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### ANTI-PLAQUE EFFECT OF CLASSICAL DANTAKASTHA - AN IN-VITRO STUDY

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#### **ABSTRACT**

Introduction: - When we see the *Ayurvedic* literature about *Dantadhavana* we found that they have a broad-spectrum health benefits as compared to modern oral hygiene practices. And it seems that prevailing oral hygiene practices covers the preventing aspect of oro-dental disease less as compared to the ancient oral hygiene practices. Here an attempt was made to study the effects of classical *Dantakashtha* for anti- plaque effect in comparison to well establish anti plaque agent named chlorhexidine by taking these two classical *Dantakashtha* for in-vitro study. Methods: - Fresh juice was collected from selected nine *Dantakashtha*. Sample of the plaque was taken from healthy individual in laboratory. Plaque was cultivated in brain

heart infusion broth medium. Every sample drug is tested thrice for one given variable to calculate more appropriate value in terms of mean value. **Result:** - All specimens show significant (p value- <0.0001) difference with negative control and positive control in terms of zone of inhibition except one specimen. **Conclusion:** - We can conclude that Dantakastha Dravyas have anti-plaque activity.

#### INTRODUCTION

Fast moving hectic lifestyle is creating many health problems. Due to faulty eating habits; consumption of junk food, fast food, ice–creams, sweets, chocolates and chewing of tobacco, *Gutakha*, smoking and alcohol consumption and improper way of tooth brushing leads to oral cavity problems. Therefore, dental diseases are a major health problem world-wide and among these dental plaque and periodontitis are most common. The use of *Dantakashtha* is well documented in *Ayurveda* to avoid these oral diseased conditions which is described under *Dinacharya* in classics. Ayurveda is most suitable and convenient for dental disorders as well as for daily use to attain oral hygiene in comparison to modern way of toothbrushing or oral wash. *Dantadhawan* plants are believed to have anti-plaque, anti-bacterial properties which are beneficial for prevention and treatment of oro-dental diseases.

In this global fast-moving economy and jet life it is utmost important to give fullest possible level of performance in our work for that we need a healthy body in terms of all physical, mental, social and spiritual aspects. To achieve this healthy state and long-life *Ayurveda* give a great importance to the prevention of disease and preservation of health. For this our acharyas also gave the proper pathway in terms of *Dincharya* regimens, *Ritucharya* and many more things. Among them *Dantadhawana*, *Kaval*, *Gandush* are also included. But the fact is that in this modern era the globally accepted oral hygiene practice are much more different then that ancient time. Now everyone is habitual of using toothbrushing with toothpaste and gargles with mouth wash which are full of chemical substances along with harmful ingredients as preservatives and many more things added to increase efficacy of these products.

No, doubts these chemical containing objects fulfil the fast-acting need of this global time but also carries side effects along with. In this series Chlorhexidine containing mouth wash has side-effect including brown discoloration of teeth, include retroactive materials and there is taste perturbation. There can be oral mucosal erosion which is an idiosyncratic reaction and is also dose dependent. On the other hand, toothbrushing habit provide a lot of mechanical force to clean the debris adhered to teeth but also proves to be harmful for the delicate gums associated with these teeth. When we see the *Ayurvedic* literature about *Dantadhavana* we found that they have a broad-spectrum health benefits as compared to modern oral hygiene practices and it seems that prevailing oral hygiene practices covers the preventing aspect of oro-dental disease less as compared to the ancient oral hygiene practices. But it is believed

that ancient Dantadhavana procedures are not sufficient to meet the need of oral health problems in this modern era.

Here an attempt was made to study the effects of classical *Dantakashtha* for anti- plaque effect in comparison to well establish anti plaque agent named chlorhexidine by taking these two classical *Dantakastha* for in-vitro study.

#### MATERIALS AND METHODS

Fresh juices were collected from selected *Dantakashtha* (Details given in table 1 at the end). Sample of the plaque was taken from healthy individual in laboratory. Plaque was cultivated in brain heart infusion broth medium.

- 1. In vitro antibacterial activity of formulations was carried out by using the Kirby-Bauer Agar Well diffusion method.
- 2. This classic method yields a zone of inhibition in mm result for the amount of antibiotic that is needed to inhibit growth of specific microorganisms.
- 3. Each formulation was used as such (100%) and in 50% diluted form with DMSO.
- 4. Sample prepared as each liquid formulation directly poured or dissolved in DMSO.
- 5. For the determination of zone of inhibition (ZOI), bacterial strain was taken and as a standard antimicrobial and control DMSO for comparison of the results.
- 6. The dilution of formulation in DMSO or as such and Chlorhexidine gluconate solution as antimicrobial, positive reference standards were prepared in double distilled water.
- 7. Muller Hinton agar plates for bacteria were seeded with liquid oral/dental plaque culture of bacterial strains and allowed to stay at 37°C for 24 hours.
- 8. The zones of growth inhibition around the wells were measured after 18 to 24 hours of incubation at 37°C.
- 9. Diameter of Well was 8 mm., volume applied in each well was 100 µl.

