DOI: 10.1111/j.1365-263X.2009.00988.x

INTERNATIONAL JOURNAL OF PAEDIATRIC DENTISTRY

REVIEW ARTICLE

The effects of orthodontic appliances on *Candida* in the human mouth

KYOKO HIBINO[1], RICKY W. K. WONG[1], URBAN HÄGG[1] & LAKSHMAN P. SAMARANAYAKE[2]

[1]Discipline of Orthodontics, Faculty of Dentistry, and [2]Department of Oral Biosciences, Faculty of Dentistry, The University of Hong Kong, Hong Kong SAR, China

International Journal of Paediatric Dentistry 2009; 19: 301–308

Background. *Candida* is an opportunistic pathogen present in about 50–60% of the healthy human population, and becomes pathogenic when the host immune defence is undermined such as in HIV infection. Adhesion and colonization of the oral cavity by *Candida albicans* is an initial step in candidosis, and the presence of orthodontic and other oral appliances seems to alter the oral ecological environment, hence may tip the balance to favour the candidal presence.

Objective. The purpose of this paper was to review the literature with specific attention to prevalence; intra-oral density of the candidal organisms; and *Candida* carriage status in orthodontic patients before, during, and after treatment.

Conclusions. The limited amount of literature demonstrated that the density of *Candida* increases; the most common *Candida* species isolated in the orthodontic patients was *C. albicans*; and that there seems to be a direct relationship between the presence of a removable appliance, *Candida*, and low salivary pH levels. No healthy patients developed *Candida* infection from the orthodontic appliances. However, there seems to be a trend that some non-*Candida* carriers converted to *Candida* carriers following the insertion of the appliances by unknown mechanism. This may indicate a more cautious approach when providing orthodontic treatments to immunocompromised children concerning the possible increased risk of candidal infection.

Introduction

Candida is an opportunistic pathogen present in about 50–60% of the human population[1]. The infection caused by *Candida*, called candidiasis or candidosis, remains an important clinical problem, especially in the immunocompromised patient population.

Many studies have been conducted to investigate the *Candida* carriage in immunocompromised patients such as those infected with HIV[2] or suffered from diabetes[3]. There are also studies conducted in healthy individuals investigating the relationship between the presence of *Candida* and caries prevalence[4] or periodontal condition[5].

It is shown that orthodontic appliances and other oral devices promote changes in oral microbiota[6,7]. With the increase in the use of immunosuppressive and corticosteroid therapies in recent years, and the AIDS epidemics, dental practitioners may encounter more patients seeking orthodontic treatment who are immunocompromised. Therefore, it is important to know how the orthodontic appliances will affect the oral candidal status as the presence of *Candida* in the oral cavity can lead to infection. There is only a small number of papers related to this topic, and so far there is no comprehensive literature review.

Therefore, the purpose of this paper was to review the literature with specific attention to prevalence; intra-oral density of the candidal organisms; and *Candida* carriage status in orthodontic patients before, during, and after treatment.

Methods

The following electronic databases were searched from their respective inceptions: MEDLINE and Cochrane Library. In addition, the databases in

Correspondence to:
Ricky W. K. Wong, Faculty of Dentistry, The University of Hong Kong, 2/F Prince Philip Dental Hospital, 34 Hospital Road, Hong Kong SAR, China.
E-mail: fyoung@hkucc.hku.hk

Table 1. Summary of literatures.

Author/year	Subjects	Appliances used	Length of study	Sampling techniques	Plaque score
Addy *et al*. 1982	12- to 16-year-old adolescents in UK; 148 subjects	Removable and fixed appliances	Cross section	Imprint culture	Yes
Arendorf and Addy 1985	8- to 17-year-old adolescents in UK; 33 subjects	Removable appliances	9 months	Imprint culture	Yes
Hägg *et al*. 2004	Cohort 50 consecutive cases in Hong Kong; 27 Chinese subjects followed. Mean age 15.5 ± 2.4 years	Fixed appliances	3 months	Oral rinse Pooled plaque Imprint culture	Yes
Brusca *et al*. 2007 (*in vitro* study)	N/A	Fixed appliance brackets: metal, ceramic, and Morelli composite brackets	48 hours (incubation)	N/A	N/A
Arslan *et al*. 2008	72 subjects in Turkey; 42 subjects were *Candida* carriers and were followed. Mean age 19.8 years	Fixed appliances	12 months	Swab	No
Lee *et al*. 2008	Cohort 112 Chinese in Hong Kong; 97 patients followed. Mean age 17.7 years	Fixed appliances	12 months	Oral rinse	No

