British Journal of Medicine & Medical Research
10(3): 1-7, 2015, Article no.BJMMR.19571
ISSN: 2231-0614

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Lack of Association between Halitosis and the Presence of *Streptococcus mutans* in Saliva

Jannan Ghapanchi[1], Fereshteh Kamali[2], Zahra Jalaly[3], Hooman Ebrahimi[4], Abdollah Bazargani[5], Sara Poor Shahidi[4*] and Mehrdad Vossoughi[6]

[1]Department of Oral Medicine, School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran.
[2]Department of Oral and Maxillofacial Pathology, School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran.
[3]School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran.
[4]Department of Oral Medicine, School of Dentistry, Tehran University of Medical Sciences, Tehran, Iran.
[5]Department of Bacteriology and Virology, Shiraz University of Medical Sciences, Shiraz, Iran.
[6]Department of Dental Public Health, School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJMMR/2015/19571
Editor(s):
(1) Ibrahim El-Sayed M. El-Hakim, Ain Shams University, Egypt and Riyadh College of Dentistry and Pharmacy, Riyadh, Saudi Arabia.
Reviewers:
(1) Anonymous, SMBT Dental College and Hospital, India.
(2) A. Papazafiropoulou, Department of Internal Medicine and Diabetes Center, Tzaneio General Hospital of Piraeus, Greece.
Complete Peer review History: http://sciencedomain.org/review-history/10408

Original Research Article

Received 17th June 2015
Accepted 14th July 2015
Published 4th August 2015

ABSTRACT

Background: *Streptococcus mutans* is a Gram-positive bacteria which plays a major role in tooth decay. *S. mutans* is among the bacterial agents that initiate biofilm formation on the tooth surface and other bacteria will added then to the attached bacteria to make dental plaque. Some of these secondary bacteria are important agents in halitosis
Objectives: To compare the presence of *Streptococcus mutans* in saliva of patients with halitosis and control group, using culture method.
Materials and Methods: Saliva specimens of 100 patients referring to diagnosis ward of Shiraz

**Corresponding author: Email: sarapourshahidi20@hotmail.com;*

medical school were collected. 51 patients (34 female and 17 male) complaining of halitosis were considered as study group and 49 patients (31 female and 18 male) without halitosis as control group. All specimens were cultured on MSB agar media and isolates were identified as *S. mutans* by traditional tests. The number of *S. mutans* was determined as cfu/ml in each patient saliva.
An organoleptic evaluation was carried out during the initial consultation with the distance of operator to patient (1 m =grade 3) and (30 cm =grade 2 and 10 cm =grade 1).
Results: Of 51 patients with halitosis in 11 (21.61%) patients saliva *S. mutans* were detected. In 49 patients of control group, 14 subjects (28.6%) showed growth of *S. mutans*. There was no Statistical difference between halitosis group and control group in the frequency of *Streptococcus mutans* detection (OR= 0.69, 95% C.I: 0.28-1.71, p=0.419). Statistical analysis also did not show any significant difference between the number of *S. mutans* colonies per ml of saliva between halitosis and control groups (p=0.287).
Conclusion: Our findings showed that there was no association exists between halitosis and the *Streptococcus mutans* presence in saliva.

Keywords: Halitosis; oral malodour; Streptococcus mutans.

1. INTRODUCTION

Halitosis, fetor oris, oral malodor or bad breath are the general terms used to describe the unpleasant breath emitted from a person's mouth regardless of its oral or non-oral origin.

Halitosis is a wide spread problem among the general population. Causes of bad breath can be multifactorial and longtime sufferers can be marred from deep psychological stress. Bad breath occurs as noticeably unpleasant odors are exhaled in breathing. Halitosis is estimated to be the third most frequent reason for seeking dental aid, following tooth decay and periodontal disease. As 9 out of 10 cases of bad breath have an oral origin, the initial inquiry should be with a dentis [1].

Bad breath is originated from the mouth in most cases (85–90%). The simplest way to distinguish the oral from non- oral etiologies is to compare the smell coming from the patient`s mouth with the smell exiting the nose. If the odor is primarily from the mouth, an oral origin may be inferred [2].

The intensity of bad breath differs during the day due to eating certain foods (such as garlic, onions, meat, fish, and cheese), obesity, smoking, and alcohol consumption [3,4]. Bad breath is usually worse upon awakening ("morning breath") since the mouth is exposed to less oxygen and is inactive during the night. It might be also transient, that is disappearing following eating, brushing teeth, flossing or rinsing with specialized mouthwash.

