Study Protocol

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Probiotic vs Zinc Chlorhexidine (Zn/CHX) Mouthwashes in Control Halitosis and Plaque in Fixed Orthodontic Patients

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Probiotic vs Zinc chlorhexidine Zn/CHX mouthwashes in control halitosis and plaque in fixed orthodontic patients: A randomized clinical trial

Introduction

Fixed orthodontic appliance promote orthodontic bracket-archwire assembly, which disrupts biofilm balance and amplify plaque build up in inaccessible sites which compromise tooth brushing efficiency [1-2]. These alteration enhance mature biofilm development by lengthen plaque residence period and roghen surface[1].Prospective studies reveal that fixed orthodontic appliances significantly elevate plaque indices especially in mechanically inaccessible sites [2,3],therefore specific orthodontic index which is modified bonded bracket index (mBB index) have been used to quantify plaque for orthodontic patients [4] .

Localized plaque accumulation at brackets and gingival margins promotes gingival inflammation, throughout maturation of anaerobically protected biofilms sites, this enhances anaerobic metabolism resulting in higher production of volatile sulfur compounds (VSC) associated with oral malodor specifically. Patients may exhibit signs of gingival inflammation alongside marked oral malodor [5-7].

According to the World Health Organization (WHO) and the Food and Agriculture Organization of the United Nations (FAO), probiotics are “living microorganisms that, when administered inadequate amounts, confer benefit to host health” [8]. Probiotic mouth washes are evaluated as a microbiome modulating supplement which aim to modulating orthodontic related biofilm rather than indiscriminately suppressing it, previous orthodontic studies indicate thatprobiotic interventions may reduce mutans streptococci level in patients with fixed appliance therapy, however reported effects on dental plaque and gingival parameters remain inconsistent across clinical trials [9,10].

Zinc chlorhexidine mouth washes (Zn/CHx) are widely used as clinically applicable adjuncts for controlling halitosis and plaque related inflammation without prolonged use of high concentrated antiseptic agents. Fixed appliance,clinical studies consistently demonstrate that Zn/CHx formulations significantly reduce organoleptic halitosis scores and volatile sulfur compound levels providing both short and long term benefits.Regular Zn/CHx use provides reliable control of oral malodor [11-13].

Although probiotics have garnered heightened attention during fixed orthodontic therapy, clinical studies show heterogenous outcomes, and clear clinical benefit in reduction of halitosis and plaque accumulation in F.O.T. [14] .Therefore, this study aims to determine the effect of probiotics and Zn/CHx mouthwashes on plaque accumulation and halitosis in fixed orthodontic patients.

Materials and methods

Study design:

This randomized double blind parallel groups clinical trial with two arms carried out at the postgraduate orthodontic clinic at the college of dentistry /Baghdad University. The recruitment period will be between December 2026 to May 2026.The clinical trial will be prospectively registered at clinical Trials.gov in compliance with WHO and ICMJE guidelines. Ethical approval will be obtained from the scientific and ethical committee at the college of Dentistry , University of Baghdad before to patients recruitment.

Participants:

A total of 60 participants orthodontic patients receive Fixed orthodontic treatment demonstrating effects that corresponds to the short course antiseptic effect of probiotic and Zn/CHX mouth washes on halitosis and biofilm buildup were included in this study. Participants undergo dental examination with medical and dental history. Prior to data collection, power analysis will be performed using G* power software version 3.1 to determine the minimum required sample size .

The inclusion criteria :

The participants wear active labial F.O.A in both arches (with bonded 1st molars) for at least 6 months, promoting a consistent intra oral environment with defined plaque retentive areas surrounding brackets (excluded patients with banded molar); fully erupted permanent dentition; baseline intra oral halitosis organoleptic score must be ≥2 on a 0–5 OlS scale; baseline participant have mild to moderate plaque buildup as measured by the mBB index must have score above zero. Participants aged between 18-30 years old with good medical health, with no systematic disease or consuming drugs known to influence salivary flow, halitosis, or oral microbiota, not alcohol drinking, not smokers, pregnant and lactating womens were excluded , free from any allergy or intolerance to the study ingredients. Prior 1 month of the study they don't used corticosteroid treatment, antibiotic, antimicrobial mouth rinses, not used oral spray or tongue scraper, anti-inflammatory drugs or probiotics, and no periodontal therapy. The participant with no caries, mild to moderate crowding.

The participant included in this clinical trial were divided into 2 groups ,1st group 30 participants used probiotic mouth wash,2nd group 30 participants used zinc chlorhexidine Zn/CHO mouth wash. Eligable participants were required to demonstrate adequate compliance with prescriped oral hygine instructions and provide agreement with all aspects of the study protocol, written informed consent was secured prior to their inclusion.