Control were as DMSO and Positive control or Standard as Chlorhexidine gluconate 2% solution.

Every sample drug is tested thrice for one given variable to calculate more appropriate value in terms of mean value.

#### RESULTS

In this study part the assessment criteria were the measurements of zone of inhibition. Nine Dantakastha Dravyas were taken to compare with the established gold standard anti-plaque agent named Chlorhexidine and one variable was negative control also.<sup>[4]</sup> Hence in total 11 variables were tested here. (Details given in table no 2 & 3 at the end.)

Comparison between others variable and positive control:

When all the variables were tested by using ANOVA test against positive control, they showed that there is an extremely significant difference (p value- <0.0001). To see this significance exactly Tukey's Multiple comparison test was applied.

**Intra** Variable comparison (Fig. 1 & 2)

- 1. Ziziphus sativus with others: Ziziphus sativus showed the significant difference (p value- 0.02) in the assessment criteria with the *Terminalia arjuna*, *Azadirachta indica* and **negative** control, while showed **highly significant** (p value- 0.0001) difference with the **Positive** control drug. All the other variables which are showing significant difference are better than Ziziphus sativus.
- 2. Nerium indicum with others: Nerium indicum has significant (p value- 0.02) difference with Calotropis procera and Aegle marmelos, while moderate significant (p value-0.004) difference with *Ficus bengalensis*, while **highly significant** (p value- 0.0007) with Terminalia arjuna and Azadirachta indica, and extremely significant (p value-<0.0001) difference with the positive control.
- **3.** Calotropis procera with others: Calotropis procera shows highly significant (p value-0.0001) difference with negative control and extremely significant (p value-<0.0001) difference with positive control.
- **4.** Ficus bengalensis with others: Ficus bengalensis shows extremely significant (p value-<0.0001) difference with negative and positive control.
- 5. Terminalia arjuna with others: Terminalia arjuna shows extremely significant (p value-<0.0001) difference with negative control and positive control.
- **6.** Albizzia lebbeck with others: Albizzia lebbeck shows extremely significant (p value-<0.0001) difference with positive control and **moderate significant** (p value-<0.0042) difference with negative control.

- **7.** Aegle marmelos with others: Aegle marmelos shows extremely significant (p value-<0.0001) difference with positive control and **highly significant** (p value- 0.0001) difference with negative control.
- **8.** *Azadirachta indica* with others: *Azadirachta indica* shows extremely significant (p value- <0.0001) difference with negative control and positive control.
- **9.** *Pongamia pinnata* with others: *Pongamia pinnata* shows extremely significant (p value- <0.0001) difference with positive control and highly significant (p value- 0.0007) difference with negative control.

#### DISCUSSION

All the variable in terms of *Dantakastha Dravyas* have significant difference in terms of zone of inhibition with the Positive control named Chlorhexidine. *Dravyas* also have significant difference with negative control. This proves that all these *Dravyas* have anti-plaque activity while we compare these with negative control. In this study we took fresh juice of all *Dravyas* for in-vitro study, this one may be the reason for under performance of selected variables because fresh juice has less potency with them with time in comparison of chlorhexidine due to absence of preservatives and efficacy enhancers. Performance supposed to be higher if we took the aqueous or alcoholic extracts for in-vitro study.

Katu Rasa has properties like Vaktram Shodhyati, Krimin Hinasati, Bandhan Chinnati etc. and Tikta Rasa has properties like Lekhana, Shleshma-opshoshana etc. and Kashaya Rasa has properties like Sharira-Kledsya-Upyokta etc. [5] These properties check the formation of suitable media for plaque producing micro-organisms. Thus, we can say that Anti-plaque effect of the Dantakastha Dravyas may be due to their Katu, Tikta and Kashaya properties. Beyond this they have properties like Putihara, Krimighna, Kandughna, Varnasodhana and Varnaropana properties. Its antimicrobial and anti-inflammatory activity had been proven by various research also.

Table 1: Name of selected dantakastha.

