

# Early colonization of dental implants by putative periodontal pathogens in partially edentulous patients

van Winkelhoff AJ, Goené RJ, Benschop C, Folmer T. Early colonization of dental implants by putative periodontal pathogens in partially edentulous patients.  
Clin Oral Impl Res 2000; 11: 511–520. © Munksgaard 2000.

There is limited scientific information available on the early colonization of the peri-implant pockets in partially edentulous individuals. Knowledge about this process is one step in better understanding the etiology and pathogenesis of peri-implantitis. In this study, the early colonization of the peri-implant pockets by putative periodontal pathogens was studied in 20 partially edentulous individuals using anaerobic culture techniques. At baseline, the presence and levels of putative periodontal pathogens in the microflora of periodontal pockets and saliva were established. Immediately after loading of the titanium implants and after 6 and 12 months the presence and levels of selected putative periodontal pathogens were determined in periodontal and peri-implant pockets. A second aim was to detect bacterial contamination of the implant site and the inside of the implant. At baseline, the most frequently isolated species from the periodontal pockets were *Fusobacterium nucleatum*, *Prevotella intermedia* and *Peptostreptococcus micros*. *Bacteroides forsythus*, *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis* were isolated from 9, 2 and 3 patients respectively. Six months after placing of the bridges, the majority of the implant sites had detectable levels of most periodontal bacterial species with the exception of *A. actinomycetemcomitans* which could not be isolated from any of the peri-implant samples during the experimental period, although 2 patients had this organism at baseline. In 2 patients with detectable subgingival *P. gingivalis* at baseline this species was found after 12 months in the peri-implant sites. One of these patients lost 2 implants which was associated with a high proportion of *P. gingivalis* in the peri-implant pockets. A second patient developed 2 fistulas around 2 implants at 8 months and this event was also associated with the presence of *P. gingivalis*. It is concluded that proper periodontal infection control before instalment of dental implants in partially edentulous patients may prevent early bacterial complications.

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Key words: dental implants – bacterial colonization – *Porphyromonas gingivalis*

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Accepted for publication 28 October 1999

Osseointegrated dental implants to replace lost teeth in edentulous and partially edentulous patients has become a predictable treatment modality in restorative dentistry. Favourable long-term results of dental implant systems have been reported (Albrektsson 1988). However, implant failure may occur (Esposito et al. 1998; Tanner et al. 1997). Breakdown of peri-implant tissues can occur due to mechanical stress or peri-implant infection ulti-

mately leading to loss of the implant (Rams et al. 1984; Mombelli et al. 1987; Alcoforado et al. 1991; Rosenberg et al. 1991).

Early bacterial colonization of peri-implant pockets in edentulous subjects is characterized by an increase of facultative anaerobic streptococci, whereas Gram negative strict anaerobic rods are usually isolated infrequently in low proportions (Mombelli et al. 1988). Long-term results on

colonization of the peri-implant area showed a decrease in the proportions of facultative streptococci and an increase in the percentage of Gram positive facultative rods and Gram negative strict anaerobic rods, e.g. *Fusobacterium* spp. and *Prevotella* spp. (Mombelli & Mericske-Stern 1990). Peri-implant infection in edentulous subjects is associated with bacteria that are found in adult periodontitis, however, with the exception of *Porphyromonas gingivalis* and *Actinobacillus actinomycescomitans* (Mombelli et al. 1987). This phenomenon can be explained by observations of Danser et al. (1994) who were no longer able to detect these periodontal pathogens on mucosal sites after full-mouth extraction in patients with severe periodontitis. In addition, *P. gingivalis* and *A. actinomycescomitans* could not be isolated from the peri-implant pockets of edentulous patients with a history of periodontitis (Danser et al. 1995). The composition of the peri-implant microflora seems to be related to the presence of pockets around natural teeth that seem to be an obligate ecological niche for some oral pathogens. Indeed, Apse et al. (1989) found a higher percentage of dark-pigmented anaerobic rods and wet spreaders around dental implants in partially edentulous individuals in comparison to edentulous subjects. Quirynen et al. (1996), using differential phase contrast microscopy, found that the subgingival microflora around implants harbored more spirochetes and motile rods when teeth were present in the same jaw. They found the highest proportions of spirochetes and motile rods in deepened pockets around implants in patients with refractory periodontitis. Papaioannou et al. (1996), applying phase contrast microscopy and DNA probes, investigated the prevalence of putative periodontal pathogens in severe periodontitis patients with dental implants and found similar microbiological profiles around teeth and dental implants with equal pocket depths confirming the hypothesis that pockets around teeth can act as a reservoir for putative periodontal pathogens.

Besides dark-pigmented Gram-negative anaerobic rods, other bacterial species that have been associated with peri-implantitis include the Gram negative *Bacteroides forsythus*, *Fusobacterium nucleatum*, *Campylobacter gracilis* and the Gram positive *Peptostreptococcus micros* and *Streptococcus intermedius* (Tanner et al. 1997).