A literature review was conducted and volatile sulfur compounds (VSCs) was found to be a major cause of bad breath. Bacterial enzymes can produce these compounds in the mouth by breaking down the substances such as food debris, cells, saliva, bacteria and blood. Amino acids such as cystine, methionine and cysteine are metabolized through this process and creat malodorous gases. Most common compounds are hydrogen sulfide, methyl mercaptan and dimethyl sulfide [4-6]. The most common bacteria to produce these compounds are gram–negative anaerobic bacteria such as *Porphyromonas gingivalis*, *Prevotella intermedia*, *Fusobacterium nucleatum*, *Bacteroides forsythus* and *Treponema denticola*, and actinobacilli which are commonly isolated from halitosis patients and well-characterized as VSC-producing bacteria [4,6]. Many oral sites such as teeth, periodontal pockets, faulty restorations, buccal mucosa and removable partial dentures harbor these bacteria. However, the posterior dorsal surface of the tongue is considered as the primary site. The presence of halitosis bacteria at the dorsum of the tongue play an important role in the development of bad breath [4-6]. Recognition of this condition is simple, but diseases causing halitosis may produce distinctly different smells. The distinct smell produced by each disease may offer some help in differentiating the etiology of halitosis if various factors causing this condition are understood. Halitosis can be divided into the following categories: (1) halitosis due to local factors of the pathological origin, (2) halitosis due to local factors of non-pathological origin, (3) halitosis due to systemic factors of pathologic origin, (4) halitosis due to systemic factors of non-pathologic origin, (5) halitosis due to systemic administration of drugs, and (6)

halitosis due to xerostomia. Halitosis may be caused by local conditions such as poor oral hygiene, extensive caries, gingivitis, periodontitis, open contacts allowing food impaction, Vincent's disease, hairy or coated tongue, fissured tongue, excessive smoking, healing extraction wounds and necrotic tissues from ulceration [7,8]. Chronic periodontal disease is a major cause of halitosis in adults. Periodontal pockets produce hydrogen sulfides which give off an offensive odor and these pockets encourage trapping of foods [6,7]. Halitosis may also be related to the increase of gram-negative filamentous organisms, the increase of pH to 7.2 and the formation of indoles and amines in the oral cavity [9]. Some studies suggest that kakidrosis may be caused by gram-positive bacteria and especially coryneform bacteria [10]. Recent studies have demonstrated a close association between halitosis and Gram-positive, non-sporeforming anaerobic bacillus which can produce VSC. This bacterium is called Solo bacterium moorei [11-13]. A study in UK showed that in persons suffering halitosis, Lysobacter-type species, *S. salivarius*, *Prevotella melaninogenica*, unidentified oral bacterium, *Prevotella veroralis* and *Prevotella pallens* are the most commonly found species [14]. Much of the previous studies regarding bad breath have concentrated on the characterization of the tongue microflora using conventional microbiological culture methods. It has been estimated according to some studies that almost 50% of the oral microflora is uncultivable in routine methods [15].

Streptococcus mutans (*S. mutans*) is facultatively anaerobic coccus-shaped, Gram-positive bacteria which is commonly found in the oral cavities and play a major role in tooth decay formation and metabolizing sucrose to lactic acid [16,17]. There are twenty-five species of oral streptococci in human oral cavity, each one develops specialized properties for colonization in oral sites and constantly changing conditions to fight competing bacteria. Oral diseases can be initiated by imbalances in the microbial flora. In specific conditions, commensal streptococci can be changed in to opportunistic pathogen, which can initiate the disease and produce damage in the host. So, oral streptococci may be harmless or harmful bacteria according to oral environmental condition.

A study in US showed that there is a positive correlation between the concentrations of *S. mutans* in saliva and its isolation from the smooth surface of the teeth, on the other hand no positive relationship was found between the concentrations of *S. mutans* in the plaque and in saliva [18].

As we know *S. mutans* is among the bacterial agents that initiate biofilm formation on the tooth surface and other bacteria will added then to the attached bacteria to make dental plaque. Some of these secondary bacteria are important agents in halitosis such as *Fusobacterium*, *Porphyromonas*, *Prevotella* and etc. So there may be a possible relation between the number of *S. mutans* present in the mouth and biofilm formation and halitosis. There are few reports in the literature evaluating the association between *S. mutans* and halitosis. Our aim was to find any association between halitosis and the presence of *S. mutans* in saliva of patients who attended Shiraz Dentistry School, Shiraz, Iran.

