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Effect of Chlorhexidine Mouthwash on Gingival Health around Orthodontic Miniscrew Implants: A Pilot Placebo-Controlled Randomized Trial

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The data that support the findings of this study are available from the corresponding author upon reasonable request.

Effect of Chlorhexidine Mouthwash on Gingival Health around Orthodontic Miniscrew Implants: A Pilot Placebo-Controlled Randomized Trial

Abstract

Objective: This randomized controlled trial (RCT) aims to investigate the short-term effects of chlorhexidine mouthwash (MW) on gingival health surrounding orthodontic miniscrew implants (OMIs) and their overall survivability.

Materials and Methods: Thirty-two participants (mean age, 22.8 years) undergoing fixed orthodontic appliance treatment after maxillary premolar extraction were randomly allocated in a parallel fashion to either receive (1) MW with an active component of chlorhexidine, or (2) a placebo. Each participant received two maxillary buccal OMIs for anchorage reinforcement purposes. Participants were assessed for their gingival oral health status around all inserted OMIs and had their OMI survivability recorded at three time points; T1=1 month, T2=3 months, and T3=6 months after OMI placement. A Kaplan Meier plot was used to estimate the survival function of OMIs.

Results: All randomized participants completed the follow-up period. In terms of gingival oral health, there were no statistically significant differences at any time point

between the chlorhexidine MW group and the placebo-controlled group (P > 0.05). One OMI was lost in the chlorhexidine MW group and another two OMIs in the control group. There was no significant difference between both groups in terms of survivability (P = 0.585).

Conclusion: The use of chlorhexidine MW does not seem to have a significant clinical impact on the gingival health around OMIs or their survivability in this pilot study.

Keywords:

Temporary Anchorage Devices, Orthodontic Miniscrew, Mouth wash, Chlorhexidine, Oral health, Periodontal health

Introduction

More patients are seeking orthodontic treatment due to the rise in interest for oral health and functional benefits, as well as psychosocial well-being, all contributing to an improved quality of life [1]. Thus, considerable account is given to address these demands during the early stages when anchorage planning arises as a key domain in orthodontic treatment provision. In recent years, the introduction of orthodontic miniscrew implants (OMIs) allowed effective utilization of extraction spaces and management of complex orthodontic tooth movement. OMIs are flexible in terms of their placement locations in the oral cavity, their relatively small size, as well as their ease of placement [2]. However, OMIs do have some limitations ranging from tissue irritation, infection, injury to adjacent structure, mobility, and OMI fracture [3].

In cases where OMIs are used, optimum oral health maintenance and meticulous oral hygiene measures are required, which may be challenging for the patients [4]. Retention of food remnants and subsequent bacterial plaque could promote gingival inflammation in the early phases of orthodontic treatment and onwards [5]. In turn, bacterial proliferation may increase implant failure rates, as the presence of small, stagnated proteins and carbohydrate molecules in anaerobic atmosphere would act as an enriched environment for peri-implantitis and subsequent implant failure [6].

In addition, other underlining factors increase the susceptibility of individuals to periodontal disease either directly or indirectly, such as: hormonal changes, some diseases (cancer, HIV and diabetes), certain medications (anticonvulsant and anti-angina) and smoking [7,8]. Symptoms of periodontal disease vary from one patient to another, from halitosis, bleeding, swelling and gingival recession. In turn, patient education regarding these symptoms is important to advocate extra precautions in oral hygiene measures.

In the past decades, chlorhexidine was used frequently as an active component to control plaque and gingivitis [9]. Previous research illustrated that chlorhexidine mouthwash (MW) was able to provide antiseptic and antibacterial effects through its sustained release [10,11]. In addition, it decreases soft tissue proliferation and epithelialization which is commonly believed to decrease infection if used preoperatively [12]. However, side effects such as staining of teeth, taste alteration, and irritation of mucosa are also often reported [13]. Clinicians in favor of OMIs tend to implement different measures. In literature, several clinical reports advocated the preoperative use of chlorhexidine MW to control gingival health around OMIs which would further improve their survivability as anchorage reinforcement devices [14-17]. In theory, proponents of such practices often rely on the aforementioned points to promote the preoperative prescription of chlorhexidine MW in patients requiring OMIs, though there appears to be a significant shortfall of carefully conducted prospective trials to confirm these benefits. Thus, understanding the effects of chlorhexidine MW on the short-term prognosis of OMIs through a randomized design would be of great importance to clinicians utilizing them.

