

FORMULATION OF TOOTHPASTE FROM VARIOUS FORMS AND EXTRACTS OF TENDER TWIGS OF NEEM.

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ABSTRACT:

Since many centuries, Ayurveda regarded neem (*Azadirachta indica* Family: Meliaceae) as a cure for many ailments, predominantly due to its superb antimicrobial activity. It has been a practice since time immemorial to use tender twigs of neem as dentifrice. Looking as these facts the possibility of developing an authentic dentifrice from neem extract and formulations were formulated and evaluated for their antimicrobial activity and some formulations were found to have significant antimicrobial activity. For the present work the tender stems of neem were specifically chosen so that most specific results could be obtained especially when neem powder is selected for developing dentifrices. In addition to the antimicrobial activity it was thought that the small fibers in the powder may act as abrasive thereby potentiating the activity of the dentifrice. The scope of the present work is limited to testing the antibacterial activity of neem extract and neem powder in a tooth paste form.

KEYWORDS: Tender twigs of neem, Antimicrobial activity, *Azadirachta indica*.

INTRODUCTION:

Neem, a large green tree, 12 to 18 meter in height and 1.8 to 2.4 meter in girth forming a broad crown commonly found throughout the greater part of India. Almost every part of the tree is bitter and finds application in indigenous medicine. Neem extract has been reported to have antidiabetic, antibacterial and antiviral activity^{2, 3, 4}. The stem and root branches are reported to have astringent⁵, tonic and antipruritic activity¹. Stem bark contains tannins⁶. Neem oil is used in toothpastes¹. Aqueous extract of various parts of neem plant are used as gargle in stomatitis for bad gums and reduces all types of inflammations. Since ancient times, Ayurveda recommends tender twigs of neem as effective dentifrice and it has proven credential for the same. In recent times, many attempts have been made to formulate and market neem toothpaste^{7, 8, 9}. Neem oil is used in toothpaste formulation and was found to have excellent antibacterial activity. Considering the facts that various parts of neem have antimicrobial and astringent properties and that the fiber content of the tender twigs have considerable abrasive prop-

erty, it was thought to develop toothpaste which can combine all these properties in one dentifrice. Hence, present investigation was planned with tender twigs of neem plant instead of all parts of neem taken together.

MATERIALS AND METHODS:

Collection of tender stem:

Tender stem of *Azadirachta indica* were collected from Sakegaon village, Dist. Buldhana. The stems were dried in vacuum oven at 35°C to get coarse powder and fine preparation of paste. Fine neem powder (FNP) was obtained by sifting through muslin cloth.

Preparation of extracts:

The tender stem (100 grams) was extracted by two methods.

Method I: Continuous hot extraction (Soxhlet extractor) ¹⁰ with absolute alcohol at 50°C designated as SNE.

Method II: Percolation in conical percolator by standard percolation method using water, ethyl alcohol, benzene, petroleum ether and isopropyl alcohol as a solvent¹¹ and designated by PNE.

These extracts were dried by evaporation in vacuum oven.

After evaporation, the dried extracts were evaluated for antimicrobial activity.

Formulation of neem toothpaste:

The standard toothpaste base was formulated 12, 13 as given in Table 1. Extracts of neem twigs (SNE and PNE) and the dried neem twig powder (FNP) were incorporated in the base in various concentrations as given in Table 2. All the formulations were filled in regular metal tubes used for toothpastes. The storage in tubes was done to avoid the problem of crusting and drying of toothpaste during evaluation and stability studies.

Evaluation of neem extract and neem powder:

Evaluation of the extracts and the powders were carried out for antimicrobial activity. Antimicrobial activity was determined by Cup plate method^{14, 15}. The plates were prepared by using sterile nutrient agar medium and were inoculated with the bacterial cultures obtained from oral swabs by using lawn

culture technique. Microorganisms present in oral swab were *Streptococcus viridans* and *Streptococcus mutans*¹⁶. Sterile discs were used to inoculate the formulation on the plates. The plates were incubated at 37°C for 24 hours and the observations were recorded for three consecutive days. The statistical mean of three readings of zone of inhibition was determined.

Evaluation of toothpaste:

Total nine formulations with various concentrations of SNE, PNE and FNE were prepared and evaluated along with marketed neem paste (Henken India Ltd.) for antimicrobial activity same as performed for evaluation of neem extract and neem powder.

Stability studies:

The filled tubes were subjected to stability studies by storing at room temperature and normal atmospheric conditions for two months and evaluated for antimicrobial activity.