2. MATERIALS AND METHODS

The study sample consisted of 51 patients with manifestations of halitosis as study group and 49 participants without halitosis as control group. The current study was conducted between September to May 2011 and all patients were examined in oral medicine department of Shiraz University of medical sciences, Shiraz, Iran. With the beginning of the study, patients were instructed not to eat, drink coffee or perform any oral hygiene at least for 4 hours before the examination, and also to refrain from any activity that could mask their bad breath including Perfumed cosmetic products, chewing gum, candy or mouthwash. Onion and garlic should be avoided two days before the examination and any treatment with antibiotic must be completed at least four weeks or more before visiting the clinic. Organoleptic measurement was carried out simply by sniffing the patient's breath and scoring the level of oral malodor. By inserting a translucent tube (2.5 cm diameter, 10 cm length) in to the patient's mouth and having the person exhale slowly, the breath (which is now undiluted by room air) can be evaluated and assigned an organoleptic score [19]. An organoleptic evaluation was carried out during the initial consultation with the distance of operator to patient (1 m =grade 3) and (30 cm =grade 2 and 10 cm =grade 1) each patient filled out a specially designed questionnaire. The questionnaire includes many items (general and local) that may affect halitosis. History of all systemic disease & certain drug consumption upper respiratory tract disease, hormonal statue,

GI problem, constipation, *H. pylori* infection, Diabetes, psychological problems, Liver dysfunction, kidney diseases, blood borne disease & any special condition were made. Complete information was collected about type, frequency, time of day, extent of halitosis, previous therapies & psychological stress as well as typical halitosis co-factors such as dietary and smoking habits. The approval of the local ethics committee was obtained before the trial started and all the patients expressed their written informed consent. Oral cavities were examined carefully by two expert dentists. The recorded clinical findings focused on common halitosis sites including an examination of the oral and pharyngeal soft tissue coated tongue, Waldeyer's ring, salivary ducts as well as dental fillings and restorations. A periodontal screening and assessment of oral hygiene was also evaluated. If signs of periodontal disease or pericoronitis were present, an orthopantomogram (OPG) was taken for further periodontal therapy or extraction. All fillings, dentures, crown & bridges quality & hygiene were evaluated carefully. Plaque index & pocket depth were measured using periodontal probes and disclosing tablets and patients oral hygiene were categorized from poor to excellent (poor, fair, good & excellent). Patients with history of smoking, alcohol and antibiotic consumption, systemic disease, untreated periodontal disease and orthodontic appliances were excluded from the study. In the present study halitosis group consisted of patients with organoleptic score or score more than 2.

Patients' unstimulated saliva samples were collected by spitting the whole saliva into sterile containers in the educational dental clinic and transported to microbiology laboratory immediately.

For isolating and enumerating the *S. mutans*, the inoculated plates were incubated in anaerobic atmosphere by the following method (18). The collected saliva were spread on selective media by a sterile L shaped glass (spread method). So one hundred microliters of whole saliva were spread on mitis salivarius-bacitracin agar (MSB), composed of mitis salivarius agar base (Himedia, Mumbai, India), potassium tellurite 0.01/ml and 0.1 U of bacitracin (Sigma Chemical Co., St. Louis, Mo.) per ml. The plates were incubated at 37ºC for 48 hours in an atmosphere of 80% N2, 10% H2 and 10% CO2.

Colony counts with morphology typical of *S. mutans* were made on MSB agar (Emilson 1981). Microbial counts were expressed as colony-forming units (cfu) *per* ml of saliva. Colonies on the MSB agar plates were visualized by Gram's staining and subjected to the biochemical tests. The biochemical profile of *S. mutans* is: raffinose, mannitol, melibiose, trehalose and inulin positive fermentation; esculin hydrolysis negative in the presence of bile and positive in the absence of bile; negative urease; negative arginine hydrolysis; and resistance to 2U of bacitracin.

Statistical analysis was done by SPSS version 20.0 (IBM SPSS, IBM corporation, Chicago, IL, USA). Chi-square and Student's t tests were used to compare the demographic characteristics between the groups. The relationship of the presence of *S. mutans* with Halitosis, sex groups was assessed using Chi-square test, Odds ratio (OR) and corresponding 95% confidence interval (C.I). Mann-Whitney U test was performed to compare mean number of *S. mutans* colonies per ml of saliva and age between the groups. A p-value less than 0.05 was considered statistically significant.

