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The miracle gel for Down syndrome adolescents

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Abstract

Aim: The aim of this study was to determine if chlorhexidine gel (0.12%) had any effect on the levels of *Porphyromonas gingivalis* in subgingival plaque of DS children and healthy children.

Methods: Twenty Down syndrome and twenty healthy children formed Group I and Group II, respectively. In Group I, the plaque index (Silness and Loe) and gingival index (Loe and Silness) were recorded. In both groups, subgingival plaque samples were collected to determine the levels of *Porphyromonas gingivalis.* Following the collection of baseline plaque samples, oral prophylaxis was carried out in both groups. In Group I, chlorhexidine (0.12%) gel was applied over the gingiva once in two weeks for a period of 3 months. Parents were given oral hygiene counselling on method of tooth brushing for their children. A second sample of plaque was obtained at the end of 3 months.

Results: A significant difference was observed in mean *Porphyromonas gingivalis* levels between Down syndrome and healthy children ($p<0.001$). There was a significant difference observed in the mean levels of *Porphyromonas gingivalis* at different time intervals in Group I ($p<0.001$). Pairwise comparison of *Porphyromonas gingivalis* levels in plaque at different time intervals in Group I showed a significant difference between baseline and 3 months ($p<0.001$). A significant difference was observed in the mean plaque index scores and mean gingival index scores at different time intervals in Group I ($p<0.001$). Pairwise comparison of mean plaque index scores and mean gingival index scores at different time intervals in Group I showed a significant difference between baseline and 3 months ($p<0.001$).

Conclusion: The significantly higher levels of *Porphyromonas gingivalis* in subgingival plaque of Down syndrome children than that of healthy children showed

improvement following tooth brushing and use of 0.12% chlorhexidine gel. It indicates the need for continuous mechanical and chemical plaque control measures along with regular monitoring in Down syndrome children.

Keywords: Down syndrome, Prevention, Chlorhexidine, Gingival health.

Introduction

Down's syndrome (DS), which was first defined by Langdon Down, is characterized by physical and mental abnormalities due to underlying chromosomal aberrations. Patients with Down syndrome present mouth alterations such as: pseudo macroglossia, protruded tongue and malocclusions, and these alterations interfere with the quality of toothbrushing. The manual dexterity and many times, the motivation, are indispensable factors for efficient oral hygiene through mechanical means in patients with Down syndrome.1,2 This in turn leads to the accumulation of plaque and debris, hence favouring development of gingivitis and periodontitis.3 The obstacles inherent to children with Down syndrome and the difficulties faced by parents and /or people in charge for toothbrushing, lead the professional in dentistry to look for a substance capable of aiding and stimulating these patients in the mechanical control of the dental biofilm.2 The control of biofilm in an individual with Down syndrome requires a regime that is simple, easy for patient or carer to use, acceptable to both patient and carer and sparing of resources. It is acknowledged that chemical adjuncts may potentially simplify plaque control routines however, the way in which they are delivered may be critical to a successful outcome. Supervised preventive programs have been shown to be very effective in reducing plaque and gingival inflammation in people with Down Syndrome.4 The use of antimicrobial agents can be a useful aid in plaque control for these individuals.5

Chlorhexidine is a biguanide with cationic properties. It has been shown to decrease plaque bacteria by up to 62% 6,7 and can be useful in controlling dental biofilm and in the reduction of gingival bleeding.2 It is bacteriostatic in low concentrations and bacteriocidal in high concentrations and is effective against gram positive and gram negative bacteria.8

Gingivitis and periodontitis begin early in Down syndrome population and their severity increases with age. Periodontal disease is the most significant oral health problem in Down syndrome and it is often diagnosed during adolescence. Altered immune/inflammatory responses, and early colonization of periodontal pathogens in these individuals are important contributing factors to their increased susceptibility to periodontitis. *Porphyromonas gingivalis* is the predominant pathogen seen in subgingival dental plaque of DS adolescents.4 *Porphyromonas gingivalis* has been shown to significantly impair the cellular migration of cultured gingival fibroblasts in individuals with Down syndrome. It is suggested that *Porphyromonas gingivalis* readily invades gingival tissue and subsequently impairs cellular motility, resulting in prevention of healing and regeneration of periodontal tissues.

Most of the studies on oral health of DS children and adolescents have reported on their salivary parameters, dental caries and oral hygiene.6-9 Therefore, the objective of this clinical study was to evaluate the effect of an antimicrobial agent like Chlorhexidine on the gingival health of subjects with Down syndrome. The aim was also to assess whether tooth brushing, along with the use of 0.12% chlorhexidine gel, has an effect on the levels of plaque index, gingival index and *Porphyromonas gingivalis* levels of adolescents with DS.

