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# **Effect of probiotic mouthwashes in comparison to chlorhexidine on periodontal health: Systematic review and meta-analysis**

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**Abstract**---Aim: To evaluate and compare the efficacy of Chlorhexidine and Probiotic mouthwashes in reducing plaque accumulation and gingivitis. Materials and methods: PubMed, Ovid SP, Embase, and Google Scholar databases were searched from years 1980 upto April 2020 for potential papers using relevant MeSH terms and pre-specified inclusion and exclusion criteria by two independent reviewers. Studies comparing Probiotics and Chlorhexidine in mouthwash form were included and evaluated for outcomes such as plaque score, gingival score. Results: A total of eleven articles were

included for qualitative analysis, among them seven studies qualified for quantitative analysis. Random effects model was carried out to perform meta-analysis, basing on heterogeneity. Plaque index (PI): Significant reduction of plaque scores post-intervention were found when compared to baseline in both Probiotic mouthwash group [ $p=0.00001$ , MD: 0.87, 95% CI: 0.41 to 1.33] and chlorhexidine mouthwash groups [ $p=0.00001$ , MD: 0.97, 95% CI: 0.66 to 1.28]. No significant intergroup difference was found between Chlorhexidine and Probiotic mouthwash groups ( $p= 0.73$ ). Gingival index (GI): Significant reduction of gingival index scores post-intervention were found when compared to baseline in both Probiotic mouthwash group [ $p=0.001$ , MD: 0.48, 95% CI: 0.19 to 0.77] and chlorhexidine mouthwash groups [ $p=0.0001$ , MD: 0.46, 95% CI: 0.23 to 0.68]. No significant intergroup difference was found between Chlorhexidine and Probiotic mouthwash groups ( $p= 0.90$ ). Conclusion: Low-quality evidence suggests that short-term usage of Probiotic mouthwash is comparable chlorhexidine in reducing gingival inflammation plaque scores.

**Keywords**---Adults, Adolescents, Children, Chlorhexidine, Mouthwash, Probiotic.

## Introduction

Probiotics are defined as friendly bacteria, which are live, non-pathogenic, when administered in adequate amounts in foods or as dietary supplements, render health benefits to the host. Probiotics are showing encouraging results in the field of medicine, more literature is proving the favourable effects of probiotics on human health such as improving the health of gastrointestinal tract, respiratory health, and protection against allergies.<sup>1-3</sup>

Probiotics usage in Dentistry is gaining pace as there is an increase in the number of studies presenting favourable outcomes. Probiotics ingestion has numerous oral health benefits such as improvement of outcomes after non-surgical periodontal therapy,<sup>4</sup> reduction of clinical gingivitis,<sup>5,6</sup> halitosis,<sup>7</sup> reduction in the severity of cancer therapy-induced oral mucositis,<sup>8</sup> reduction of oral candidiasis in denture wearers,<sup>9</sup> reduction of *S.mutans* counts.<sup>10</sup> Periodontal benefits of consumption of probiotics might be because probiotics will reduce the load of harmful bacteria, reduce the adhesion and proliferation of bacteria on the tooth surface, inhibition of intercellular plaque matrix formation, reduction of toxins produced thereby decreasing the plaque formation and gingivitis.<sup>11</sup>

Most of the reported systematic reviews evaluated the effect of oral consumption of probiotics and its effects on gingiva, plaque, and periodontal health.<sup>12</sup> To the best of our knowledge, so far no systematic review has evaluated the efficacy of topical usage of probiotic mouthwash on periodontal health. The aim of the present systematic review and meta-analysis is to evaluate the efficacy of topically used Probiotic mouthwash in the reduction of gingivitis and plaque accumulation and compare it with Chlorhexidine.

## Materials and Methods

This study is reported according to PRISMA guidelines. **Eligibility criteria:** The search strategy was conducted using the population, intervention, comparison, and outcome [PICO] framework, based on the following question. “**Comparison of Probiotic and chlorhexidine mouthwashes in reducing gingival and plaque scores clinically**”. The Population Intervention Comparison Outcome (PICO) search strategy of the systematic review was: [P] patient: any age group; [I] intervention: any strain of probiotic in any concentration used as a mouthwash; [C] comparison: chlorhexidine mouthwash only.; [O] outcome of interest: clinical scores of plaque accumulation, gingival status.