the library of the University of Hong Kong were searched. The key words and combinations used in searching the databases were 'orthodontic appliances and *Candida* (infection)' and 'orthodontic treatment and *Candida*'.

Fixed and removable orthodontic appliances

Up to date, there are only six papers published in the English language that were written on the effect of orthodontic appliances to the *Candida* present in the mouth (Table 1)[8–13]. Five of the six studies were conducted *in vivo*, and the majority of the subjects were children and adolescents[8–12]. The other paper was conducted *in vitro*, investigating the effect of different bracket materials on adherence of microorganisms including *Candida*[13].

Prevalence of candidal carriers

Candida albicans is an opportunistic pathogen, and oral yeast carriage is found in one-third of the general population according to one major study[14]. However, the published figures on prevalence vary from 25 to 75% attributed to the population sampled and the sensitivity of the sampling technique[15].

It was reported that the presence of a prosthesis or an appliance increased the presence of *Candida* on the teeth and all mucosal sites sampled[6]. Addy *et al*.[8] reported that there was no statistical difference in the prevalence of the *Candida* carriage status, between groups of healthy adolescents who wear no appliance, fixed appliance, and removable appliance, being 46–52%, respectively. However, the prevalence of candidal recovery at some sites and candidal densities at all sites was significantly increased in fixed and removable appliance wearers. It was suggested that the presence of orthodontic appliances might lead to proliferation of *Candida* in the oral carriers, but there was no evidence that the non-carriers of *Candida* could convert to carriers. This, however, was a cross-sectional study and subsequently did not allow to investigate fluctuations which might occur during the long orthodontic treatment which often lasts for a long time.

The first long-term study[9] on patients undergoing orthodontic treatment was done on patients treated with removable appliance. It was reported that the *Candida* carriers before treatment was 39%; after 9 months, it had increased 79%, and after treatment was reduced to 14%. Thirteen of the 33 patients converted from non-carriers to carriers during the treatment, but ten patients converted back to non-carriers after the removal of the appliances. In addition, it was reported that two of the initial *Candida* carriers became non-carriers after the treatment. It was concluded that there was a

direct relationship between the presence of an acrylic appliance and *Candida*, and suggested the appliance transiently initiates the carrier state. This is the only study that had followed the subjects to explore the effects of the appliance insertion and removal in the mouth. Most likely, it is because of the fact that removable appliances can be removed anytime for the microbiological culture, whereas fixed appliances are removed only after treatment is completed.

Hägg *et al.*[10] investigated the prevalence of *Candida* using three different sampling techniques, in a group of adolescents during fixed orthodontic appliance therapy. The prevalence of *Candida* prior to the insertion of the fixed appliances was 30, 7, and 22% for oral rinse, imprint culture, and pooled plaque, respectively, which with the exception of imprint culture technique were similar to the 24% reported earlier[13]. There was considerable individual variation in the candidal counts from the subjects irrespective of the sampling technique. It was demonstrated that candidal carriage did significantly increase (27%) after the insertion of the fixed appliance detected with imprint culture technique only. One of five patients changed from non-carrier to carrier after the insertion of the fixed appliance which might suggest that the fixed appliance transiently initiated the carrier state.

Arslan *et al.*[11] reported similar findings in alterations in the carrier rate of *Candida* spp. during 1 year of fixed appliance treatment among adolescents, a comparatively high prevalence *Candida* carriers (59%). However, the patients who were followed were *Candida* carriers, so they were unable to report on the number of non-carriers converted to carriers after the insertion of the fixed appliances.