Table 1 Formulation of toothpaste base selected for study.

Sr.No.	Ingredients	Qty. Used (%)	Property
1.	Neem extract/powder	various conc.	Active Ingredient
2.	Calcium carbonate	35.00	Abrasive
3.	Sodium lauryl sulphate	01.50	Surfactant
4.	Glycerin	30.00	Anticrusting agent
5.	Methyl cellulose	01.00	Gelling agent
6.	Sodium saccharine	00.30	Sweetener
7.	Methyl paraben	00.10	Preservative
8.	Propyl paraben	00.02	Preservative
9.	Titanium dioxide	00.50	Opacifier
10.	Menthol	01.50	Flavoring agent
11.	Purified water	q.s.	Vehicle

Table 2 Formulation of toothpaste with various concentrations of neem extract and neem powder.

Sr. No.	Batch No.	Active ingredient used	Concentration (%)
1.	A	PNE	00.25
2.	B	PNE	00.50
3.	C	PNE	00.75
4.	D	SNE	00.25
5.	E	SNE	00.50
6.	F	SNE	00.75
7.	G	FNP	02.50
8.	H	FNP	05.00
9.	I	FNP	07.50
10.	J	MNP	05.00

SNE: Sox let neem extract.

PNE: Percolation neem extract

FNP: Fine neem powder.

MNP: Marketed Neem Paste. (NEEM PASTE, Henken India Ltd.)

Table 3 Antimicrobial activity of various neem extracts and powder.

Sr. No.	Active ingredient Used (cm)	Concentration %	Statistical Mean of Zone of inhibition
1	PNE	00.25	00.70 ± 0.05
2	PNE	00.50	00.90 ± 0.10
3	PNE	00.75	01.30 ± 0.10
4	SNE	00.25	00.60 ± 0.10
5	SNE	00.50	00.90 ± 0.15
6	SNE	00.75	01.10 ± 0.05
7	FNP	02.50	00.30 ± 0.06
8	FNP	05.00	00.50 ± 0.04
9	FNP	07.50	00.50 ± 0.03

Table 4 Antimicrobial activity of finished formulations and formulations after two months storage at room temperature.

Sr. No.	Active Ingredient	Statistical Mean of Zone of Inhibition (cm) [Finished formulations]	Statistical Mean of Zone of Inhibition (cm) [Stored formulations]
1.	PNE	00.70	00.70 ± 0.05
2.	PNE	00.90	00.90 ± 0.10
3.	PNE	01.30	01.30 ± 0.10
4.	SNE	00.60	00.60 ± 0.10
5.	SNE	00.90	00.90 ± 0.90
6.	SNE	01.10	01.10 ± 0.05
7.	FNP	00.30	00.30 ± 0.06
8.	FNP	00.50	00.50 ± 0.04
9.	FNP	00.50	00.50 ± 0.03
10.	MNP	02.50	02.50 ± 0.10

RESULTS AND DISCUSSION:

The primary objective of this study was to determine efficacy in terms of antimicrobial properties of tender twigs of neem in toothpaste formulation. Various extracts of tender twigs of neem were prepared with a view to preserve its antimicrobial property. SNE and PNE were evaluated for antimicrobial property as given in Table 3. Zone of inhibition by bore cup technique suggests that the extracts obtained by cold percolation method were having maximum antimicrobial activity. Toothpaste with SNE were having white to off white color and with that of FNE were having pale greenish color. All the toothpastes were having mint odor and smooth texture. Regarding taste, toothpastes with SNE and PNE were having sweet to mint taste and with that of FNE were having slightly bitter taste. Antimicrobial activity of toothpaste formulations with various concentrations of extracts and the powder and marketed paste were as given in Table 4. Amongst the nine formulations batch C (containing 0.75% of PNE) was most effective against oral swab culture indicating that formulations containing extract by cold percolation method is most effective. Comparison of antimicrobial activity of marketed paste with all formulations suggests that all formulations of SNE, PNE and FNP were found to have comparable antimicrobial activity as that of marketed paste. Antimicrobial activi-

ties after stability study at room temperature for two months were as given in Table 4. It shows that no significant change was found in antimicrobial activity of all toothpastes. However, it is felt important to mention here that formulations containing powdered neem twigs were equally effective. Moreover the powder contains tannins and fibers which are expected to potentiate the tooth cleaning property and ultimately the antimicrobial activity. The powder form is expected to be commercially more viable and effective alternative without significant compromise on the teeth cleaning effect.

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