellulose gum, providing a stable and user-friendly activator gel. All these ingredients will be added in tested quantities so as to enhance the properties of the serum.

Preparation of Activated Charcoal

Coconut shells are cooked in an open metal pot for three to five hours until no more gas vents out from the pot and only charcoal remains. The charcoal is allowed to cool down, transferred to a clean container, and rinsed with cold water to remove any debris or ash [8].

The cleaned charcoal is transferred to a mortar and pestle and ground into a fine powder. The dry charcoal powder is transferred to a stainless steel or glass mixing bowl, where calcium chloride solution is added to the powder in small increments with continuous stirring till a paste consistency is obtained. The remaining moisture is drained from the bowl and the charcoal is cooked again for three hours to activate it.

Preparation of *Cocos nucifera* extract

Cocos nucifera was sourced from a local coconut field, and the pulp extract was obtained by grating. Extracts were blended without the addition of any water, and the extracts were obtained using a muslin cloth. The coconut milk extract obtained was stored at -4 degrees. [10]

Preparation of the herbal tooth serum

The herbal tooth serum was prepared by using the dry gum method. Firstly, gum is mixed with humectants for proper dispersion. Other powdered ingredients are shifted together and added gradually to the mucilaginous mixture with continuously gentle stirring in a conical flask.

The primary active ingredients of the entire tooth serum play a crucial role in strengthening enamel and reversing demineralization effects, ensuring optimal dental health. Additionally, the formulation includes an abrasive component, calcium silicate, that also binds the active ingredients. The humectant properties are provided by sorbitol. Peppermint oil adds flavor, while additional antibacterial properties are achieved with thymol.

This comprehensive formulation serves as a natural alternative to conventional products for enhancing enamel remineralization and empowers users to proactively embrace oral hygiene, knowing they're benefiting from nature's bounty to protect and strengthen their teeth.

Packing

It is packed in a collapsible tube and at last sealed with the help of a collapsible tube sealing machine.

3. Evaluation of herbal tooth serum

The physical examination of the charcoal formulation involved visual assessment of its color and evaluation of smoothness by rubbing the paste between the fingertips. Homogeneity was tested at 27–20°C by observing the serum as it exuded a uniform mass from a collapsible tube or other suitable container, with the bulk of the contents gradually rolling out from the crimp. Spreadability was assessed by weighing 1-2 g of charcoal and placing it between two glass slides (10 x 10 cm), which were dragged in opposite directions without sliding; the spreadability was measured in centimeters after three minutes. To detect sharp or abrasive particles, the contents were applied to the finger and scratched along a 6-inch length of butter paper. Foamability was determined by mixing 0.5 g of charcoal with 10 ml of water in a measuring cylinder, noting the initial volume, and shaking the mixture 10 times to calculate the total volume of foam produced.

4. Results

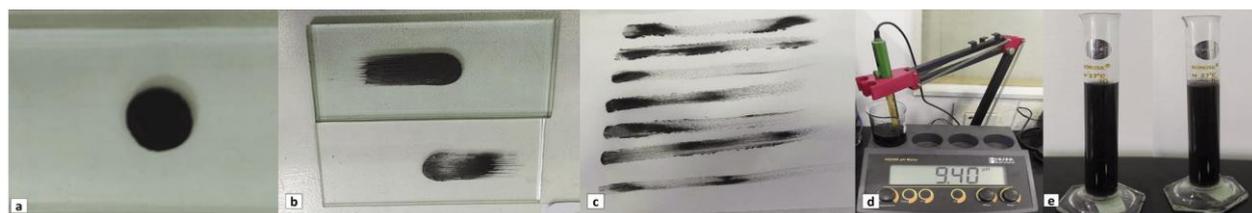


Figure 1: In-vitro physical testing of the Novel tooth serum formulation: a) Color and Homogeneity of the serum; b) Sharp edge or abrasive particle determination, c) Spreadability, d) pH measurement, e) Foamability

Physical Examination: The serum charcoal was dark black with a smooth texture.

Homogeneity: The charcoal paste had good consistency and showed no signs of deterioration at $34 \pm 30^\circ\text{C}$ (Figure 1a).

Sharp Edge and Abrasive Particle Determination: No sharp or abrasive particles were detected (Figure 1b).

