

Microbial flora surrounding orthodontic temporary skeletal anchorage devices: A systematic review

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ABSTRACT

Aim: The oral cavity harbours distinct microorganisms, which create a unique microenvironment. These microorganisms might trigger inflammatory reactions in the host, potentially leading to inflammation that can question the stability of temporary skeletal anchorage devices (TSADs). This study aimed to systematically review the literature on the type of microorganisms around TSADs.

Methods: A search of studies in six electronic databases – Cochrane Library, PubMed, OVID, Scopus, LILACS and Web of Science were performed until 30 May 2024 without any restriction in date or language of publication. The selection of articles was limited to studies evaluating the microorganisms around TSADs during orthodontic treatment. Two reviewers independently performed eligibility screening, study selection, and data extraction. The Newcastle Ottawa scale was used to assess the Risk of bias in all the included studies. Meta-analysis could not be performed because of the heterogeneity of the studies.

Results: From 7020 articles, seven prospective studies were included for the qualitative analysis. *Porphyromonas gingivalis* and *Treponema denticola* were found around all TSADs used in orthodontic therapy. There was a significant difference in the type of microorganisms around successful and failed TSADs.

Conclusions: There was an overall colonization of diverse microorganisms around TSADs. Failed TSADs showed greater *Porphyromonas gingivalis*, *Parvimonas micra* and facultative anaerobic enteric commensal *Enterobacter*.

1. Introduction

Orthodontic anchorage is fundamental to orthodontic treatment. It is defined as the ability to resist unwanted reactive tooth movements. It can be provided by teeth, palate, head, neck or bone implants. Temporary Skeletal Anchorage Devices (TSADs) provide persistent orthodontic forces and anchorage placed in the alveolar bone. Mini-implants can be used as TSADs; unlike traditional anchorage, it has several advantages like the need for good control of tooth movements, convenience of the operator, shortening of treatment time and patient cooperation.^{1,2}

Though TSADs have a high success rate (>80 %),³ implant-related and site-related factors contribute to the failure.⁴ The most important reason for early failure is inflammation or Peri-implantitis. They are exposed to all types of microorganisms in the oral cavity since they are placed trans-gingivally, including bacteria associated with periodontitis and peri-implantitis.⁵ The microbiota that is observed in periodontitis, such as *Enterococcus faecalis*, *Streptococcus oralis*, *Aggregatibacter*

actinomycetemcomitans, *Streptococcus sanguinis*, *Prevotella intermedia*, *Porphyromonas gingivalis*, which causes peri implantitis that leads to implant mobility and loss.^{6,7} The healing process after the insertion of TSADs could be impeded by microbiota dysbiosis. Stability is questionable if there is a bacterial invasion during the healing process, as the insertion process triggers an inflammatory response.⁸

Comprehending the qualitative and quantitative aspects of these microorganisms will help reduce inflammation, improve oral hygiene and subsequently increase the stability of mini-implants. The available literature exhibited diverse outcomes, requiring a thorough review to assess and synthesize findings systematically and establish a more conclusive understanding of the subject matter. The review question was, "Is there a difference in the types of microorganisms between successful and failed TSADs?"

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2. AIM

This systematic review aims to assess the distribution and diversity of microorganisms around TSADs.

3. Objective

The primary objective of this systematic was to find the difference in the type of microorganisms between successful and failed TSADs.

The secondary objective of this systematic was to find if the diversity in microorganisms contributed to the failure of TSADs.

4. Materials and methods

4.1. Protocol and registration

The systematic review was prepared as per the Preferred Reporting Items for Systematic Review (PRISMA) guidelines.⁹. This systematic review was registered with PROSPERO under the ID number CRD42024527818.

4.2. Eligibility criteria

According to the PICO (participants, intervention, comparison, and outcome) design schema: (P) human participants undergoing orthodontic treatment of any ethnicity, malocclusion, age or sex who had good oral and general health and not under anti-biotics or anti-inflammatory drugs three months before the intervention, (I) who require TSADs for orthodontic treatment, (C) between successful and failed TSADs (O) assessing their diversity of microorganisms around TSADs. Excluded studies were abstracts, author debates, book chapters, case reports, case series, conferences, summary articles, investigations on animals and non-clinical studies, commentaries and interviews. The prime outcome of this review was to determine the difference in the type of microorganisms between successful and failed TSADs, while the secondary outcome was to find if the diversity in microorganisms contributed to the failure of TSADs.

5. Information sources and search strategy

A systematic search was performed across six electronic databases: Cochrane Library, PubMed, OVID, Scopus, LILACS and Web of Science using variations of the following keywords: “Oral microbiome, Temporary anchorage device, Mini-implants, Orthodontics”. A grey literature search in Clinical Trials, Google Scholar and OpenGrey was done. The search approach used MeSH (Medical Subject Headings) terms and the Boolean Operators "AND" and "OR" with no publication year limitations until 30 May 2024 (Table 1). However, the search was limited to articles written in English. A manual search of the reference lists of the included articles was also done.

6. Study selection

Study selection was performed in two phases by two investigators, which included an independent initial screening of articles based on the research question and against the eligibility criteria. In the initial screening process, titles and abstracts were screened, and a full-text review was done for incomplete information provided in the abstract and title. Furthermore, to ensure any exclusion of relevant articles, a hand search was performed using the references of the included articles. The authors were contacted if there was any lack of information. The pool of articles was finally assessed for eligibility for qualitative and quantitative reviews. The two reviewers settled discrepancies through discussion. Any disagreements were resolved by a third reviewer (VK).