3. RESULTS

The study sample consisted of 51 patients (34 female and 17 male) with manifestations of halitosis as study group and 49 subjects (31 female and 18 male) without halitosis as control group. The mean age of participants was 25.59 ± 5.15 years. Table 1 shows demographic characteristics of participants. There was no significant difference between the two groups in terms of mean age (p=0.301) and sex ratio (p=0.721). In halitosis group, 11 (21.61%) patients showed growth of *S. mutans*. In control group, 14 subjects (28.6%) *S. mutans* and was detected. There was no Statistical difference between halitosis group and control group in the frequency of *S. mutans* detection (OR= 0.69, 95% C.I: 0.28-1.71, p=0.419). There was also no significant difference between males (10 subjects, 28.6%) and females (15 subjects, 23.1%) in the frequency of *S. mutans* detection (OR= 0.75, 95% C.I: 0.30-1.91, p=0.545). However, the mean age of participants with a *S. mutans* detection (33.45±11.63) was significantly greater that of participants without *S. mutans* (26.73±7.83) (p=0.001). Table 2 summarizes the relationship of the *S. mutans* detection with halitosis, sex, and age.

Table 1. Demographic characteristics of participants

Variable		Group		p
		Halitosis (n=51)	Control (n=49)	
Age		26.11±5.00	25.05±5.20	0.301
Sex	F	34 (67)	31 (63)	0.721
	M	17 (33)	18 (37)	

The qualitative variables are presented as mean±SD and qualitative variables are summarized using frequency (%)

Table 2. The association of presence of *S. mutans* with halitosis and demographic characteristics

Variable		n	*S. mutans*		p	OR	95% C.I
			No	Yes			
Group	Control	49	35 (71.40)	14 (28.60)	0.419	1	-
	Halitosis	51	40 (78.39)	11 (21.61)		0.69	0.28-1.71
Sex	M	35	25 (71.40)	11 (28.60)	0.545	1	-
	F	65	51 (76.90)	14 (23.10)		0.75	0.30-1.91
Age	-	-	26.73±7.83	33.45±11.63	0.001	-	-

The qualitative variables are summarized using mean±SD and qualitative variables are summarized using frequency (%)

Statistical analysis also did not show any significant difference between the number of *S. mutans* colonies per ml of saliva in halitosis (3.49±7.53) and control (4.84±7.98) groups (p=0.287).

4. DISCUSSION

Bad breath has been recorded in the literature for thousands of years. It is estimated that more than 85% of bad breath originates from oral cavities and numerous reports have proved this finding [19]. Halitosis is estimated to be the third most frequent reason for seeking dental aid, following tooth decay and periodontal disease. The etiology of halitosis can be of systemic (extra–oral) or intra–oral origins. Halitosis is often caused by food debris and biofilm buildup on the teeth and tongue. Oral bad breath originates from bacterial flora with high capability to produce volatile sulfur compounds. Halitosis may be also related to the increase of Gram-negative filamentous organisms, the increase of pH to 7.2 and the formation of indoles and amines in the oral cavity [10]. Periodontopathogenic bacteria can produce volatile sulfur compounds (VSCs) including hydrogen sulfide (H2S) and methyl mercaptan (CH3SH) [20]. The H2S and CH3SH in exhaled air of the mouth were found to be the major elements of oral malodor. These elements are produced primarily through the putrefaction of the salivary bacteria, periodontal pockets, tongue surface and in other oral sites [21].

The oral microorganisms which are most likely to cause oral malodor include Gram-negative bacteria species including *Treponema denticola*, *Porphyromonas gingivalis*, *Porphyromonas endodontalis*, *Prevotella intermedia*, *Bacteroides loescheii*, Enterobacteriaceae, *Tannerella forsythensis*, *Centipeda periodontii*, *Eikenella corrodens*, *Fusobacterium nucleatum* [22].

However, there exists no significant association between halitosis and any specific bacterial infection. These findings suggest that halitosis reflects complex interactions between numerous species of oral bacteria. The volatile sulfide compounds, diamines and short chain fatty acids are considered as the main agents involved in oral malodor [5]. This finding may be associated with changing in oral bacterial flora and decrease in salivary flow in this group.