Information on the early colonization of the peri-implant pocket in partially edentulous patients with a history of periodontitis is scarce. More specifically, the role of *P. gingivalis* in peri-implant infections is poorly documented and this periodontal pathogen is only occasionally described in peri-implant infection (Becker et al.

1990; Papaioannou et al. 1996; Rosenberg et al. 1991). Very little longitudinal information is available on the colonization of dental implants in partially edentulous patients.

The aim of this study was to study the early peri-implant colonization in partially edentulous patients. A second aim of the study was to monitor the prevalence and proportions of putative periodontal pathogens pre-operatively and during the operative phase.

## Material and methods

### Patients

For this study, 20 patients were recruited from the Clinic of Periodontology and Implantology Amsterdam and the Clinic of Periodontology and Implantology, Hoofddorp, The Netherlands. All participants were no older than 65 years of age without clinical signs of caries or periodontitis and candidate for one or two 2–3 unit bridges supported by two implants in the premolar and/or molar regions in the upper or lower jaw. A period of 9–12 months had to be passed since the loss of the teeth. Exclusion criteria were: alcohol or drug abuse, health conditions not permitting surgical procedures, psychiatric disorders, patients that had previously failing implants in the selected insertion locations, replacement of non-integrated implants, grafts of any kind in the future implant sites, disorders in the planned implant site such as tumors, chronic bone disease or previous irradiation. Patients participated in the study on the basis of informed consent.

### Clinical protocol

Pre-operative radiographic examination including orthopantomogram and intra-oral radiographs were taken from the future implant sites of the patients that matched the inclusion criteria. The implants used in this study were 5 mm Ø (Brånemark System<sup>®</sup>, NobelBiocare AB). According to the manufacturer's recommendation antibiotic prophylaxis was administered and included 3 gram of amoxicillin 1 h prior to implant placement (Dent et al. 1997). The surgical procedures were carried out according to the Brånemark System<sup>®</sup> protocol (Adell et al. 1981). After a healing period of 6 months, abutments (EsthetiCone<sup>®</sup>) were connected.

Finally, bridges were placed approximately 4 weeks after abutment connection. The total number of upper and lower bridges was 9 and 11 respectively. The status of the gingival tissues at 1, 6 and 12 months was monitored according to Mühlemann & Son (1971) and recorded as no

symptoms (0) or symptoms of inflammation (1) at four sites of each implant. Parameters for successful and failing implants were based on proposed criteria according to Albrektsson et al. (1986). Radiographs were taken at baseline and at 12 months after loading of the implants.

#### Microbiological sampling

Samples for microbiological testing were obtained pre-operatively, during surgical treatment and at three time-points after loading of the implants (Table 1). Pre-operative microbiological testing involved a saliva sample (see below) and a pooled paper point (Fine, West Palm beach, USA) sample from the deepest pocket in each quadrant of the dentition using two sterile paper points per site (Mombelli et al. 1991). At these sites the plaque index (0, 1, 2), the bleeding index (0, 1, 2) and the probing pocket depth was determined (Winkel et al. 1997).

Approximately 1.5 ml unstimulated saliva was collected in 2.0 ml of reduced transport fluid at three different time points. Immediately prior to implant placement, the prepared site was sampled with two sterile paper points. Prior to the abutment connection, the fixture site was isolated with gauze. After the incision was made, care was taken to avoid contamination of the inner part of the implant using sterile gauzes at the lingual and buccal sites and by constant suction. Immediately after removal of the cover screw, the inner part of the implant was sampled with two paper points. After placement of the suprastructures, approximately 1 month later, the four same selected periodontal pockets and two peri-implant pockets were sampled with two paper points at each site. This procedure was repeated at 6 and 12 months. Samples were stored at 4°C and processed within 4 h.

#### Microbiological procedures

Samples were vortexed for 30 s and 10-fold serially dilutions were prepared in RTF; 100 µl of each di-

lution was plated on non-selective 5% horse blood agar plates (Oxoid no. 2, Oxoid Ltd., Basingstoke, England) supplemented with haemin (5 mg/l) and menadione (1 mg/l) for determination of the total anaerobic bacterial counts and determination of specific periodontal pathogens (see below). Samples were also plated on trypticase soy serum-bacitracin-vancomycin plates (TSBV, Slots 1982) for isolation and counting of *Actinobacillus actinomycetemcomitans*. TSBV plates were incubated in air with 5% CO<sub>2</sub> at 37°C for 3 days, blood agar plates were incubated for 14 days at 37°C in 80% N<sub>2</sub>, 10% CO<sub>2</sub> and 10% H<sub>2</sub>. Presence and numbers of the putative periodontal pathogens *Porphyromonas gingivalis*, *Prevotella intermedia*, *Bacteroides forsythus*, *Fusobacterium nucleatum*, *Peptostreptococcus micros* and *Campylobacter rectus* were determined on the anaerobic blood agar plates. Identifications of these bacterial species were based on Gram stain and cell morphology, aerotolerance, production of catalase and on a number of biochemical reactions (Rapid ID 32A, BioMerieux, SA, France; van Winkelhoff et al. 1988; Winkel et al. 1997).

