

Review on Moringa Based Herbal Toothpaste against Biofilm Forming Pathogen

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Submitted: 20-04-2023

ABSTRACT:

In this study, Formulating an Moringa leaves based herbal toothpaste which product from the formation of biofilm in mouth mostly by the oral organism Streptococcus mutans. Dental caries are very common problem in human health and dental caries are mostly affected by bacteria and fungi microorganisms. The main aim of this study to overcome the forming biofilm in mouth which leads to cause human infections, and sometimes it may cause periodontal diseases. Plaque contain a variety of microorganisms. The pathogenicity of certain microbial species such as Streptococcus mutans, Staphylococcus epidermidis, Candida albicans, Lactobacillus, Legionella pneumophilia, Pseudomonas aeruginosa, Escherichia coli. Here the Streptococcus mutans has the capable of colonizing the oral cavity and also forms the major biofilm formation. Biofilm formation has the process of protein-bacterium interaction. By using Moringa oliefera as a herbal plant to overcome from the biofilm formation in mouth. Moringa oliefera is are used as traditional system of medicine. This species is known as "The Miracle tree" because it is useful in the curing various diseases. Each and every part of Moringa plant like root, leaves, bark, stem, seed, flower are responsible for curing various diseases in human. Moringa leaves has good antibacterial activity which inhibit the growth of bacterial organisms. Moringa leaves has the both Anti-bacterial and Anti-inflammatory activity. It mostly inhibits the Streptococcus species organisms. By formulating the antimicrobial toothpaste was evaluated by the antimicrobial sensitivity test against the selected

Accepted: 30-04-2023

bacteria. The Moringa leaves based toothpaste showed high inhibition against the selected bacteria. This Moringa based toothpaste were carried out with various phytochemical analysis like flavonoids, alkaloids, tannins, saponins, phenols, amino acids, proteins etc., And the Moringa leaves based herbal toothpaste carried out the acceptability and long and shelf life.

Key words: Biofilm formation-Streptococcus mutans-Moringa leaves- antibacterial -herbal toothpaste.

I. INTRODUCTION

Dental caries (tooth decay) which leads to damage the tooth that happens when decay-causing by bacteria in your mouth. That makes acids that attack the tooth's surface. Due to the involvement of high acids content in mouth which leads to the small holes in tooth which expressed as cavity in mouth. If the tooth decay is not treated immediately it cause harmful infections, or it leads to diseases in oral care. By simple word the dental caries is the breakdown of teeth due to the microbial activity like bacteria and fungi. Dental caries are most common health problem in the world, is a chronic disease that destroys tooth tissue and that can adversely affect chewing and aesthetic appearance (9). Nearly 60-70% of the child population is affected by this dental caries. More or less the dental caries or tooth decay are effected by both gram positive and negative microorganisms. The main affecting agents of this dental caries are pathogenic microorganisms like Streptococcus mutans. Streptococcus mitis, Streptococcus sanguis, Streptococcus salivarius and Streptococcus



sobrinus are the microbial bacteria which causes tooth decay in mouth(2). The most common oral and dental health diseases like tooth decay, gingivitits, and periodontitis which causes frequently in humans in different stages of age groups(1). In this development of tooth decay or dental caries diseases, number of microorganisms, such as Streptococcus mutans and Lactobacillus are the two microbes which has the significant effects in causing dental caries in human, because they play a key role in fermenting the sugar molecules like sucrose which results in producing the lactic acids which involve in demineralization of cavity and dental plaque. And some other microbes like Staphylococcus aureus, Streptococcus mitis, and Candida albicans have been also involved in dental diseases. A majority of the involvement on the epidemiology of dental caries suggests that Streptococcus mutans is the one of the major pathogenic and carcinogenic bacteria in initial stage of formation of biofilm in mouth of dental caries(19).

This study mainly aims to reveals the most pathogenic biofilm forming microorganism in mouth which is inhibited by using the herbal based toothpaste to overcome the common oral care problem arising in human generation. Biofilm formation are formed by many of the microbes like Streptococcus mutans, Staphylococcus aureus, Lactobacillus, Klebsiella, Escherichia coli etc., In this Streptococcus mutants has the major involvement in the process of forming biofilm formation in mouth. The additional properties of S.mutants has the capable of colonizing in oral cavity and able to survive in acidic environment (13). Streptococcus mutants are responsible for bacteria adhesion and the formation of microbial colonies on the host cell surface specially when it is influences by human saliva. When this structure are engaged in pathogenic interaction with host cells and they are responsible for formation of clustering of bacteria in the form of biofilm-like structure (24). Most of the studies in article suggesting that Streptococcus mutans are the only organism are isolate have a greater ability to form biofilm than the other Streptococcus species involved in the formation of biofilm, which colonize in the human tooth decay or dental caries(21, 22).

