

# Antimicrobial Activity of Chewing Sticks of Jimma – Ethiopia against *Streptococcus pyogenes*

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Article Info	Summary
<p><b>Article History</b></p> <p>Received : 19-05-2011                  Revised : 03-07-2011                  Accepted : 07-07-2011</p> <p><b>*Corresponding Author</b></p> <p>Tel : +91-94431 45690 (M)                  +91-431 2407086 (O)                  Fax : +91-431 24070 45</p> <p>Email:                  drttns@bdu.ac.in                  kothaiseshathri@yahoo.co.in</p>	<p>Chewing sticks were used to maintain oral hygiene / infection in the study area Jimma. Its use is a re-emerging science because of wide range of antibiotic resistant microbes and adverse effect of the dentifrices. Hence the objective of the study was to identify an oral health problem, common in the study area, the probable organism responsible for the disease and to screen the medicinal plants identified against the pathogen. Ethanobotanical survey was used as a tool to identify the oral problem and the medicinal plants. The causative organism was identified as <i>Streptococcus pyogenes</i> by throat swab culture. The medicinal plants reported in the survey to manage oral hygiene / infection were screened for their antimicrobial activity (Perez et al., 1991) against the isolated pathogen. Among the tested plants, <i>Juniperus procera</i> which was not documented as a chewing stick and tested against <i>S.pyogenes</i> before, showed maximum activity. The current research validates the use of <i>Juniperus procera</i> as chewing stick.</p> <p><b>Key Words:</b> Tonsillitis, Chewing sticks, <i>Juniperus procera</i>, Antimicrobial property and <i>Streptococcus pyogenes</i></p>

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## Introduction

Ethnobotany, the largest sub-discipline of ethnobiology, is generally defined as the "science of people's interaction with plants"(1). It also stands as a crossroad between social and biological sciences (2). Generally local communities worldwide are extremely knowledgeable about local plants and natural resources, on which they are so immediately and intimately dependent. Unfortunately much of this wealth of knowledge is becoming lost as traditional cultures become eroded. The traditional healers who were the custodians of the fore said knowledge were advanced in age and reluctant in transferring their knowledge. Quite surprisingly little attention has been paid to the historical development of such orally transmitted, indigenous knowledge systems (3). Through ethnobotanical survey several researchers have discovered and rediscovered many plant based therapies which are in practice or at the verge of extinction. Many scientists have realized that the study of native culture can provide valuable clues for improved health. To uncover the hidden events, ethnobotanical study is mandatory. Rich biodiversity of an area is used by local people for various activities like, traditional health care, cultural, and religious purposes throughout their life (4). Antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world (5). Similarly the use of variety of plants to manage oral hygiene / infection is common in the study area Jimma, Ethiopia which was not exploited for their antimicrobial activity. Hence the focus of this research was to conduct ethnobotanical survey in Jimma a town of Ethiopia, to explore the medicinal plants used for oral hygiene / infection and to scientifically validate their effect against *S.pyogenes*, the causative agent of tonsillitis through antimicrobial study.

## Materials and Methods

Ethnobotanical study was conducted during October 2006 to November 2007 using semi-structured questionnaire. A total of 516 informants were selected using purposive and random sampling methods. The information regarding their culture, personal information, plants used for common illness / infection management, their names in local languages were collected by repeated interviews in Amharic and Oromo i.e. the local languages spoken in Jimma, where the study was conducted & later translated to English.

The selected plants were collected and the herbarium sheets were sent to the Herbarium department in Addis Ababa University and the voucher specimen number were obtained.

Plant materials collected were extracted with ethanol (6). A known quantity of the dried extract was reconstituted with ethanol and working concentration prepared and used in the antimicrobial study.

The causative organism of tonsillitis was confirmed as *Streptococcus pyogenes* by throat swab culture of representative volunteers. The clinical strain isolated from throat swab and the referral strain *Streptococcus pyogenes* (ATCC 19615) obtained from Ethiopian Health and Nutrition Research Institute, Addis Ababa were used in the antimicrobial study. Muller Hinton agar plates were prepared and swabbed with four hours culture of *S.pyogenes* at 0.5 Mc Farland standards. 6mm wells were bored in the centre of the Muller Hinton agar plates and loaded with known concentration of plant extract.

