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CONCENTRATION AND SPECIES COMPOSITION OF AEROBIC AND FACULTATIVELY ANAEROBIC BACTERIA RELEASED TO THE AIR OF A DENTAL OPERATION AREA BEFORE AND AFTER DISINFECTION OF DENTAL UNIT WATERLINES

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Abstract: Bacteriological air sampling was conducted at 25 dental units during restorative treatment sessions before and after disinfection of dental unit waterlines (DUWL) with hydrogen peroxide. Air samples for determining the concentration and species composition of aerobic and facultatively anaerobic bacteria were collected with the portable Reuter Centrifugal Sampler (RCS Plus) in the dental operation area close to patient's mouth. Large concentrations of airborne bacteria in the range of 0.35–40.08 × 10^3 cfu/m^3 (median = 1.63 × 10^3 cfu/m^3) were recorded before DUWL disinfection. After disinfection, the concentrations were significantly lower ($p<0.05$), ranging from 0.51–3.82 × 10^3 cfu/m^3 (median = 0.9 × 10^3 cfu/m^3). Streptococci were most numerous among airborne bacteria before DUWL disinfection, forming 79.23% of total isolates. The remaining isolates were staphylococci/micrococci (15.7%), corynebacteria (2.3%), endospore-forming bacilli (1.45%), Gram-negative bacteria (1.31%), and actinomycetes (0.01%). After DUWL disinfection, a significant decrease in the numbers of streptococci ($p<0.05$) and Gram-negative bacteria ($p<0.01$) was noted, while the numbers of other types of bacteria were unaffected. Altogether, 50 species or genera of bacteria were identified in the examined air samples before and after DUWL disinfection. Of these, 36 species or genera are considered potentially pathogenic, as a potential cause of infection, allergic disease or intoxication. In conclusion, the high pollution of dental operation area with bacteria indicates a need for use of preventive measures protecting dental staff and patients, such as DUWL disinfection that proved efficient in decrease of exposure in the present study.

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INTRODUCTION

Aerosols released during various dental treatment procedures, such as drilling and scaling, may contain potentially pathogenic bacteria posing a risk of infection for dental staff and patients [8, 9, 11, 13, 16, 18, 22]. Bacteria are spread mainly with the particles of saliva, nasopharyngeal secretions, blood, plaque and tooth debris from patient's oral cavity, or with droplets of coolant water from dental unit waterlines (DUWL) sprayed by the dental handpieces [9, 11].

To date, most of the studies on bacterial aerosols released during dental treatment were carried out with the sedimentation method [1, 5, 15, 17] which does not allow for proper determination of the concentration and species composition of bacteria in 1 m^3 of air. The mean concentrations

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of airborne bacteria measured during dental treatment at the distance of 1–2 m from a patient's mouth by a few authors using a volumetric method were in the range of 0.3–9.2 × 10^2 cfu/m^3 [7, 10, 12], with maximum peaks exceeding 10^3 cfu/m^3 [3, 12]. However, the species composition of bacteria in the air of dental surgeries has not been determined until recently with a volumetric method and remains largely unknown, as the only available data on this subject were obtained with the inaccurate sedimentation method [1, 15, 17].

To fill this gap, examinations of the concentration and species composition of bacteria in volumetric air samples collected during various dental treatment procedures were carried out in the present work at 25 dental units. For better assessment of the infection risk, the air samples were taken in the respiratory area of the dentist, much closer to mouth of the dentist and patient (circa 25 cm) than in hitherto done studies. In order to evaluate the effects of proper maintenance of water in dental unit reservoirs on bacterial pollution of air, the samples were taken at all units before and after disinfection of water with hydrogen peroxide, and the results compared using statistical methods.

MATERIALS AND METHODS

Study area. Bacteriological air sampling was conducted during the year 2006 at 25 dental units located in public dental care facilities. Each unit was equipped with reservoir of distilled water and tubes conducting water to handpieces (high-speed handpiece, low-speed handpiece, air-water syringe, scaler). Air samples were collected during restorative dental treatment sessions, each for one patient, with the use of a high-speed handpiece. Besides air samples, from each dental unit were taken, before and after disinfection, samples of water and sections of waterline tubes for detection of bacteria, using the techniques described elsewhere [19, 21, 22].