S. N.	Drug Name	<b>Botanical Name</b>	Rasa (Predominant)
1)	Arka	Calatropis procera	Katu, Tikt
2)	Vata	Ficus bengalensis	Kashay, Madhur
3)	Karanja	Pongamia pinnata	Katu, Tikt, Kashay
4)	Badar	Zizyphus Sativa	Madhur, Amla, Kashay
5)	Sirish	Albizzia lebbeck	Kashay, Tikt, Madhur
6)	Arjuna	Terminalia arjuna	Kashay
7)	Karveer	Nerium indicum	Tikt, Katu, Kashay

8)	Nimb	Azardirachta indica	Tikt
9)	Bilva	Aegle marmelos	Kashaya, Tikt

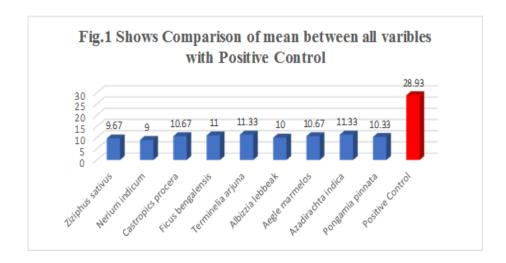
Table 2: Showing anti microbial senstivity data sheet ZOI in mm.

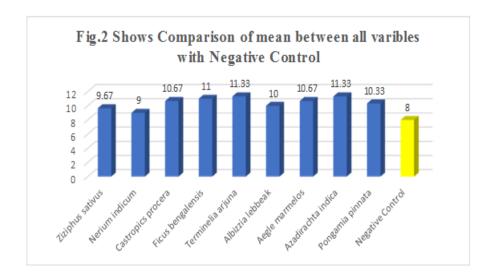
Sr. no.	Oral/ Dental Plaque culture	Std. (Positive control)	Concentration		DMSO (Negative control)
			50%	100%	-
1	Ziziphus sativa- 1	26	8	10	8
2	Ziziphus sativa- 2	27	8	10	8
3	Ziziphus sativa- 3	26	8	9	8
4	Nerium indicum- 1	30	8	9	8
5	Nerium indicum- 2	30	8	9	8
6	Nerium indicum- 3	31	8	9	8
7	Calotropis procera-1	30	8	11	8
8	Calotropis procera -2	29	8	10	8
9	Calotropis procera -3	30	8	11	8
10	Ficus bengalensis-1	27	8	11	8
11	Ficus bengalensis-2	27	8	10	8
12	Ficus bengalensis-3	28	8	12	8
13	Terminelia arjuna-1	32	8	11	8
14	Terminelia arjuna-2	32	8	12	8
15	Terminelia arjuna-3	31	8	11	8
16	Albizzia lebbeak-1	28	9	10	8
17	Albizzia lebbeak-2	28	9	10	8
18	Albizzia lebbeak-3	29	9	10	8
19	Aegle marmelos-1	30	8	10	8
20	Aegle marmelos-2	29	8	11	8
21	Aegle marmelos-3	30	8	11	8
22	Azadirachta indica-1	29	9	12	8
23	Azadirachta indica-2	30	9	11	8
24	Azadirachta indica-3	29	10	11	8
25	Pongamia pinnata-1	27	8	10	8
26	Pongamia pinnata-2	28	8	10	8
27	Pongamia pinnata-3	28	8	11	8

Table 3: Showing mean value of zone of inhibition observed in In-vitro study.

Sr. no.	Dental Plaque culture	Concentration Mean Value	
		50%	100%
1	Ziziphus sativus	8	9.67
2	Nerium indicum	8	9
3	Castropics procera	8	10.67
4	Ficus bengalensis	8	11
5	Terminelia arjuna	8	11.33
6	Albizzia lebbeak	9	10
7	Aegle marmelos	8	10.67

8	Azadirachta indica	9.33	11.33
9	Pongamia pinnata	8	10.33
10	Chlorhexidine gluconate	28.93	
11	Negative Control	8.00	





#### **CONCLUSION**

In in-vitro study all the selected *Dantakastha Dravyas* are significantly less effective against positive control variable under given condition. Again, in in-vitro study, except Nerium indicum all the selected Dantakastha Dravyas are significantly effective against negative control in terms of anti-plaque agent. Neem and Arjuna tree both have the highest anti-plaque activity among all selected Dantakastha Dravyas having 11.33mm zone of inhibition. Ficus bengalensis has anti plaque activity having 11mm zone of inhibition, And Arka and Bilva both have the anti-plaque activity having collectively 10.67mm zone of inhibition. Karanja has 10.33mm, Shirish 10mm, Badar 9.67mm and Karveera has least 9mm zone of inhibition. Neem and Arjuna have highest anti-plaque activity among selected Dantakastha Dravyas followed by Vata (Ficus bengalensis), Arka, Bilva, Karanja, Shirisha, Badar and Karveera respectively.

Here, we can conclude that Dantakastha Dravyas have anti-plaque activity.

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