Table 1

Search strategy.

KEYWORDS USED	SEARCH ENGINE	NO.OF ARTICLES RETRIVED
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	PUBMED	1134
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	SCOPUS	3
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	OVID	5367
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	LILAC	51
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	WEB OF SCIENCE	314
((oral microbiome) AND (temporary anchorage device)) OR (mini implants) AND (orthodontics)	Cochrane Library	151
	TOTAL	7020

6.1. Data collection process and data items

The two reviewers (NJ and HP) extracted pertinent data independently. In the process, any concerns about a specific study were answered by contacting the lead author (VK). Each reviewer put the data into a Microsoft Word document on their own before discussing it to reach an agreement. The produced data was deduplicated using an automation technique (Zotero). 7020 papers were obtained, and after removing duplicates, 4701 were included for title reading, followed by 64 articles for abstract reading, with seven publications meeting the inclusion and exclusion criteria. 306 articles were obtained from the grey literature search, and these articles were excluded after the title and abstract had been screened (Fig. 1). After individual examination of the included studies, the main characteristics were extracted and stored in a standardized form in Microsoft Office Excel: author's name, study design, participants, age, methods of assessing, microbes assessed, results, control group, observation time and outcome. This data was then shared with the senior reviewers to streamline and finalize.

6.2. Risk of bias assessment in individual studies

Risk of Bias (RoB) assessment of individual studies was performed according to the Newcastle-Ottawa Scale for cross-sectional studies under the domains of – selection, comparability and outcome/exposure. The included studies were graded as good, fair and poor quality using the Newcastle -Ottawa Scale.¹⁰. The number of studies graded as “Fair” was seven, and the graphical representation of the ‘Risk of bias summary for each included study’ is portrayed in a graphical representation (Fig. 2). The risk of bias assessment of all included studies was carried out independently by two reviewers, and dissent was settled through discussion with the third reviewer.

7. Results

7.1. Study selection

The search of the six databases revealed 7020 records. Following the removal of 2319 duplicates, 4638 irrelevant studies were eliminated based on the title. The abstracts were read for 63 articles, and 49 articles were eliminated as they did not meet the inclusion and exclusion criteria. Seven articles had to be discarded for the following reasons: no comparison group was present in the study. Ultimately, seven studies were included in this systematic review, all prospective studies. Due to the heterogeneity and methodological diversity of the interventions and

PRISMA 2020 flow diagram for new systematic reviews which included searches of databases, registers and other sources

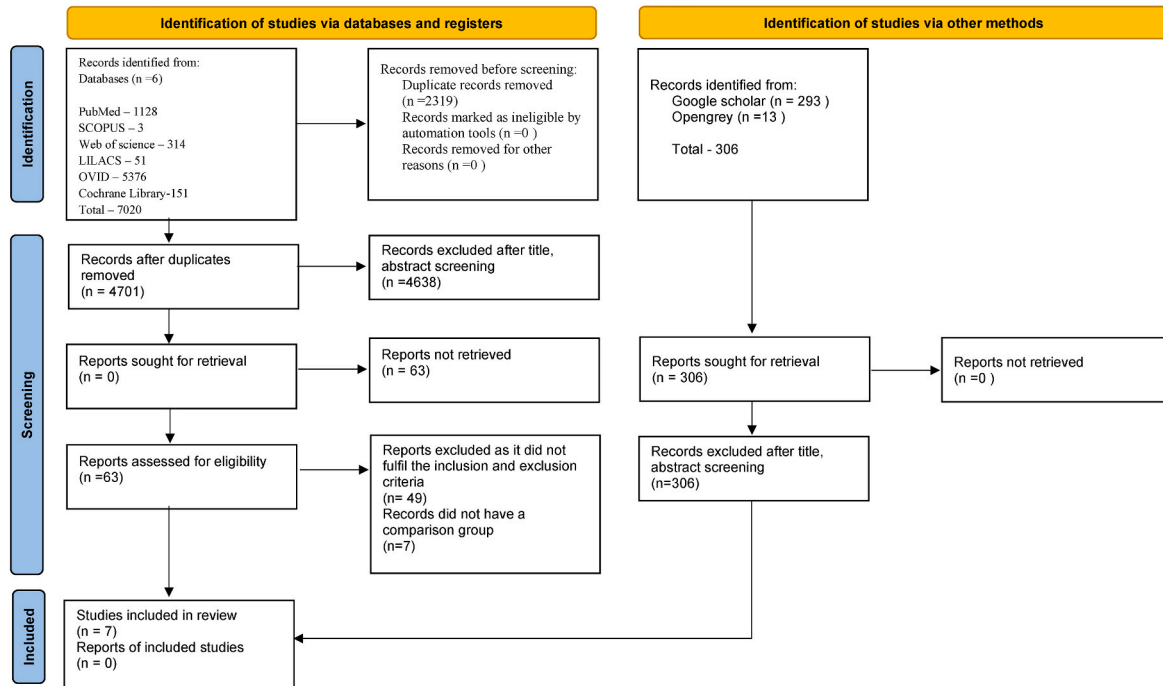


Fig. 1. Prisma flow chart.