Methodology

This preliminary study was carried out on 30 adolescents, aged 15-18 years, with Down Syndrome selected from an institution for special children in Bangalore. Prior to the study, ethical approval and clearance was obtained from the Institutional Ethics Review Board of our institution. Written permission was obtained from the concerned authorities of the institution for special children. The nature of the study was explained to the concerned authorities and prior informed written consent was taken from their parents/caretakers of all the subjects. A proforma was used to gather demographic data, medical and drug history. The exclusion criteria were: (1) those on long term medication, (2) very uncooperative and inability to cooperate, (3) severe intellectual disability present along with Down syndrome, (4) association with any other medically compromised conditions and (5) those who had undergone oral prophylaxis in the preceding 6 months.

Oral examination was done by a single trained and calibrated examiner under artificial light using a sterile dental mirror and WHO CPITN probe. The plaque index (PlI) was recorded using Silness and Loe index10 and gingival health was assessed using Loe and Silness gingival index (GI).11

Collection of baseline samples: Subgingival plaque samples were collected from the mesial and buccal sites of first permanent molars by means of sterile curettes. The plaque samples were then transferred into sterile eppendorf tubes containing buffer solution and were placed in an icebox (0˚C). It was then immediately transferred to Credora Life Sciences, an ISO certified laboratory and stored at -80º C until further analysis.12

Assessment of *Porphyromonas gingivalis*

Microbiological evaluation of the dental plaque samples for the presence of *Porphyromonas gingivalis* was done by suspending the plaque sample in 1ml of saline. Aseptically 0.1ml. of the suspension was transferred to Tryptic soy broth (TSB culture medium) and incubated under anaerobic conditions overnight for 24 hours at 37ºC. The growth of the bacteria was measured spectrophotometrically by reading its optical density (OD) at 600 nanometer wavelength and expressed as colony forming units (cfu) per ml.13

Following the collection of baseline plaque samples, oral prophylaxis was carried out in all the subjects.

Preparation of the Chlorhexidine gel (0.12%): A blank gel consisting of hydroxy propyl methylcellulose (HPMC), glycerine and water was prepared. Water was added to HPMC followed by vigorous mixing until the HPMC became completely miscible with water. Glycerine was then added to this mixture and mixed well in order to form the blank gel. Further, a commercially available chlorhexidine (CHX) gel (2%) (Unilab Chemicals and Pharmaceuticals Pvt. Ltd., India) was added to the blank gel and stirred using a magnetic stirrer to obtain 60 g of 0.12% CHX gel.

Application of the gel: This indigenously prepared CHX gel (0.12%) was applied in subjects with Down syndrome, by gently massaging 0.3-0.5g of the gel over the buccal and palatal/lingual surfaces of gingiva using sterile cotton swabs. The time of application was between 10am to 11am. They were instructed not to drink, eat or rinse their mouth for 30 minutes following the gel application. Regular application of CHX gel was carried out by the same examiner once in 15 days over a period of 3 months. A total of 6 applications was done. During this period, parents were given oral hygiene counselling on method of tooth brushing for their children.

At the end of 3 months, plaque index and gingival index were measured, as well as a second sample of subgingival plaque was collected from the Down syndrome subjects in the same manner as described earlier.

Data obtained was tabulated and subjected to statistical analysis using repeated measures of ANOVA Test for the comparison of mean *Porphyromonas gingivalis* levels based on optical density between both the time intervals in Down syndrome group.

Results

The mean levels of *Porphyromonas gingivalis* at baseline was 0.317±0.035 cfu/ml. There was a significant difference observed in the mean levels of *Porphyromonas gingivalis* between baseline and at 3 months ($p<0.001$) (Tables 2).

A significant difference was observed in the mean plaque index (PlI) and mean gingival index (GI) scores in all the subjects between baseline and 3 months.

Discussion

Individuals with Down syndrome are more likely to develop aggressive periodontal disease at an earlier age than the general population.4,14,15 Subjects with severe intellectual disability and very uncooperative children were excluded from our study so as to facilitate adequate collection of dental plaque samples and proper assessment of gingival health. Sampling was done only after establishing a friendly relationship between the examiner and subjects. Alterations in levels of *Porphyromonas gingivalis* were avoided by including only those DS individuals who had not undergone oral prophylaxis in the preceding 6 months. Dental plaque was recorded using Silness and Loe plaque index because it ignores the coronal extent of plaque and assesses only the thickness of plaque at the gingival area of the tooth. It has good validity and reliability for both mechanical anti-plaque procedures and chemical agents. Loe and Silness gingival index was used because it is simple to use, reliable and can determine the severity of gingivitis. 10,11

Spectrophotometric analysis was employed to assess growth of *Porphyromonas gingivalis.* It is estimated by optical density which is based on the fact that an increase in the number of bacteria, results in less light transmitted. It is a simple, rapid, economical and non-destructive method.13

