# The effect of oral health education on dental plaque development and the level of caries-related *Streptococcus mutans* and *Lactobacillus* spp.

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SUMMARY The aim of this study was to determine the influence of oral hygiene instruction (OHI) on oral hygiene status and on the reduction of inducing bacteria (*Streptococcus mutans* and *Lactobacillus* spp.) in 30 subjects (12 males and 18 females) with an age range from 10 to 14 years. The patients were instructed on how to carry out effective oral hygiene close to brackets and ligatures, and in the use of dental floss for cleaning spaces around brackets, between the teeth and under archwires. The plaque index (PI) was used to measure oral hygiene status, and the level of *S. mutans* and *Lactobacillus* spp. was determined using the colour reaction time test before and after 1 month of OHI. Statistical analysis included a Student's *t*-test and chi-square test.

The results showed that OHI significantly improved oral hygiene. The percentage of patients with a PI exceeding 1 decreased from 23.3 to 10 per cent and for those with a PI below 0.1 it increased from 20 to 60 per cent. The level of *S. mutans* and *Lactobacillus* spp. was not reduced. The percentage of subjects with excessive levels of *S. mutans* (above 10<sup>6</sup> bacteria in 1 ml of saliva) decreased after OHI but only from 73.3 to 70 per cent. For *Lactobacillus* spp., a level above 10<sup>5</sup> of bacteria in 1 ml of saliva was found for all subjects at both time points. Patients with fixed appliances, trained in scrupulous tooth cleaning, remain at risk of developing caries and should be carefully monitored.

#### Introduction

The human oral cavity is a complex ecosystem inhabited by more than 300 bacterial species, mycoplasmas, protozoa, and yeasts (Marcotte and Lavoie, 1998). Any external interference could disturb the delicate balance between components of microflora in this environment. Fixed orthodontic appliances are an example of such interference. Bonding of brackets usually includes acid etching of enamel, which results in changes in the morphology and chemical nature of the oral cavity. It has been found that decalcified enamel constitutes good support for adhesion and proliferation of Streptococcus mutans, Veillonella spp., and Actinomyces viscosus (Boyar et al., 1989). It is also known that living cells easily adhere and colonize polymeric surfaces (Griesser et al., 1993; Röder et al., 1993; Tamada and Ikada, 1993; Zühlke et al., 1993; Langer, 1995; West and Hubbell, 1999). Thus, composite resins containing polymers used for attaching brackets to etched enamel provide surfaces especially prone to adhesion and growth of microorganisms (Weitman and Eames, 1975; Gwinnett and Ceen, 1979; Sukontapatipark et al., 2001). Moreover, the configuration of fixed appliances promotes retention of food and reduces efficiency of self-cleaning by saliva (Balenseifien and Madonia, 1970). In effect, fixed dental appliances induce development and retention of bacterial plaque (Balenseifien and Madonia, 1970; Zachrisson, 1976; Gorelick et al., 1982; O'Reilly and Featherstone, 1987;

Ögaard *et al.*, 1988; Mitchell, 1992). Development of dental plaque usually leads to an increased level of caries inducing bacteria in the oral cavity, e.g. *S. mutans* and *Lactobacillus* spp. (Balenseifien and Madonia, 1970, Diamandi-Kiopioti *et al.*, 1987; Boyar *et al.*, 1989). However, according to Bloom and Brown (1964), *Lactobacillus* spp. may not be the primary constituent of dental plaque.

The above observations indicate that fixed orthodontic appliances induce a certain risk for the development of caries. Therefore, orthodontists commonly recommend scrupulous tooth cleaning. This subject has been discussed by many authors (Zachrisson, 1974; Gold, 1975; Schwaninger and Vickiers-Schwaninger, 1979; Trombelli *et al.*, 1995) who, in addition to regular tooth brushing, often suggest measures such as the use of an electric toothbrush (Trombelli *et al.*, 1995) and irrigation (Schwaninger and Vickiers-Schwaninger, 1979). To decrease the bacterial level and plaque, various mouthwashes and varnishes have been used (Harrap and Best, 1984; Addy, 1986; Axelsson and Lindhe, 1987; Schaeken *et al.*, 1989; Sandham *et al.*, 1991; Machuca *et al.*, 1997; Twetman and Petersson, 1997; Van Lunsen *et al.*, 2000).