Here the formation of biofilm can also be inhibited by the herbal plant which should have the anti-bacterial and anti- inflammatory activity against the dental caries. Herbal medicine is an alternative formulation to chemically prepared products in every aspects in pharmaceutical

products and fields with no exceptions (1). Herbal medicine have high demands due to their problem arising with artificial medicine such as their cost and side effects to human health that is why the herbal medicine has the huge demands. Utilization of the herbal based products which may be more helpful and harmless for the human health care(4). Moringa oliefera belongs to the family, Moringaceae and order Brassicales (1). Moringa is also known as drumstick or horse radish in English. This Moringa plant has several medicinal and nutritional values in each and every part of the plant. And this Moringa is very healthy nutritive vegetables(1). This Moringa plants is also known as "The miracle tree", because it has the ability to cure the various diseases involved in mouth and dental caries. Some of the research has began to isolate the bioactive compound from the Moringa plant to known about their applications in various field(6). This Moringa leaves are usually used for food, traditional medicine and traditional ritual ingredients. It is noticed that Moringa leaves contains more vitamin A than carrots, more calcium from milk, more iron from spinach, more vitamin C from oranges and potassium from banana[3,4]

Moringa oliefera demonstrates immeasurable potential in their view of the various biological activities which are reported from its secondary compounds, such as alkaloids, flavonols, steroids, saponins, coumarins, quinones, resins, lectins among others(6). Based on the information and results acquired in other studies, this review aims to expose the potential of Moringa leaves benefits which associates with remedy for biofilm formation in dental decay. Now this Moringa leaves has the good side like rich in their vitamins, antioxidants, and minerals. Moringa leaves also contain the characteristics of fight against inflammation and it protects the liver. Moringa leaves is good for detoxification. Moringa leaves has interaction with thyroid medicine, medicine breakdown by the liver, diabetes and blood pressure. The involvement of Moringa leaves in dental caries is, it can be used as an ingredients in toothpaste, mouthwash, root canal irrigation, wound healing after tooth extraction are can be used to prevent the dental caries(8). Here the Moringa leaves has been used as main ingredient in the formulation of toothpaste against the formation of biofilm. Moringa leaves can enhance the rebuilding of enamel surface lesions and this Moringa gas the ability to deposits a new layer that resembles the structure of healthy enamel. So this



extraction of Moringa leaves against high proportion of Streptococcus mutans forming carcinogenic biofilm. This herbal leave has the antimicrobial activity against the pathogenic bacteria present in tooth decay. If this benifical herbal leaves as an main ingredient used in the toothpaste to avoid the biofilm formation by bacteria, the toothpaste inhibit the formation of cariogenic biofilm by Steptococcus mutans due to the good antibacterial activity of Moringa leaves and the combination of water-insoluble essential oils such as peppermint, eucalyptus, clove, cinnamon are used as flavouring agents in toothpaste. This flavouring agents also called as "masking agents" or "bitter blockers".

The essential leave extract has prepared by using the phytochemicals and most of the volatile substances has taken as peppermint as a flavouring agents and smell. In this review, the paper aimed to provide information on formulating the toothpaste against the formation of biofilm in mouth by pathogenic bacteria by essential extraction leaves against the selected microorganism(1).

Dental caries

Dental caries is a biofilm-induced disease, resulting from interactions of microorganisms, host-related factors and diet which form into formation of biofilm on tooth surface(35).The common oral caries are caused by the both bacteria and fungi. Dental caries are mainly caused due to no proper cleaning teeth well, frequent snacking and sipping sugary drink are main reason behind the cavities. Due to this reasons, the microorganisms are more helpful for the further dental caries leads to the oral diseases[15]. Cavities are permanently damages the hard surface of the teeth which develops into tiny openings or small holes. Mostly the initial stage for the dental caries starts from the formation of biofilm in your mouth by pathogenic microbes.

Dental caries is always localized in the destruction of susceptible dental tissues by the products produced from bacterial fermentation of dietary carbohydrates(32). The significance of demineralisation are found in hard dental tissues, but the diseases may cause by the initiation of the bacterial biofilm which covers the whole tooth surface in mouth(33). This dental caries is the multifactorial diseases that are initially starts within the microbiological shifts with biofilm and then it is affected by salivary flow and which are preventing by hygiene behaviour like cleaning teeth(33).