Some studies suggest that *Solobacterium moorei* is associated with oral halitosis. Haraszthy et al. [12] showed that *S. moorei* can be isolated from dorsal surface of the tongue in 100% of the halitosis patients and 14% of subjects without halitosis. Infection with this bacteria is correlated with level of produced volatile sulfur compounds. Apparently, some oral bacteria can inhibit the growth of *S. moorei*. L. Masdea reported that *Streptococcus salivarius* K12 has an inhibitory effect on halitosis associated bacterial growth. These bacteria include *Solobacterium moorei* CCUG39336, four clinical *S. moorei* isolates, *Atopobium parvulum* ATCC33793 and

Eubacterium sulci ATCC35585. This study demonstrated that *S. salivarius* K12 inhibits the growth of all Gram-positive bacteria tested, but the extent bacterial inhibition was varied. *E. sulci* ATCC35585 was the most sensitive strain, while all five *S. moorei* isolates were inhibited to a lesser extent. This researcher recommended that using antimicrobial agents such as tea tree oil and alpha-bisabolol in oral health care products may reduce the halitosis due to *S. moorei* growth inhibition [23].

A study on oral microflora in 20 patients suffering from halitosis and 12 subjects without it in UK showed that uncultured *Veillonella* spp. are the most uncultured species in both the halitosis samples and the control group.

P. pallens was unique to the halitosis group which was found in nine halitosis samples (2.7% of the totalclones). The next prevalent species only in the halitosis group was *Bulleidia* (*Solobacterium moorei*) which was found in five samples (1.1% of the clones). The most important finding of the present study was the higher microbial diversity on the dorsal surface of the tongue in halitosis subjects compared to control group [14]. Kazor study presented *that S. salivarius*, *Rothia mucilaginosa* (*Stomatococcus mucilaginosus*) and an uncharacterized cultivable species of *Eubacterium* (strain FTB41) were the most prevalent microorganisms [13]. Chen et al. [24] reported that the administration of *Lactobacillus salivarius* TI2711 (LS1) neutralized salivary pH and decreased the numbers of black-pigmented anaerobic rods in the saliva of healthy subjects. Many studies found that consumption of products containing probiotic lactobacilli will reduce the number of *S. mutans* in the oral cavity and decrease caries rate [25,26]. Iwamoto et al. concluded that physiologic halitosis is decreased significantly after consumption of probiotic tablets containing *L. salivarius* WB21 And according to the current study the level of H2S, CH3SH, VSC and finally bad breath is reduced prominently [27].

A key finding of our study was that *S. mutans* detection from the saliva of patients with halitosis did not show any statistical difference with control group and presence of this microorganism is not a marker of halitosis. However, further studies are required to prove this finding and also to establish the relationship between VSCs production and these gram positive bacteria.

5. CONCLUSION

Halitosis has multifactorial origin and our findings showed that there was no association exists between halitosis and the Streptococcus mutans presence in the saliva.

ACKNOWLEDGMENTS

The authors thank the Vice –Chancellory of Shiraz University of Medical Sciences for suuporting this research. This article is based on the thesis by Dr Zahra Jallali. The authors also thank Dr Vossoughi for the statistcal analysis.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Loesche WJ, Kazor C. Microbiology and treatment of halitosis. Periodontology. 2002;28:256-79.
2. Rosenberg M. The science of bad breath. Scientific American. 2002;286(4):72-9.
3. Rosenberg M, Knaan T, Cohen D. Association among bad breath, body mass index, and alcohol intake. Journal of Dental Research. 2007;86(10):997-1000.
4. Porter SR, Scully C. Oral malodour (halitosis). BMJ (Clinical Research ed). 2006;333(7569):632-5.
5. Oral malodor. Journal of the American Dental Association (1939). 2003;134(2): 209-14.
6. Scully C, Felix DH. Oral medicine--update for the dental practitioner: oral malodour. British Dental Journal. 2005;199(8):498-500.
7. Massler M, Emslie RD, Bolden TE. Fetor ex ore; A review. Oral Surgery, Oral Medicine, and Oral Pathology. 1951;4(1): 110-25.
8. Hine MK. Halitosis. Journal of the American Dental Association (1939). 1957;55(1):37-46.
9. McNamara TF, Alexander JF, Lee M. The role of microorganisms in the production of oral malodor. Oral surgery, oral medicine, and oral pathology. 1972;34(1):41-8.
10. Lukacs A, Korting HC, Ruckdeschel G, Ehret W. Do cutaneous coryneform bacteria produce short-chain fatty acids in vitro? Dermatologica. 1991;182(1):32-4.

