Received: 12th June 2015 Accepted: 15th September 2015 Conflicts of Interest: None

Source of Support: Nil

Original Research

A Randomized Control Trial to Evaluate Efficacy of Anti-bacterial and Anti-inflammatory Effect of *Aloe vera*, Pomegranate and Chlorhexidine Gel against Periodontopathogens

Abhilasha Sahgal¹, Shailendra Singh Chaturvedi², Hiroj Bagde³, Poonam Agrawal³, Rutuj Suruna⁴, Mrinal Limaye⁵

Contributors:

¹Reader, Department of Department of Periodontology, Sardar Patel Dental College, Lucknow, Uttar Pradesh, India; ²Reader, Department of Department of Periodontology, New Horizon Dental College, Bilaspur, Chhattisgarh, India; ³Sr Lecturer, New Horizon Dental College, Bilaspur; Chhattisgarh, India; ⁴Private Practitioner, Pune, Marahastra, India; ⁵Sr Lecturer, Dental College, CSMSS Dental College, Aurangabad, Maharastra

Correspondence:

Dr. Bagde H. New Horizon Dental College, Bilaspur - 495 001, Chhattisgarh, India. Phone: +91-9766105900. Email: hiroj.bagde@gmail.com

How to cite the article:

Sahgal A, Chaturvedi SS, Bagde H, Agrawal P, Suruna R, Limaye M. A randomized control trial to evaluate efficacy of antibacterial and anti-inflammatory effect of *Aloe vera*, pomegranate and chlorhexidine gel against periodontopathogens. J Int Oral Health 2015;7(11):33-36.

Abstract:

Background: The anti-bacterial and anti-inflammatory effects of a gel *Aloe vera*, pomegranate and chlorhexidine (Chx) gel were evaluated for 7 days.

Materials and Methods: Forty subjects were included the experiment. Test gels and placebo gels were used in this randomized trial. They were randomized into placebo, Chx, *A. vera* and pomegranate gel groups. On days 0 and 7, the plaque index and gingival index were recorded. Furthermore, the microscopic analysis to detect various organisms of the plaque was done pre and post application of gels.

Results: The results showed significant reduction in the quantitative bacterial levels in the pomegranate as well as Chx group. *A. vera* gel showed least reduction amongst all.

Conclusion: The gel containing *Punica granatum* extract was equally efficient in preventing plaque formation and gingivitis as compared to gold standard Chx gel.

Key Words: *Aloe vera* gel, chlorhexidine gel, gingivitis, pomegranate gel

Introduction

Plaque control helps in the prevention of periodontal diseases¹ hence the main aim of a professional is to arrest the ongoing active disease. Plaque control can be carried with the help of either reducing the periodontopathogens or making microbes beneficial to tissues.

Mechanical and chemical agents are available for plaque control. Chlorhexidine (Chx) being used effectively for plaque control it is considered as a gold standard for chemical plaque control. Apart from all the routinely used plaque control measures some alternative therapies such as herbal products for the treatment of inflammation and for reduction of the plaque bacteria have been tried.² Amongst them *Aloe vera* has been extensively used and has shown to significantly reduce gingivitis and plaque accumulation.³ So, pomegranate and its products have been tested and proven to be effective against the plaque bacteria and some viruses.⁴

Pomegranate (*Punica granatum*) also has been used as a medicinal fruit extensively in many cultures.⁵ The edible parts of the fruit are consumed or used in the preparation of fresh juices, canned food and also in the production of tooth paste^{6,7} as well as in therapeutic formulas. Fruit methanol and peel extracts have a potent antimicrobial effect⁸⁻¹⁴ *A. vera* is a cactus plant with some anti-inflammatory activity¹⁵⁻¹⁷ antiulcer activity,^{18,19} astringent effect, and enhancing wound healing^{20,21} properties which are to be used to either reduce the plaque bacteria or to reduce the inflammation.

Studies have proven that *A. vera* gel as well as pomegranate has reduced the bacterial pathogenicity and also the inflammatory component.

Both these gels were compared with the gold standard of Chx gel, which has been proven clinically beneficial to reduce the plaque bacteria.

Hence, we compared the efficacy of anti-plaque and the antiinflammatory activity of pomegranate, *A. vera* and Chx gel as compared to a control formulation in an experimental gingivitis model.

Materials and Methods

This study was designed in the Department of Periodontics at ACPM Dental College, Dhule. The clinical and microbiological study was designed as a comparative evaluation of a group of patients over a period of 1-week in an experimental gingivitis model for 40 patients with chronic periodontitis. Informed consent was taken from the patient for undergoing the trial. The following inclusion and exclusion criteria were made.

Inclusion criteria

- Patients within the age group of 30-40 years and with chronic periodontitis
- Patients without any history of systemic disease

 Patients should not have been under antibiotic coverage for last 6 months.