Poor oral hygiene is the causes of biofilm accumulation and there will specific group of bacteria responsible for tooth decay. Due to this the bacteria may causes like plaque-induced inflammatory periodontal diseases(34).

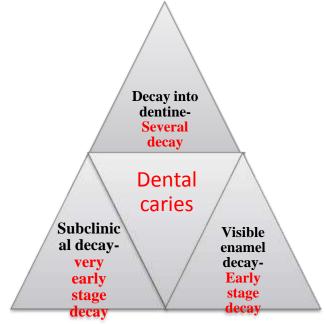


Diagram 1: Diagram represents the stages of dental caries (32).



World of Oral microbiota

There are about 700 different kinds of microorganisms exists in human mouth(28).Oral microbiota can be used as targets to treat oral and systemic diseases(28). In human oral there is the presence of both bacteria and fungi.In fungi alone there are 85 species of fungi can be found in mouth. Among these fungi, the most important fungi involved in mouth is Candida(29). Candida is the neutral organism when the oral microbiota is normal, when this oral microbiota is balance is broken Candida will have opportunity to attack oral tissue(30).

The oral microbiota is a important part in human microbiota. This oral microbiota includes several hundred and several thousands of different species, which comprises several facets (14). Mostly the oral microbes loaded in human mouth saliva and soft tissues of tongue, mucosa membrane and hard tissue of teeth(37). Detection of oral microbes are changes according to their age-related microbiological changes in mouth. Complex formation of composition of oral microbes on oral hard and soft tissues in mouth develops into the biofilm formation(37). This problem is arise due to microbial settlement shedding surfaces in the soft tissues of mucosal membrane like lips, cheek and tongue which have complex formation of microbial load uncountable in mouth(38).

Here 30% of microbial load can be detectable by using molecular process. The shedding surfaces, where the monolayer which forms bacterial originated and which regularly presented in tongue with its stable multi-layer of formation of biofilm bacteria. Tongue has more oral microbiota than the other mucosal tissues(39). Oral microbiota plays a important role in maintaining the homeostasis of oral cavity (40). Three major hypotheses have been developed in the dental caries. The specific plaque hypothesis, the non-specific hypothesis and ecological plaque hypothesis(41-43). Oral caries can also causes for both child and addults. Oral caries of microbes have no age limitations. As most of the common chronic infections or diseases in children is caused due to their child's life style quality is responsible(44).

	Tuble 1. Tuble shows the unterent types of bucteria and range in oral cartes(+).		
	Bacteria	Fungi	
	Streptococcus mutans	Candida albicans	
Oral	Staphylococcus aureus	Aspergillus fumigatus	
microbes	Escherichia coli	Cryptococcus	
	Lactobacillus		
	Porphyromonas gingivalis		

 Table 1: Table shows the different types of bacteria and fungi in oral caries(44).

Common oral microbes include both bacteria and fungi. Dental caries can be caused due to either aerobic or anaerobic microbial bacteria. Oral bacteria include Steptococcus mutans, Porphyromonas gingivalis, Staphylococcus aureus, Candida albicans. and Lactobacillus. Streptococcus mutants is the main component in oral microbiota in biofilm formation(28). Porphyromonas gingivalis is a non-glycolytic gram negative anaerobic bacteria involved in periodontal diseases. Lactobacillus organisms is a bacterium that can ferment the sugar molecules to produce lactic acid(31).

Dental caries is usually associated with increased numbers of streptococcus mutans and lactobacillus are the most bacterial organisms at the sites of disease which initiate by formation of biofilm, estimation of salivary levels of these organisms may be useful for assessing caries risk in the patients and monitoring their response to preventive measures(45). Pathogenesis of dental caries and evaluation of inhibitory concentration of herbal plants can be detected against oral pathogen (36)

Virulence of biofilm formation

Virulence is described as an ability of the microbes to infect the host and cause a disease. Virulence factors are the essential molecules that assist the bacterium colonize the host the cellular level[16]. Biofilm helps bacteria to evade the host defence mechanisms and persist for a longer time



and causes several oral-associated diseases (17). Biofilm formation can be formed in three different stages like attachment, maturation and dispersion(46). Biofilm formation are formed by most common oral cavity infectious disease is caused and plays an important role in forming biofilm by the microorganisms on teeth and gum surfaces in dental caries(47). The extracellular polysaccharides(EPS) also affect the physical and biochemical properties of the biofilm (48). The primary sources of extracellular polysaccharides are glycosyltransferase(GTF) and fructosyltransferase(FTF), are forms interaction with sucrose and starch(49).