The primary aim of this study was to evaluate whether using 0.2% chlorhexidine MW prescribed during a one-week period to orthodontic patients requiring OMI placement would affect the short-term gingival health status around OMIs in comparison to using a placebo. In addition, this report would also assess whether prescribing 0.2% chlorhexidine MW would influence the overall OMI survivability recorded in the short-term.

Methods and materials

Trial design

This is a double-arm parallel-group prospective randomized placebo-controlled trial (RCT) performed in a single-center at the first affiliated hospital of Zhengzhou University from September 2, 2019, and March 19, 2021. Prior to study commencement, ethical approval was obtained from the ethical committee at Zhengzhou University (ss-2019-009). Reporting and presenting data from this trial was done following the Consolidated Standards of Reporting Trials (CONSORT guidelines) [18]. All participants were informed about the study objectives and signed the respective consent forms.

Participants, Eligibility Criteria, and Sample

Forty orthodontic patients with upper and lower fixed appliances who were scheduled for OMI placement between maxillary second premolars and first molars for anchorage purposes were assessed for trial eligibility at the Zhengzhou university hospital. Eligibility criteria included: 1) planned bilateral extraction of maxillary first premolars, 2) maximum anchorage demand in the upper arch, 3) adolescents and young adults up to 35 years old, 4) non-smokers, 5) good oral hygiene, 6) non-syndromic / no systemic diseases, 7) did not receive OMIs before, and 8) not currently on medications.

Random sequence generation, concealment, and masking

A total of thirty-two patients each receiving two OMIs (i.e., a total of sixty-four OMIs) were randomly assigned to either (1) MW with active component of chlorhexidine (Corsodyl®), or (2) placebo-controlled group without the active ingredient. A block randomization with the size of 4 ensured an equal allocation ratio of 1:1. Random sequence generation was performed by independent personnel from the faculty's pharmaceutical laboratory in Zhengzhou University using a random table number. The chlorhexidine MW and the placebo were both prepared and dispensed in identical containers with sequentially numbered container codes.

These codes were only broken after the termination of the study and following data analysis; thus, randomization was concealed and both clinicians and study participants were unaware of the allocation. The placebo and the chlorhexidine MW containers were distributed in an indistinguishable manner. The placebo was prepared to match the MW in color, odor, taste, and quantity except for the active ingredient. Thus, all study participants and outcome assessors were masked to the original group assignment.

Study Procedures

All patients were treated with upper and lower 0.022" x0.028" slot Roth prescription fixed appliances [Oramco®]; 0.2% chlorhexidine MW or placebo were given to patients an hour before insertion of OMIs, and on the following consecutive six days. Patients were instructed to rinse for 30 seconds twice daily and to maintain good oral hygiene by brushing twice-a-day using 1450 ppm fluoride toothpaste. In addition to baseline assessments, data were recorded in three distinct time points, (T1) one month following OMI placement, (T2) after 3 months, (T3) after 6 months from the start. If patients were unable to come on time, another appointment was scheduled within the same week.

After reaching the main archwire (0.019" x 0.025" stainless steel [Oramco®]) and at the start of space closure stage, a single experienced operator inserted self-tapping OMIs bilaterally with diameter 1.6 mm and length of 8 mm (MAS, Titanium Biological Products), between the maxillary second premolars and first molars buccally in the keratinized mucosa under local anesthesia. Late loading on OMIs with orthodontic forces ranging between 150-200 grams, using active lace back (tie back wire) as a force delivery system was performed one month later. Active lace back was run from the OMIs directly to crimpable hooks placed distal to the lateral incisors **(Figure 1).** Participants were initially assessed for gingival health around the OMIs on the day of OMI loading.

The primary outcome was to assess the degree of gingival inflammation around OMIs using a modification of the original gingival index [19] **(Table 1)**. Clinical evaluation of gingival health was performed every visit accompanied with full photographical

records. The main investigator performed the follow up assessments for all patients, and a second investigator also recorded the observations around which half of the sample was randomly assessed for inter-observer reliability and measurement error. The secondary outcome of interest was to assess the failure rate of OMIs for six months from placement day. Failure was judged if OMIs did not serve their function including being lost due to excessive mobility, overgrowth of gingival tissues requiring premature removal of the OMIs, or other reasons [15,20]. Patients were instructed to contact the department directly if there was an emergency, or the OMI fell or became loose. During the trial, COVID-19 pandemic occurred and the government in Zhengzhou city issued a 2-month enforced quarantine. Thus, patients were instructed to directly contact the operator in case they had an emergency or any questions. Follow up of patients continued as usual after the quarantine was lifted.