The aims of this investigation were to study the effect of oral hygiene instruction (OHI), the use of floss, and a manual tooth brush designed specifically for orthodontic patients, and to determine plaque and *S. mutans* and *Lactobacillus* spp. levels.

## Subjects and methods

Thirty patients (12 males, 18 females) with an age range of 10 to 14 years, treated with fixed appliances at the Department of Orthodontics, Medical University of Lodz, Poland, were included in the study. The levels of S. mutans and Lactobacillus spp. were assessed using the colour reaction time test (Vivadent, Schaan Liechtenstein). The kit contains agar plates for cultivation of S. mutans and Lactobacillus spp. The plates were inoculated with saliva obtained from the patients and incubated at 37°C for 48 hours. Bacteria levels were then evaluated to compare the density of colonies of S. mutans and/or Lactobacillus spp. with the chart provided by the supplier. In addition, each patient's plaque index (PI) according to Silness and Löe (1964) was determined. Briefly, determination of PI was performed as follows: visualization of dental plaque was achieved using plaque colourant disclosing tablets (RedCote, J.O. Butler Co., Chicago, Illinois, USA). A score of 0 to 3 was assigned for the four sides of each tooth; score 0 when there was no plague on the tooth wall, 1 when plaque was invisible to the naked eye but could be collected using a probe, 2 when plaque was visible at the gingival margin, and 3 when plaque not only was visible at the gingival margin but also covered a significant part of the tooth wall. The average score for an individual tooth was equal to the sum of the scores for all sides divided by four, and the PI for each patient was equal to the sum of the average scores for all teeth divided by the number of teeth.

According to Bratthall (1980) and Loesche (1986), levels of *S. mutans* exceeding 10<sup>6</sup> and/or *Lactobacillus* spp. exceeding 10<sup>5</sup> of bacteria per 1 ml of saliva indicate a high caries risk. On the basis of the above criteria, the patients were assigned to a group with either acceptable or excessive levels of *S. mutans* and *Lactobacillus* spp.

PI was determined for the incisors and first molars in the upper and lower arch. The average value of these 12 teeth was used to register oral hygiene: 'very good' corresponded to PI < 0.1, 'good' to  $0.1 \le PI < 1$ , 'poor' to  $1 \le PI < 2$ , and 'very poor' to  $2 \le PI \le 3$ .

After determination of the average PI and *S. mutans* and *Lactobacillus* spp. levels, the subjects were provided with manual toothbrushes specifically designed for orthodontic patients (Ortho P35, Oral-B Laboratories, Kronberg im Taunus, Germany) and were instructed on how to efficiently clean their teeth. The heads of these toothbrushes are shaped in such a way that they fit the brackets. The manufacturers suggest an additional period of intensive brushing in the horizontal direction with brackets and ligatures fitting rifts on the brush and scrupulous cleaning of the spaces around brackets, under the arch and between teeth with dental floss. One month after OHI, oral hygiene and *S. mutans* and *Lactobacillus* spp. levels were again determined.

# Statistical analysis

The hypothesis that the average PI values and the difference in average values before and after OHI ( $\Delta$ PI) differ significantly was verified, assuming that the analysed variables had a normal distribution. The hypothesis that the distribution of patients into groups with different hygiene status does not depend on OHI was verified using the chi-square goodness-of-fit test for contingency tables. The calculations were made according to Green and Margerison (1978).

# Results

The average PI values  $[\overline{PI} = \sum_i PI_i/N]$  where  $PI_i$  denotes the PI for patient 'i' and N is the total number of patients] before and after OHI for females, males, and the genders combined are shown in Table 1 together with the difference in average PI values. The data indicate that the  $\overline{PI}$  for males was higher than for females. However, at the confidence level of  $\alpha = 0.05$ , this difference was not significant.