Biofilm can be referred as the communities of microorganisms which attached to the surface of the mouth which is approached by the bacteria and fungi(56). Oral biofilm formation are said to be mixed species of microbial communities, and their outgrowth and overgrowth can be expressed as oral diseases(57). The formation of biofilm is a multistep and more complicated process in oral cavity. Main interaction are begins with the salivary agglutinations and loaded microbes in mouth mostly by Streptococcus mutans(20). The interaction between the salivary and pathogenic oral microbes which may cause the formation of further composed of cells which starts beginning the bacterial colonization in mouth(50). The microbial species like Actionmyces specie, Lactobacillus Strepococcus species, species, Candida species. Now these microbes get

transforms into several different types of biofilm formation in the first layer of oral plaque. Biofilm formation begins the maturation process by the aggregation of bacterial growth(50).

The formation of biofilm begins with the coating of tooth surface of the salivary(50). This salivary components such as proteins, histamine, lysozyme, peroxidase, mucin are involved in mouth salivary. The bacterial components such as glycosyltransferase, fructosyltransferase and lipoteichoic acid (51,23). Formation of biofilm process, the bacterial surface adhesions is played by salivary agglutination(52). The initial stages of biofilm formation and adhesion by Streptococcus mutans may be stimulated by the salivary proteins and salivary agglutination(53). The expression of virulence factors of biofilm formation in the oral cavity may be modulated in two ways such as by the bacterial growth in environment and interaction between them (54).

Interaction between the oral cavity with their colonizing microorganisms are the major factors affecting the development of biofilm (54). The virulence of the oral microbes ia not depends on the environmental condition, but also it may cause due to the composition of bacterial flora(55). High range of microbes involved in biofilm formation by the Streptococcus mutans (55). This biofilm formation can be generally treated with effective antibiotics like Chlorohexidine, gluconate, penicillin, methicillin, ampicillin and erythromycin(58).

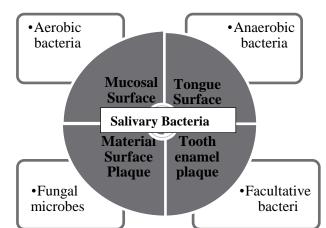


Diagram 2: Flow chart shows the microbes involved in types of dental caries(3).

The communities of oral bacteria are like Streptococcus mutans, Staphylococcus aureus, Lactobacillus, Porphyromonas gingivalis, Escherichiacoiland Candida albicans are plays a major role in dental caries(28). Dental caries is dynamic, and active structure. The role of sugars as the risk factors in the initiation process of dental caries (59).



The preventive measures should be taken for the most common dental disease occurred by the accumulation of bacterial growth in human mouth. Chemical agents are available in market areas, but by using herbal materials against the bacteria can be more benefit for oral health. Herbal materials should have their own anti-bacterial activity and anti-inflammatory activity which indicates oral health of human. Antibiotics like hexetidine, delmopinol, tricolsan, phenolic compounds which may inhibits the maturation and development of biofilm formation as well as effect of bacterial metabolism(60).

Moringa oliefera plant

Moringa (Moringa oliefera) are used as traditional and medicinal plant(4). Moringa plant is a natural-gift for several issues arises by microorganisms[25].Moringa plant is also called as "the miracle tree" because each part of Moringa plant like plant from leaves, bark, seeds, fruits, root are used by the human in their daily life(8). Moringa belongs to the family of Moringaceae . Moringa is fast growing soft wood tree which reach about 12m in height. This plant can grow in both dry and moist condition. Flower and fruits of the Moringa oliefera originates twice in a year(4). This herbal plant are known to horse radish, drumstick, Moringa are the nicknames for Moringa oliefera called in several names. The leaves of Moringa are usually used for food, traditional medicine and traditional ritual ingredients[18]. One of the treatment used by leaves is jaundice. This Moringa leaves contains more vitamin A, calcium, iron, vitamin C and more potassium(27). Moringa leaves and seeds has both anti-bacterial, antiinflammatory and anti-fungal activity against the dental caries which also protect against diabetes[12].