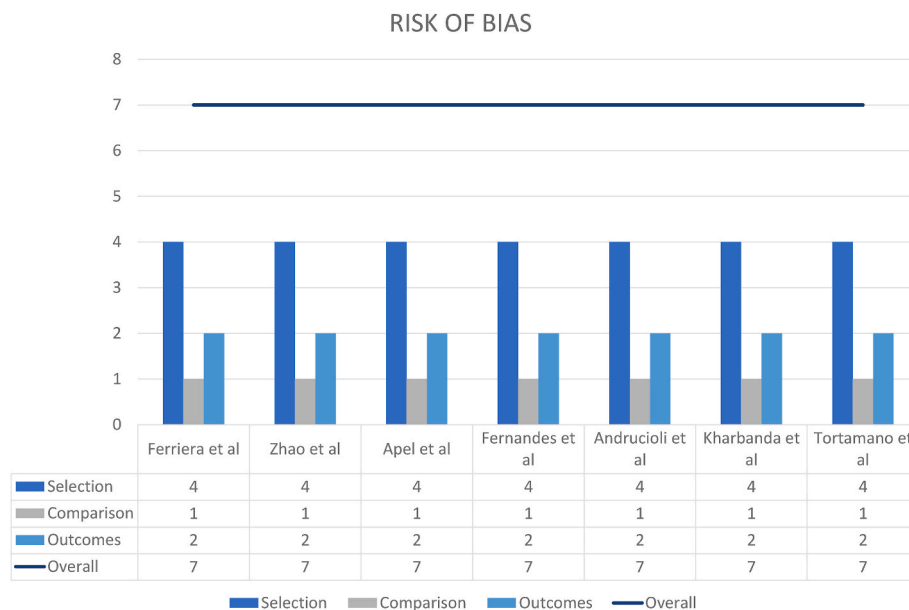


Fig. 2. RISK OF BIAS.

outcomes measured, we could not combine the results into a meta-analysis. Thus, only a narrative synthesis of the data has been presented. The search strategy results are presented in the PRISMA flow chart (Fig. 1).

7.2. Methods

The included seven studies consisted of prospective studies published between 2009 and 2023. All seven studies included both genders. Four out of seven studies used RT-PCR to quantify the presence of microbial flora around the TSADs.^{3,11–13} Along with RT-PCR, 16S rRNA gene sequencing, Metagenomic sequencing, and autofluorescence were

used to evaluate the difference in microorganisms around successful and failed TSADs.^{12,13} While one study validated the presence of microorganisms around TSADs using scanning electron microscopy,¹⁴ Another study quantified the bacterial endotoxin by DNA-DNA hybridization. It used DNA probes for forty microbial species to evaluate the microbial contamination.¹⁵ The microbial changes around TSADs were assessed using microbiologic culture and biochemical techniques in one of the studies (Table 2).¹⁶

This systematic review evaluated the difference in the type of microorganisms between successful and failed TSADs. All seven studies determined if the difference in microorganisms between successful and failed TSADs contributed to the failure of TSADs. The studies included

Table 2
– Study characteristics of each study.

S. NO	Author and year	Study design	Age	Sample size	Methods	Microbes assessed	Result	Control group	Observation time	Journal published	Funding
1.	Apel et al. 2009 (Apel et al., 2009)	Prospective study	16–19 years	12mini implants-(8 FTSAD +4 STSAD)	RT-PCR	Actinomyces viscosus - four and Campylobacter gracilis -three successful, whereas in failed implants both the species were found rarely (12.5 %)	There was no difference between both groups	Successful TSADs-4	Different from case to case as well as the time point of failure	Clinical Oral implant research	Nil
2.	Tortamano et al., 2012 (Tortamano et al., 2012)	Prospective study	16–40 years	31mini-implants (15 F + 16S)	Polymerase Chain Reaction	Prevotella intermedia(Pi), Actinobacillus actinomycetemcomitans (Aa), and Porphyromonas gingivalis (Pg)	Mobility of mini-implants is not associated with presence of Aa,Pi and Pg	TSADs without mobility and without clinical signs of inflammation - 16	Successful TSADs- 169 and 1023 days Failed TSADs-7 and 731 days	Angle orthodontist	Nil
3.	Ferriera et al., 2015 (Ferriera et al., 2015)	Prospective study	18 to 34 years	12 mini-implants(5 FTSAD and 7 STSAD)	Scanning electron microscopy	None of the group except one successful group-Rods, Filamentous and Coccoid bacteria	No relationship of microorganisms between failure and successful group	Successful TSADs- 7	Successful mini-implants was 9–24 months (mean 15.8 months, SD 7.40) Failed mini-implants was 2–3 months (mean 2.4 months, SD50.22)	Microscopy research and technique	Nil
4.	Andrucioi et al., 2018 (Andrucioi et al., 2018)	Prospective study	11–49 years	25 TSADs 10 F + 15 S	DNA-DNA hybridization technique and quantify bacterial endotoxin	Endotoxin is produced in the cell wall of Gram-negative microorganisms(also known as LPS due to its lipopolysaccharide nature)	Neither microbial contamination nor endotoxin quantification was determinant for the instability of the mini-implants.	Successful TSADs- 15	Mean time- 26.1 months for successful TSADs and 6.7 months for failed TSADs	Journal of oral applied science	São Paulo Research Foundation (FAPESP – Process number 2011/23822-0) and a research postgraduate scholarship granted by the Coordination of Higher Education and Graduate Training (CAPES)
5.	Garcez et al., 2020 (Garcez et al., 2020)	Prospective study	25–38 years	60 mini screws (6 FTSADs+54 STSADs)	Quantitative Light Fluorescence q-PCR	Porphyromonas gingivalis.	Higher number of P. gingivalis contamination around inflamed miniscrews	Successful TSADs-54	After the end of the treatment or due to replacement caused by inflammation	Photodiagnosis and Photodynamic Therapy	Nil
6.	Zhao et al., 2023 (Zhao et al., 2023)	Prospective study	12–45 year old	29 TSADs(15 FTSAD+14 STSAD) 135 TSADs (62F + 73S) 34TSAD(18F + 16S)	16S rRNA gene sequencing Metagenomic sequencing RT-PCR	Eikenella corrodens, Prevotella intermedia, Neisseria elongata, Parvimonas spp and Catonella morbi	There was difference in the microorganisms between both the group	Successful TSADs-14 Successful TSADs-73 Successful TSADs-16	Not given	BMC oral health	National Program for Multidisciplinary Cooperative Treatment on Major Diseases
7.	Kharbanda et al., 2023 (Kharbanda et al., 2023)	Prospective study	12–27 years	102 mini-implants (28 FTSADs +74STSADs)	Microbial culture and biochemical techniques	Streptococcus spp, anaerobic gram-negative rods and Staphylococci, Parvimonas micra and Veillonella	Failed MSIs were characterized by a higher proportion of Staphylococci, facultative anaerobic enteric commensal Enterobacter, and obligate anaerobe Parvimonas micra.	Successful TSADs-74	Not given	American Journal of orthodontics and Dentofacial orthopedics	Nil