In a recent study comprising 112 orthodontic patients (mean age 17.7 years) treated with fixed appliance and with multiple samples obtained during 12 months, 32% carried *Candida* before treatment, and it increased gradually and significantly to 50% at the fifth month and remained on that level for the rest of the observation period, similar to that reported in the study by Arslan *et al.*[11] However, among the orthodontic patients were 11% consistent *Candida* carriers, and 25% consistent non-carriers regardless of the presence of the fixed orthodontic appliance, whereas 14% of the patients converted from being a non-*Candida* carrier to *Candida* carrier during the study period. This indicates that fixed orthodontic appliance might be an initiator for the conversion of the candidal carriage state, but it is not the only factor. It is not understood what the possible differences in the individual's oral environment are which lead to that some healthy subjects are candidal carriers and others are not.

The presence of fixed orthodontic appliance alters the oral environment so that the proliferation of organisms such as *Candida* species might occur. However, in the study conducted by Lee *et al.*[12], the increase in frequency of candidal carriers was not significant and the actual results do not allow concluding that orthodontic treatment neither increases the frequency of candidal carriage in a healthy population nor change candidal non-carrier state into candidal carrier state.

However, the removable and fixed orthodontic appliances may transiently initiate candidal carrier state. Many factors might affect the results, for example: (i) the sample size may not be sufficiently large to show any statistical significance; (ii) if the observation period allows to study the sample before, during, and after treatment, or just at one occasion or a limited period; and (iii) the sampling methods used may not be sensitive enough to detect low density of *Candida*, leading to 'exclusion' of the carriers. Only one study[9] followed-up the orthodontic patients until the completion of the treatment, and that treatment was with a removable appliance, whereas the majority of orthodontic treatments today are provided with fixed orthodontic appliances. Longitudinal prospective studies following a sufficiently large sample of orthodontic patients some time prior, during, and after treatment, using sensitive tests to indentify *Candida* and measure the conditions in oral cavity, are necessary to provide the information needed to explore further the impact of orthodontic appliances have on *Candida*.

Site prevalence

The most frequent site for candidal isolation in non-appliance and fixed appliance wearers[8,9]

was the posterior part of the tongue, whereas for removable appliance wearers, the highest prevalence was the posterior and anterior palatal sites[9]. At all sites sampled, *Candida* was recovered from a greater percentage of subjects wearing fixed or removable orthodontic appliances than those not wearing appliances[8,9]. After the completion of treatment and removal of the orthodontic appliance, the prevalence of *Candida* returned to levels not significantly different from those before treatment. The effect of the removable appliance on the site prevalence of *Candida* may be explained by that the removable appliances protect the *Candida* from the natural and mechanical removal of the saliva and the defensive system. In general, fixed appliances do not cover the mucosa and as expected have the candidal distribution similar to that of non-appliance wearers.

Site density

Studies[8,9,11] have shown that there are considerable individual variation and gross skewness in site counts for all the three groups, but there was an increase in candidal density at all sites in the fixed and removable appliance wearers. This observation is similar to the increase in oral colonization by the *C. albicans* in individuals wearing either full or partial removable dentures[16]. Therefore, the presence of appliances both fixed and removable will increase the density of the *Candida*.

Prevalence of different Candida *species found*

Candida albicans is the predominant *Candida* species found pre- and post-insertion of orthodontic appliances using oral rinse and pooled plaque techniques[10,11], oral swabs, and saliva[11]. Other *Candida* species isolated less frequently were *Candida tropicalis*, *Candida krusei* and *Candida kefyr*[11].

The pH and Candida

A low salivary pH level is associated with increased frequency of *Candida* in the mouth[16–18]. It was suggested that a direct relationship between yeasts and acid production exists[19].