## Results

Ethnobotanical data concerned about the cultural beliefs, diseases and traditional hygiene practices were categorized and found that medicinal plants used to manage oral infection / hygiene occupied the major portion. The information collected regarding oral hygiene/infection information was analyzed and found that among the five oral complaints (Fig. 1), tonsillitis was reported by 30.04% which included 14.34% of women, 8.14% of male and 7.56% children. In case of oral hygiene 47.87% were chewing stick users with high number of males (25.99%). There was no report on the use of chewing stick or modern brush by children. 29.26% of inhabitants did not respond to questions related to oral hygiene / infection (Fig. 2).

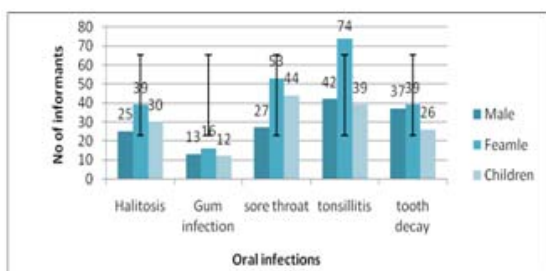


Fig 1 Ethnobotanical data on oral infection (N=516)

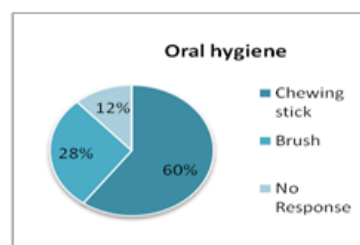


Fig. 2 Oral hygiene practices in Jimma N = (516)

Plant names reported in local languages, botanical identity, family, voucher specimen number, part of the plant and purpose for which it was used were tabulated (Table 1). The results revealed that nine plants viz *Clausena anisata*, *Clematis simensis*, *Juniperus procera*, *Olea europaea*, *Phoenix reclinata*, *Rubus apetalus*, *Sida rhombifolia* and *Stereospermum kunthianum* were commonly used as chewing sticks. *Clerodendrum myricoides*, *Sesbania sesban* and *Vernonia amygdalina* were recommended as chewing sticks by herbalists / traditional healers in case of gum and tooth infection. Clove, garlic and pepper were mentioned for use as self medication to cure tooth ache, sore throat and bad breath.

Table 1 Plants used for oral hygiene / infection in Jimma

Local name	Botanical name	Family	Voucher Number	Part used	Purpose
Aratha	<i>Sida rhombifolia</i> L	Malvaceae	KSMW 3461	Stem	Tooth brush
Atefaris	<i>Datura stramonium</i> L	Solanaceae	KSMW 3476	Seed	Tooth ache
Botoro	<i>Stereospermum kunthianum</i> Cham	Bignoniaceae	**	Stem	Tooth brush
Digitta	<i>Sesbania sesban</i> (L)Merr.(1912)	Fabaceae	*	Stem	Tooth brush / tooth pain
Grawa	<i>Vernonia amygdalina</i> Del.(1826)	Asteraceae	KSMW 3465	Stem	Tooth brush /tooth ache,
Hareg	<i>Clematis simensis</i> Fresen.(1837)	Ranunculaceae	**	Stem	Tooth brush
Injori	<i>Rubus apitalus</i> Poir.(1804)	Rosaceae	**	Stem	Tooth brush
Kondu berbere	<i>Piper nigrum</i> L.	Piperaceae	*	Fruit	Throat infection
Krum food	<i>Syzygium aromaticum</i> L	Myrtaceae	*		Tooth ache
Limmich	<i>Clausenia anisata</i> Burm.f (1769)	Rutaceae	KSMW 3486	Stem	Tooth brush
Mesirich	<i>Clerodendrum myricoides</i> (Hochst).R.Br.ex.Vatke	Verbanaceae	KSMW 3478	Stem	Tooth brush/tooth ache,
Nech shrinkurt	<i>Allium sativum</i> L.(1762)	Alliaceae	*	Bulb	Tooth ache
Sansel	<i>Justicia schimperiana</i> (Hochst).Nees	Acanthaceae	KSMW 3470	Stem	Tooth brush
Tsid	<i>Juniperus procera</i> Hochst ex Endlicher	Cupressaceae	KSMW 3485	Stem	Tooth brush
Weira	<i>Olea europea</i> Miller (1768)	Oleaceae	KSMW 3487	Stem	Tooth brush
Yemder berbere	<i>Spilanthes mauritiana</i> (Rich.ex.Pers)DC	Compositae	KSMW 3473	Flower	Tonsil
Zambaba	<i>Phoenix reclinata</i> Jacq (1801)	Arecaceae	KSMW 3484	Petiole	Tooth brush