(STSAD- Successful Temporary skeletal anchorage device, FTSAD- Failed Temporary skeletal anchorage device, rRNA- Ribosomal ribonucleic acid, RT-PCR- Reverse transcription polymerase chain reaction, DNA- Deoxyribonucleic acid,MSI-Miniscrew implants, qPCR- Quantitative polymerase chain reaction).

patients of both genders with ages ranging from 19 to 43 years. The study group consisted of patients who required TSADs for orthodontic treatment. The dimensions of mini-implants used in all six studies were 1.6 mm in diameter and 8 mm length, and in the study by Garcez et al. the dimensions used were not mentioned¹³. The TSADs were used to retract the anterior segment after extraction in three studies.^{3,11,16}. Among the studies included, one study used TSADs to perform distalization of the maxillary posterior teeth, en-masse retraction of the six anterior teeth in both arches, intrusion of the maxillary incisors and molars, mesialization of the maxillary and mandibular second molars¹⁴ and three studies did not mention the purpose of using TSADs.^{12,13,15}. The interpretations were made using data from 484 TSADs classified according to the stability of the TSADs (318 successful and 166 failed), originating from seven prospective studies.^{3,11–16} intending to achieve an adequate DNA quantity for metagenomic sequencing in the Zhao et al. study, they had six samples in the successful and six samples in the failed group by combining 10–12 TSADs in one sample.¹² Successful TSADs were defined as TSADs that maintained stability until the objective was accomplished and no signs of inflammation were noted. Failed TSADs, conversely, were unable to serve as anchorage devices because of mobility and signs of inflammation were observed.

7.3. Risk of bias within studies

Risk of Bias (RoB) assessment of individual studies was carried out using the Newcastle–Ottawa scale (NOS)¹⁰ for all the seven studies under the domains of – selection of samples, comparability and exposure/outcome. In NOS scoring, a maximum of four points for selection, two points for comparability and three points for the outcome were assigned. Studies that reached a score of seven or more were considered low RoB, five to six as moderate RoB and up to four as high RoB. The Risk of bias for all seven studies was graded as "Fair (Table 3).^{3,11–16}. The diagrammatic representation of the percentage risk of bias about different domains/parameters such as 'selection bias, performance bias, detection bias, attrition bias, reporting bias, other biases' assessed across all included studies are presented in traffic-light plot (Fig. 3).

Two out of the seven studies were aided by an external organization. The study by Zhao et al. was supported by the National Program for Multidisciplinary Cooperative Treatment on Major Diseases¹². The study by Andruccioli et al. was supported by a grant-in-aid from the São Paulo Research Foundation and a research postgraduate scholarship granted by the Coordination of Higher Education and Graduate Training (CAPES).¹⁵.

7.3.1. A qualitative synthesis of the result

Garcez et al. showed a significant difference between 12 successful and 28 failed mini-screws based on the fluorescence intensity, area of fluorescence and the number of bacteria ($p < 0.05$). Successful mini-

screws presented lower CFU count and less fluorescence intensity when compared to failed mini-screws and in fluorescent biofilm, identified the presence of *Porphyromonas gingivalis*, which correlated fluorescence intensity with the number of bacteria, comparing successful orthodontic mini-screw with failed ones.¹³

Zhao et al. found that between successful and failed TSADs, there was no difference in the bacterial load ($p = 0.251$), but in the case of *Prevotella intermedia*, the sensitivity was 37.5 % in the successful group and 50 % in the failed group ($p = 0.510$). In the failed group, there was a higher quantification of *Prevotella intermedia* ($p = 0.0048$). β -diversity based on Manhattan distances and Bray Curtis showed different group clusters ($p = 0.015$, $p = 0.044$). Amplicon sequencing analyzed the microbial composition at the phyla and genera levels. Proteobacteria, Fusobacteria, Bacteroidetes and Firmicutes constitute the major part of the microorganisms on the TSADs at the phyla level. At the genus level, *Fusobacterium*, *Prevotella*, *Streptococcus*, *Veillonella*, *Leptotrichia*, and *Selenomonas* were dominant. In the failed group of TSADs, taxa that are associated with periodontal diseases, such as *Filifactor alocis*, *Fusobacterium nucleatum*, *Prevotella nigrescens* and *Porphyromonas gingivalis*, observed a strong correlation.¹²

According to Apel et al., an average cell number of 1.6×10^7 was observed in the quantitative bacterial analysis of eight failed mini-implants. In contrast, four successful mini-implants had an average cell number of 2×10^7 . The disparity was higher within the failed group than between both successful and failed groups. The peri-implantitis-associated or classic periodontopathogenic species of oral origins, such as *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis*, were not or seldom found (*Tannerella forsythia*) in failed group and was absent in successful group.³.