Arendorf and Addy[9] noted a significant fall in salivary pH in the presence of the acrylic removable appliance, and after appliance removal, salivary pH rose significantly ($P < 0.001$) back to a level almost identical to a level pre-treatment. There was a demonstrable association between the fall in salivary pH and the increase in both the frequency and density of candidal colonization. The decrease in candidal counts following the removal of the appliance probably leads to the increase in salivary pH to the pre-insertion levels. They concluded that there is a direct relationship between the presence of a removable appliance, *Candida*, and low salivary pH levels. Unfortunately, none of the studies carried out on fixed appliances measure the salivary pH to confirm this relationship[8,10–12]. Further studies will be needed.

The effect of fixed and removable appliances on oral hygiene of the patients

It is shown that the insertion of an orthodontic appliance into the oral cavity increases the number of plaque retention areas[20,21]. It was also found that the presence of a fixed appliances greatly inhibited oral hygiene and created new retentive areas for plaque and debris, which in turn predisposed to increased carriage of microbes and subsequent infection[22].

Addy *et al.*[8] showed that the total mouth plaque scores for the non-appliance and removable appliance wearers were not significantly different. For the removable appliance wearers, however, the palatal plaque scores were significantly increased, whereas the buccal scores significantly decreased when compared with the respective surfaces in non-appliance wearers. The effects of removable orthodontic appliances on plaque accumulation were similar to the finding for partial denture wearers[23]. It was mainly in an upper removable appliance that leads to an altered distribution of plaque on the teeth with the palatal side covered by the appliances that showed significant increase in plaque accumulation. In non-appliance wearers, the buccal plaque score is higher than the palatal aspects. It may be that the removable appliance protects the plaque from natural and mechanical removal. Similar, but reduced,

effect was noted by Arendorf and Addy[9]. They showed an increase in palatal plaque score during therapy, although not significant. The plaque scores were reduced in other sites and continued to fall after appliance removal. This may be because of the oral hygiene instructions from the dental teams. Therefore, they have suggested that the appliance has an effect on candidal carriage; this would not appear to have related to a decrease in oral hygiene.

Hägg *et al.*[10] also noted the increase in the plaque score with 10% increase after the insertion of fixed appliance ($P < 0.05$), however, only after the second and third visits. This could be caused by the presence of orthodontic attachments on the buccal and lingual surfaces of the teeth, thus the difficulty in brushing the teeth well with the orthodontic attachments on the tooth surfaces.

However, some studies show that there was no significant difference in plaque accumulation between pre-treatment and the insertion of fixed orthodontic appliances[24,25]. It was mentioned that the behavioural factor in maintaining good oral hygiene may be a more important factor. A study conducted in Hong Kong[26] looked at 760 adolescents from a lower-income group. It reported a significant increase in plaque index after insertion of fixed orthodontic appliances. Thus, it was concluded that the presence of an appliance can hinder the patients from maintaining good oral hygiene other than the behavioural factor.

It seems that the presence of both removable and fixed appliances may increase the amount of plaque in the mouth. However, there is no direct evidence to demonstrate that poor oral hygiene leads to candidal carriage as not all studies show the insertion of appliances leads to an increase in plaque accumulation, but there is an increase in the density of *Candida*. Further studies will be needed to explore this relationship.

Different sampling techniques

The wide range of prevalence of *C. albicans* can be caused by the differences in sampling techniques. Each technique has its own limitations and advantages.

In epithelial smears[27], a yeast-like form may be observed, but they need further identification as *C. albicans*. Salivary samples[28–30] detect *Candida* more frequently than oral swabs[31]. However, it did not appear to yield the highest frequency of carriers in the study by Arendorf and Walker[6]. The imprint culture technique[32] allows quantification of candidal organisms on the mucosal surface, so it allows establishing a normal range for the prevalence and density of *C. albicans* at various sites in the subjects[6]. It detects *Candida* inefficiently, but does show the distribution of the organism on the teeth, mucosa, and fitting surface of the denture, when the mouth is heavily colonized[6]. Arendorf and Walker[6] compared the four methods – epithelial smears, salivary samples, oral swabs, and imprint technique – and showed that the imprint cultures showed the increased yield in various sites which merits the adaptation of this procedure as the method of choice for detecting carriers of *C. albicans*. Oral rinse technique is the most sensitive for evaluation of oral yeast and coliform carriage, and imprint technique is the most sensitive for the localization of yeast growth. More variant species can be isolated using the oral rinse technique than the imprint culture or pooled plaque technique[33]. Hägg *et al.*[10] demonstrated the difference in the detection of *Candida* prevalence using these three techniques showing the importance of the sampling methods. Therefore, when interpreting the above studies, it is important to know which technique they have used to interpret the findings.