11. Haraszthy VI, Zambon JJ, Sreenivasan PK, Zambon MM, Gerber D, Rego R, et al. Identification of oral bacterial species associated with halitosis. Journal of the American Dental Association (1939). 2007;138(8):1113-20.
12. Haraszthy VI, Gerber D, Clark B, Moses P, Parker C, Sreenivasan PK, et al. Characterization and prevalence of Solobacterium moorei associated with oral halitosis. Journal of Breath Research. 2008;2(1):017002.
13. Kazor CE, Mitchell PM, Lee AM, Stokes LN, Loesche WJ, Dewhirst FE, et al. Diversity of bacterial populations on the tongue dorsa of patients with halitosis and healthy patients. Journal of Clinical Microbiology. 2003;41(2):558-63.
14. Riggio MP, Lennon A, Rolph HJ, Hodge PJ, Donaldson A, Maxwell AJ, et al. Molecular identification of bacteria on the tongue dorsum of subjects with and without halitosis. Oral Diseases. 2008;14(3):251-8.
15. Socransky SS, Gibbons RJ, Dale AC, Bortnick L, Rosenthal E, Macdonald JB. The microbiota of the gingival crevice area of man. I. Total microscopic and viable counts and counts of specific organisms. Archives of Oral Biology. 1963;8:275-80.
16. Loesche WJ. Microbiology of Dental Decay and Periodontal Disease. In: Baron S, editor. Medical Microbiology. Galveston TX: The University of Texas Medical Branch at Galveston; 1996.
17. Nicolas GG, Lavoie MC. [Streptococcus mutans and oral streptococci in dental plaque]. Canadian Journal of Microbiology. 2011;57(1):1-20.
18. Duchin S, van Houte J. Colonization of teeth in humans by Streptococcus mutans as related to its concentration in saliva and host age. Infection and Immunity. 1978;20(1):120-5.
19. Rosenberg M. Clinical assessment of bad breath: current concepts. Journal of the American Dental Association (1939). 1996;127(4):475-82.
20. Persson S, Edlund MB, Claesson R, Carlsson J. The formation of hydrogen sulfide and methyl mercaptan by oral bacteria. Oral Microbiology and Immunology. 1990;5(4):195-201.
21. Bosy A, Kulkarni GV, Rosenberg M, McCulloch CA. Relationship of oral malodor to periodontitis: evidence of independence in discrete subpopulations. Journal of Periodontology. 1994;65(1):37-46.
22. Awano S, Gohara K, Kurihara E, Ansai T, Takehara T. The relationship between the presence of periodontopathogenic bacteria in saliva and halitosis. International Dental Journal. 2002;52(Suppl 3):212-6.
23. Masdea L, Kulik EM, Hauser-Gerspach I, Ramseier AM, Filippi A, Waltimo T. Antimicrobial activity of Streptococcus salivarius K12 on bacteria involved in oral malodour. Archives of Oral Biology. 2012;57(8):1041-7.
24. Chen LJ, Tsai HT, Chen WJ, Hsieh CY, Wang PC, Chen CS, et al. In vitro antagonistic growth effects of Lactobacillus fermentum and lactobacillus salivarius and their fermentative broth on periodontal pathogens. Brazilian Journal of Microbiology: [Publication of the Brazilian Society for Microbiology]. 2012;43(4): 1376-84.
25. Ahola AJ, Yli-Knuuttila H, Suomalainen T, Poussa T, Ahlstrom A, Meurman JH, et al. Short-term consumption of probiotic-containing cheese and its effect on dental caries risk factors. Archives of Oral Biology. 2002;47(11):799-804.
26. Caglar E, Cildir SK, Ergeneli S, Sandalli N, Twetman S. Salivary mutans streptococci and lactobacilli levels after ingestion of the probiotic bacterium Lactobacillus reuteri ATCC 55730 by straws or tablets. Acta Odontologica Scandinavica. 2006;64(5): 314-8.
27. Iwamoto T, Suzuki N, Tanabe K, Takeshita T, Hirofuji T. Effects of probiotic Lactobacillus salivarius WB21 on halitosis and oral health: An open-label pilot trial. Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontics. 2010;110(2):201-8.

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/10408