Spreadability: The charcoal spread evenly between glass slides, with measurements taken after three minutes and averaged from three readings (Figure 1c).

pH Measurement: The pH was 9.40, suitable for intraoral use (Figure 1d).

Foamability: The presence of foam was confirmed after shaking the mixture with water (Figure 1e).

5. Discussion

The herbal tooth serum prepared using activated charcoal and *Cocos nucifera* extracts was evaluated for its physical properties such as homogeneity, spreadability, pH, abrasiveness, and formability. The study aimed to formulate a natural, herbal alternative to conventional synthetic tooth serum with ideal physical properties suitable for intraoral usage [14].

An ideal oral remineralizing agent should possess a high remineralization potential, effectively depositing bioavailable calcium and phosphate ions, such as those found in CPP-ACP or nano-hydroxyapatite, to restore demineralized enamel. It should work synergistically with fluoride to enhance enamel resistance against acid attacks and caries, maintaining a neutral or slightly basic pH that supports remineralization while preventing further demineralization. The agent must adhere well to the tooth surface for sustained release of ions and should be non-toxic, biocompatible, and safe for daily use across all age groups. Low abrasivity is essential to protect the enamel and dentin, while mild antibacterial properties can help reduce plaque formation and maintain a healthy oral environment. By promoting hydroxyapatite formation, preventing demineralization, and being gentle on oral tissues, an ideal remineralizing agent plays a crucial role in restoring and protecting dental health.

Activated charcoal itself does not directly aid in remineralization because it lacks the essential minerals, like calcium and phosphate, necessary for restoring demineralized enamel. However, it can play an indirect role by improving the oral environment, which supports remineralization. Charcoal helps maintain a neutral pH in the mouth, reducing acidity that typically contributes to enamel demineralization. By binding with acids, toxins, and bacteria, charcoal can help lower the risk of decay, creating conditions that are more favorable for remineralization processes facilitated by other agents, such as fluoride or calcium-phosphate-containing products. Activated charcoal's nanopores increase the surface area and make ion exchange in the mouth easier. The product can remineralize the enamel surface and absorb and remove stains, pigments, and chromophores that cause teeth to discolor. [15],[16]

Incorporating coconut milk extracts into tooth serum formulations can also offer benefits for preventing and managing white spot lesions (WSLs). Coconut milk contains various nutrients and compounds beneficial for dental health, including lauric acid, which possesses antibacterial properties similar to those found in coconut oil [17,18]. These antibacterial properties can help inhibit the growth of harmful bacteria in the mouth, reducing plaque formation and the risk of enamel demineralization, thereby contributing to WSL prevention [18].

This study has incorporated activated charcoal and *Cocos nucifera* extracts, the combined efficiency of which has been reflected in enhanced physical and biological properties. The lack of any synthetic ingredient in the tooth serum formulation has also improved the overall usage and efficacy of the serum.

Previous studies have shown that toothpastes containing *Cocos nucifera* extracts show significantly higher antibacterial activity against common pathogens, effectively combating *S. mutans*, *S. aureus*, *C. albicans*, and *L. rhamnosus*. [17]

Contrary to previous studies, the results of this study confirmed that the herbal serum containing activated charcoal showed reduced abrasiveness and was safe for intraoral usage on hard tissues [14]. The use of herbal ingredients in the tooth serum also reduces the harmful effects of oral microbes, thereby promoting the overall periodontal health of the patients. [19]

Although previous studies on tooth pastes containing *Cocos nucifera* and activated charcoal extracts were carried out independently [15,18], this study is the first of its kind to combine the beneficial effects of both the extracts in preparing a completely herbal tooth serum as a natural alternative to conventional synthetic tooth serums.

Through the incorporation of complete herbal ingredients like activated charcoal and *Cocos nucifera* extracts, the invention seeks to offer a natural and sustainable approach to remineralization, ensuring improved oral health outcomes and contributing to the prevention and mitigation of white spot lesions, a common concern in dental care [20]

6. Limitations of the study

The study's limitations include the absence of clinical trials to validate the intraoral effectiveness of the serum charcoal, potential variations in results due to uncontrolled environmental factors, and a lack of long-term stability data under varying storage conditions.