The isolation of *C. albicans* or other *Candida* species from the oral cavity, in the absence of lesions, does not constitute evidence of clinical candidiasis. It is mentioned that none of the sampling techniques usefully locates or confirms colonization[34]. On the other hand, it has been suggested that the pathogenicity of *C. albicans* depends upon the number of organisms present[35]. Arendorf and Walker[6] found out that imprint culture technique may be useful in discriminating between the carrier state and oral candidosis, as there was an apparent limit to the candidal density in healthy dentate and denture-wearing subjects. Colony counts in excess of 30 colonies per cm^2 of mucosa in the dentate, and 49 colonies per cm^2 of mucosa in denture wearers suggest a *Candida* infection.

Candidal colonization and candidosis

Candida albicans is frequently found in human mouth, but only few carriers develop clinical signs of candidosis. The pathogenesis of *Candida* infections is complex, involving the interaction of yeast and host factors[36].

Oral candidosis results from yeast overgrowth and penetration of the oral tissues when the host's physical and immunological defences have been undermined. The most critical thing that determines whether clearance, colonization, or candidosis is the host's immune competence.

According to Cannon *et al.*[36], the ability of a *Candida* strain to overcome the host clearance mechanisms and to colonize surfaces depends on the effectiveness of adherence mechanisms, the avidity of the yeast adherence, and the yeast growth rate. Without attachment, the growth rate of *C. albicans* is insufficient to maintain carriage in the mouth. Progression from adherent replicating yeast to a mucosal infection depends on adherence and growth rate, but also involves tissue penetration. For an infection to persist, the host immune system must fail to contain the growth of the yeast. The balance among clearance, colonization, or candidosis therefore depends on the ability of *Candida* strains to modulate expression of virulence factors in response to environmental change, combined with competence of the host immune system. Therefore, in the immunocompetent orthodontic patients, the increase in the prevalence of density of *Candida* may not tip the balance to *Candida* infection, but in the presence of other local or systemic factors, the increase in *Candida* may lead to candidosis.

Host factors that reported to be associated with increased oral carriage rates of *Candida* include: xerostomia or reduced saliva flow rate[37], low saliva pH[38], smoking[38], and increased saliva glucose concentration[39]. However, there are conflicting reports on these factors. The immune competence of individuals and/or the presence of other predisposing factors can affect colonization by *C. albicans*[40]. Old age is shown to be one of the factors that is related to the prevalence of candidal carriage[41,42], and that the prevalence of *Candida* species is higher in the immunocompromised patients[2].

Many factors can predispose individuals to oral candidosis[14,43]. Factors that affect host immunity such as AIDS, malignancy, anticancer treatment, and long-term antibiotic therapy are some examples. Ill-fitting denture is a local factor that will also predispose an individual to candidosis. The yeast/host cell interaction is also affected by external factors such as drug treatment. Antibiotic treatment can cause *C. albicans* overgrowth in the oral cavity by eliminating competing microorganisms and exposing additional sites suitable for colonization.

None of the studies showed the subjects developing candidosis. This may indicate that as long as the subjects are medically healthy, there will only be an increase in *Candida* colonization but no candidosis.