DUWL disinfection. After taking the first series of air samples, waterlines in all 25 dental units under study were disinfected with the preparation Oxygenal 6 (KaVo, Biberach, Germany), containing 6% hydrogen peroxide (H_2O_2) as an active substance. The effects of H_2O_2 were enhanced by the presence of silver ions. The disinfectant was applied according to producer's instructions. After disinfection, air samples were collected during dental treatment procedures at all 25 units.

Method of bacteriological air sampling. Air samples were collected with the portable Air Sampler RCS Plus (Biotest AG, Dreieich, Germany). This is a miniature Reuter Centrifugal Sampler (RCS) in which a high-speed rotor collects air sample on strips coated with agar media located on the inner walls of a mini-centrifuge. The samples were taken on TC (Total Count) Strips for isolation and identification of total aerobic and facultatively anaerobic mesophilic bacteria, provided by the manufacturer of the sampling device. They were coated with a medium containing casein digested with pancreatin, soy peptone, NaCl, KH_2PO_4, K_2HPO_4, agar-agar. The TC Strips also contained disinfectant-neutralizing substances. The sampler was placed within 25 cm of the patient's mouth. A volume of 100 litres of air was drawn on each sample.

Isolation and identification of airborne bacteria. The air samples were incubated for 48 hrs at 33°C. Then, the grown colonies were counted and differentiated on the basis of colony morphology, and Gram staining and the concentration of particular morphological types of bacteria and total mesophilic bacteria in cfu per 1 m^3 of air was estimated.

Bacterial isolates were subcultured on tryptic soy agar slants and identified to species or genus level with the use of metabolic microtests: API 20E and API 20NE (bioMérieux, Marcy l'Etoile, France) for identification of, respectively, fermenting and non-fermenting Gram-negative bacteria, and GP2 Microplate™ (BIOLOG, Inc., Hayward, CA, USA) for identification of Gram-positive bacteria. Prior to microtests the test for oxidase (Bactident Oxidase, Merck, KGaA, Germany) with strains of Gram-negative bacteria or the test for catalase (with the use of 3% H_2O_2) with strains of Gram-positive bacteria were carried out.

Statistical analysis. The analyses were performed with use of the Statistica for Windows v. 5.0 package (Statsoft, Inc., Tulsa, OH, USA). The data distribution was checked for normality by the Kolmogorov-Smirnov test. The significance of differences between variables was tested by the Wilcoxon matched pairs test. Results were considered significant for "p" values of ≤0.05.

RESULTS

Concentration and composition of airborne bacteria before DUWL disinfection. The total concentrations of aerobic and facultatively anaerobic bacteria measured in the operational areas of 25 dental units showed highly variable, non-normal distribution ranging between 0.35–40.08 × 10^3 cfu/m^3 (median 1.63 × 10^3 cfu/m^3) (Tab. 1). Gram-positive cocci evidently prevailed in the examined air samples, of which streptococci formed 79.23% and staphylococci/micrococci 15.7% of total isolates. The prevalence of streptococci was mainly due to large concentrations of *Streptococcus mutans*/*ratti* at unit no.14 and of *Lactococcus lactis ss lactis* at unit no. 15 (21.7 × 10^3 cfu/m^3 and 32.13 × 10^3 cfu/m^3, respectively). The percentages of Gram-negative bacteria, endospore-forming bacilli and corynebacteria were within a range of 1.31–2.3%, and actinomycetes formed only 0.01% of the total count.

Effect of DUWL disinfection on concentration and composition of airborne bacteria. DUWL disinfection

Table 1. Concentration and species composition of airborne bacteria in dental treatment area before disinfection of unit waterlines (cfu/m^3).