An electronic search was performed in Four databases such as PubMed, Ovid SP, Embase, Google Scholar. The search was conducted till April 2020. Articles published in English are only included. The search was based on the pre-specified question using relevant MeSH terms. *((mouthwash) OR (mouth rinse)) AND (and) (Probiotic)* for three databases (PubMed, Ovid SP, Embase). Term “Probiotic Mouth rinse” was used in google scholar and the first 300 papers were screened for relevant papers.

### Eligibility criteria

In-vivo studies that compared any probiotic mouthwash (any concentration) and chlorhexidine for evaluation gingival and plaque scores. Studies in which probiotic was ingested in the form of tablets or(,) food products,(or) toothpaste form were excluded as we aim to evaluate only probiotic mouthwashes (topical only). Comparative studies, case reports, narrative reviews, and systematic reviews and articles that cannot be translated into English were excluded. Initially studies obtained after comprehensive MeSH terms search were imported to Zotero([www.zotero.org](http://www.zotero.org)) from all the databases and exclusion of duplicates was performed and then screening of titles and abstract was carried out. Potential articles were then included for a full review.

Data extraction and data analysis was performed by two independent reviewers and was recorded on excel. The data form contained the information regarding the author, year of publication, age of the participants, number participated in the study, intervention used, duration of intervention, comparison, and outcome variables. The outcome measures of interest were plaque score, gingival inflammation.

**Data synthesis:** Qualitative data; all studies were included for qualitative data analysis. Quantitative data was presented only in seven studies.

**Risk-of bias (RoB) assessment:** The methodological quality assessment of the included articles was conducted independently by two review team members using the Cochrane Collaboration's criteria. Quality of all selected trials was assessed for risk of bias under seven domains: sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, completeness of outcome data, selective reporting of outcomes, and other sources of bias. Studies with seven domains of low bias risk were classified

into a low risk of bias group. If one or more domains presented a high risk of bias, these studies were categorized as having a high risk of bias; otherwise, the study was categorized as having an unclear risk of bias.

## Results

In all the databases, 964 records were found, of which 11 were duplicates. Removing the duplicate articles, 953 records were screened by title and abstract. Full text of 18 potentially relevant papers were evaluated, among them eight were excluded (Reasons for exclusion is given in *Table-1*).<sup>13-20</sup> Consequently, 11 studies were included for qualitative analysis,<sup>21-31</sup> among them seven studies were included for meta-analysis.<sup>21-25,28,29</sup> A flowchart of the search results is presented in *figure-1*.

Table-1: Table showing Excluded articles and reasons for exclusion

Sno	Excluded articles	Reasons for Exclusion
1.	Sajedinejad 2018	Control was not Chlorhexidine, plaque score was not measured.
2.	Klarin 2018	Plaque and gingival scores were not measured.
3.	Yousuf 2017	Control was not Chlorhexidine.
4.	Sujlana 2017	Control was not Chlorhexidine.
5.	Jindal 2016	Control was not Chlorhexidine.
6.	Jyothika 2015	Plaque and gingival scores were not measured.
7.	Dhawan 2013	Control was not Chlorhexidine
8.	Noordin 2007	Control was not Chlorhexidine