Orthodontic bracket materials and Candida *carriage*

Brusca *et al.*[13] found that the adherence of *C. albicans* was increased by the composite bracket, whereas the use of metallic brackets decreased the number of colony-forming units. The adherence was most with composite brackets, followed by ceramic then metallic. The microorganisms had the highest adherence to the esthetic brackets because they find a highly favourable ecological niche in the more porous and less smooth structure of the bracket material. It was reported that *Candida* adheres directly to plastic, forming a fine layer of biofilm on the surface of the synthetic device[44]. How this difference in the adherence to different bracket materials influence the candidal carriage rate and *Candida* density has not been studied and may need further study to investigate this.

The presence of elastomers, metal ligatures, or nickel titanium or steel arch wires and adhesives forms a critical interface because they facilitate microbial adherence. It was, however, found that no differences in microbial adherence when comparing brackets ligated with rubber bands and those ligated with metal ligatures, suggesting that probably ligating materials do not influence the *Candida* population much, and the bracket materials[45] are more important.

Summary

Adhesion and colonization of the oral cavity by *C. albicans* is an initial step in candidosis. It seems that any foreign objects in the mouth, whether they are fixed orthodontic appliances and removable appliances, seem to alter the microbiological environment by providing suitable surfaces for the adherence of *Candida*, inhibiting the patient to maintain good oral hygiene, altering the protection function of the saliva perhaps by reducing its flushing effect, which leads to the increase in the prevalence of *Candida* in the mouth.

Oral hygiene is one of the important factors that could be associated to the prevalence of *Candida*. However, no study has shown clear relationship between them.

Up to date, there is a limited number of studies investigating the effect of orthodontic appliances on *Candida* carriage status and *Candida* density in the oral cavity. Long-term studies will be required to confirm the trend that has been observed. In addition, no studies are available on the effect of the long-term wear of removable retainers and the full-time use of functional appliances. These appliances, especially functional appliances, cover a large area of mucosa with acrylic for full-time for at least 6–12 months. This may affect the microbiological environment and lead to changes in the *Candida* prevalence and density. Further research should be conducted to investigate this possibility.

There seems to be some susceptible patients who changed from non-*Candida* carriers to *Candida* carrier status after the wearing of the orthodontic appliances. The exact explanation for this conversion is still unclear, and similarly it is unknown why certain subjects do not carry *Candida* in their mouth despite the presence of orthodontic appliances. It will be of our interest to understand why certain patients converted from non-*Candida* carriers to *Candida* carriers or did not convert to *Candida* carriers so that we can try to minimize the possibility of the patients mainly the immunocompromised patients to harbour more *Candida*.

Finally, the increase in colonization of *Candida* in these orthodontic patients does not mean that they will develop candidosis, but there will be an increase risk for infection especially if their immune defence has been undermined by some factors such as antibiotic usage and local trauma from the appliances. A more cautious approach when providing orthodontic treatments to immunocompromised children concerning the possible increased risk of candidal infection should be taken.

What this paper adds

- This paper has provided a summary of all the literature available on the effects of orthodontic appliances to the *Candida* in the mouth.
- It has shown that the orthodontic appliances alter the microbiological environment that leads to the increase in the prevalence of *Candida* in the mouth, and some susceptible patients may change from non-*Candida* carriers to *Candida* carrier status.

Why this paper is important to paediatric dentists

- It is important for us to know the microbiological effects from the insertion of the appliances, and can take a more cautious approach when providing orthodontic treatments to immunocompromised children concerning the possible increased risk of candidal infection.