Unit No.	Gram-negative bacteria	Staphylococci and other catalase-positive cocci	Streptococci and other catalase-negative cocci	Endospore--forming bacilli	Corynebacteria and related organisms	Actinomycetes	Total
1	80	540	790	100	320	0	1,830
2	130	350	3,500	40	10	0	4,030
3	50	440	270	70	40	0	870
4	40	1,750	2,100	110	0	0	4,000
5	60	790	1,450	60	80	10	2,450
6	20	750	60	120	0	0	950
7	90	650	770	220	0	0	1,730
8	20	800	130	50	0	0	1,000
9	30	3,230	380	30	40	0	3,710
10	30	260	50	50	120	0	510
11	230	240	70	60	0	0	600
12	0	780	0	30	40	0	850
13	90	550	310	10	710	0	1,670
14	0	210	21,700	40	30	0	21,980
15	0	90	39,970	0	20	0	40,080
16	0	420	880	70	420	0	1,790
17	80	310	1,720	50	30	0	2,190
18	20	250	100	40	30	0	440
19	0	350	1,230	50	0	0	1,630
20	0	480	500	80	130	0	1,190
21	110	580	480	20	30	0	1,220
22	20	300	2,040	40	130	0	2,530
23	90	300	10	0	50	0	450
24	30	280	10	30	0	0	350
25	80	900	210	70	60	0	1,320
Total	1300	15,600	78,730	1,440	2,290	10	99,370
Percent	1.31%	15.70%	79.23%	1.45%	2.30%	0.01%	100%
Median	30.0	440.0	480.0	50.0	30.0	0.0	1,630.0
Range	0–230	90–3,230	0–39,970	0–220	0–710	0–10	350–40,080
Mean	52.0	624.0	3,149.2	57.6	91.6	0.4	3,967.6
SD	53.9	639.0	8,781.6	45.6	163.6	2.0	8,618.0
Total positive	19 (76%)	25 (100%)	24 (96%)	23 (92%)	18 (72%)	1 (4%)	25 (100%)

with H_2O_2 resulted with a statistically significant ($p = 0.04$) decrease of the concentration of airborne bacteria by nearly 50%, to the median level of 0.9×10^3 cfu/m^3 (Tab. 2). This drop was due to the significant decrease of the numbers of streptococci ($p = 0.031$) and Gram-negative bacteria ($p = 0.0023$). The numbers of staphylococci/micrococci, endospore-forming bacilli and corynebacteria did not show a significant change, and the number of actinomycetes even significantly increased after disinfection ($p = 0.024$). The most common airborne bacteria after DUWL disinfection were staphylococci/micrococci (61.19% of total isolates), followed by streptococci (24.28%), endospore-forming bacilli (7.92%), and corynebacteria (4.18%) (Tab. 2).

Identified species of airborne bacteria. As many as 43 species or genera of the aerobic and facultatively anaerobic bacteria were identified in the examined air samples before DUWL disinfection (Tab. 3), while 33 species or genera of bacteria were identified after DUWL disinfection (Tab. 4). Altogether, 50 species or genera of bacteria were identified in the examined air samples before and after DUWL disinfection. Of these, 15 species belonged to streptococci, 13 – to staphylococci/micrococci, 9 – to Gram-negative bacteria, 8 – to corynebacteria, 3 – to actinomycetes, and 2 – to endospore-forming bacilli.

Of the total number of 50 species or genera identified in air samples, 10 were identified in water samples from

Table 2. Concentration and species composition of airborne bacteria in the dental treatment area after disinfection of unit waterlines (cfu/m^3).