 $\overline{\text{PI}}$  values were always lower following OHI (Table 1). To determine the statistical importance of this observation, the hypothesis that  $\overline{\Delta \text{PI}} = 0$  (i.e. that there is no difference in PI for patients before and after OHI) was tested. Confirmation of the hypothesis was undertaken using the Student's *t*-test, where  $t = (\overline{\Delta \text{PI}}/s)\sqrt{N-1}$  in which *s* denotes the standard deviation and *n* the number of patients. The *t* values for females, males, and both genders combined were 3.94, 3.82, and 4.87, respectively, and were larger than the critical values of *t* at the confidence level  $\alpha = 0.05$ . Thus, the hypothesis that  $\overline{\Delta \text{PI}} = 0$  was rejected and an alternative hypothesis that OHI led to a significant reduction in the PI was accepted.

However, OHI did not significantly affect the number of patients with acceptable levels of *S. mutans* and *Lactobacillus* spp. (below 10<sup>6</sup> and 10<sup>5</sup> of bacteria in 1 ml of saliva for *S. mutans* and *Lactobacillus* spp., respectively). The number of female patients with an acceptable level of *S. mutans* increased after OHI from 6 to 7. For male patients no change was observed.

The level of *Lactobacillus* spp. exceeded the acceptable level (10<sup>5</sup> in 1 ml of saliva) in all examined patients and did not depend on OHI.

**Table 1** Average plaque indices  $(\overline{PI})$  before and 1 month after oral hygiene instruction (OHI) and the average changes in plaque indices  $(\overline{\Delta PI})$ .

	PI before OHI (standard deviation)	PI after OHI (standard deviation)	$\overline{\Delta PI}$ (standard deviation)
Females	0.414 (0.315)	0.192 (0.265)	0.223 (0.233)
Males	1.099 (0.957)	0.496 (0.670)	0.603 (0.523)
Combined	0.688 (0.722)	0.313 (0.484)	0.375 (0.414)

## Discussion

The results of the study were compared with the findings of Borutta et al. (2002). However, in their research plaque accumulation was scored on a scale from 0 to 5 using the Quigley-Hein Index (QHI), whereas in the present investigation it was scored on a scale from 0 to 3. Thus, for comparison of the average  $\overline{QHI}$  and  $\overline{PI}$ , the latter should be multiplied by 5/3. Borutta et al. (2002) found that for patients using the same type of toothbrush, QHI was from 1.32 to 1.71 (1.33 at the initial check-up), whereas for those using powered toothbrushes it ranged from 0.42 to 0.51. In the present investigation before OHI,  $\overline{PI} = 0.688$ , which corresponded to a it of 1.15. Thus, at the beginning of both studies the degree of plaque formation was similar. However, 1 month after OHI, there was a decrease in  $\overline{PI}$  to 0.375 in the present study, i.e. to a value corresponding to a QHI of 0.52 which is close to that found by Borutta et al. (2002) for patients using powered brushes ( $\overline{QHI} = 0.42$ ). The above finding indicates that correctly performed manual oral hygiene and cleaning with dental floss give similar results to those obtained with a powered toothbrush.

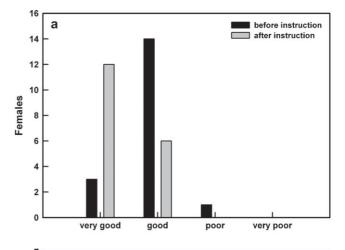
The beneficial result of correct manual tooth cleaning was also supported by analysis of the distribution of the patients into groups with various hygiene status determined before and after OHI. The distribution of patients into groups with very good, good, poor, and very poor oral hygiene, performed on the basis of their individual PI, is shown in Figure 1. It is worth noting that the percentage of female patients with very good oral hygiene increased from 16.7 to 66.7 per cent. For males, the corresponding figures were 25 to 50 per cent, respectively.

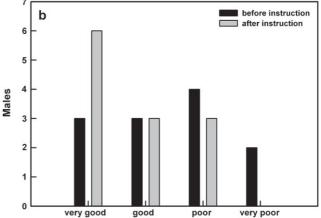
The hypothesis that the distribution of patients into various hygiene status groups does not depend on OHI was verified. Since the calculated value of  $\chi^2 = 10.961$  was higher than  $\chi^2_{a=0.05} = 7.815$ , the hypothesis formulated above had to be rejected, and the converse that the observed improvement in the oral hygiene status following OHI was accepted.