Exclusion criteria

- Antimicrobial therapy for patients at least 1-month prior to the study
- Individuals using mouthrinses or dentifrices or gelcontaining substances with anti-inflammatory properties and antiplaque properties
- Individuals allergic to drugs or chemicals used in the study products
- Pregnant or lactating mothers
- · Smokers.

Patients initially received thorough oral hygiene instructions. Scaling and root planning using hand and ultrasonic instruments was completed at the baseline visit. Forty patients were randomly divided into four groups with ten patients in each group:

Group 1 (n = 10): Intra oral pomegranate gel application

Group 2 (n = 10): Intra oral placebo gel

Group 3 (n = 10): Intra oral *A. vera* gel application

Group 4 (n = 10): Intra oral Chx gel application.

The pomegranate gel was prepared from drying the seeds of the fruit and then further processing it with the cellulose to make a gel formulation.

In the study, all the individuals were refrained from oral hygiene procedures for 7 days and plaque samples were collected from them. The following indices were recorded for the evaluation of the tissue condition.

- a. Gingival index (GI)
- b. Plaque index (PI)

The quantitative analysis of the bacteria was done by the collection of sub-gingival plaque sample and fixing it on the slide and measuring it in the field on the microscope. The qualitative analysis of the bacteria was done by culturing of the sub gingival plaque samples and evaluating them for *Porphyromonas gingivalis, Actinobacillus* species and *Prevotella intermedia*.

Results

For the quantitative analysis of mean quantitative analysis of bacteria as measured in the fields on the slide pre and post application of the gels intraorally at baseline and 7 days after the therapy (Table 1).

There was a significant reduction in the quantitative bacterial levels in the pomegranate as well as Chx group.

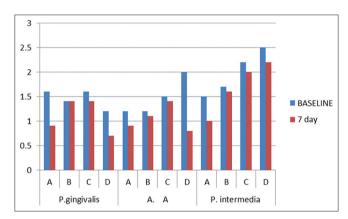
For the qualitative analysis of the bacteria *P. gingivalis,* Aggregatibacter actinomycetemcomitans, *P. intermedia* were found to be significantly reduced in the Group A and D as

compared to Group C and the placebo group showed minimal reductions (Graph 1, Table 2).

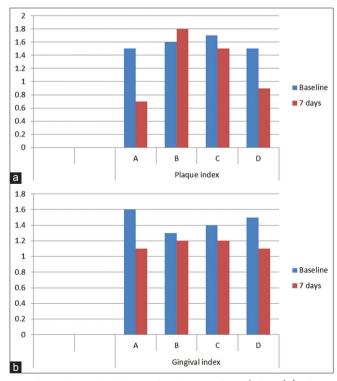
The GI and the PI for the Group A and D reduced significantly as compared to Group B and C, but the pomegranate group was nearby the values for reduction as compared to Chx group (Graph 2a and b, Table 3). Thus, the clinical manifestation of plaque accumulation and inflammatory activity was minimized more by Chx and pomegranate gel application.

Discussion

Though around 6000 plants in India are used in herbal medicines, little research has been conducted on efficacy, safety, and properties of herbal products. Over the decades, very few studies have been conducted to show the clinical



Graph 1: Preapplication and postapplication qualitative bacterial analysis.



Graph 2: Clinical indices at baseline and at 7th day. (a) Plaque index. (b) Gingival index.

Table 1: Bacterial count in fields of microscope for different groups.						
Bacterial count in fields	1	2	3	4	5	
Pre application						
Pomegranate (Group A)	+++ Bacilli	+	++	+	+++ Cocci	
Placebo (Group B)	++++ Cocci	++++ Bacilli	++++ Bacilli	++++ Bacilli	+++ Filamentous	
Aloe vera (Group C)	++++ Bacilli	++++	++++	+++ Bacilli	+++ Bacilli	
				+++ Cocci		
Chx gel (Group D)	++ Filamentous	+	-	+	++ Filamentous	
Post application						
Pomegranate (Group A)	+ Bacilli	-	+	-	++ Cocci	
Placebo (Group B)	+++ Cocci	++++ Bacilli	+++ Bacilli	++++ Bacilli	+++ Filamentous	
Aloe vera (Group C)	+++ Bacilli	+++	+++	+++ Bacilli	+++ Bacilli	
				+++ Cocci		
Chx gel (Group D)	+ Filamentous	-	-	-	+ Filamentous	

Name of bacteria	Groups	lication qualitative bacterial analysis. Bacteria count (*105)		
	_	Pre-operative	Post-operative	
P. gingivalis	A	1.6	0.9	
	В	1.4	1.4	
	С	1.6	1.4	
	D	1.2	0.7	
A. actinomycetemcomitans	A	1.2	0.9	
	В	1.2	1.1	
	С	1.5	1.4	
	D	2.0	0.8	
P. intermedia	A	1.5	1.0	
	В	1.7	1.6	
	С	2.2	2.0	
	D	2.5	2.2	