In individuals with DS, there is a need for an oral hygiene regime that is simple, easy, economical and acceptable to both patient and caregiver.16 Cumbersome tooth brushing techniques and flossing, may be difficult to practice, due to reduced manual dexterity.17 Anti-plaque chemical agents such as chlorhexidine gluconate along with tooth brushing, has proved to be useful in reducing plaque and gingivitis.17 However, the manner in which it is delivered may be critical to a successful outcome.18 Mouth rinses may not be suitable for use in DS, due to their inability to rinse the mouth and low gag reflex. Application of CHX gel in trays has not been well accepted in children with learning disabilities.17 Higher concentrations of CHX (0.2% or 1%) have been reported to cause mucositis, superficial mucosal erosions and burning sensation.19 In this study, a structured plaque control regime was implemented in subjects with DS. Tooth brushing which is a simple yet effective method for reducing plaque and gingivitis was followed. Good compliance was achieved by using a convenient and simple technique of massaging 0.12% CHX over the gingiva. CHX, a cation, interacts and forms salts of low solubility with anions, such as sodium lauryl sulfate (SLS) and sodium monofluorophosphate (MFP) present in dentifrices. To optimize the anti-plaque effect of CHX, an interval of 2 hours was given between tooth brushing and application of CHX gel.20 Long term use of CHX has side effects of extrinsic staining,19 offensive taste and altered taste sensation. Hence, the gel application was carried out fortnightly, and only for a period of 3 months.

Several studies have shown higher plaque accumulation and greater severity of gingivitis in DS children compared

to healthy children.6,7,21 In the present study, there was a significant decrease of 35.4% in plaque scores from 1.47 to 0.95, at the end of 3 months. Similarly, moderate gingivitis (GI score=1.48) that was seen at the beginning showed a significant reduction in inflammation resulting in mild gingivitis (0.99) after 3 months.

Our study found the levels of *Porphyromonas gingivalis* to be significantly higher in DS. This microorganism thrives in individuals with poor oral hygiene and high plaque accumulation. In the present study, tooth brushing along with CHX gel application in subjects with DS decreased *Porphyromonas gingivalis* by 38.9% (from 0.316 cfu/ml to 0.193 cfu/ml) at the end of 3 months.

Gingival massaging of CHX gel can mechanically disrupt the biofilm on teeth, dispersing the agents throughout the gingiva, stimulating blood circulation to the gingival tissues and thereby strengthening its immune response.22 The most important unique property of CHX is its substantivity or oral retentiveness.23 CHX also has the ability to neutralize *Porphyromonas gingivalis.* The dicationic positively charged CHX is attracted to the negatively charged phosphate containing compounds in the bacterial cell wall. This alters the integrity of the bacterial cell membrane and makes CHX get attracted to the inner cell membrane and binds to phospholipids causing leakage of low molecular weight compounds like potassium ions. Cytoplasm of the cells get coagulated and chemically precipitated due to the formation of phosphate complexes which include adenosine triphosphate and nucleic acids leading to bacterial death.24,25

Preventive care has shown to be effective in suppressing the onset and progression of periodontal disease in this population.26 Individuals with DS need more assistance from caretakers with their daily oral health care. In our study, reinforcement of tooth brushing through instructions, monitoring and continuous motivation was carried out in the presence of parents/ caregivers and school teachers. The visits were interactive and parents discussed their child's oral health.

The results of this study indicated that professional treatment along with regular tooth brushing and CHX gel (0.12%) application for a short duration brought about an improvement in gingival health. However, in order to obtain long lasting effects, periodic application of CHX gel in low concentration may be necessary in addition to mechanical plaque control.

Conclusions

1. The mean levels of *Porphyromonas gingivalis* in subgingival plaque of Down syndrome at baseline was 0.317 ± 0.035 cfu/ml ($p<0.001$).
2. In Down syndrome, following tooth brushing and the use of 0.12% chlorhexidine gel for 3 months, there was a significant reduction in mean *Porphyromonas gingivalis* levels (0.1936±0.051 cfu/ml) ($p<0.001$).
3. The mean plaque index and mean gingival index scores in Down syndrome children at baseline were 1.47 ± 0.36 and 1.48 ± 0.35, respectively. Tooth brushing and the use of 0.12% chlorhexidine gel for 3 months, showed a significant reduction in mean plaque index (0.95±0.28) and mean gingival index scores (0.99 ± 0.38) ($p<0.001$).

Legends of Tables

Table 1: Comparison of mean *Porphyromonas gingivalis* levels between both the time intervals in DS adolescents

Time	*Porphyromonas gingivalis* levels Mean ± SD (cfu/ml)	H	p value
Baseline	0.3211±0.037	61.82	p<0.001*
At 3 months	0.1973±0.04		

*p<0.001 is significant

Table 2: Comparison of mean plaque index (PlI) and gingival index (GI) scores between both the time intervals in DS adolescents

Index	Time		H	p value
	Baseline	At 3 months		
Plaque Index Score (Mean± SD)	1.48 ± 0.2	0.95±0.3	38.100	<0.001*
Gingival Index Score (Mean± SD)	1.46 ± 0.1	0.95 ± 0.1	39.077	<0.001*

*p<0.001 is significant

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