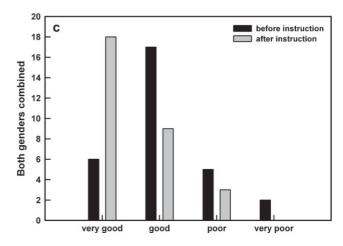
Corbett *et al.* (1981) reported that patients with banded orthodontic appliances had significantly higher levels of *S. mutans* than subjects wearing unbanded appliances. One could expect that OHI should lead to an improvement in oral hygiene and eventually to an increase in the number of patients with acceptable levels of *S. mutans* and *Lactobacillus* spp. However, according to the present findings OHI did not have any beneficial influence on bacterial level, regardless of patient gender or type of bacteria, and the number of patients with an acceptable bacteria level was quite low.

# Conclusions

The findings of this study indicate that in patients with fixed orthodontic appliances, intensive brushing and careful







**Figure 1** Distribution of the number of females (a), males (b), and both genders combined (c) with very good, good, poor, and very poor hygiene before and after oral hygiene instruction.

cleaning with dental floss of the spaces around brackets, under archwires and between the teeth, leads to a significant increase in the level of oral hygiene characterized by the PI. However, these measures were insufficient to decrease *S. mutans* and *Lactobacillus* spp., levels. Thus, patients with fixed appliances have a higher caries risk.

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#### References

- Addy M 1986 Chlorhexidine compared with other locally delivered antimicrobials. A short review. Journal of Clinical Periodontology 13: 957–964
- Axelsson P, Lindhe J 1987 Efficacy of mouthrinses in inhibiting dental plaque and gingivitis in man. Journal of Clinical Periodontology 14: 205–212
- Balenseifien J W, Madonia J V 1970 Study of dental plaque in orthodontic patients. Journal of Dental Research 49: 320–324
- Bloom R H, Brown Jr L R 1964 A study of the effects of orthodontic appliances on the oral microbial flora. Oral Surgery, Oral Medicine, and Oral Pathology 17: 658–667
- Borutta A, Pala E, Fischer T 2002 Effectiveness of a powered toothbrush compared with a manual toothbrush for orthodontic patients with fixed appliances. Journal of Clinical Dentistry 13: 131–137
- Boyar R M, Thylstrup A, Holmen L, Bowden G H 1989 The microflora associated with the development of initial enamel decalcification below orthodontic bands *in vivo* in children living in fluorinated-water area. Journal of Dental Research 68: 1734–1738
- Bratthall D 1980 Selection for prevention of high caries risk group. Journal of Dental Research 59: 2178–2182
- Corbett J A, Brown L R, Keene H J, Horton I M 1981 Comparison of Streptococcus mutans concentrations in non-banded and banded orthodontic patients. Journal of Dental Research 60: 1936–1942
- Diamandi-Kiopioti A, Gusberti F A, Lang N P 1987 Clinical and microbiological effects of fixed orthodontic appliances. Journal of Clinical Periodontology 14: 326–333
- Gold S L 1975 Plaque-control motivation in orthodontic practice. American Journal of Orthodontics 68: 8–14
- Gorelick L, Geiger A M, Gwinnett A J 1982 Incidence of white spot formation after bonding and banding. American Journal of Orthodontics 81:93–98
- Green J R, Margerison D 1978 Statistical treatment of experimental data. Elsevier, Amsterdam, Chapters 8 and 10
- Griesser H J, Chatelier R C, Gegenbach T R, Johnson G, Steele J G 1993 Attachment of human cells on plasma polymers: interactions between surface properties and adhesive glycoproteins. American Chemical Society Polymer Preprints 34: 657–658
- Gwinnett A J, Ceen R F 1979 Plaque distribution on bonded brackets: a scanning microscope study. American Journal of Orthodontics 84: 667–677
- Harrap G J, Best J S 1984 Human oral retention of zinc from mouthwashes containing zinc salts and its relevance to dental plaque control. Archives of Oral Biology 29: 87–91
- Langer R 1995 Polymers for drug delivery and tissue engineering. Annals of Biomedical Engineering 23: 101–111
- Loesche W J 1986 The identification of bacteria associated with periodontal disease and dental caries by enzymatic methods. Oral Microbiology and Immunology 1: 65–70