### Flow Diagram

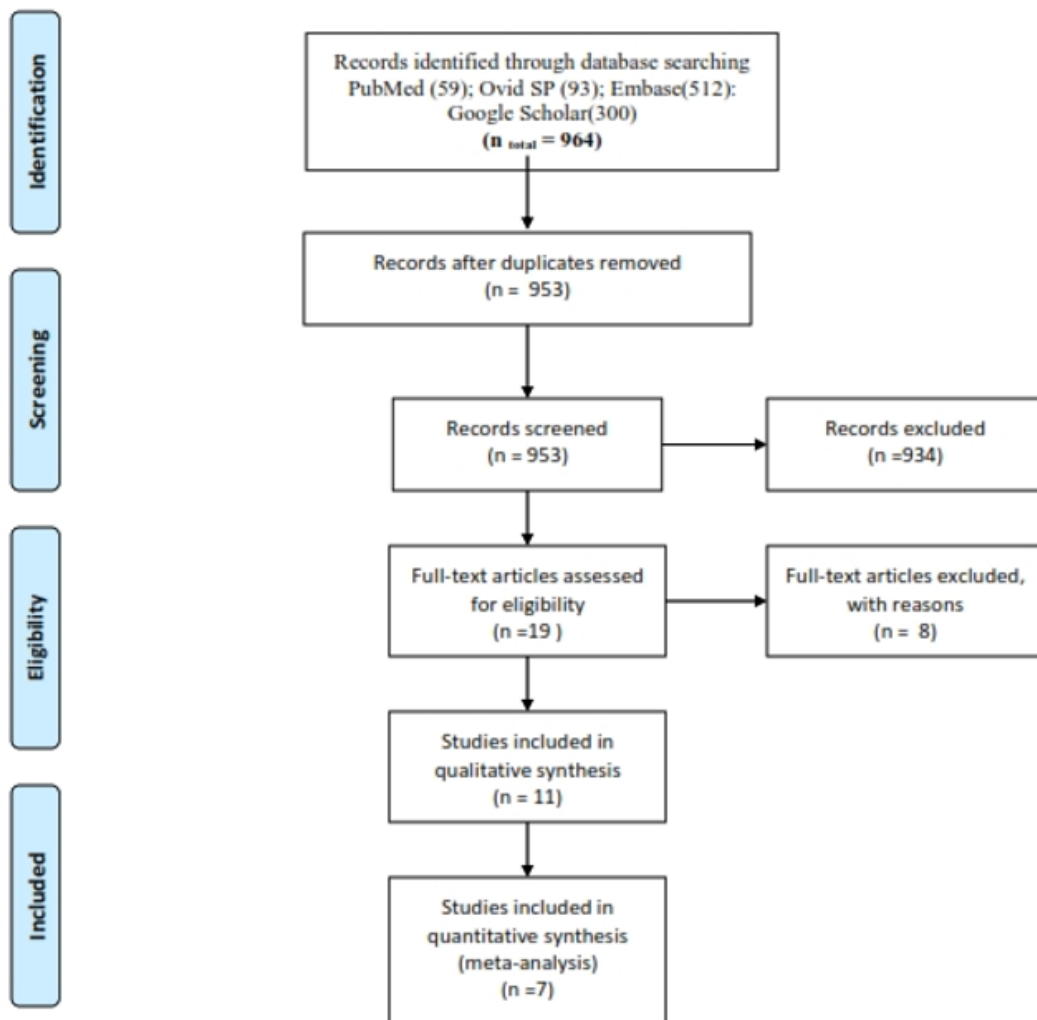


Figure-1: Flowchart

*Characteristics of included studies:* The characteristics of the included studies are shown in Table-2. All the studies were published between 2010 to 2019. Among eleven studies, six studies were performed in children aged below 16 years,<sup>22,24,27-29,31</sup> and the rest of the five studies were performed in adults (> 18 years).<sup>21,25-27,30</sup> The duration of mouthwash used across studies ranged from 7 to 30 days. 7 days in one study,<sup>28</sup> 14 days in five studies,<sup>21,22,25,30,31</sup> 15 days in two studies,<sup>26,29</sup> 28 days in two studies,<sup>23,27</sup> 30 days in one study.<sup>24</sup> Frequency of mouthwash usage was once<sup>22,24,29,31</sup> or twice daily.<sup>21,23,25-28,30</sup> In all the studies 0.2% concentration of chlorhexidine was compared with probiotic mouthwashes. In all the studies probiotics were freshly prepared before usage. All eleven included studies evaluated Plaque scores, but gingival index was only evaluated in nine studies.

The gingival index was not evaluated in two studies.<sup>28,29</sup> Qualitative analysis: All the eleven included studies were included for qualitative analysis. Quantitative analysis: Seven studies were included in quantitative data synthesis. Four studies were excluded from quantitative data analysis (missing data).<sup>26,27,30,31</sup>

Table-2: Characteristics of Included studies.