## Results

### Clinical findings

The mean age of the 20 patients participating in the study was 52.8(±8.4) years and the group consisted of 13 males and 7 females, 6 patients were current smokers. All selected participants concluded the study. The four selected periodontal sites had a mean probing pocket depth of 4.6 (±1.2) mm, a mean bleeding index of 0.64 (±0.47) and a mean plaque index of 0.37 (±0.54). No significant changes in these parameters were observed during the experimental period. The mean gingival index at the implant sites at 6 and 12 months was low and amounted 0.33 and 0.26 respect.

In 18 of the 20 patients the treatment was uneventful. None of the implants showed spontaneous soft tissue penetration at the time of abutment connection. In 2 patients, complications occurred during the experimental period. In patient

Table 1. Time points on which sampling for microbiological testing was performed

	Pre-operative phase	Surgical phase		Post-operative phase		
		Implant placement	Abutment connection	0 months	6 months	12 months
Pockets	+			+	+	+
Saliva	+	+	+			
Implant site		+				
Inside of the implant			+			
Abutment				+	+	+

#13, mobility of the two implants was noticed at 12 months indicating loss of osseointegration. Both implants, located in the left upper molar region were lost at 13 months. At baseline, this patient had 50% of *P. gingivalis* in the periodontal pocket sample. This species was first detected in the peri-implant pockets at 6 and 12 months after loading. There was no occlusal overload articulation and disclusion was over the canines. Comparison between the X-rays taken at the moment when the

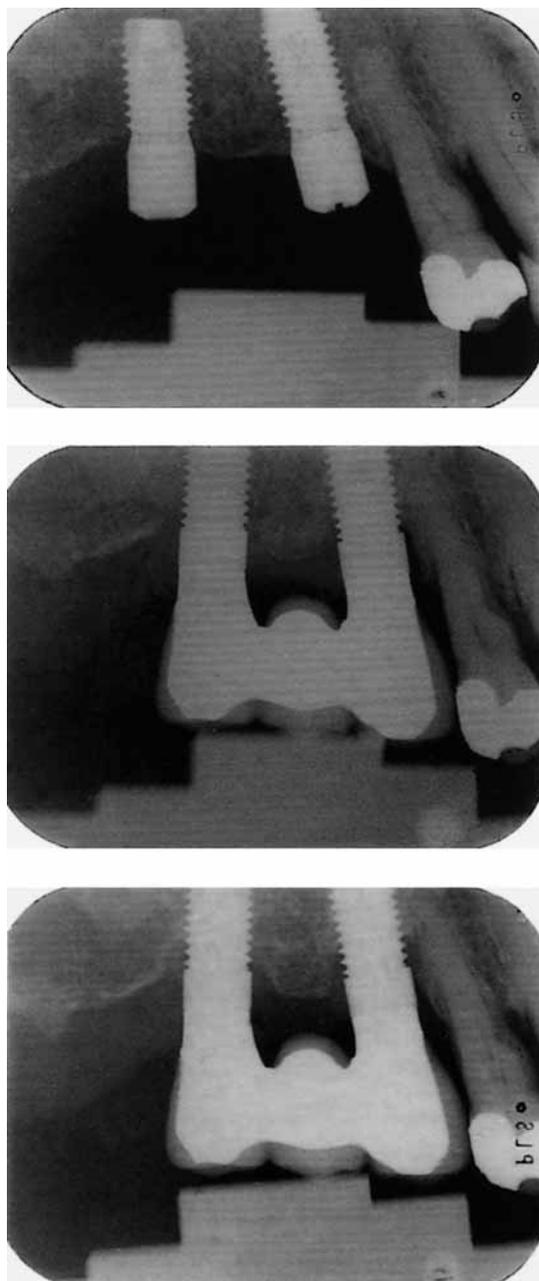


Fig. 1. X-rays of two implants in patient #8 after connection of the healing abutments (A), after connection of the bridge (B) and 8 months after loading (C). Bone loss is evident around both implants at 8 months.



Fig. 2. Two fistulas present at the vestibular side of two implants in the upper jaw visualized by two paper points in patient #8.

A

bridge was placed and shortly before the implants were removed showed no clear differences.