It was concluded by Tortamano et al. that the presence of *Prevotella intermedia* (Pi), *Actinobacillus actinomycetemcomitans* (Aa), and *Porphyromonas gingivalis* (Pg) around the mini-implants did not contribute to the mobility of the TSADs. The presence of Aa was around 13.33 % in 15 failed TSADs group and 31.25 % in 16 successful TSADs group, whereas Pg was 33.33 % in the failed group and 37.4 % in the successful group.¹¹

Bacterial contamination and medians of endotoxin were determined for the stability of TSADs by Andruccioli et al. In both the 15 successful and 10 failed TSADs groups, the presence of all 40 microbial species of the Actinomycetes group, yellow, purple, green, red, orange complexes and other species (100 %) were significant. A median value of 65,750 EU/mL of endotoxin was revealed in the healthy mini-implants and 43,500 EU/mL in the inflamed mini-implants. There were no significant differences between the two groups ($p = 0.63613$).¹⁵

In a study by Ferriera et al., SEM analysis revealed substantial biofilm formation on the transmucosal profile and head of all mini-implants of both groups with a large number of microorganisms. In the successful group, only one mini-implant exhibited rods, filamentous and coccoid

Table 3
Risk of bias of included studies.

Authors	Selection	Comparability	Outcome	Total	Interpretation
Ferriera et al., 2015(Ferriera et al., 2015)	4	1	2	7	Fair
Zhao et al., 2023 (Zhao et al., 2023)	4	1	2	7	Fair
Apel et al., 2009 (Apel et al., 2009)	4	1	2	7	Fair
Garcez et al., 2020 (Garcez et al., 2020)	4	1	2	7	Fair
Andruccioli et al. 2018 (Andruccioli et al., 2018)	4	1	2	7	Fair
Tortamano et al. 2012 (Tortamano et al., 2012)	4	1	2	7	Fair
Kharbanda et al., 2023 (Kharbanda et al., 2023)	4	1	2	7	Fair

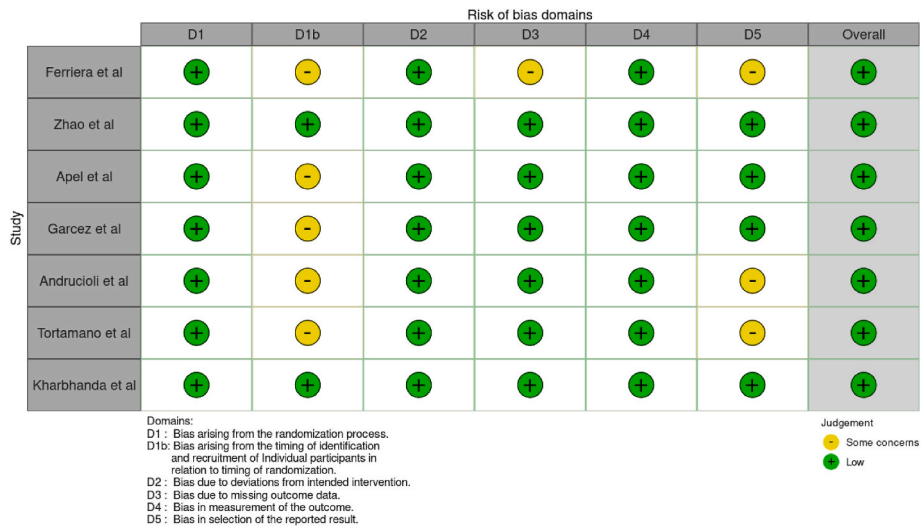


Fig. 3. Risk of bias for each included study.

Table 4
Microorganisms around successful and failed TSADs.