7. Future scope of the research

Future research on the herbal tooth serum should focus on confirming its effectiveness and safety through clinical trials and long-term studies. Optimizing ingredient concentrations, evaluating consumer acceptance, and assessing environmental impact will enhance the formulation and marketability. Additionally, developing variations with specific functional additives and conducting comparative studies will broaden its application. Investigating the mechanistic pathways of the serum's action on enamel remineralization will provide further insights into its benefits and guide improvements.

8. Conclusion

The herbal tooth serum formulation combining activated charcoal and *Cocos nucifera* extracts demonstrated excellent physical properties such as homogeneity, smoothness, absence of sharp or abrasive particles, optimum pH, and foamability to be used as a herbal alternative to synthetic tooth serums commercially available in the market.

Its physical properties highlight its potential for clinical usage, thereby enhancing oral hygiene routines. Further research and development could optimize its utilization in various dental formulations.

Additional Information

Disclosures

Human subjects: All authors have confirmed that this study did not involve human participants or tissue.

Animal subjects: All authors have confirmed that this study did not involve animal subjects or tissue.

Conflicts of interest: In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations

that might have an interest in the submitted work. **Intellectual property info:** The Novel Herbal Tooth Serum with Activated Charcoal and Cocos nucifera Extracts that has been mentioned in the study has been filed for a patent and the procedure is in progress. **Application number is as follows: 202441061170.**

Other relationships: All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

References

- [1] Marya, Anand, et al. "White spot lesions: A serious but often ignored complication of orthodontic treatment." *The Open Dentistry Journal* 16.1 (2022).
- [2] Jain RK, Verma P. Visual assessment of extent of White Spot lesions in subjects treated with fixed orthodontic appliances: A retrospective study. *World J Dent [Internet]*. 2022 Apr 11;13(3):245–9. Available from: <https://www.wjoud.com/doi/10.5005/jp-journals-10015-2042>
- [3] Janakiram C, Venkitachalam R, Fontelo P, Iafolla TJ, Dye BA. Effectiveness of herbal oral care products in reducing dental plaque & gingivitis - a systematic review and meta-analysis. *BMC Complement Med Ther [Internet]*. 2020 Feb 11;20(1):43. Available from: <http://dx.doi.org/10.1186/s12906-020-2812-1>
- [4] Muthuswamy Pandian S, Subramanian AK, Ravikumar PA, Adel SM. Biomaterial Testing in Contemporary Orthodontics: Scope, Protocol and Testing Apparatus. *Semin Orthod [Internet]*. 2023 Mar 1;29(1):101–8. Available from: <https://www.sciencedirect.com/science/article/pii/S1073874622000846>
- [5] Devi BV, Rajasekar A. Effect of herbal and nonherbal dentifrice on gingival health - A clinical study. *J Adv Pharm Technol Res [Internet]*. 2022 Nov;13(Suppl 1):S207–11. Available from: http://dx.doi.org/10.4103/japtr.japtr_177_22
- [6] Khera S, Gupta S. THE MAGIC OF AYURVEDA IN DENTISTRY: A REVIEW. *International Journal of Ayurveda and Pharma Research [Internet]*. 2020 Oct 24 [cited 2024 Aug 24];82–90. Available from: <https://ijapr.in/index.php/ijapr/article/view/1652>
- [7] Moghadam ET, Yazdani M, Tahmasebi E, Tebyanian H, Ranjbar R, Yazdani A, et al. Current herbal medicine as an alternative treatment in dentistry: In vitro, in vivo and clinical studies. *Eur J Pharmacol [Internet]*. 2020 Dec 15;889:173665. Available from: <http://dx.doi.org/10.1016/j.ejphar.2020.173665>
- [8] da Silva Palandi S, Kury M, Picolo MZD, Coelho CSS, Cavalli V. Effects of activated charcoal powder combined with toothpastes on enamel color change and surface properties. *J Esthet Restor Dent [Internet]*. 2020 Dec 1 [cited 2024 Aug 24];32(8):783–90. Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1111/jerd.12646>
- [9] Afshari M. *Electrospun Nanofibers [Internet]*. Woodhead Publishing; 2016. 650 p. Available from: <https://play.google.com/store/books/details?id=pTyXCgAAQBAJ>
- [10] Balakrishnan N, Kumar Subramanian A. Comparative analysis of antimicrobial efficacy: Harnessing Cocos nucifera extracts in novel toothpaste formulations. *Texila International Journal of Public Health [Internet]*. 2024 Apr 30;12(special1). Available from: <https://www.texilajournal.com/public-health/article/2531-comparative-analysis-of>
- [11] Kumar R, Nagesh S, Mani SP. Preparation and Assessment of Antimicrobial Effect of Strontium and Copper Co-substituted Hydroxyapatite Nanoparticle-Incorporated Orthodontic Composite: A Preliminary In Vitro Study. *Cureus [Internet]*. 2023 Oct;15(10):e47495. Available from: <http://dx.doi.org/10.7759/cureus.47495>
- [12] Rajendran R, Antony S DP, Ashik P M, Bharath S, Thomas AJ, Heboyan A. Remineralization potential of strontium-doped nano-hydroxyapatite dentifrice and casein phosphopeptide-amorphous calcium phosphate cream on white spot lesions in enamel following orthodontic debonding - a randomized controlled trial. *SAGE Open Med [Internet]*. 2024;12:20503121231221634. Available from: <http://dx.doi.org/10.1177/20503121231221634>
- [13] Govindaraj A, Paulpandian SS, Shanmugam R. Effect of chlorhexidine and fluoride varnish on the incidence of White Spot lesion in orthodontic patients. *Ann Dent Spec [Internet]*. 2023;1(11):35–9. Available from: <https://annalsofdentalspecialty.net.in/article/effect-of-chlorhexidine-and-fluoride-varnish-on-the-incidence-of-white-spot-lesion-in-orthodontic-pa-vmkxqbkngq73t>