Statistical analysis

All statistical tests were performed using SPSS software 22.0 (Statistical Package for Social Science, Armonk, NY: IBM Corp) at significant levels 0.05 (p-value $\leqslant 0.5$). A normality test (Kolmogorov-Smirnov) was done to check the normal distribution of the sample. Descriptive statistics were calculated in the form of Mean ± Standard deviation (SD) for parametric data and the student t-test was used to compare both groups. Gingival oral health parameters in both study groups were presented as frequency (n,%), and a nonparametric Chi-square and Mann-Whitney U test were used to compare both groups.

Post hoc calculation was performed using G*power 3.1.9.6 for mac OS. The primary outcome denoting the degree of gingival inflammation using a modification of the gingival index at different time points (T0, T1, T2, and T3) was included. A total sample size of 30 participants was sufficient to detect an effect size of 0.25 with the corresponding statistical analyses for nonparametric data, at a power of 0.9 (i.e., 90%) and partial eta square (0.06).

To calculate the inter-observer reliability between both investigators and measurement error, kappa test was used. A Kaplan Meier plot was used to demonstrate the survival function of OMIs.

Results

Thirty-two participants (mean age 22.8; range 16 to 35 years) were randomly assigned to either the chlorhexidine MW (16 patients, 32 OMIs) or the placebo-controlled group (16 patients, 32 OMIs), with a total of sixty-four assigned OMIs. All participants completed the six-month follow up period. All participants were from the Han Chinese ethnicity. Females represented almost two thirds of the sample (65.6%) in comparison to males (34.4%). The participant flow chart and trial stages are presented in **Figure 2**. Baseline characteristics and demographic data were similar across both groups **(Table 2)**.

Gingival health around OMIs

In terms of gingival oral health, there was no statistical significance between both groups at any given time point (**Table 3**). The frequencies (n, %) of occurrences of different grades in both right and left sides between 0-6 months are also presented (**Table 3**).

Table 4 shows the gingival oral health parameters in both study groups presented as median with its interquartile range (IQR) without any significant detected differences between both groups (Mann-Whitney U).

Repeated measure ANOVA (**Supplementary material**) and MANOVA were applied to ranked data over the whole sample to check the overall differences in gingival oral health parameters. Significant values were noted (corrected model: $F = 2.51$; p-<0.001, time: $F = 10.26$, p-value <0.001).

Regression trendline revealed an increase in gingival scores with time. However, these changes were non-significant between both groups with a determination coefficient (0.09) and (0.1), in the chlorhexidine and placebo groups; respectively.

Survivability of OMIs

No OMIs were lost on the right side in any group. On the left side, one OMI failed after one month in the chlorhexidine MW group. In the control group, two OMIs failed after 3

months. The survival function of OMIs which failed on the left side are plotted on the Kaplan Meier graph, where no statistically significant differences are found between both groups (Log Rank = 0.299, P = 0.585) **(Supplementary material)**.

Measurement error

Kappa test was used to assess the interrater reliability of the clinical assessment of gingival oral health on a random sample of 16 participants. Results showed substantial agreement (0.83) between both investigators.

Discussion

This pilot study aimed to evaluate the effect of chlorhexidine MW on gingival health status around OMIs and overall implant survivability. The findings of the current study show that utilization of chlorhexidine MW in orthodontic patients with OMIs had no significant clinical impact either on the gingival oral health surrounding OMIs or their survivability in the short-term.

In recent years, OMIs have been advocated as a simple and efficient anchorage device that has revolutionized the entire scope of orthodontics. The steady and gradual modifications in their designs and techniques have widened their scope of clinical applications [21]. To our knowledge, this is the first RCT that investigates the clinical influence of chlorhexidine MW to improve the clinical outcomes in cases of orthodontic anchorage implantation. Previous research highlighted inflammation around implants to be one of the main factors influencing successful outcomes with OMIs [22]. Hence, the idea to evaluate the potential effects of using an antiseptic to reduce the risk of inflammation and subsequent survivability of OMIs.