P. gingivalis: Porphyromonas gingivalis, A. actinomycetemcomitans: Aggregatibacter actinomycetemcomitans, P. intermedia: Prevotella intermedia

Table 3: Clinical indices (plaque index and gingival index) at baseline and at $7^{\rm th}$ day.						
Index	Group	Baseline	Post application 7th day			
Plaque index	A	1.5	0.7			
	В	1.6	1.8			
	С	1.7	1.5			
	D	1.5	0.9			
Gingival index	A	1.6	1.1			
	В	1.3	1.2			
	С	1.4	1.2			
	D	1.5	1.1			

efficacy of *P. granatum* and *A. vera*. In the present study, it was demonstrated that hydroalcoholic extract of pomegranate seed in the form of a gel exerted a significant reduction in clinical parameters as compared to Chx gel.

Evaluation of plaque scores showed that all the three groups except placebo showed reduction however, it was more significant in pomegranate gel group. These findings are contrary with the studies by Overholser *et al.*²² and Haffajee *et al.*²³ where Chx showed a significant reduction. In a microbiological study, Menezes *et al.* (2006)showed that following 1-min mouth rinsing significant reduction in plaque was observed with *P. granatum* (84%)as compared to Chx (79%).²⁴

Analysis of GI scores revealed that *P. granatum* was more efficient as compared to Chx gel and *A. vera* gel which may be due to its strong styptic action. Similar results were reported by Hafajee *et al.*²³

A study by Salgado *et al.* in 2006 on 10% *P. granatum* gel does not support our finding where it was less helpful to control plaque and gingivitis.²⁵ Gel was placed into tooth shield in a non-diluted form; it may be speculated that gel solubilization with saliva would be necessary for its antimicrobial action to take place. May be direct interaction of saliva to *P. granatum* led to acceptable results in our study.

Despite it's free commercial use, *A. vera* has limited antigingivitis and antiplaque action claims.²⁶ To the best of our knowledge, the present study is the first report about the effect of a gel containing *A. vera* and pomegranate on plaque and gingivitis. However, studies²⁷ have been conducted to check the efficiency of dentifrice containing *A. vera* on plaque and gingivitis wherein they showed reduced gingivitis.

Villalobos $et\ al.^2$ who observed reduction in plaque and gingivitis by use of $A.\ vera$ mouthrinse is in accordance with this study.

In the present study, the concentration of *A. vera* used is 98 % which is much higher than another study,²⁸ which has 50% concentration and this could be the reason for the superior effect of this phytotherapic effect.

Qualitative analysis of the bacteria *P. gingivalis, A. actinomycetemcomitans, P. intermedia* were found to be significantly reduced in the Group A and D as compared to Group C and the placebo group showed minimal reductions. Plaque scores, gingival scores, quantitative, and qualitative analysis of microbes showed reduction in pomegranate gel group. Hence, it can be concluded that herbal products can also be used as antiplaque agents in order to overcome side effects associated with the use of Chx.

Oral health and related impact on quality of life is still controversial.²⁸ Risk factors might be many but to reduce risk

factors cannot directly cure the disease hence adjunct use of different agents on a large scale can prove to be useful.^{29,30}