B

The second complication occurred in patient #8. Alveolar bone loss was noticed around two implants located at position 15/16, 10 months after loading of the implants (Fig. 1). Two fistulas vestibular at these locations had developed (Fig. 2). Microbiological testing of the periodontal pockets at that time point revealed a low percentage (0.08%) of *P. gingivalis*. In the 5–6 mm deep peri-implant pockets 14% *P. gingivalis* was found. Samples from the fistulas did not reveal detectable *P. gingivalis*. Through surgical access, granulation tissue was removed and the implant surface was rinsed with chlorhexidine (0.2%) and sterile saline. After this treatment the patient was prescribed metronidazole (500 mg TID; Winkel et al. 1997). This therapy was effective in controlling the infection and in halting bone loss. No further pathological event occurred in this patient during the course of the study.

C

Microbiological findings

Table 2 summarizes the prevalence and occurrence of selected putative periodontal pathogens in pockets and saliva at baseline. *Fusobacterium nu-*

Table 2. Number of patients with detectable putative periodontal pathogens in pockets and saliva at baseline

Bacterial species	Pockets	Saliva	Both samples positive
Aa	2	3	2
<i>P. gingivalis</i>	3	0	0
<i>P. intermedia</i>	15	9	9
<i>B. forsythus</i>	9	5	3
<i>P. micros</i>	10	4	1
<i>F. nucleatum</i>	18	18	17

## Colonization of implants in partially edentulous patients

Table 3. Detection of selected periodontal bacteria in the implant site and saliva during the implant placement

Bacterial species	Implant site	Saliva fixture placement	Both samples positive
Aa	0	3	0
<i>P. gingivalis</i>	0	0	0
<i>P. intermedia</i>	1	5	0
<i>B. forsythus</i>	0	0	0
<i>P. micros</i>	0	4	0
<i>F. nucleatum</i>	2	13	2

Table 4. Frequency of detection of periodontal bacteria in the inside of the implant and the saliva at the abutment operation

Bacterial species	Inside implant	Saliva abutment placement	Both positive
Aa	0	0	0
<i>P. gingivalis</i>	0	2	0
<i>P. intermedia</i>	2	5	1
<i>B. forsythus</i>	2	3	1
<i>P. micros</i>	8	7	2
<i>F. nucleatum</i>	9	15	4

*cleatum* and *Prevotella intermedia* were the most frequently isolated bacterial species in the periodontal pocket samples. *P. gingivalis* and *A. actinomycetemcomitans* were found in 3 patients. The relationship between the subgingival presence and isolation from the saliva was rather poor for most test species, with the exception of *F. nucleatum*, which was found in both samples in 17 of the 18 positive patients. Table 3 summarizes the microbiological findings of the implant sites and saliva during the implant placement. In three occasions bacteria were detected in the implant site and involved *P. intermedia* or *F. nucleatum*. The total number of salivary bacteria at baseline and during

the fixture placement, i.e. after the prophylactic use of amoxicillin, was not statistically significantly different. Table 4 summarizes the microbiological findings of the sampling of the inside of the implant during the abutment operation. In 9 patients, samples from the inside of the implants revealed detectable numbers of bacteria. The most frequently isolated bacterial species from the inside of the implants were *F. nucleatum* and *P. micros*, 9 and 8 patients respectively; *P. intermedia* and *B. forsythus* were both isolated from 2 implants. At baseline and at the fixture operation, *A. actinomycetemcomitans* and *P. gingivalis* were detected in saliva samples in three and zero patients respectively. In contrast, at the abutment operation, *A. actinomycetemcomitans* was not found in any of the patients whereas *P. gingivalis* was isolated from two saliva samples.

In Table 5, the isolation of *P. gingivalis* and *A. actinomycetemcomitans* at baseline and at several time points after loading of the implants is summarized. In patient 1, *P. gingivalis* was isolated from the periodontal pockets at baseline but was no longer found during the rest of the experimental period. In patients 11, 15 and 17 the organism was detected once in the periodontal pockets at different time points, but not in the peri-implant pockets. In patients 14 and 18, *P. gingivalis* was isolated both in the periodontal and the peri-implant pockets at 12 months. In patient 13 *P. gingivalis* was found at baseline in the periodontal pockets and in the peri-implant pockets at 6 and 12 months. *A. actinomycetemcomitans* was isolated from the periodontal pockets from 2 patients but was not recovered from the peri-implant sites at any time point.