The overall assessment of gingival oral health around OMIs across different time points demonstrated no difference in the chlorhexidine MW group compared to the placebo. An in-depth view of the results showed some marginal non-significant improvement during the first month in the chlorhexidine group which comes as no surprise considering the potential antiseptic properties of the MW during the first week of OMI placement. However, these potential benefits did not seem to translate

into a clinically detectable effect to impact the survivability of OMIs or the gingival health around them. An important factor for consideration could as well be related to the utilization of active tie-backs instead of elastomeric chains, which are more cleansable and less prone to food stagnation [23].

Chlorhexidine MW is a broad-spectrum biocide that works against gram positive and negative bacteria and fungi. It has rapid mechanism of action that disrupts the cell membrane within 30 seconds. For instance, previous literature showed that sterilization of the surgical field using chlorhexidine was found to play a vital role in decreasing inflammation across the entire treatment [24,25]. For intra-oral application, it has the capability to bind to proteins found in mucosa, which allows slow and long-term release. Therefore, a longer and sustained release of chlorhexidine MW during the 6-month study period may have influenced the overall outcomes in our trial. However, chlorhexidine MW is commonly prescribed for specified periods in many implant procedures [14-17].

On the other hand, findings showed that there were no lost OMIs on the right side, but three OMIs failed on the left side during the 6-month observation period. This might indicate better survivability on the right side, but the difference was non-significant. All included participants stated that they were right-handed. In the context of prior research, it would be expected that better brushing would be more prevalent on the left side [26]. However, differences were not significant and the number of failed OMIs would make it difficult to reach definite conclusions. Also, being right-handed might have exerted higher forces on the left side during brushing which could have possibly influenced the stability of the OMIs. In the placebo group, two OMIs failed as compared to just one in the chlorhexidine group. Though it is tempting to attribute such effect to the MW, the results were not statistically significant and the clinical significance from such difference is highly doubtful. The overall OMI failure rate in this trial accounted for only 4.7%, which is less than those commonly reported failure rates in previous systematic reviews (13.5% - 16.4 %) [27,28]. However, these promising figures could be easily explained by many influential factors such as the expertise of the operator, intra-operative management, the population group, and the site of chosen implantation [29].

Limitations

This is the first randomized report to shed some light on the clinical efficacy of using chlorhexidine MW prior to OMI placement and the overall clinical expectations from such procedures. However, there are some limitations that should be considered and some obstacles, which were encountered during the trial stages. It is important to note that this trial was completed during the Covid-19 pandemic. There were some disruptions and challenges during the follow up of some patients, which could have had some effect on their oral health maintenance and hygiene practices [30]. Nevertheless, these pandemic-related effects would have been expected to apply to both recruited groups. A further limitation is related to the lack of *a priori* trial registration. In addition, this trial followed participants for a 6-month observation period which is commonly reported in literature in cases of OMI anchorage reinforcement in premolar extraction-based cases [31]. The extended follow-up period would allow sufficient understanding whether the results could be linked to compliance-related treatment factors. In addition, this would also adhere to the recommendations that encourage adequate extension of RCT period for comprehensive assessment of the short- and long-term effects of interventions [32]. Finally, this report should only be considered as a pilot due to its sample size. Future trials in this area could expand their sample, explore OMI placement in different locations, and assess other populations to establish some form of generalizability.

Conclusions

- Within the limitations of this pilot randomized report, the utilization of chlorhexidine MW prior to OMI insertion and during the first week of placement does not appear to have a significant impact on the gingival health status around OMIs.
- The prescription of chlorhexidine MW to orthodontic patients requiring OMI placement does not seem to influence the overall survivability of OMIs in the short-term.

Conflict of interest

The authors declare no conflict of interest or financial interest.

References

1. Taylor KR, Kiyak A, Huang GJ, Greenlee GM, Jolley CJ, King GJ. Effects of malocclusion and its treatment on the quality of life of adolescents. *Am J Orthod Dentofacial Orthop*. 2009;136:382-92.

2. Jambi S, Walsh T, Sandler J, Benson PE, Skeggs RM, O'Brien KD. Reinforcement of anchorage during orthodontic brace treatment with implants or other surgical methods. *Cochrane Database Syst Rev*. 2014(8):Cd005098.