References

- 1. Wu CD, Savitt ED. Evaluation of the safety and efficacy of over-the-counter oral hygiene products for the reduction and control of plaque and gingivitis. Periodontol 2000 2002;28:91-105.
- 2. Villalobos OJ, Salazar CR, Sanchez GR. Effect of a compound mouthwash *Aloe vera* in plaque and gingival inflammation. Acta Odontol Venez 2002;39:16-24.
- 3. Gracious Ross R, Selvasubramanian S, Jayasundar S. Immunomodulatory activity of *Punica granatum* in rabbits A preliminary study. J Ethnopharmacol 2001;78(1):85-7.
- 4. Howell AB, D'Souza DH. The pomegranate: Effects on bacteria and viruses that influence human health. Evid Based Complement Alternat Med 2013;2013:606212.
- 5. Gracious RR, Selvasuramanian S, Jayasundar S. Pharmacology of pomegranate J Ethnopharmacol 2002;79:75-8.
- 6. Fadavi A, Barzegar M, Azizi MH, Bayat M. Physicochemical composition of ten pomegranate cultivars (*Punica granatum* L.) Grown in Iran. Food Sci Technol Int 2005;11(2):113-9.
- 7. Mousavinejad G, Emam-Djomeh Z, Rezaei K, Khodaparast MH. Food Chem 2009;115:1274-8.
- 8. Prashanth D, Asha MK, Amit A. Antibacterial activity of *Punica granatum*. Fitoterapia 2001;72(2):171-3.
- 9. Ismail T, Sestili P, Akhtar S. Pomegranate peel and fruit extracts: A review of potential anti-inflammatory and anti-infective effects. J Ethnopharmacol 2012;143(2):397-405.
- 10. Dahham SS, Ali MN, Tabassum H. Studies on antibacterial and antifungal activity of pomegranate (*Punica granatum* L.). Am Eurasian J Agric Environ Sci 2010;9(3):273-81.
- 11. Fawole OA, Makunga NP, Opara UL. Antibacterial, antioxidant and tyrosinase-inhibition activities of pomegranate fruit peel methanolic extract. BMC Complement Altern Med 2012;12:200.
- 12. Al-Zoreky NS. Antimicrobial activity of pomegranate (*Punica granatum* L.)fruit peels. Int J Food Microbiol 2009;134(3):244-8.
- 13. Naz S, Siddiqi R, Ahmad S, Rasool SA, Sayeed SA. Antibacterial activity directed isolation of compounds from *Punica granatum*. J Food Sci 2007;72(9):M341-5.
- 14. Duman AD, Ozgen M, Dayisoylu KS, Erbil N, Durgac C. Antimicrobial activity of six pomegranate (*Punica granatum* L.) varieties and their relation to some of their pomological and phytonutrient characteristics. Molecules 2009;14(5):1808-17.
- 15. Davis RH, Rosenthal KY, Cesario LR, Rouw GA. Processed *Aloe vera* administered topically inhibits inflammation. J Am Podiatr Med Assoc 1989;79(8):395-7.

- 16. Davis RH, Leitner MG, Russo JM, Byrne ME. Antiinflammatory activity of *Aloe vera* against a spectrum of irritants. J Am Podiatr Med Assoc 1989;79(6):263-76.
- 17. Vázquez B, Avila G, Segura D, Escalante B. Antiinflammatory activity of extracts from *Aloe vera* gel. J Ethnopharmacol 1996;55(1):69-75.
- 18. Imanishi K, Aloctin A. Active substance of *Aloe arborescens* Milleras an immunomodulator. Phytother Res 1993;7:20-2.
- 19. Saito K, Hiroko A. Purification of active substances of *Aloe arborescens* Miller and their biological and pharmacological activity. Phytother Res 1993;7:14-9.
- 20. Davis RH, Leitner MG, Russo JM, Byrne ME. Wound healing. Oral and topical activity of *Aloe vera*. J Am Podiatr Med Assoc 1989;79:559-62.
- 21. Davis RH, Donato JJ, Hartman GM, Haas RC. Antiinflammatory and wound healing activity of a growth substance in *Aloe vera*. J Am Podiatr Med Assoc 1994;84(2):77-81.
- 22. Maenthaisong R, Chaiyakunapruk N, Niruntraporn S, Kongkaew C. The efficacy of *Aloe vera* used for burn wound healing: A systematic review. Burns 2007;33(6):713-8.
- 23. Chithra P, Sajithlal GB, Chandrakasan G. Influence of *Aloe vera* on the healing of dermal wounds in diabetic rats. J Ethnopharmacol 1998;59(3):195-201.
- 24. Overholser CD, Meiller TF, DePaola LG, Minah GE, Niehaus C. Comparative effects of 2 chemotherapeutic mouthrinses on the development of supragingival dental plaque and gingivitis. J Clin Periodontol 1990;17(8):575-9.
- 25. Haffajee AD, Yaskell T, Socransky SS. Antimicrobial effectiveness of an herbal mouthrinse compared with an essential oil and a chlorhexidine mouthrinse. J Am Dent Assoc 2008;139(5):606-11.
- 26. Menezes SM, Cordeiro LN, Viana GS. *Punica granatum* (pomegranate) extract is active against dental plaque. J Herb Pharmacother 2006;6(2):79-92.
- 27. Salgado AD, Maia JL, Pereira SL, de Lemos TL, Mota OM. Antiplaque and antigingivitis effects of a gel containing *Punica granatum* Linn extract: A double-blind clinical study in humans. J Appl Oral Sci 2006;14(3):162-6.
- 28. Nategh B, Moghaddam MA, Nodehi D, Sharbaf DA. Periodontitis and oral health. Int J Contemp Dent Med Rev 2015;2015:Article ID: 020515. doi: 10.15713/ins. ijcdmr.76.
- 29. Mehta A. Risk factors associated with periodontal diseases and their clinical considerations. Int J Contemp Dent Med Rev 2015;2015:Article ID: 040115. doi: 10.15713/ins.ijcdmr.31.
- 30. Hoseinishad M, Nosratipour A, Moghaddam SM. Homeopathy in dentistry: A review. Int J Contemp Dent Med Rev 2015;2015:Article ID: 030815. doi: 10.15713/ins.ijcdmr.87.