Study	Methods	Microbes assessed Successful TSADs	Microbes assessed Failed TSADs
Apel et al. (Apel et al., 2009)	RT-PCR	1.9 x10 ⁷ to 4x 10 ⁷ (standard deviation: 1.3x 10 ⁷) Treponema denticola (75 %) Actinomyces viscosus (100 %) Campylobacter gracilis (75 %)	3.4 x10 ⁶ to 2x 10 ⁸ (standard deviation: 1.7x 10 ⁷) Treponema denticola (50 %) Actinomyces viscosus (12.5 %) Campylobacter gracilis (12.5 %)
Tortamano et al. (Tortamano et al., 2012)	PCR	Porphyromonas gingivalis was detected in six of 16 (37.4 %) Aggregatibacter actinomycetemcomitans five of the 16 samples (31.25 %)	Porphyromonas gingivalis was detected in four of 15 samples(33 %) Aggregatibacter actinomycetemcomitans was present in two of the 15 experimental samples (13.33 %)
Ferriera et al. (Ferriera et al., 2015)	Scanning electron microscopy	One TSADs showed presence of rods, filamentous and coccoid bacteria	No microbes assessed in the body of the implant
Andruccioli et al. (Andruccioli et al., 2018)	DNA-DNA hybridization	All 40 species of Actinomyces group were found with an increase in Parvimonas micra, Treponema denticola and Eubacterium saburreum	All 40 species of Actinomyces group were found
Garcez et al. (Garcez et al., 2020)	Median no.of microorganisims	12,950,000	8,490,000
	Bacterial endotoxin	65,750 EU/mL	43,500 EU/mL
Zhao et al. (Zhao et al., 2023)	Autofluorescence	Less fluorescence intensity and lower CFU at miniscrew threads	More fluorescence intensity and more CFU at mini screw threads
	RT-PCR	Lesser number of Porphyromonas gingivalis	Higher number of Porphyromonas gingivalis
Kharbhandha et al. (Kharbhandha et al., 2023)	Taxonomic composition	Propionibacterium acidifaciens, Anaeroglobus geminatus, Actinomyces dentalis, Prevotella oulorum, Actinomyces massiliensis, Cardiobacterium hominis, Shewanella sediminis, Ferrimonas balearica, Olsenella Profusa, and Prevotella salivae	Prevotella intermedia, Eikenella corrodens, Parvimonas spp, Neisseria elongata, and Catonella morbi
	16S rRNA gene sequencing	α- diversity showed no significant difference β-diversity was less	α- diversity showed no significant difference β-diversity was high
	Metagenomic sequencing	Staphylococcus aureus Propionibacterium acidifaciens Bifidobacterium dentium Anaeroglobus geminatus Actinomyces dentalis Neisseria bacilliformis Prevotella pallens	Neisseria sicca Parvimonas micra Serratia spp Actinomyces cardiffensis Selenomonas sp Alloprevotella tannerae Aggregatibacter actinomycetemcomitans Eubacterium brachy Eubacterium nodatum
	RT-PCR	Veillonella parvula, Haemophilus parainfluenzae, Actinomyces odontolyticus, Actinomyces israelii, and Streptococcus gordonii Prevotella intermedia, the detection rate was 37.5 % Actinomyces odontolyticus, Streptococcus gordonii, Streptococcus mitis and Veillonella parvula,	Prevotella intermedia, the detection rate was 50 %
Kharbhandha et al. (Kharbhandha et al., 2023)	Microbiologic culture and biochemical techniques	Facultative anaerobic gram-negative rods (Klebsiella, Enterobacter, Pseudo monas, Acinetobacter, Citrobacter), Staphylococci, Parvimonas micra and Veillonella parvula	Staphylococci, facultative anaerobic enteric commensal Enterobacter, and obligate anaerobe Parvimonas micra.

(rRNA- Ribosomal ribonucleic acid, RT-PCR- Reverse transcription polymerase chain reaction, DNA- Deoxyribonucleic acid, MSI-Miniscrew implants, qPCR- Quantitative polymerase chain reaction).

bacteria on the body segment in addition to a large number of fibres and no mini-implants in the failed group exhibited bacteria on their body.¹⁴

A predominant presence of Streptococci colony was found in both Gingival crevicular fluid(GCF) and Per-mini-implant crevicular fluid (PMCIF) obtained from the region of 102 TSADs. The colonization of Staphylococci was found to be five times more in the age group below 14 years and three times more in the age group above 14 years compared to the gingival sulci, whereas in the PMCIF, a predominance of anaerobic gram-negative cocci and facultative anaerobic gram-negative bacteria was observed. The aerobes decreased from 78.1 % at T0 to 66.7 % in the age group below 14 after 12 weeks of placement of TSADs, whereas the anaerobes increased from 21.8 % to 33.3 % in the GCF samples. However, the aerobic population decreased from 95.6 % to 61.5 % in the PMCIF samples, and the anaerobic population increased from 4.3 % to 38.5 % in this age group. There was a decrease from 70.7 % to 60.8 % in the age group above 14 after 12 weeks in the aerobic population observed in the GCF samples, and in the PMCIF samples, it decreased from 79.8 % to 59.1 %. The anaerobic population increased from 20.8 % to 40.2 %. There was a significant difference in the aerobic microorganisms with a strong association of Staphylococci with failed mini-implants between the successful and failed mini-implants.¹⁶

Out of the seven studies included, four concluded that there was a difference in the microorganisms between successful and failed TSADs. There was an increased colony forming unit (CFU) and the presence of *Prevotella intermedia* in the failed group. Microorganisms, especially the anaerobic species like Staphylococci, facultative anaerobic enteric commensals and anaerobic cocci that contribute to the periodontal problems, were identified among the failed TSADs. Bacterial endotoxin was found more in the failed TSADs, indicating greater microbial colonization in this group.

8. Result

Porphyromonas gingivalis was found in approximately 80 % of failed TSADs in RT-PCR and 37.4 % of failed TSADs, with a good correlation between the number of bacteria and fluorescence intensity.^{11,13,15} *Parvimonas micra* was observed in both failed TSADs^{12,16} and successful TSADs. *Treponema denticola* was found to be 75 % in successful TSADs compared with failed TSADs (50 %).¹⁵ Another study,¹⁵ concluded that there was no exhibition of bacteria on the body of TSADs and only one in the successful group exhibited rods, filamentous and coccoid bacteria on this region in addition to a large number of fibres.¹⁴ (Table 4).

9. Discussion

The use of absolute anchorage has revolutionized orthodontic practice by offering precise control and predictability in tooth movement, ultimately leading to more efficient and effective treatment outcomes. They are positioned directly on the gingival tissue, making them vulnerable to various oral microorganisms. The failure rate of mini-implants varies from 6.6 to 16.15 %, which is more when compared to dental implants (3 %) and other anchorage devices like mini plates (2.6–7.3 %).¹⁷

The included studies consisted of seven prospective studies published between 2009 and 2023. The quality assessment of these studies was assessed using the Newcastle–Ottawa Scale (NOS).¹⁰ All seven studies were graded as "Fair", with five studies scoring seven and two studies scoring six.