3. Kravitz ND, Kusnoto B. Risks and complications of orthodontic miniscrews. *Am J Orthod Dentofacial Orthop*. 2007;131:S43-51.

4. Yeung SC, Howell S, Fahey P. Oral hygiene program for orthodontic patients. *Am J Orthod Dentofacial Orthop*. 1989;96:208-13.

5. Zachrisson S, Zachrisson BU. Gingival condition associated with orthodontic treatment. *Angle Orthod*. 1972;42:26-34.

6. Sato R, Sato T, Takahashi I, Sugawara J, Takahashi N. Profiling of bacterial flora in crevices around titanium orthodontic anchor plates. *Clin Oral Implants Res*. 2007;18:21-6.

7. Kinane DF, Chestnutt IG. Smoking and periodontal disease. *Crit. rev. oral biol. med*. 2000;11:356-65.

8. Heasman PA, Hughes FJ. Drugs, medications and periodontal disease. *Br Dent J*. 2014;217:411-9.

9. Löe H, Schiott CR. The effect of mouthrinses and topical application of chlorhexidine on the development of dental plaque and gingivitis in man. *J Periodontal Res*. 1970;5:79-83.

10. Hugo WB, Longworth AR. Some aspects of the mode of action of chlorhexidine. *J. Pharm. Pharmacol*. 1964;16:655-62.

11. Zanatta FB, Antoniazzi RP, Rösing CK. The effect of 0.12% chlorhexidine gluconate rinsing on previously plaque-free and plaque-covered surfaces: a randomized, controlled clinical trial. *J Periodontol*. 2007;78:2127-34.

12. Giannelli M, Chellini F, Margheri M, Tonelli P, Tani A. Effect of chlorhexidine digluconate on different cell types: a molecular and ultrastructural investigation. *Toxicol In Vitro*. 2008;22:308-17.

13. McCoy LC, Wehler CJ, Rich SE, Garcia RI, Miller DR, Jones JA. Adverse events associated with chlorhexidine use: results from the Department of Veterans Affairs Dental Diabetes Study. *J Am Dent Assoc*. 2008;139:178-83.

14. Topouzelis N, Tsaousoglou P. Clinical factors correlated with the success rate of miniscrews in orthodontic treatment. *Int J Oral Sci*. 2012;4:38-44.

15. Uesugi S, Kokai S, Kanno Z, Ono T. Prognosis of primary and secondary insertions of orthodontic miniscrews: What we have learned from 500 implants. *Am J Orthod Dentofacial Orthop*. 2017;152:224-231.

16. Pjetursson BE, Asgeirsson AG, Zwahlen M, Sailer I. Improvements in implant dentistry over the last decade: comparison of survival and complication rates in older and newer publications. *Int J Oral Maxillofac Implants*. 2014;29 Suppl:308-24.

17. Young MP, Korachi M, Carter DH, Worthington HV, McCord JF, Drucker DB. The effects of an immediately pre-surgical chlorhexidine oral rinse on the bacterial contaminants of bone debris collected during dental implant surgery. *Clin Oral Implants Res.* 2002;13:20-9.

18. Moher D, Schulz KF, Altman DG. The CONSORT statement: revised recommendations for improving the quality of reports of parallel-group randomized trials. *Ann. Intern*. 2001;134:657-62.

19. 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;19:1188-96.

20. Wu TY, Kuang SH, Wu CH. Factors associated with the stability of mini-implants for orthodontic anchorage: a study of 414 samples in Taiwan. *Int J Oral Maxillofac Surg.* 2009;67:1595-9.

21. Chang HP, Tseng YC. Miniscrew implant applications in contemporary orthodontics. *Kaohsiung J Med Sci.* 2014;30:111-5.

22. Jasoria G, Shamim W, Rathore S, Kalra A, Manchanda M, Jaggi N. Miniscrew implants as temporary anchorage devices in orthodontics: a comprehensive review. *J. Contemp. Dent.* 2013;14:993-9.

23. Türkkahraman H, Sayin MO, Bozkurt FY, Yetkin Z, Kaya S, Onal S. Archwire ligation techniques, microbial colonization, and periodontal status in orthodontically treated patients. *Angle Orthod.* 2005;75:231-6.