In Table 6, the isolation frequency of periodontal bacteria, other than *A. actinomycetem-*

Table 5. Subgingival occurrence of *A. actinomycetemcomitans* (Aa) and *P. gingivalis* at baseline and in the peri-implant pockets at 1, 6 and 12 months after loading of the implants

Species	Patient number	Baseline	1 month		6 months		12 months	
		Pockets	Pockets	Abutment	Pockets	Abutment	Pockets	Abutment
AA	9	0.005	0	0	0	0	0	0
	17	0.7	0	0	0	0	0	0
<i>P. gingivalis</i>	1	3.5*	0	0	0	0	0	0
	2	0	0	0.9	0	0	0	0
	8	0	0	0	0	9.1	0	0
	11	0	2.4	0	0	0	0	0
	13	50.0	0	0	0	2.0	0	11.0
	14	0	6.4	0	1.6	0	7.1	1.5
	15	0	0	0	1.1	0	0	0
	16	0	0.9	0	0	18.0	0	0
	17	0	0	0	51.3	0	0	0
	18	4.2	0	0	0	0	11	14.0

\*, mean proportion of anaerobic counts.

Table 6. Number of patients positive for selected periodontal bacteria pre-operative and 0, 6 and 12 months after loading of the implants

	Pre-operative Pockets	1 month		6 months		12 months	
		Pockets	Abutment	Pockets	Abutment	Pockets	Abutment
Pi	<b>15*</b>	<b>14</b>	<b>12</b>	<b>11</b>	<b>12</b>	<b>13</b>	<b>13</b>
mean %	3.1	2.3	4.2	5.0	5.4	10.5	4.8
SD	3.0	2.9	5.5	5.6	5.0	20.8	7.9
Bf	<b>9</b>	<b>13</b>	<b>4</b>	<b>11</b>	<b>11</b>	<b>17</b>	<b>16</b>
mean %	8.0	3.1	2.1	6.3	4.7	6.3	3.7
SD	7.5	2.3	1.3	5.4	3.7	6.1	2.8
Pm	<b>10</b>	<b>18</b>	<b>18</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>20</b>
mean %	9.4	6.3	8.5	10.1	9.5	5.9	7.9
SD	8.1	6.4	7.6	13.3	9.2	6.0	6.5
Fn	<b>18</b>	<b>20</b>	<b>17</b>	<b>19</b>	<b>18</b>	<b>20</b>	<b>20</b>
mean %	9.3	6.3	6.7	5.9	4.8	7.3	7.0
SD	6.9	7.9	8.3	7.0	5.2	7.6	5.9

\*, number of patients with detectable organism; Pi, *P. intermedia*; Bf, *B. forsythus*; Pm, *P. micros*; Fn, *F. nucleatum*. Figures in bold denote the number of patients positive for the given microorganism. Mean %: mean proportion of the anaerobic cultivable microflora. SD: standard deviation.

*comitans* and *P. gingivalis*, is summarized. At 1 month, the percentage of implant sites with detectable *P. intermedia*, *P. micros* and *F. nucleatum* were 60, 90 and 85% resp. The prevalence of *B. forsythus* in the peri-implant pocket at 1 month was low (4/20, 20%), but increased to 16/20 (80%) at 12 months. Also, the number of patients with detectable *B. forsythus* in the periodontal pockets increased from 9/20 (45%) to 17/20 (85%).

At 6 months, the prevalences of the target organisms in the periodontal pockets and the peri-implant pocket showed great similarity indicating that after 6 months equilibrium had been established.

## Discussion

Previous studies have clearly indicated that the dental microflora can be an important source of bacteria colonizing dental implants (Apse et al. 1989; Papaioannou et al. 1996; Quirynen & Listgarten 1990; Quirynen et al. 1996). In this study we have longitudinally investigated the bacterial colonization of titanium dental implants in partially edentulous individuals. At 1 month, the majority of implant sites had detectable levels of *P. intermedia*, *P. micros* and *F. nucleatum* indicating that these commensal oral bacteria are early colonizers of the peri-implant pockets in partially edentulous patients. In contrast, only after 6 months 11 of 20 patients had detectable *B. forsythus*. We also observed an increase in patients with *B. forsythus* positive periodontal pockets. This may reflect a true biological phenomenon or may be the result of the microbiological techniques used in this study. Our observations are different from those reported by Mombelli et al. (1988) studying the colonization

of osseointegrated titanium implants in 5 edentulous patients. At 6 months, they found *P. intermedia* and *F. nucleatum* in one (20%) and 3 (60%) patients resp., whereas in the present study these organisms were isolated from 55% and 95% of the patients. In addition, *P. intermedia* was found in the present study in high frequency (14 of 20 patients) already 1 month after loading of the implants whereas Mombelli et al. (1988) were able to isolate *P. intermedia* only after approximately 4 months. These data indicate that colonization of the implant sulcus by strict anaerobic gram negative rods is different in partially edentulous patients in comparison to fully edentulous patients. One other possible important difference was the frequent detection of *P. gingivalis* in the present study. A few authors have shown that only dentate patients may have detectable *P. gingivalis* in the peri-implant pockets. Koka et al. (1993) studied the early colonization of Brånemark dental implants in 4 partially edentulous patients and, using a slot immunoblot assay, reported colonization *P. gingivalis*, *P. intermedia* and *F. nucleatum* within 28 days after second-stage surgery. In the present study we confirm their observation, 1 patient had detectable *P. gingivalis* 1 month after loading of the implant. In a 3-year study, Leonardt et al. (1993) found similar bacterial profiles, including *A. actinomycetemcomitans* and *P. gingivalis*, in periodontal and titanium implant pockets 6 months after abutment connection in 19 partially edentulous patients. At abutment connection 2 implants were considered failures due to lack of osseointegration. At the time of bridge connection, 25% of the patients had detectable *P. gingivalis* and 18.8% of the patients had detectable *A. actino-*