The definition of successful or failed TSADs is clear to the clinician. However, the parameters that determine the success or failure of TSADs may include peri-implantitis, tissue inflammation or mobility of TSADs. This systematic review assessed all these factors under the umbrella of successful/failed TSADs, and the etiology of the failure of TSADs is not within the scope of this systematic review. The objective of this systematic review was limited to identifying the diversity in microorganisms between successful and failed TSADs. Microorganisms could be the

critical factor in the success/failure of TSADs, as microbial colonization occurs within 24 h of placement in the oral cavity.¹⁸

All seven studies showed an overall colonization of microorganisms. Four of these studies showed no specific aggressive microorganisms around failed TSADs to prove an association between the failure of TSADs and microorganisms.^{3,11,14,15} Three studies showed that different microorganisms like *Porphyromonas gingivalis*, Staphylococci, *Parvimonas micra* and facultative anaerobic enteric commensal *Enterobacter* were observed around the failed TSADs.^{12,13,16}

The difference in flora could be one of the reasons for the failure of TSADs. However, it is clinically important to observe that many of these are common commensals in the oral cavity. A possible explanation could be that with changes in the oral environment, lowered immune response, type of nutrient and atmospheric gradient, these commensals are likely to be pathogenic.¹⁹

Pathogens could contribute to the failure of TSADs. Therefore, it is critical to comprehend the microbiological factors underlying TSAD failure and develop methods to reduce bacterial adhesion.¹² The healthy periodontium has species that maintain internal harmony, and the lack of these species in the failed TSADs can also indicate one reason for the failure of TSADs. In failed TSADs, there was a wide variability in the prevalence of microorganisms. The authors also reported a weak correlation between these microorganisms and failure. Thus, the inference could be that various pathogens and their concentration might each contribute to the failure of TSADs.

Failed TSADs had a greater prevalence of bacteria, which had genes involved in flagellar assembly, bacterial chemotaxis, and oxidative phosphorylation. This is because these genes are important for bacterial colonization and infection.¹² *Prevotella intermedia* in lower concentrations/minimal numbers could produce marked effects leading to failure.²⁰ *Porphyromonas gingivalis* can function as either a harmless commensal or a harmful pathogen, and its presence alone does not reliably predict any diseases. They also found that *Tannerella forsythia*, *Aggregatibacter actinomycetemcomitans* and *Porphyromonas gingivalis* are prevalent in the buccal epithelial cells of healthy mouths. This presence creates a reservoir for these periodontal pathogens, which can facilitate their recolonization in the subgingival area of patients with periodontitis.²¹ *Parvimonas micra* is a gram-positive anaerobic coccus that is typically found as a commensal organism in the oral cavity and is associated with many dentoalveolar infections, periodontitis and endodontic infections. Its presence around failed TSADs was observed in two studies,^{12,16} and in one study, it was observed around successful TSADs.¹⁵ Characteristics of periodontitis and periimplantitis, gingival inflammation, vascular disruption and bleeding provide an iron/heme-rich environment for bacterial growth like *Porphyromonas gingivalis* and for the development of biofilm.

Meta-analysis could not be performed as there were diverse microorganisms around TSADs, and different methods were used to assess these microorganisms.

10. Limitations and future directions

In each study, different methods were used to assess the microorganisms around TSADs. Standardized methods to assess these microorganisms can aid in precise evaluation. Microorganisms around bone screws might differ as the type of material, amount of force applied, and site of insertion are not similar to mini-implants.

Clinicians should have knowledge of the healthy commensals that adhere to the skeletal anchorage, and this can help in the effective management of microbial flora to improve patient outcomes. Future research can implicate identifying more high-quality, randomized, controlled, and multi-centric trials with larger samples and gender specificity, along with long-term follow-up data to substantiate the results. It might also enable personalized orthodontic treatments based on an individual's microbial profile.

11. Conclusions

The seven studies included in this systematic review were graded as "Fair". Most of the studies had convenient samples and a comparable control group that reported complete statistical data with relevant analysis. The review elucidated the overall colonization of microorganisms, predominantly *Porphyromonas gingivalis*, *Parvimonas micra* and facultative anaerobic enteric commensal *Enterobacter* around the TSADs. There was a difference in the amount and type of microorganisms between the successful and failed TSADs. Ensuring oral hygiene or suitable medications for these microorganisms can help the orthodontist avoid this colonization to prevent failure of TSADs.

Patient consent

Patient consent not applicable. No patients were included in the study since it is a systematic review.

Ethical clearance

This paper did not use experimental data from human subjects.