24. Paolantonio M, Perinetti G, D'Ercole S, et al. Internal decontamination of dental implants: an in vivo randomized microbiologic 6-month trial on the effects of a chlorhexidine gel. *J Periodontol.* 2008;79:1419-25.

25. James P, Worthington HV, Parnell C, et al. Chlorhexidine mouthrinse as an adjunctive treatment for gingival health. *Cochrane Database Syst Rev.* 2017;3:Cd008676.

26. Tezel A, Canakçi V, Ciçek Y, Demir T. Evaluation of gingival recession in left- and right-handed adults. *Int. J. Neurosci.* 2001;110:135-46.

27. Schätzle M, Männchen R, Zwahlen M, Lang NP. Survival and failure rates of orthodontic temporary anchorage devices: a systematic review. *Clin Oral Implants Res.* 2009;20:1351-9.

28. Papageorgiou SN, Zogakis IP, Papadopoulos MA. Failure rates and associated risk factors of orthodontic miniscrew implants: a meta-analysis. *Am J Orthod Dentofacial Orthop.* 2012;142:577-595.e7.

29. Mohammed H, Wafaie K, Rizk MZ, Almuzian M, Sosly R, Bearn DR. Role of anatomical sites and correlated risk factors on the survival of orthodontic miniscrew implants: a systematic review and meta-analysis. *Prog Orthod*. 2018;19:36.

30. Wafaie K, Mohammed H, Mohamed AMA, Zhou J, Daniel B, Yiqiang Q. A qualitative study of orthodontic patients' experiences in quarantine during the COVID-19 pandemic outbreak. *Am J Orthod Dentofacial Orthop*. 2022;161:e498-e506

31. Motoyoshi M, Hirabayashi M, Uemura M, Shimizu N. Recommended placement torque when tightening an orthodontic mini-implant. *Clin Oral Implants Res.* 2006;17:109-14.

32. Cobourne MT, Huang GJ. Orthodontic clinical trials: Evaluating outcomes that actually matter. *Am J Orthod Dentofacial Orthop*. 2022;161:1-2.

Figure legends and Tables

Figure 1: Intra-oral photos of the used appliances and OMIs.

Figure 2: Flowchart showing the participant recruitment process.

Table 1: The modified gingival index.

Table 2: Baseline characteristics of the participants.

Table 3: Gingival oral health parameters in both study groups presented as frequency (n, %). Differences between right and left sides assessed by Wilcoxon's signed rank, and between-groups with Mann-Whitney U.

Table 4: Gingival oral health parameters in both study groups presented as minimum, maximum, median and IQR. Differences between right and left sides assessed by Wilcoxon's signed rank, and between-groups with Mann-Whitney U.

Table 1: The modified gingival index

Degree	Description
G0	Absence of any signs of inflammation
G1	Mild inflammation (redness) around OMIs without swelling or bleeding
G2	Moderate to severe inflammation around OMIs with marked redness, swelling, and/or with a tendency to bleed

G: Grade; OMI: Orthodontic miniscrew implant

Table 2: Baseline characteristics of the participants

Parameter		Chlorhexidine (n=16)	Placebo (n=16)	Significance
Age	Mean ± SD	24.1 ± 6.5	21.4 ± 6.2	$t = 1.165^{\S}$
	Range (min-max)	(16 – 33)	(16 – 35)	
Gender	Male; n (%)	5 (31.25%)	6 (37.5%)	$\chi^2 = .139^{\S}$
	Female; n (%)	11 (68.75%)	10 (62.5%)	

§non-significant at p>.05; t: independent t-test; χ^2: Chi- square test; n: number; SD: Standard deviation

Table 3: Gingival oral health parameters in both study groups presented as frequency (n, %). Differences between right and left sides assessed by Wilcoxon's signed rank, and between-groups with Mann-Whitney U

		Chlorhexidine			Placebo			Mann-Whitney U sign.
		Frequency n (%)			Frequency n (%)			
		R	L	Sign. (W)	R	L	Sign. (W)	
Baseline (insertion day)	G0	16 (100)	16 (100)	>1.00 ns	16 (100)	16 (100)	>1.00 ns	1.00 ns
	G1	0 (0.0)	0 (0.0)		0 (0.0)	0 (0.0)		
	G2	0 (0.0)	0 (0.0)		0 (0.0)	0 (0.0)		