S no	Author-year	Age	Number	Duration of intervention	Intervention	Gingival index score		Plaque index Score		Other measures evaluated in the study but not included in the current review	Result
						Probiotic	Chx	Probiotic	Chx		
1.	Shah 2019	Not mentioned	30 divided into three groups G1: 10 G2: 10 G3: 10	Twice daily after brushing for 28 days.	G1: 0.2% Chlorhexidine G2: Probiotic mouthwash (Sporlac dissolved in water) G3: Control	Sporlac-B (0.78 ± 0.34) to PI (0.18 ± 0.22)	Chx group - B(0.88 ± 0.46) to PI(0.34 ± 0.44)	Sporlac-B(1.03 ± 0.58) to PI(0.16 ± 0.18)	Chx group-B(1.09 ± 0.48) to PI(0.55 ± 0.52)	S.mutans count in saliva	Plaque score both chx and probiotic are comparable  Gingival index better with Probiotic significantly than Chx.
2	Vivek and mahender 2019	25-35 years	45 divided into three groups G1: 15 G2: 15 G3: 15	Twice daily after brushing for 14 days	G1: Probiotic mouthwash (Darolac dissolved in water) G2: 0.2% Chlorhexidine G3:	Darolac-B(0.12 ± 0.08) to PI (0.07 ± 0.04)	Chx group - B(0.18 ± 0.09) to PI(0.06 ± 0.03)	Darolac-B(0) to PI (0.08 ± 0.03)	Chx group-B(0) to PI(0.06 ± 0.03)	Oral hygiene index-simplified	No significant difference between Chx and Probiotic mouth wash in terms of both Plaque and Gingival

			15		Control							scores reduction Both were equally efficacious.
3	Sharma 2019	6 to 9 years old.	60 divided into three groups G1: 20 G2: 20 G3: 20	Once /twice daily after brushing for 14 days	G1: control distilled water G2: Chx G3: Probiotic mouthwash (Darolac dissolved in water)	Darolac- B(2.08±0.46) to PI(0.66±0.14)	Chx group - B(1.98±0.31) to PI(0.86±0.24)	Darolac- B(2.30±0.45) to PI(0.72±0.21)	Chx group- B(2.14±0.27) to PI(0.81±0.22)	-	Probiotic group was better than Chx in both Plaque and Gingival scores reduction.	
4	Kandaswamy 2018	10-12 years	45 divided into three groups G1: 15 G2: 15 G3: 15	Once daily after brushing for 30 days	G1: Probiotic mouthwash (Bifilac sachets) dissolved in water) G2: 0.2% Chlorhexidine G3: Sesame seed oil	Bifilac - B(0.32 ± 0.24) to PI(0.04 ± 0.83)	Chx group - B(0.37 ± 0.26) to PI(0.05 ± 0.09)	Bifilac - B(0.96 ± 0.31) to PI(0.36 ± 0.26)	Chx group- B(0.86 ± 0.25) To PI(0.18 ± 0.19)	-	No significant difference between Chx and Probiotic mouth wash in terms of both Plaque and Gingival scores reduction. Both were equally efficacious.	
5	Deshmukh 2017	18-21 years	45 divided into three	Twice daily after brushing for 14	G1: Herbal mouth rinse. G2: Chlorhexidine	Darolac- B(0.18±0.11) to	Chx group - B(0.19±0.0	Darolac- B(0) to PI(0.04±	Chx group- B(0) to PI(0.04±0.0	Oral hygiene index was measure	No significant difference between Chx and	

			groups G1: 15 G2: 15 G3: 15	days.	chlorhexidine G3: Probiotic mouthwash (Darolac dissolved in water)	PI (0.04±0.09)	9) to PI(0.04±0.02)	0.06)	7)	d.	Probiotic mouthwash in terms of both Plaque and Gingival scores reduction Both were equally efficacious.
6	Nadkerny 2015	20 to 30 years	45 divided into three groups G1: 15 G2: 15 G3: 15	Twice daily after brushing for 15 days	G1: Probiotic mouthwash (Sporlac Plus dissolved in water) G2: 0.2% Chlorhexidine G3: Control(saline)	Mean difference was presented. Mean difference between baseline and post intervention was (0.45±0.174)	Mean difference between baseline and post intervention was (0.40±0.124)	Mean difference between baseline and post intervention was (0.36±0.14)	Mean difference between baseline and post intervention was (0.21±0.15)	Oral hygiene index was measured.	No significant difference between Chx and Probiotic mouthwash in terms of both Plaque and Gingival scores reduction Both were equally efficacious.
7	Mishra 2014	6 to 14 years	60 divided into three groups G1: 20 G2: 20 G3:	Twice daily after brushing for 7 days	G1: Probiotic mouthwash (Evora Plus dissolved in water) G2: 0.2% Chlorhexidine	Not measured	Not measured	Evora Plus- B(1.86±0.32) to PI(1.58±0.29)	Chx group- B(1.68±0.46) To PI(0.32±0.25)	S.viridans count was measured.	Chx was better than probiotic in terms of plaque score significantly.