Unit No.	Gram-negative bacteria	Staphylococci and other catalase-positive cocci	Streptococci and other catalase-negative cocci	Endospore--forming bacilli	Corynebacteria and related organisms	Actinomycetes	Total
1	0	780	70	100	20	0	970
2	0	470	50	0	10	10	540
3	0	850	780	60	20	180	1,890
4	0	960	20	100	20	10	1,110
5	80	870	50	50	50	0	1,100
6	20	410	0	40	40	0	510
7	50	710	1,260	130	40	10	2,200
8	20	460	110	10	130	10	740
9	80	1,390	1,800	550	0	0	3,820
10	10	310	90	170	30	0	610
11	20	800	10	100	20	20	970
12	0	480	0	10	30	0	520
13	0	1,710	1,770	30	110	0	3,620
14	0	810	110	290	50	0	1,260
15	0	620	190	10	0	10	830
16	20	540	100	30	210	0	900
17	0	590	80	0	210	0	880
18	0	640	50	40	60	150	940
19	0	480	0	150	40	0	670
20	0	590	100	60	50	0	800
21	0	540	90	40	20	0	690
22	0	1,140	70	90	20	0	1,320
23	0	800	180	250	60	0	1,290
24	0	690	140	20	0	0	850
25	20	510	80	20	0	0	630
Total	320	18,150	7,200	2,350	1,240	400	29,660
Percent	1.08%	61.19%	24.28%	7.92%	4.18%	1.35%	100%
Median	0.0**	640.0	90.0*	50.0	30.0	0.0	900.0*
Range	0–80**	310–1,710	0–1,800*	0–550	0–210	0–180	510–3,820*
Mean	12.8	726.0	288.0	94.0	49.6	16.0	1,186.4
SD	23.5	315.9	527.4	121.1	57.6	45.4	861.7
Total positive	9 (36%)	25 (100%)	22 (88%)	23 (92%)	21 (84%)	8 (32%)	25 (100%)

* Significantly less ($p<0.05$) compared to values recorded before DUWL disinfection. **Significantly less ($p<0.01$) compared to values recorded before DUWL disinfection.

dental unit reservoirs, as reported earlier [21]. 36 species or genera are considered potentially pathogenic, as a potential cause of infection, allergic disease or intoxication [6, 14, 22].

DISCUSSION

The level of bacterial contamination of air found at the examined dental units during restorative treatment sessions was high. It was distinctly greater compared to the data reported by earlier authors who also used a volumetric sampling method [3, 7, 10, 12]. At 17 out of 25 dental units examined before DUWL disinfection, the level of 10^3 cfu/m^3 was exceeded, and at 2 units the level of 10^4 cfu/m^3 was exceeded. After DUWL disinfection, at 9 units the level of 10^3 cfu/m^3 was exceeded and nowhere the level of 10^4 cfu/m^3 was exceeded.

At 24 out of 25 dental units examined the level of 3.75×10^2 cfu/m^3 was exceeded, considered by Legnani *et al.* [12] as "very bad". At all 25 dental units the level of 2.0×10^2 cfu/m^3 proposed as a ceiling limit for bacteria in clean rooms and hospitals [4], was exceeded. The relatively large

Table 3. Species and genera of bacteria isolated from air of dental unit operation areas before DUWL disinfection.

Gram-negative bacteria
Acinetobacter lwoffii (9); ***Aeromonas* spp.** (5); ***Brevundimonas vesicularis*** (1, 3, 10, 11, 13, 18, 24); ***Empedobacter brevis*** (2, 7, 11, 22); ***Pantoea agglomerans*** (2, 25); ***Pseudomonas* spp.** (25); ***Ralstonia pickettii*** (1, 2, 3, 4, 6, 7, 9, 11, 13, 21, 23, 24); *Sphingomonas multivorum* (17); ***Sphingomonas paucimobilis*** (2, 6, 8, 17, 21)

Staphylococci and other catalase-positive cocci
Kocuria rosea/erythromyxa (5, 6, 8, 9, 13, 17, 20, 21, 22, 23, 24); *Kytococcus sedentarius* (2, 6, 7, 8, 11, 12, 13); *Macrococcus carouselicus* (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 16, 17, 18, 20, 21, 22, 24, 25); ***Micrococcus luteus*** (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 23, 24, 25); ***Micrococcus lylae*** (4, 12, 23); ***Micrococcus* spp.** (1, 2, 3, 4, 5, 6, 7, 8, 10, 16, 18, 19, 21, 25); *Staphylococcus arlettae* (1, 3, 4, 5, 6, 7, 8, 10, 13, 14, 15, 16, 17, 18, 19, 21, 23, 25); *Staphylococcus gallinarum* (6, 12, 19); ***Staphylococcus haemolyticus*** (1, 5, 6, 8, 12, 13, 20, 21, 24); ***Staphylococcus hominis*** (2, 4, 13, 15, 16, 18, 22, 23); ***Staphylococcus hominis/novobiosepticus*** (8, 9); ***Stomatococcus mucilaginosus*** (3, 9, 11, 13); Unidentified catalase-positive cocci (1, 2, 7, 10, 11, 12, 17, 18, 21, 22, 25)