Table 2: Antimicrobial activity of plants used for oral hygiene / infection against *S.pyogenes*

Botanical name of the chewing sticks	Extract / Zone of inhibition in mm				
	Streptococcus pyogenes (clinical isolate)		Streptococcus pyogenes (Referral strain) ATCC 19615		
	Water	Ethanol	Water	Ethanol	MIC
<i>Clausenia anisata</i>	7.33 ± 0.57	0.00 ± 0.00	6.33 ± 0.57	0.00 ± 0.00	-
<i>Clematis simensis</i>	0.00 ± 0.00	8.66 ± 0.57	0.00 ± 0.00	9.33 ± 0.57	50mg/ml
<i>Cleodendrum myricoides</i>	6.66 ± 0.57	0.00 ± 0.00	7.33 ± 0.57	0.00 ± 0.00	-
<i>Juniperus procera</i>	7.66 ± 0.57	20.00 ± 1.00	8.00 ± 1.00	20.33±0.57	25mg/ml
<i>Justicia schimperiana</i>	6.66 ± 0.57	11.00 ± 1.00	6.33± 0.57	11.00±1.00	50mg/ml
<i>Olea europea</i>	7.00 ± 0.00	14.00 ± 1.73	7.33 ± 0.57	15.33±0.57	25mg/ml
<i>Phoenix reclinata</i>	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	8.33 ±0.57	100mg/ml
<i>Rubus apitalus</i>	7.66 ± 0.57	15.33 ± 0.57	8.00 ± 0.00	15.66±0.57	50mg/ml
<i>Sesbania sesban</i>	0.00 ± 0.00	10.33 ± 0.57	8.00 ± 0.00	11.00±1.00	25mg/ml
<i>Sida rhombifolia</i>	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	50mg/ml
<i>Spilanthes mauritiana</i>	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	-
<i>Stereospermum kunthianum</i>	0.00 ± 0.00	14.00 ± 1.00	0.00 ± .00	14.66±1.15	25mg/ml
<i>Vernonia amygdalina</i>	0.00 ± 0.00	8.00 ± 0.00	0.00 ± 0.00	7.66 ± 0.57	100mg/ml

Ethanol –negative control –showed no activity.

Penicillin and tetracycline served as positive control and produced 29 & 34 mm of zone of inhibition respectively.

Fig.3 *Spilanthes mauritiana*

Fig. 4 Brushing with chewing stick



Fig. 5 Chewing sticks



Fig. 6 Juniperus in hedge



*Spilanthes mauritiana* was the only plant reported for tonsillitis. *Justicia schimperiana* leaves were often collected for cleaning the 'tella' brewing pots and the sticks were chewed by few women. 'Tella' is a local alcohol consumed widely by the inhabitants in the study area.

Antimicrobial activity of twelve selected chewing stick plants and *Spilanthes* was studied and found that ethanol extract of *Juniperus procera* showed highest zone of inhibition i.e 20mm at 100mg/ml concentration. Aqueous extract of the chewing sticks showed weak activity except *Clematis simensis*, *Sida rhombifolia* and *Stereospermum kunthianum*. Ethanol extract of *Juniperus procera* was most effective against *S.pyogenes* (Table 2). The minimum inhibitory concentration ranged from 25 – 100 mg.ml.