			20		G3: Herbal mouth rinse						
8	Shah 2014	6 to 10 year s.	40 divi ded into four gro ups . G1: 10 G2: 10 G3: 10 G4: 10	Twice daily after brus hing for 28 days	G1: Probiotic mouthw ash (Evora Plus dissolve d in water) G2: 0.2% Chlorhe xidine  G3: Fluoride mouth rinse G4: Control	Only mean gingiv al score given B(2.5) to PI(0.5)	Only mean gingiv al score given B(2.4) to PI(0.6)	Only mean plaque score given B(2.5) to PI(0.5).	Only mean plaque score given B(2.4) to PI(0.6) .	-	No significan t difference between Chx and Probiotic mouth wash in terms of both Plaque and Gingival scores reduction . Both were equally efficaciou s.
9	Thakk ar 2013	13- 15 year s	90 chil dre n divi ded into thre e gro ups . G1: 30 G2: 30 G3: 30	Once daily for 15 days	G1: control distilled water G2: Chx G3: Probiotic mouthw ash (Darolac dissolve d in water)	Not meas ured	Not meas ured	Darola c-  B(1.09 ±0.51) to PI(0.07 ±0.01)	Chx group-  B (0.92± 0.48) to PI(0.1 1±0.0 2)	-	Probiotic mouth rinse was better than Chx in terms of plaque score reduction .
1 0	Priyad arshin i 2013	21- 45 year s.	45 divi ded into thre e	Twice daily after brus hing for 14	G1: Probiotic mouthw ash (Sporlac Plus	Only mean gingiv al score given	Only mean gingiv al score given	Only mean plaque score given B(2.5)	Only mean plaque score given B(2.65	S.mutan s count in saliva	Probiotic better than Chx in both plaque and

			groups G1: 15 G2: 15 G3: 15	days	dissolved in water) G2: 0.2% Chlorhexidine  G3: Control(saline)	B(0.38) to PI(0.18).	B(0.4) to PI(0.3).	to PI(1.51).	) to PI(1.95).		gingival scores.
11	Harini 2010	6-8 years	45 divided into three groups G1: 15 G2: 15 G3: 15	Once daily after brushing for 14 days	G1: control distilled water G2: Probiotic mouthwash G3: Chx	Only mean gingival score given B(2.3) to PI(0.20).	Only mean gingival score given B(2.2) to PI(0.3).	Only mean plaque score given B(2.0) to PI(0.2).	Only mean plaque score given B(2.4) to PI(0.4).	-	Gingival index better with probiotic group than Chx  Plaque index both probiotic and Chx similar.

\*Abbreviations used in this table: G1=group 1; G2=group 2; G3=group 3 G4=group 4: B=baseline : PI= post intervention; m= mean; sd=standard deviation

### **Risk of Bias**

Risk of bias (figure-2) was evaluated according to Cochrane guidelines. Randomization was not mentioned in two studies.<sup>27,31</sup> Allocation concealment was mentioned only in four studies.<sup>25,28-30</sup> Blinding of participants and personnel and blinding of outcome assessment was carried out in only two studies.<sup>29,30</sup> attrition and selective reporting bias was not present in any of the included studies.

	Random sequence generation (selection bias)	Allocation concealment (selection bias)	Blinding of participants and personnel (performance bias)	Blinding of outcome assessment (detection bias)	Incomplete outcome data (attrition bias)	Selective reporting (reporting bias)	Other bias
Deshmukh 2017	+	+	-	-	+	+	+
Harini 2010	-	-	-	?	+	+	+
Ingle 2013	+	+	+	+	+	+	+
Kandaswamy 2018	+	-	-	-	+	+	+
Mishra 2014	+	+	-	-	+	+	+
Nadkerny 2015	+	-	-	-	+	+	+
Shah 2014	-	-	-	-	+	+	+
Shah 2019	+	-	-	-	+	+	+
Sharma 2019	+	-	-	-	+	+	+
Thakkar 2013	+	+	+	+	+	+	+
Vivek 2019	+	-	-	-	+	+	+

Figure-2: Risk of Bias

**Overall risk of Bias**

Only two studies presented low risk of bias in all the seven domains.<sup>29,30</sup> remaining all studies (n=9) presented high risk of bias. The overall risk of bias is high.