	Failed	0 (0.0)	0 (0.0)		0 (0.0)	0 (0.0)		
1 month	**G0**	13 (81.3)	14 (87.5)	>1.00 ns	12 (75.0)	12 (75.0)	1.00 ns	0.724 ns
	G1	3 (18.8)	1 (6.3)		3 (18.8)	3 (18.8)		
	G2	0 (0.0)	0 (0.0)		1 (6.3)	1 (6.3)		
	Failed	0 (0.0)	1 (6.3)		0 (0.0)	0 (0.0)		
3 months	**G0**	11 (68.8)	12 (75.0)	0.713 ns	11 (68.8)	8 (50.0)	0.129 ns	0.867 ns
	G1	2 (12.5)	2 (12.5)		4 (25.0)	5 (31.3)		
	G2	3 (18.8)	1 (6.3)		1 (6.3)	1 (6.3)		
	Failed	0 (0.0)	1 (6.3)		0 (0.0)	2 (12.5)		
6 months	**G0**	10 (62.5)	10 (62.5)	0.891 ns	10 (62.5)	8 (50.0)	0.131 ns	0.838 ns
	G1	3 (18.8)	4 (25.0)		5 (31.3)	5 (31.3)		
	G2	3 (18.8)	1 (6.3)		1 (6.3)	1 (6.3)		
	Failed	0 (0.0)	1 (6.3)		0 (0.0)	2 (12.5)		

ns: non-significant at p>0.05; W: Wilcoxon's signed rank; n: number; sign: significance; R: right; L: left; G: group

Table 4: Gingival oral health parameters in both study groups presented as minimum, maximum, median and IQR. Differences between right and left sides assessed by Wilcoxon's signed rank, and between-groups with Mann-Whitney U

Follow-upTime		**Chlorhexidine**			**Placebo**			**Mann-Whitney U sign.**
		Median (IQR), min, max.			**Median (IQR), min, max.**			
		R	**L**	**Sign.^{W}**	**R**	**L**	**Sign.^{W}**	
Baseline	**Median (Q1-Q3)**	0.0(0.0-0.0)	0.0 (0.0-0.0)	1.00 ns	0.0 (0.0-0.0)	0.0 (0.0-0.0)	1.00 ns	1.00 ns
	Min-max	0.0 - 0.0	0.0 - 0.0		0.0 - 0.0	0.0 - 0.0		
1 month	**Median (Q1-Q3)**	0.0 (0.0-0.0)	0.0 (0.0-0.0)	1.00 ns	0.0 (0.0-0.75)	0.0 (0.0-0.75)	1.00 ns	0.724 ns
	Min-max	0.0 - 1.0	0.0 - 3.0		0.0 - 2.0	0.0 - 2.0		
3 months	**Median (Q1-Q3)**	0.0 (0.0-1.0)	0.0 (0.0-0.75)	0.713 ns	0.0 (0.0-1.0)	0.5 (0.0-1.0)	0.129 ns	0.867 ns

	Min-max	0.0 - 2.0	0.0 - 3.0		0.0 - 2.0	0.0 - 3.0		
6 months	**Median (Q1-Q3)**	0.0 (0.0-1.0)	0.0 (0.0-1.0)	0.891 ns	0.0 (0.0-1.0)	0.5 (0.0-1.0)	0.131 ns	0.838 ns
	Min-max	0.0 - 2.0	0.0 - 3.0		0.0 - 2.0	0.0 - 3.0		

IQR: interquartile range; min: minimum; max: maximum; ns: non-significant at p>0.05; sign: significance; R: right; L: left