## Discussions

Oral health is the total health of an individual to maintain personal & social well being. Oral diseases directly affect the quality of life by having a serious impact on an individual's well-being and ability to fulfil desired socioeconomic functions (7). Moreover the use of chewing sticks to manage oral infections is in danger of being irrevocably lost if quick efforts are not exerted to document this invaluable knowledge. Recently it was noted that chemical dentifrices slowly and steadily

replaced the chewing sticks in most developing and developed countries but Ethiopia is an exception to it. The results of this study revealed that nearly fifty percentages of the informants use chewing sticks to maintain oral hygiene. Halitosis was observed and reported could be as a result of severe tonsillitis. The recurrent infection may be due to malnutrition, chemotherapy, poor development of immune system.

*Spilanthes mauritiana* (Fig. 3) is a common weed used to cure tonsillitis and known to all in the study area. According to them *Spilanthes* flowers were collected in odd numbers, chewed and the extract was swallowed. The medication was continued until cure. In spite of the foresaid traditional cure, 30.04% were reported to be infected with tonsillitis. The results of this research revealed that the ethanol and water extract of *Spilanthes* was not effective against *S.pyogenes* the causative organism of strep throat (tonsillitis). This indicated that the *Spilanthes* flower does not have any activity against *S. pyogenes*. This might be one of the reasons for high percentage of strep throat caused by *S.pyogenes*.

*Streptococcus pyogenes* is one of the normal floras of animals and found in some patients with severe tooth decay. Tonsillitis is transmitted from one person to another by coughing and sneezing.

The study identified that chewing sticks were used to maintain oral hygiene and manage oral infections (Fig. 4). Chewing sticks made from *Clausenia anisata*, *Clematis simensis*, *Phoenix reclinata* and *Stereospermum kunthianum* were available in the market (Fig. 5). Vendors reported that *C. simensis* and *S. kunthianum* were supplied by people who collect them from nearby forest. *Phoenix reclinata* leaves were collected for basket making in the New Year season (September), the petiole which comes as a by-product was used as a chewing stick which is not reported elsewhere and thereby this is the first report of its kind. *Juniperus* (Fig. 6) and *Rubus apetalus* were found as hedge plants and thereby people use as and when required.

*Juniperus procera*, commonly known African Juniper is a coniferous tree native to the mountains of eastern Africa from eastern Sudan south to Zimbabwe, and the southwest of the Arabian Peninsula. It is an endangered tree of Ethiopia (8). The species was pushed to such position as the tree is cut for fuel, construction and land clearing for residence. In traditional medicine the plant was documented to cure intestinal worms (9), fever, stimulant, birth control agent, sudorific, emmenagogue and rheumatism (10), bladder infections, chronic arthritis, gout, fluid retention, kidney disease, menstrual irregularities, heartburn, and dyspepsia (11). Plant based antimicrobials have enormous therapeutic potential as they can serve the purpose with lesser side effect that are often associated with synthetic antimicrobials (5). Previous studies have not documented *Juniperus procera* as a chewing stick (of Jimma town) or its antimicrobial property against *Streptococcus pyogenes*. Moreover, in the study area, the plant was not given a status of medicinal plant rather than a hedge plant. But for the first time, this study reports the use of *Juniperus procera* as a chewing stick and as a cure for tonsillitis.

Wu, suggested that antimicrobial substances that naturally protect plants against various invading microorganisms or other parasites may leach out into the oral cavity from the chewing sticks and these compounds may benefit the users by protection against bacteria. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body (12). The same reason can be accounted for the activity of *Juniperus procera* against *S.pyogenes*.

Ilias Muhammed, studied the bark of *Juniperus* which yielded three antibacterial diterpenoids namely (+)- E-Communic acid, (+)-Z- Communic acid and (+)- Totarol. Of the three, Totarol exhibited potent activity against *Mycobacterium* and (+)-Z- Communic acid exhibited activity against many gram +ve bacteria (13). The current study also revealed the antimicrobial activity of *Juniperus procera* against *Streptococcus pyogenes*, a gram positive organism. Further experiments need to be undertaken to find the active ingredient in *Juniperus procera*, responsible for the antimicrobial activity against *S.pyogenes*.

## Conclusion

The research concludes that scientific exploration is mandatory to select a chewing stick to manage a disease. Antimicrobial screening also supports the use of *Juniperus procera* which offer protection against strep throat caused by *S. pyogenes*.

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