**Comparative effectiveness of Probiotic and Chlorhexidine mouthwashes in reducing gingival inflammation**

Gingival status was evaluated in nine studies,<sup>21-27,30,31</sup> among the eleven included studies. Gingival status in all the studies included were evaluated at baseline and follow up visit with Gingival index (GI). Among the nine studies only five studies presented data (mean and standard deviations) clearly,<sup>21-25</sup> other four studies data was not presented clearly so were excluded from meta-analysis.<sup>26,27,30,31</sup> In the nine studies that evaluated gingival scores, Five studies reported no significant difference in the reduction of gingival score from baseline to post-intervention in both Probiotic and Chlorhexidine.<sup>21,23-26</sup> Four studies reported better gingival index outcome with Probiotic group in comparison to Chlorhexidine group.<sup>22,23,30,31</sup> *Quantitative analysis of included studies (n=5):* Probiotic mouthwashes resulted in a significant reduction of GI scores when compared to baseline [p=0.001, MD: 0.48, 95% CI: 0.19 to 0.77]. Chlorhexidine mouthwash group also resulted in a significant reduction of GI scores when post-intervention scores were compared to baseline [p=0.0001, MD: 0.46, 95% CI: 0.23 to 0.68]. There were no significant differences between Probiotic and chlorhexidine mouthwash groups in terms of gingival index score reduction (p= 0.90). *figure-3*

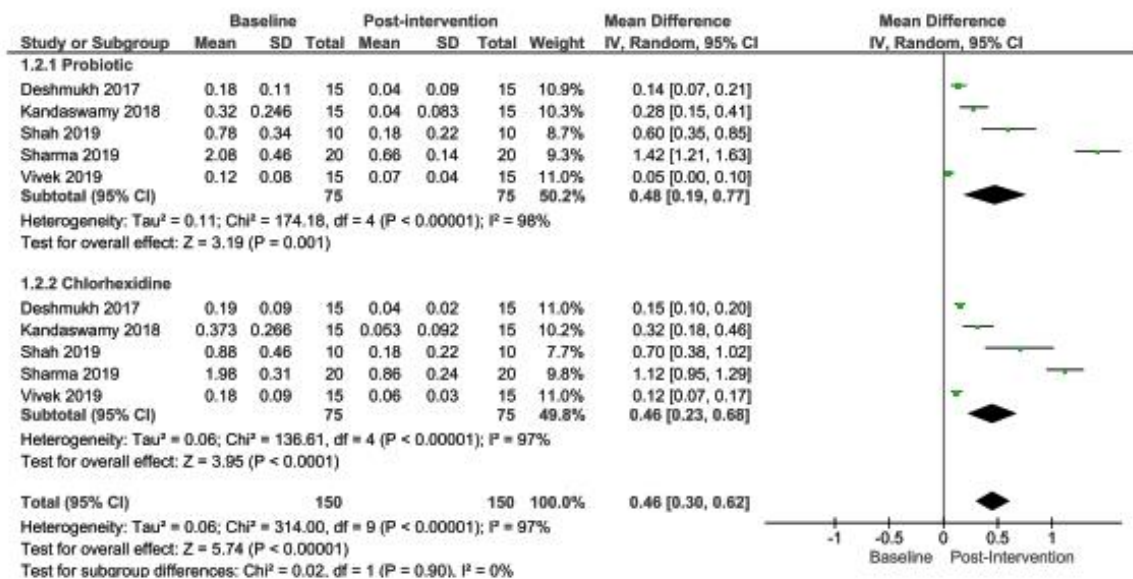


Figure-3: Gingival Index

### Comparative effectiveness of Probiotic and Chlorhexidine mouthwashes in reducing Plaque accumulation

All the eleven included studies, evaluated Plaque Index scores. In the eleven studies that evaluated plaque scores, Seven studies reported no significant difference in the reduction of plaque score from baseline to post-intervention in both Probiotic and Chlorhexidine.<sup>21,23-27,31</sup> Three studies reported better Plaque Index outcome with Probiotic group in comparison to Chlorhexidine group.<sup>22,29,30</sup> One study reported better Plaque Index outcome with Chlorhexidine group in comparison to Probiotic group.<sup>28</sup> Among eleven studies, only five studies were included in meta-analysis,<sup>22-24,28,29</sup> remaining six studies were excluded as data was not clear, or presented as mean difference.<sup>21,25-27,30,31</sup> *Quantitative analysis of included studies (n=5):* Probiotic mouthwashes resulted in a significant reduction of PI scores when compared to baseline [p=0.00001, MD: 0.87, 95% CI: 0.41 to 1.33]. Chlorhexidine mouthwash group also resulted in a significant reduction of PI scores when post-intervention scores were compared to baseline [p=0.00001, MD: 0.97, 95% CI: 0.66 to 1.28]. There were no significant differences between Probiotic and Chlorhexidine mouthwash groups in terms of Plaque Index score reduction (p= 0.73). *figure-4*