- [14] Mamatha A, Sagar M, Pallavi B, SharathTP, Yashwanth S, Raj P, et al. Herbal Effervescent Powder For Gastritis Using Shankabhasma, Yashadbhasma, Triphala And Others. SFS [Internet]. 2023 Jun 20 [cited 2024 Aug 26];547–58. Available from: <https://sifisheressciences.com/index.php/journal/article/view/1144>
- [15] Kumaravel, Sakthivel, et al. "Preparation and characterization of activated carbon from corn cob by chemical activation and their adsorption of brilliant green dye from wastewater." *Process Safety and Environmental Protection* (2024).
- [16] Effectiveness and abrasiveness of activated charcoal as a whitening agent: A systematic review of in vitro studies. *Annals of Anatomy - Anatomischer Anzeiger* [Internet]. 2023 Jan 1 [cited 2024 Aug 26];245:151998. Available from: <http://dx.doi.org/10.1016/j.aanat.2022.151998>
- [17] Balakrishnan N, Kumar Subramanian A. Evaluation of biological response elicited by two novel tooth cream formulations of Cocos nucifera- cell line studies and MTT assay on human gingival fibroblast. *Texila International Journal of Public Health* [Internet]. 2024 Apr 30;12(special1). Available from: <https://www.texilajournal.com/public-health/article/2532-evaluation-of-biological>
- [18] Enamel subsurface remineralization potential of virgin coconut oil, coconut milk and coconut water. *Materials Today: Proceedings* [Internet]. 2019 Jan 1 [cited 2024 Aug 24];16:2238–44. Available from: <http://dx.doi.org/10.1016/j.matpr.2019.06.116>
- [19] Bedre AS, Arjunkumar R, Muralidharan NP. Evaluation of Concentration Dependent Antimicrobial Efficacy of Herbal and Non Herbal Dentifrices Against Salivary Microflora – An In Vitro Study. *Biomedical and Pharmacology Journal* [Internet]. 2018 Jun 25 [cited 2024 Aug 26];11(2):711–8. Available from: <https://biomedpharmajournal.org/vol11no2/evaluation-of-concentration-dependent-antimicrobial-efficacy-of-herbal-and-non-herbal-dentifrices-against-salivary-microflora-an-in-vitro-study/>
- [20] Mazur M, Ndokaj A, Bietolini S, Nisii V, Duś-Ilnicka I, Ottolenghi L. Green dentistry: Organic toothpaste formulations. A literature review. *Dent Med Probl* [Internet]. 2022 Jul-Sep;59(3):461–74. Available from: <http://dx.doi.org/10.17219/dmp/146133>
- [21] Chhabria, Devanshi Rajesh, et al. "Understanding the spatial and topographic characteristics of enamel white spot lesions for targeted remineralization." *Journal of Oral Biology and Craniofacial Research* 14.5 (2024): 594-599.