Moringa plant is high in polyphenols like flavonoid and phenolic acids are prominent(4). Moringa is a traditional system of medicine which is used to cure the diseases like malaria, typhoid fever, parasitic diseases, arthritis, hypertension and diabetes. Moringa leaves which includes the antioxidant. anti-inflammatory anti-microbial, anticancer, anti hypertensive and anti-proliferation activities(61). Moringa demonstrates the uncountable various biological activities which are reported from secondary components like alkaloids, tannin, flavonoid, steroids, saponins, coumarins, quinones, resins, lectins among others(6). In the traditional system of medicine, the Moringa is mainly involves in the treatment of skin diseases, anemia, cholera, headaches, anti hypertensive, regulator of thyroid hormones, laxatives and as antibiotics(6).

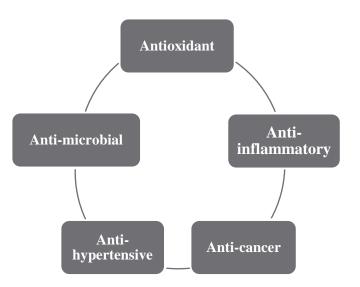


Diagram 3: Flow charts represents the the properties of Moringa leaves(9)

Moringa leaves at the flowering stage can be used against microbial strains causing oral and wound

infections (4). Moringa plant in the community and research regarding he usefulness in the human oral



cavity. Moringa leaves has the more active ingredients can be used as an antibacterial agent in oral cavity. Moringa oliefera has more potential anti-inflammatory in human oral cavity (8). Moringa plant can be consumed as a vegetable and drink, fruits and seeds. Moringa leaves has used for the treatment and management of different diseases in traditional medicine. Moringa leaves has three important character in oral caries.

Role of Moringa leaf in oral caries

The main role of Moringa leaf is antibacterial and anti-inflammatory activities which play important role in oral caries. Moringa leaves is an alternative herbal medicine for use an anticancer agent. Moringa leaves are known to cure the wound healing infections. Moringa leaves extraction are used as herbal medicine in the oral product for the oral health benefits. The ethanol extract of Moringa leaves has an anti-inflammatory effect on male white rats. Moringa leaf ethanol extract is antiinflammatory.

This Moringa leaves has helps in suppressing the growth of oral bacteria like Staphylococcus aureus and E.coil. Moringa leaves has antimicrobial properties on the leaves and seeds. This herbal Moringa leaves acts as an antibacterial agent in oral caries or dental caries, namely bacteria like Porphyromonas gingivalis, Prevotella intermedia, Staphylococcus aureus and Streptococcus mutans. Hence the Moringa leaves extract has an antimicrobial activity against the pathogenic bacteria and biofilm formation. Here proving the properties of Moringa leaves against the biofilm formation by the oral carcinogenic bacteria(8).

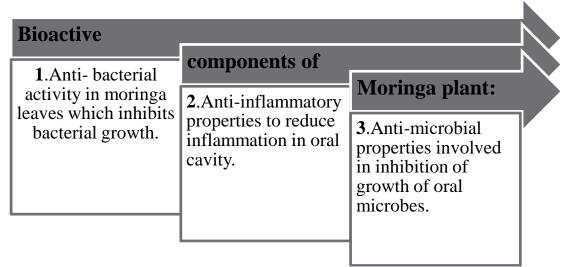


Diagram 4: Main bio active components of Moringa plants(10)

Anti-bacterial activity of Moringa plants

Moringa leaves has rich in antibacterial activity against the oral pathogenic microbes. The use of medicinal plant as an natural ingredient to cure the diseases related to human oral cavity. Plants are used as medicine for human health benefits and also they contain beneficals chemicals. These activities prevent against the pathogenic bacteria and fungi(8).

The antibacterial properties found in this herbal Moringa leaves which makes this plants widely used in dentistry. Moringa plants from leaves, seeds, flowers, bark to root have their tremendous benefits for human. The use of herbal based Moringa plant used in dentistry acts as an antibacterial agent in oral bacteria(8). The main reason for the Moringa leaves is used for the prevention of inhibiting the biofilm formation by bacteria. The biofilm in the oral cavity is structurally and functionally related with bacteria. The proposition of Streptococcus mutans growth is increased due to environmental changes, the oral biofilm which becomes carcinogenic biofilm when it get overgrowth.