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Declaration of competing interest

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References

- Reynders R, Ronchi L, Bipat S. Mini-implants in orthodontics: a systematic review of the literature. *Am J Orthod Dentofacial Orthop.* 2009;135(5):564.e1565. official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics <https://pubmed.ncbi.nlm.nih.gov/19409331/>.
- Chen YJ, Chang HH, Lin HY, Lai EH, Hung HC, Yao CC. Stability of miniplates and miniscrews used for orthodontic anchorage: experience with 492 temporary anchorage devices. *Clin Oral Implants Res.* 2008;19(11):1188–1196. <https://pubmed.ncbi.nlm.nih.gov/18983323/>.
- Apel S, Apel C, Morea C, Tortamano A, Dominguez GC, Conrads G. Microflora associated with successful and failed orthodontic mini-implants. *Clin Oral Implants Res.* 2009;20(11):1186–1190. <https://pubmed.ncbi.nlm.nih.gov/19719743/>.
- Hong SB, Kusnoto B, Kim EJ, BeGole EA, Hwang HS, Lim HJ. Prognostic factors associated with the success rates of posterior orthodontic miniscrew implants: a subgroup meta-analysis. *Kor J Orthod.* 2016;46(2):111–126. <https://pubmed.ncbi.nlm.nih.gov/27019826/>.
- Kirst ME, Li EC, Alfant B, et al. Dysbiosis and alterations in predicted functions of the subgingival microbiome in chronic periodontitis. *Appl Environ Microbiol.* 2015;81(2):783–793. <https://pubmed.ncbi.nlm.nih.gov/25398868/>.
- Genco RJ, LaMonte MJ, McSkimming DI, et al. The subgingival microbiome relationship to periodontal disease in older women. *J Dent Res.* 2019;98(9):975–984. <https://pubmed.ncbi.nlm.nih.gov/31329044/>.
- Lakhssassi N, Elhajoui N, Lodter JP, Pineill JL, Sixou M. Antimicrobial susceptibility variation of 50 anaerobic periopathogens in aggressive periodontitis: an interindividual variability study. *Oral Microbiol Immunol.* 2005;20(4):244–252. <https://pubmed.ncbi.nlm.nih.gov/15943770/>.
- He W, Zhu H, Liu C. Profiles of inflammation factors and inflammatory pathways around the peri-miniscrew implant. *Histol Histopathol.* 2021;36(9):899–906. <https://pubmed.ncbi.nlm.nih.gov/33834451/>.
- Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *BMJ (Clinical research ed.).* 2021;372:n71. <https://pubmed.ncbi.nlm.nih.gov/33782057/>.
- Wells GA, Shea B, O'Connell D, et al. The Newcastle–Ottawa Scale (NOS) for assessing the quality of nonrandomized studies in meta-analysis. *Appl Eng Agric.* 2014;18:727–734.
- Tortamano A, Dominguez GC, Haddad AC, Nunes FD, Nacao M, Morea C. Periodontopathogens around the surface of mini-implants removed from orthodontic patients. *Angle Orthod.* 2012;82(4):591–595. <https://pubmed.ncbi.nlm.nih.gov/22839769/>.
- Zhao N, Zhang Q, Guo Y, et al. Oral microbiome contributes to the failure of orthodontic temporary anchorage devices (TADs). *BMC Oral Health.* 2023;23(1):22. <https://doi.org/10.1186/s12903-023-02715-7>.
- Garcez AS, Barros LC, Fernandes MRU, Fujii DN, Suzuki SS, Nepomuceno R. Fluorescence image and microbiological analysis of biofilm retained around healthy and inflamed orthodontic miniscrews. *Photodiagnosis Photodyn Ther.* 2020;30, 101707. <https://pubmed.ncbi.nlm.nih.gov/32126307/>.
- Ferreira NO, Andruccioli MC, Nelson-Filho P, et al. Bacterial biofilm on successful and failed orthodontic mini-implants—a scanning electron microscopy study. *Microsc Res Tech.* 2015;78(12):1112–1116. <https://pubmed.ncbi.nlm.nih.gov/26515408/>.
- Andruccioli MCD, Matsumoto MAN, Saraiva MCP, et al. Successful and failed mini-implants: microbiological evaluation and quantification of bacterial endotoxin. *J Appl Oral Sci : Rev FOB.* 2018;26, e20170631. <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6025887/>.
- Mishra G, Kharbanda OP, Chaudhry R, Duggal R. A pattern of microbiological colonization of orthodontic miniscrew implants. *Am J Orthod Dentofacial Orthop.* 2023;164(4):554–566. official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics <https://pubmed.ncbi.nlm.nih.gov/37204351/>.
- Miyawaki S, Koyama I, Inoue M, Mishima K, Sugahara T, Takano-Yamamoto T. Factors associated with the stability of titanium screws placed in the posterior region for orthodontic anchorage. *Am J Orthod Dentofacial Orthop.* 2003;124(4):373–378. [https://doi.org/10.1016/s0889-5406\(03\)00565-1](https://doi.org/10.1016/s0889-5406(03)00565-1). official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics.
- de Freitas AO, Alviano CS, Alviano DS, Siqueira Jr JF, Nojima LI, Nojima MdaC. Microbial colonization in orthodontic mini-implants. *Braz Dent J.* 2012;23(4):422–427. <https://doi.org/10.1590/s0103-64402012000400019>.
- Sbordone L, Bortolaia C. Oral microbial biofilms and plaque-related diseases: microbial communities and their role in the shift from oral health to disease. *Clin Oral Invest.* 2003;7(4):181–188. <https://doi.org/10.1007/s00784-003-0236-1>.
- Abusleme L, Dupuy AK, Dutzan N, et al. The subgingival microbiome in health and periodontitis and its relationship with community biomass and inflammation. *ISME J.* 2013;7(5):1016–1025. <https://pubmed.ncbi.nlm.nih.gov/23303375/>.
- Rudney JD, Chen R, Zhang G. Streptococci dominate the diverse flora within buccal cells. *J Dent Res.* 2005;84(12):1165–1171. <https://doi.org/10.1177/154405910508401214>.