References

1 Samaranayake LP, MacFarlane TW. An *in vitro* study of the adherence of *Candida albicans* to acrylic surfaces. *Arch Oral Biol* 1980; **25**: 603–609.
2 Lin AL, Johnston DA, Patterson TF, *et al*. Salivary anticandidal acitivity and saliva composition in an HIV-infected cohort. *Oral Microbiol Immunol* 2001; **16**: 270–278.
3 Karjalainen KM, Kunnuttila ML, Kaar ML. Salivary factors in children and adolescents with insulin-dependent diabetes mellitus. *Pediatr Dent* 1996; **18**: 306–311.
4 Uygun-Can B, Kadir T, Akyuz S. Oral candidal carriage in children with and without dental caries. *Quintessence Int* 2007; **38**: 45–49.
5 Urzua B, Hermosilla G, Gamonal J, *et al*. Yeast diversity in the oral microbiota of subjects with periodontitis: *Candida albicans* and *Candida dubliniensis* colonize the periodontal pockets. *Med Mycol* 2008; **8**: 1–11.
6 Arendorf TM, Walker DM. Oral candidal populations in health and disease. *Br Dent J* 1979; **147**: 267–272.
7 Traoré O, Springthorpe VS, Sattar SA. A quantitative study of the survival of two species of *Candida* on porous and non-porous environmental surfaces and hands. *J Appl Microbiol* 2002; **92**: 549–555.
8 Addy M, Shaw WC, Hansford P, Hopkins M. The effect of orthodontic appliances on the distribution of *Candida* and plaque in adolescents. *Br J Orthod* 1982; **9**: 158–163.

9 Arendorf TM, Addy M. Candidal carriage and plaque distribution before, during and after removable orthodontic appliance therapy. *J Clin Periodontol* 1985; **12**: 360–368.

10 Hägg U, Kaveewatcharanont P, Samaranayake YH, Samaranayake LP. The effect of fixed orthodontic appliances on the oral carriage of *Candida* species and Enterobacteriaceae. *Eur J Orthod* 2004; **26**: 623–629.

11 Arslan SG, Akpolat N, Kama JD, Ozer T, Hamamci O. One-year follow-up of the effect of fixed orthodontic treatment on colonization by oral *Candida*. *J Oral Pathol Med* 2008; **37**: 26–29.

12 Lee W, Low BKM, Samaranayake LP, Hagg U. Genotypic variation of *Candida albicans* during orthodontic therapy. *Front Biosci* 2008; **13**: 3814–3824.

13 Brusca MI, Chara O, Sterin-Borda L, Rosa AC. Influence of different orthodontic brackets on adherence of microorganisms *in vitro*. *Angle Orthod* 2007; **77**: 331–336.

14 Samaranayake LP, MacFarlane TW, eds. *Oral Candidosis*. London: Wright, 1990.

15 Odd FC. *Candida and Candidosis*, 2nd edn. London: Ballière-Tindall, 1988.

16 Shipman B. Clinical evaluation of oral *Candida* in cancer chemotherapy patients. *J Prosthet Dent* 1979; **41**: 63–67.

17 Parvinen T, Larmas M. The relation of stimulated salivary flow rate and pH to *Lactobacillus* and yeast concentrations in saliva. *J Dent Res* 1981; **60**: 1929–1935.

18 Kullaa-Mikkonen A, Kotilainen R. The prevalence of oral carriers of *Candida* in patients with tongue abnormalities. *J Dent* 1983; **11**: 313–317.

19 Fosdick LS, Hansen HL. Theoretical considerations of carbohydrate degradation in relation to dental caries. *J Am Dent Assoc* 1936; **23**: 401–407.

20 Cannon RD, Chaffin WL. Oral colonization by *Candida albicans*. *Crit Rev Oral Biol Med* 1999; **10**: 359–383.

21 Calderone R, Suzuki S, Cannon R, Cho T. *Candida albicans* adherence, signaling and virulence. *Med Mycol* 2008; **38**: 125–137.

22 Atack NE, Sandy JR, Addy M. Periodontal and microbiological changes associated with the placement of orthodontic appliances. *Rev J Periodontol* 1996; **67**: 78–85.

23 Addy M, Bates JF. Plaque accumulation following the wearing of different types of removable partial dentures. *J Oral Rehabil* 1979; **6**: 111–117.

24 Sinclair PM, Berry CW, Bennett CL, Israeison H. Changes in gingiva and gingival flora with bonding and banding. *Angle Orthod* 1987; **57**: 271–278.

25 Davies TM, Shaw MC, Worthington HV, Addy M, Dummer P, Kingdon A. The effect of orthodontic treatment on plaque and gingivitis. *Am J Orthod Dentofacial Orthop* 199; **99**: 155–161.