*mycetemcomitans* in the peri-implant pockets. However, no measurable loss of implant-supporting tissues was noted in these patients over a period of 3 years. The observations of Leonard et al. (1993) show that the mere presence of small numbers of these periodontal pathogens does not necessarily lead to peri-implant infection or implant failure. In the present study, *A. actinomycetemcomitans* was not found in the peri-implant pockets despite the detection of this organism in the periodontal pockets and in saliva of 2 patients. Other studies have associated *P. gingivalis* and *A. actinomycetemcomitans* with peri-implantitis lesions (Becker et al. 1990; Rosenberg et al. 1991; Tanner et al. 1997; van Winkelhoff & Wolf 2000).

In the present study, 3 of 20 patients (15%) had detectable *P. gingivalis* 12 months after loading of the implants. One of these patients (#13) lost 2 implants within 12 months after abutment connection. Mobility of both implants was the most obvious clinical sign of loss of osseointegration. No clinical evidence for overloading of the implants could be noted. The X-ray taken prior to removal of both implants did not reveal a clear bone loss around the implants, although osseointegration was clearly lost. These findings do not prove that peri-implant infection with *P. gingivalis* was the sole reason for implant loss in this patient. It is possible that a combination of mechanical factors and the presence and level of *P. gingivalis* has led to loss of the previous established osseointegration. A striking observation in patient #13 was the presence of several periodontal bacteria in the inside of the implant from which *P. intermedia* (3.6%), *B. forsythus* (3.6%), *P. micros* (10.7%) and *F. nucleatum* (7.1%) were recovered. Three of these 4 species were also recovered from the saliva sample taken at the abutment operation. We have recovered bacteria from the inside of the implant in 9 of the 20 patients, but in no patient did we find four different species. The origin of these contaminating bacteria cannot be determined with certainty. During sampling, care was taken to avoid contamination of the inner part of the implant by saliva. One argument to consider saliva as the source of the contamination may be the observation that the same bacterial species were frequently isolated from the saliva at the same time.

Pathological events also occurred in patient #8, who presented with fistula formation between months 6 and 12. At 8 months, *P. gingivalis* was found in the deepened and bleeding peri-implant pockets. At that time, the sampled periodontal pockets appeared stable with no increasing probing depths, absence of bleeding upon probing and a low level (0.08%) of *P. gingivalis*. Surgical treat-

ment, followed by systemic metronidazole (500 mg t.i.d., for 7 days) controlled the infection and at 12 months *P. gingivalis* was no longer detectable. In both patients 8 and 13, the early (<12 months) complications were associated *P. gingivalis*. Rosenberg et al. (1991) studied the microbiology of failing implants and distinguished between infectious and traumatic failures on the basis of clinical parameters. They found *P. gingivalis* in 7 of 12 infectious implant failures.

Interestingly, patients 14 and 18 had detectable *P. gingivalis* both in the periodontal and the peri-implant pockets at 12 months. However, no pathological changes were noted in these patients during the experimental period. This indicates that the mere presence of this organism does not necessarily lead to peri-implantitis. Clinical and microbiological monitoring of these patients seems necessary to determine the long-term stability of the implants and the teeth. This study also revealed that detection of *P. gingivalis* by using a pooled periodontal pocket sample from four pockets is not always predictable in detecting this organism. In 5 patients *P. gingivalis* was isolated in the post-operative phase but not in the periodontal pocket sample at baseline. *A. actinomycetemcomitans* and *P. gingivalis* were also infrequently isolated from saliva, an observation for which we have no explanation.

In conclusion, in contrast to fully edentulous patients, colonization of peri-implant pockets in partially edentulous patients is characterized by rapid appearance of gram negative anaerobic rods that are associated with periodontal and peri-implant infections. This is probably caused by the presence of periodontal pockets that serve as a reservoir for these bacteria. Two of 20 patients in this study experienced a peri-implant infection associated with *P. gingivalis*, one of which lost both implants. It seems that proper periodontal infection control may help to prevent early bacterial complications in implant dentistry (Malmstrom et al. 1990). Infection control should involve suppression of commensal periodontal bacteria below certain thresholds and elimination of putative exogenous periodontal pathogens, i.e. *P. gingivalis*. This may be of special importance in patients with a history of periodontitis (Mombelli 1997). Microbiological testing in partially edentulous subjects with a history of periodontitis may be one measure to prevent peri-implantitis by employing appropriate antimicrobial therapy before dental implants are placed (van Winkelhoff & Winkel 1997).