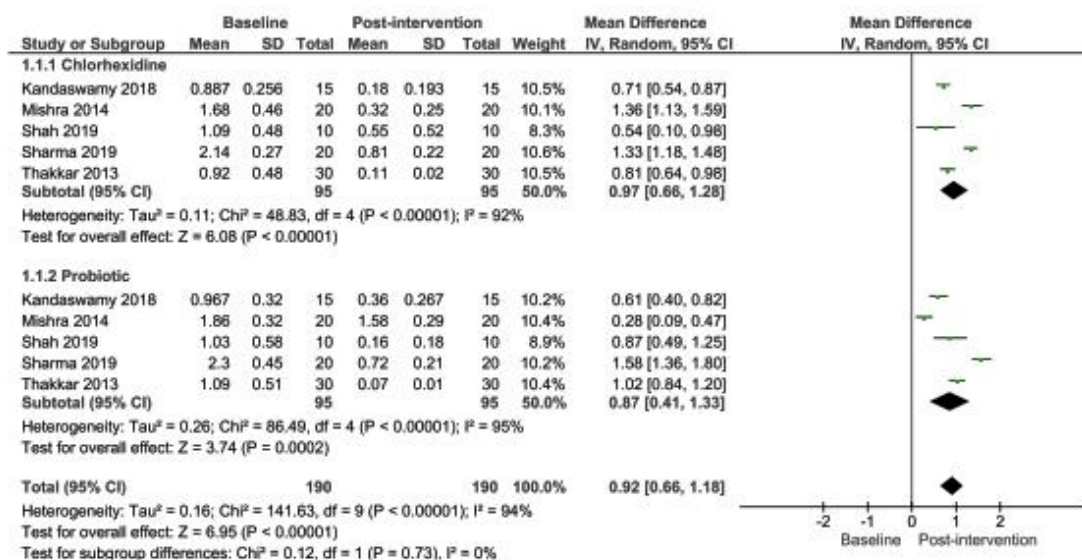


Figure-4: Plaque Index

### Discussion

Reduction of plaque accumulation and maintaining good gingival health is a prime requisite for a healthy periodontium. Chemical plaque control methods, when used in conjunction with mechanical plaque control methods, complement the health of the periodontium. Even though Chlorhexidine is a gold standard material for its remarkable anti-plaque, anti-gingivitis efficacy, its disadvantages

on long term usage such as bitter taste, staining, burning of oral mucosa, etc prompt researchers to search for alternative materials which are safe without side effects.

Probiotic mouthwashes, on the other hand, lack the side effects and are very safe for usage in all the age groups. The current meta-analysis evaluated anti-plaque, and anti-gingivitis efficacy of Probiotic mouthwash in comparison to Chlorhexidine. As heterogeneity was more in included studies, random-effects model was followed. Results reveal that there is no significant difference between Probiotic and Chlorhexidine mouthwashes in terms of Plaque Index and Gingival Index scores when post-intervention scores were compared to baseline.

**Limitations of this review:** Number of studies that presented data that could be used for meta-analysis were low.

**Directions for future research:** Most of the studies were carried out for a short duration (less than 30 days), also most of the studies were of low quality. Good quality study with adequate sample size, and for adequate duration comparing Probiotic, Chlorhexidine mouthwashes in the general population, subjects undergoing fixed orthodontic treatment can be an interesting topic for future research. Comparing the efficacy of both these mouthwashes in established gingivitis and periodontitis cases will also be an interesting topic for future research.

## Conclusion

Within the limits of this systematic review and meta-analysis, low-quality evidence suggests that Probiotic mouthwash has good anti-plaque and anti-gingivitis actions and its efficacy is comparable to Chlorhexidine mouthwashes on short term usage only (less than 30 days), more high-quality studies with sufficient sample size and longer duration are needed to validate the same.

**Source of support:** nil

**Conflicts of interest:** none

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