- Machuca G, Valencia S, Lacalle J R, Machuca C, Bullon P 1997 A clinical assessment of the effectiveness of a mouthwash based trilosan and on Zea mays L used as supplements to brushing. Quintessence International 28: 67–72
- Marcotte H, Lavoie M C 1998 Oral microbial ecology and the role of salivary immunoglobulin A. Microbiology and Molecular Biology Review 63: 71–109
- Mitchell L 1992 Decalcification during orthodontic treatment with fixed appliances—an overview. British Journal of Orthodontics 19: 199–205
- O'Reilly M M, Featherstone J D B 1987 Demineralization and remineralization around orthodontic appliances: an *in vivo* study. American Journal of Orthodontics 92: 33–40
- Ögaard B, Rölla G, Arends J 1988 Orthodontic appliances and enamel demineralization: part 1. Lesion development. American Journal of Orthodontics and Dentofacial Orthopedics. 94: 68–73.
- Röder A, Zühlke A, Widdecke H, Klein J 1993 Synthesis and application of new microcarriers for animal cell culture. Part II: application of polystyrene microcarriers. Journal of Biomaterials Science, Polymer Edition 5: 79–88
- Sandham H J, Brown J, Chan K H, Philips H I, Burgess R C, Stokli A J 1991 Clinical trial in adults of an antimicrobial varnish for reducing *Mutans streptococci*. Journal of Dental Research 70: 1401–1408
- Schaeken M J M, van der Hoeven J S, Hendriks J C M 1989 Effects of vanishes containing chlorhexidine on the human dental plaque flora. Journal of Dental Research 68: 1786–1789
- Schwaninger B, Vickiers-Schwaninger N 1979 Developing an effective oral hygiene program for the orthodontic patient: review, rationale, and recommendations. American Journal of Orthodontics 75: 447–452
- Silness J, Löe H 1964 Periodontal disease in pregnancy. II. Correlation between hygiene and periodontal condition. Acta Odontologica Scandinavica 22: 121–135
- Sukontapatipark W, El-Agroudi M A, Selliseth N J, Thunold K, Selvig K A 2001 Bacterial colonization associated with fixed orthodontic appliances. A scanning electron microscopy study. European Journal of Orthodontics 23: 475–484
- Tamada Y, Ikada Y 1993 Cell adhesion to plasma-treated polymer surfaces. Polymer 34: 2208–2212
- Trombelli L, Scabbia A, Griselli A, Zangari F, Calura G 1995 Clinical evaluation of plaque removal by counterrotation electrical toothbrush in orthodontic patients. Quintessence International 26: 199–202
- Twetman S, Petersson L G 1997 Efficacy of chlorhexidine and a chlorhexidine-fluoride varnish mixture to decrease interdental levels of *Mutans streptococci*. Caries Research 31: 361–365
- Van Lunsen D M, de Soet J J, Weerheijm K L, Groen H J 2000 Effects of dental treatment and single application of a 40% chlorhexidine vanish on *Mutans Streptococci* in young children under intravenous anaesthesia. Caries Research 34: 268–274
- Weitman R T, Eames W B 1975 Plaque accumulation on composite surfaces after various finishing procedures. Journal of the American Dental Association 91: 101–106
- West J L, Hubbell J A 1999 Polymeric biomaterials with degradation sites for proteases involved in cell migration. Macromolecules 32: 241-244
- Zachrisson B U 1974 Oral hygiene for orthodontic patients: current concepts and practical advices. American Journal of Orthodontics 66: 487–497
- Zachrisson B U 1976 Cause and prevention of injuries to teeth and supporting structures during orthodontic treatment. American Journal of Orthodontics 69: 285–300
- Zühlke A, Röder B, Widdecke H, Klein J 1993 Synthesis and application of new microcarriers for animal cell culture. Part I: design of polystyrene based microcarriers. Journal of Biomaterials Science, Polymer Edition 5: 65–78