## Résumé

Il y a peu de renseignement en ce qui concerne la colonisation initiale des poches parodontales chez les individus partiel-

ment édentés. Une connaissance dans ce domaine est une étape supplémentaire dans la connaissance de l'étiologie et la pathogénèse de la paroiimplantite. Dans cette étude, la colonisation primaire des poches paroiimplantaires par des pathogènes parodontaux putatifs a été étudiée chez vingt individus partiellement édentés en utilisant des techniques de culture anaérobie. Lors de l'examen initial, la présence et les niveaux de pathogènes parodontaux putatifs dans la microflore de poches parodontales et la salive ont été établis. Immédiatement après l'ancrage des implants en titane, et six et douze mois après, la présence et les niveaux de pathogènes parodontaux putatifs ont été déterminés dans les poches parodontales et paroiimplantaires. Le deuxième but a été de détecter la contamination bactérienne au niveau du site de l'implant et de l'intérieur de l'implant. Lors de l'examen initial, les espèces isolées le plus fréquemment des poches parodontales étaient le *Fusobacterium nucleatum*, le *Prevotella intermedia* et les *Peptostreptococcus micros*. Le *Bacteroides forsythus*, l'*Actinobacillus actinomycetemcomitans* (*A.a.*) et le *Porphyromonas gingivalis* (*P.g.*) ont été isolés respectivement chez neuf, deux et trois patients. Six mois après le placement des bridges, la majorité des sites implantaires avaient des niveaux détectables de la plupart des espèces bactériennes à l'exception de *A.a.* qui ne pouvait être isolé d'aucun des sites paroiimplantaires durant la période expérimentale, bien que deux patients avaient cet organisme lors de l'examen de départ. Chez deux patients ayant des niveaux détectables sous-gingivaux de *P.g.* lors de l'examen initial cette espèce a été retrouvée après douze mois dans les sites implantaires. Un des ces patients a perdu deux implants qui étaient associés avec une plus grande proportion de *P.g.* dans les poches paroiimplantaires. Un second patient développait deux fistules autour de deux implants après huit mois et ceci était également associé à la présence de *P.g.* Un contrôle de l'infection parodontale avant le placement des implants dentaires chez les patients partiellement édentés peut prévenir les complications de colonisation bactérienne.

## Zusammenfassung

Es bestehen nur beschränkte wissenschaftliche Informationen über die frühe Besiedelung von peri-implantären Taschen bei teilbezahnten Patienten. Das Wissen über diesen Prozess stellt einen Schritt zum besseren Verständnis der Aetiologie und Pathogenese der Periimplantitis dar. In dieser Studie wurde die frühe Besiedelung der peri-implantären Taschen durch potentielle Parodontalpathogene bei 20 teilbezahnten Individuen mittels anaeroben Kultivierungstechniken untersucht. Bei der Ausgangsuntersuchung wurde die Präsenz und die Menge von potentiellen Parodontalpathogenen in der Mikroflora der Parodontaltaschen und im Speichel bestimmt. Sofort nach der Belastung der Titanimplantate und nach 6 und 12 Monaten wurden die Präsenz und die Mengen der ausgewählten potentiellen Parodontalpathogene in den parodontalen und peri-implantären Taschen bestimmt. Ein zweites Ziel war, eine bakterielle Kontamination bei den Implantaten und auf der Innenseite der Implantate aufzudecken. Bei der Ausgangsuntersuchung waren *Fusobacterium nucleatum*, *Prevotella intermedia* und *Peptostreptococcus micros* die am meisten isolierten Arten aus den Parodontaltaschen. *Bacteroides forsythus*, *Actinobacillus actinomycetemcomitans* und *Porphyromonas gingivalis* wurden bei 9, 2 bzw. 3 Patienten isoliert. Sechs Monate nach dem Einsetzen der Brücken wies die Mehrzahl der Implantate nachweisbare Mengen der meisten parodontalen bakteriellen Arten mit Ausnahme von *A. actinomycetemcomitans* auf. *A. actinomycetemcomitans* konnte bei keiner der peri-implantären Proben isoliert werden, obwohl 2 Patienten diesen Keim bei der Ausgangsuntersuchung aufwiesen. Bei 2 Patienten mit nachweisbaren subgingivalen *P. gingivalis* bei der Ausgangsuntersuchung konnte diese Art nach 12 Monaten in den peri-implantären Stellen gefunden werden. Einer dieser Patienten verlor 2 Implantate. Der

Verlust war mit einer grossen Anzahl an *P. gingivalis* in den peri-implantären Taschen assoziiert. Ein zweiter Patient entwickelte nach 8 Monaten zwei Fisteln bei zwei Implantaten. Dieses Geschehen war ebenfalls mit der Präsenz von *P. gingivalis* assoziiert. Es wird die Schlussfolgerung gezogen, dass durch eine adäquate Beseitigung der parodontalen Infekte vor der Eingliederung von dentalen Implantaten bei teilbezahnten Patienten frühen bakteriellen Komplikationen vorgebeugt werden kann.