26 Kwan E. *Oral Health Status of 13 and 15 Year-old Secondary School in Hong Kong*. Hong Kong: University of Hong Kong, 1992.

27 Budtz-Jørgensen E. The significance of *Candida albicans* in denture stomatitis. *Scand J Dent Res* 1974; **82**: 151–190.

28 Lehner T. Immunofluorescent investigation of *Candida albicans* antibodies in human saliva. *Arch Oral Biol* 1965; **10**: 975–980.

29 Lehner T. Immunofluorescence study of *Candida albicans* in candidiasis, carriers and controls. *J Pathol Bacteriol* 1966; **91**: 97–104.

30 Sharon A, Berdicevsky I, Ben-Aryeh H, Gutman D. The effect of chlorhexidine mouth rinses on oral *Candida* in a group of leukemic patients. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 1977; **44**: 201–205.

31 Lilienthal B. Studies of the flora of the mouth. III. Yeast-like organisms: some observations on their incidence in the mouth. *Aust J Exp Biol Med Sci* 1950; **28**: 279–286.

32 Davenport JC. The oral distribution of *Candida* in denture stomatitis. *Br Dent J* 1970; **129**: 151–156.

33 Samaranayake LP, MacFarlane TW, Lamey PJ, Ferguson MM. A comparison of oral rinse and imprint sampling techniques for the detection of yeast, coliform and *Staphylococcus aureus* carriage in the oral cavity. *J Oral Pathol Med* 1986; **15**: 386–388.

34 Olsen I, Stenderup A. Clinical–mycologic diagnosis of oral yeast infections. *Acta Odontol Scand* 1990; **48**: 11–18.

35 Campbell PJ, Heseltine WW. An apparent growth stimulant for *Candida albicans* released from tetracycline-treated bacterial flora. *J Hyg* 1960; **58**: 95.

36 Cannon RD, Holmes AR, Mason AB, Monk BC. Oral *Candida*: clearance, colonization, or candidiasis? *J Dent Res* 1995; **74**: 1152–1161.

37 Parvinen T, Larmas M. The relation of stimulated salivary flow rate and pH to *Lactobacillus* and yeast concentrations in saliva. *J Dent Res* 1981; **60**: 1929–1935.

38 Arendorf TM, Walker DM. The prevalence and intra-oral distribution of *Candida albicans* in man. *Arch Oral Biol* 1980; **25**: 1–10.

39 Knight L, Fletcher J. Growth of *Candida albicans* in saliva: stimulation by glucose assocated with antibiotics, corticosteroids, and diabetes mellitus. *J Infect Dis* 1971; **123**: 371–377.

40 Hauman CHJ, Thompson IOC, Theunissen F, Wofaardt P. Oral carriage of *Candida* in healthy and HIV-seropositive persons. *Oral Surg Oral Med Oral Pathol* 1993; **76**: 570–572.

41 Sugimoto J, Kanehira T, Mizugai H, Chiba I, Morita M. Relationship between salivary histatin 5 levels and *Candida* CFU counts in healthy elderly. *Gerodontology* 2006; **23**: 164–169.

42 Shimizu C, Kuriyama T, Williams DW, *et al*. Association of oral yeast carriage with specific host factors and altered mouth sensation. *Oral Surg Oral Med Oral Pathol* 2008; **105**: 445–451.

43 Scully C, El-Kabir M, Samaranayake LP. *Candida* and oral candidosis: a review. *Crit Rev Oral Biol Med* 1994; **5**: 125–157.

44 Tronchin G, Bouchara JP, Robert R, Senet JM. Adherence of *Candida albicans* germ tubes to plastic: ultrastructural and molecular studies of fibrillar adhesins. *Infect Immun* 1988; **56**: 1987–1993.

45 Sukontapatipark W, el-Agroudi MA, Selliseth NJ, Thunold K, Selvig KA. Bacterial colonization associated with fixed orthodontic appliances. A scanning electron microscopy study. *Eur J Orthod* 2001; **23**: 475–484.