At the beginning of the study all participants received a standardized soft tooth brush and fluoridated tooth paste provided from the examiner . Prior to clinical assessment, the participant not chewing gum, not using tobacco for 12 hours, not eating highly potent odor (onion, garlic, alcohol, and coffee for 24 hours. At the day of scoring plaque and halitosis(at morning 2 hours prior to measurements), not brushing and not using mouth washes. During intervention period the particpitant instructed to rinsing his mouth with 10-15 ml of mouth wash for 60 seconds for each group daily at morning and night after tooth brushing for 4 weeks for each group.

Study procedure:

1. **Plaque accumulation:** plaque build up on bracket surfaces was evaluated by using modified bonded bracket index (mbb index) which is intended for multi bonded orthodontic patients, also it merged two plaque indices dental and orthodontic [4]. The mbb index used for orthodontic researches owing to consistent reliability and quantitive correspondence [15-17] . The mbb index will conducted in the morning at 10 AM and prior to brushing teeth .1st using disclosing agent(erythrosine) then rinsing gently to eliminate excessive amount of disclosing agent. After that scoring instantly without delay tooth by tooth in sequence pattern. bonded teeth included in this index from anterior teeth, premolars, and 1st molar (banded molar excluded). Collect data in scoring sheet. The modified bonded bracket index was applied to assess plaque level through a percentage-based scoring system consistent with the mBB index. the labial surface of each tooth carrying a bracket was sectioned into four evaluation zones (incisal, distal, mesial, gingival)[16]. Plaque accumulation was classified into the following assessment grades: grade 0 - no plaque, grade 1 - plaque covers ≤ 1/3 of the tooth surface, grade 2 - plaque covers ≤ 2/3 of the tooth surface, grade 3 - plaque covers > 2/3 of the tooth surface [15-17].
2. **Halitosis:** subjective assessment of halitosis determined by using organoleptic score (OLS) by single calibrated examiner who was blinded to group allocation [18], which determined for all participants at the standardized constant distance (10cm) [18], and the same time (morning) with adequate room ventilation. The examiner refrain from parfum, coffee, smoking, and eating food for 30 minutes prior to scoring. OLS were taken at three readings (with 1 minute interval) by well-trained single examiner then calculate the mean of 3 readings to increase reliability and determine intra examiner consistency. OLS by using Rosen berg scale from zero (no odor) to 5 (highly potent odor), score≥2 considered clinically significant. Participant sit comfortably in adequately ventilated room without detectable odor, and instructed to closing the lips and breathing through the nostrils for 1-2 minutes before test to permit volatile sulfur compounds (VSC) to collect intra-orally. Then participants were performed a slow oral exhalation (with closing the nostrils), then a gentle inhalation and finally the examiner determined the degree of odor and rates the score directly after odor evaluation.

Organoleptic score (OLS) 0-5 of halitosis[18-22]

0-No odor

1-Very mild odor

2- Moderate malodor

3-strong malodor

4-Extremely strong malodor

5-light malodor

Probiotic Vs Zn/CHX Mouth washes

Probiotic :composed of powerful strain of s.salivarius that preserve innate immune system of the main entry point of the body by taken on a regular basis and establish colonization of the throat and mouth which provide clinical care and support of nose, ear, mouth and throat health and provide fresh breath.

Zinc chlorhexidine mouth wash (Zn/CHX) composed of CHX concentrated 0.025% with Zinc acetate 0.3 %, the low concentration of CHX to maintain optimal efficacy with lower incidence of side effects. Zn /CHX was chosen as the active control due to the fact the mixture of Zn salts with low concentration of CHX have shown superior and sustained intra-oral efficacy in managing intra oral halitosis in randomized clinical trial [23].

An independent researcher prepared and coded the mouth wash bottles A&B separately, who was not involved in clinical examination, so both the participant and examiner were blinded to the type of mouth wash assigned to each code until data collection and statistical analysis were completed.

Timetable for data collection:

T0 (Day 0): Baseline prior to intervention began, check dental and medical history, inclusion and exclusion criteria, control oral hygiene routine and record mbb index, OLS.

T1 (Day 14): short term effect on plaque accumulation and halitosis, verify participant adherence with instructions, adverse effect (taste alteration, teeth staining, irritative mucosal response), record mbb index and OLS. Participant were reminded and reinforced regarding oral hygiene and mouth wash instructions.

T2(Day 30): study end point in plaque accumulation and halitosis, verify participant compliance, adverse effect, record mbb index and OLS.

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