The antimicrobial in leaves and antioxidant in flower and bark properties of the Moringa plant is used to formulated for the dental caries to control the oral pathogens[26]. This Moringa based herbal product may be toothpaste or mouthwash which inhibit the growth of bacteria which is responsible for the formation of biofilm. Root canal treatment requires ingredients like antibacterial



properties which gain successful results in the treatment. The medicinal plant of Moringa plants which have their antimicrobial properties and active components has valuable purpose I curing disease or infection of the oral cavity(8).

Anti-inflammatory activity of Moringa plant

Health factors is one of major important role which plays in human daily life. The health of the oral cavity often neglected. Oral health is also an important factor which plays major role in determining the quality of human life and this oral cavity infection is more common diseases in human life, but its causes major defects in healthy life(8).

To avoid the oral issues, herbal based things can be used as an medicine. Moringa leaves has distinct properties which acts as an medicine for health. Moringa plant has the anti-inflammatory properties which is normal protective response mechanism against tissue caused by microbial agents. Moringa leaves has anti-inflammatory effect due to the activity of secondary metabolitescontains in ethanol extract of Moringa leaves, namely flavonoid. Moringa leaves can inhibit the activity of inflammation by the enzymatic activity. Moringa leaves itself has its own anti-inflammatory range upto140 mg/ kgBW. Moringa leaves extraction has the good action against as anti-inflammatory activity and useful in health care in oral cavity or tooth decay.

Phytochemical analysis

The yield obtained by Moringa based toothpaste was 100gm. The result is concluded by the confirmatory phytochemical analysis of Moringa leaves extraction was presented in table1.

Table 2: Showing the phytochemical composition of Moringa leaves extract(5).

S.No	Phytochemicals	Values	
1.	Alkaloids	80%	
2.	Flavonoid	90%	
3.	Saponins	60%	
4.	Tannis	75%	
5.	Steroids	58%	
6.	Phenolics	90%	

Formulation of herbal toothpaste

Formulating herbal based toothpaste against the biofilm formation by carcinogenic oral bacteria[5]. All herbals were dried and grounded using suitable mixer and remaining ingredients were weighed and taken into mortar and mix well until the ingredients is mixed fully[10].Toothpaste are formed by composition of Moringa leaves extraction and the remaining materials were added and mix well until it becomes paste consistency was formed. After the production of herbal toothpaste, the consistency were analyzed by the physio chemical analysis[7,11].

Physio chemical analysis

This Photochemical analysis helps in finding the particular compound present in the Moringa leaves. This analysis which helps in finding their properties. The physio chemicalproperties of herbal based toothpaste based on their parameter the herbal based products should be valid. The physio chemical analysis is shown in table 2.

Table 3 :This table shows the physio chemical analysis of for formulated toothpaste and control
toothpaste

S.No	Parameters	Herbal toothpaste	Control toothpaste	
1.	Moisture	49%	54%	
2	Color	Green	Blue	
3	Homogenicity	Homogenous	Homogenous	
4	Texture	Smooth	Smooth	



5	pН	7.70	7.40
6	Clogging	Absent	Absent

II. CONCLUSION

The presenting literature shows that the Moringa leaves has good natural antibacterial and anti-inflammatory agents in oral cavity. Moringa leaves can be formulated as a toothpaste products which inhibits the growth of bacteria in dental caries. Oral health is most important role in human health. To overcome the dental cavity, herbal based toothpaste is formulated by using Moringa leaves extraction. Oral cavity or tooth decay can be inhibited by the ingredient which has the properties of anti-bacterial and anti-inflammatory activities(36).

This review gives the basic information regrading the antibacterial and anti-inflammatory activities of Moringa leaves act as an herbal which is formulated as oral toothpaste. Oral cavity begins with the formation of biofilm by bacteria. To inhibit the growth of bacterial microbes in oral, the herbal products(43).This study shows promises for a new benefit able herbal based product which inhibit the oral decay responsible microorganisms to overcome the oral issues. The usage of herbal based products which has both anti-bacterial, Antifungal and anti-inflammatory properties in Moringa leaves helps in overcoming from the several oral cavity problem arises in the human world(62).

The quality of the herbal based product must be non toxic due the herbal properties in the natural elements in Moringa leaves. This Moringa leaves shows the positive anti bacterial activity against he oral cavity or oral decay in human mouth. Hence this study concludes that the product formed by formulating the herbal Moringa based toothpaste against the oral cavity responsible microbes which helps in devloping the strength of oral teeth and good quality of teeth to consume any kind of food without any issues or infections in oral teeth(62).

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DOI: 10.35629/7781-080219932004 | Impact Factor value 7.429 | ISO 9001: 2008 Certified Journal Page 2002



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