Figure 1: Intra-oral photos showing the fixed orthodontic appliances and OMIs

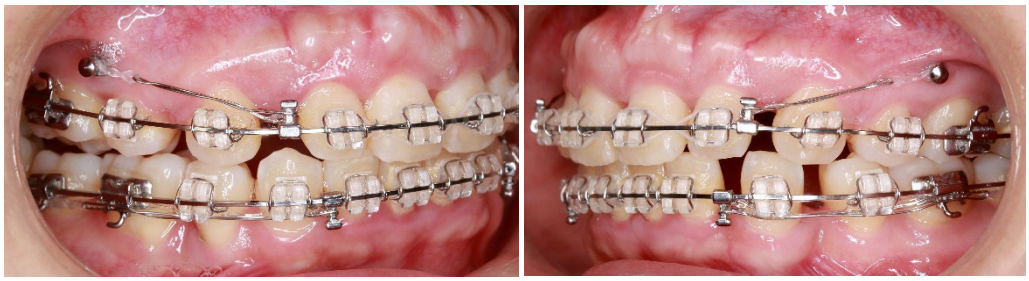
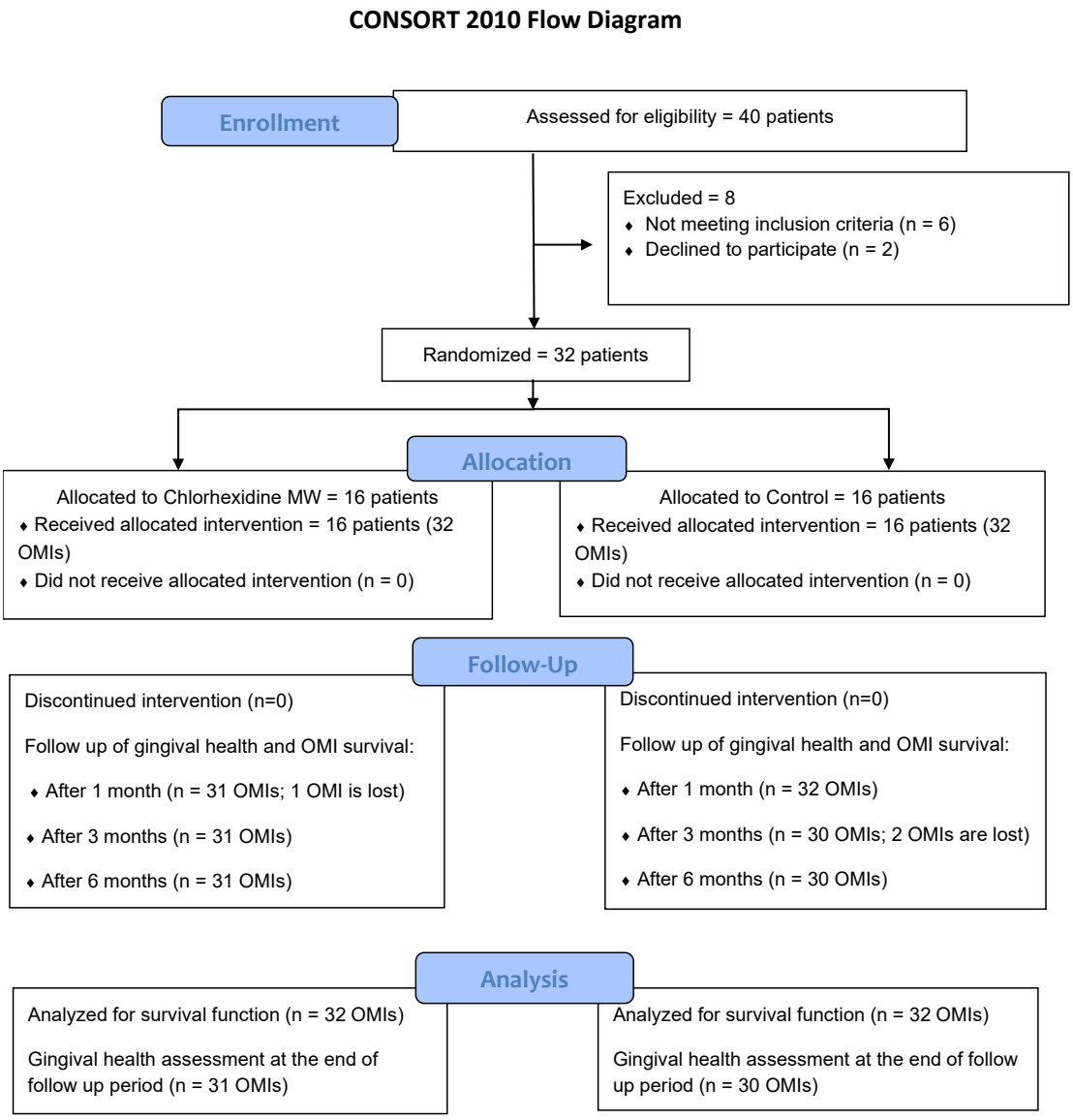


Figure 2: Flowchart showing the overall participant recruitment process



OMI: Orthodontic Miniscrew Implant; n: number; MW: Mouthwash