## Resumen

Existe una información científica limitada disponible sobre la colonización temprana de bolsas periimplantarias en individuos parcialmente edéntulos. El conocimiento de este proceso es un paso en el mejor entendimiento de la etiología y la patogénesis de la periimplantitis. En este estudio se investigó la colonización temprana de las bolsas periimplantarias por patógenos periodontales putativos en 20 individuos parcialmente edéntulos usando técnicas de cultivo anaerobio. En el punto inicial, se estableció la presencia y niveles de patógenos periodontales putativos en la microflore de las bolsas periodontales y de la saliva. Inmediatamente tras la carga de los implantes de titanio y después de 6 y 12 meses se determinó la presencia y niveles de los patógenos periodontales putativos seleccionados en las bolsas periodontales y periimplantarias. Una segunda intención fue detectar contaminación bacteriana en el lugar del implante y en el interior del implante. En el punto inicial, las especies más frecuentemente aisladas de las bolsas periodontales fueron *Fusobacterium nucleatum*, *Prevotella intermedia* y *Peptostreptococcus micros*. *Bacteroides forsythus*, *Actinobacillus actinomycetemcomitans* y *Porphyromonas gingivalis* se aislaron de 9, 2 y 3 pacientes respectivamente. Seis meses tras la colocación de los puentes, la mayoría de los lugares de implantes tenían niveles detectables de la mayoría de especies bacterianas periodontales con la excepción de *A. actinomycetemcomitans* que no pudo ser aislado de ninguna de las muestras periimplantarias durante el periodo experimental, aunque dos pacientes tuvieron este organismo en el punto inicial. En dos pacientes con *P. gingivalis* detectable en el punto inicial, esta especie se encontró después de 12 meses en los lugares periimplantarios. Uno de estos pacientes perdió dos implantes que se asociaron con una alta proporción de *P. gingivalis* en las bolsas periimplantarias. Un segundo paciente desarrolló dos fistulas alrededor de dos implantes a los ocho meses y este evento también se asoció a la presencia de *P. gingivalis*. Se concluye que el control adecuado de la infección periodontal antes de la colocación de implantes dentales en pacientes parcialmente edéntulos puede prevenir complicaciones bacterianas tempranas.

## 要旨

部分無歯顎者においてインプラント周囲ポケットの中の早期の細菌コロニー化について、科学的な情報は余りないが、このプロセスを知ることは、インプラント周囲炎の病因や病理発生に対する理解を深めるための第一歩である。本研究では、20名の部分無歯顎患者において、歯周病原菌とされる細菌のインプラント周囲ポケット内の早期コロニー化を、嫌気性培養法を用いて検討した。ベースライン時に歯周ポケットと唾液中の歯周病原菌の存在と濃度を記録した。チタン製インプラントに荷重をかけた直後、6ヶ月後、12ヶ月後に歯周ポケット及びインプラント周囲ポケットにお

いて、幾つかの歯周病原菌の存在と濃度を測定した。本研究の第2の目的は、インプラント部位及びインプラント内部において細菌汚染を検出することであった。ベースライン時に歯周ポケットから最も頻繁に分離された細菌は *Fusobacterium nucleatum*、*Prevotella intermedia* と *Peptostreptococcus micros* であった。*Bacteroides forsythus*、*Actinobacillus actinomycetemcomitans* 及び *Porphyromonas gingivalis* は各々9名、2名及び3名の患者から分離された。ブリッジ装着6ヶ月後に、大半のインプラント部位には *A.actinomycetemcomitans* 以外の大半の歯周細菌が検出可能な濃度で存在していた。*A.a* は実験期間中どのインプラント周囲の標本からも分離できなかったが、ベースライン時に2名の患者は同細菌を保有していた。ベースライン時に歯肉縁下で *P.gingivalis* が検出された患者2名において、同細菌がインプラント周囲部位で12ヶ月後に検出された。2名のうち1名は2本のインプラントを失ったが、これは高濃度の *P.gingivalis* と関連していた。2人目の患者は8ヶ月後に2本のインプラント周囲に2箇所のフィステルができたが、これも *P.gingivalis* の存在と関連していた。結論として、部分無歯顎患者にインプラントを埋入する前に、適切な歯周組織の感染コントロールを行うと、早期の細菌による合併症を予防しうると思われる。

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