Streptococci and other catalase-negative cocci
Aerococcus viridans (10, 21, 25); ***Enterococcus durans*** (22); ***Enterococcus flavescens*** (9); *Enterococcus sulfureus* (9); ***Lactococcus lactis ss lactis*** (15, 16, 17); *Leuconostoc fallax* (1, 3, 5, 6, 9, 10, 13, 19, 20, 21, 22, 23, 24, 25); ***Streptococcus acidominimus*** (13); ***Streptococcus mutans/ratti*** (1, 2, 3, 4, 5, 7, 11, 14, 15, 17, 19, 20, 22); ***Streptococcus porcinus*** (15); ***Streptococcus salivarius*** (17); ***Streptococcus suis*** (1); ***Streptococcus vestibularis*** (9); ***Streptococcus* spp.** (1, 6, 8, 13, 15, 16, 18, 21, 25)

Endospore-forming bacilli
Bacillus amyloliquefaciens (7); ***Bacillus* spp.** (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 16, 17, 18, 19, 20, 21, 22, 24, 25)

Corynebacteria and related organisms
Aureobacterium flavescens (10, 20, 22); *Brevibacterium epidermidis* (9); *Brevibacterium* spp. (5, 12, 14, 15, 17, 18); ***Corynebacterium lipophiloflavum*** (25); ***Corynebacterium* spp.** (1, 2, 3, 10, 16, 20, 21, 22, 23); ***Rothia dentocariosa*** (13)

Actinomycetes
Streptomyces albus (5)

Boldface: species potentially pathogenic. Shaded: species indigenous for oral cavity. Underlined: species isolated also from DUWL before disinfection [20, 21]. Numbers in parentheses indicate dental units at which species was isolated.

concentrations of airborne bacteria found in the present study may be due, at least to some extent, to the fact that the small dimensions of the sampler enabled collection of air samples close to a patient's mouth, much nearer than in hitherto done studies. Nevertheless, such a location of the sampler was postulated by Grenier [7] as appropriate for better assessment of the infection risk for dental staff and patient. The real exposure to bacterial aerosols in the examined surgeries could be even greater, as in the present work the obligatory anaerobic bacteria were not determined. Hence, the results of the present study are not fully comparable with earlier ones in which either aerobic or anaerobic bacteria were determined [3, 7, 10, 12]. In this study, the determination of aerobic and facultatively anaerobic bacteria was chosen as, in the opinion of some authors, they are considered as more numerous than anaerobic ones [8], comprise most potentially pathogenic species [11], and pose a much better index of waterborne infection [7] and of the effectiveness of DUWL disinfection. Nevertheless, it must be stressed that many strict anaerobes are important dental pathogens causing caries and other oral cavity diseases; therefore a similar study on the species composition of anaerobic bacteria in dental operation area would be highly desirable.

The present study demonstrates that the disinfection of water in dental unit reservoirs with H_2O_2 significantly reduces the exposure of dental staff and patients to airborne bacteria during dental treatment procedures. In DUWL tubes biofilms may develop that pose a rich source of aerobic bacteria [2, 19, 21, 22, 23] which are dispersed into air with water droplets during dental treatment. The efficiency of H_2O_2 in the reduction of the number of bacteria in DUWL biofilm and water has been proved in earlier papers [20, 22]. Thus, the significant decrease of the airborne Gram-negative bacteria is directly related to killing these organisms in DUWL by H_2O_2. On the other hand, the significant decrease in the number of airborne streptococci could be explained by presumptive inhibition of their growth by disinfectant-containing coolant water during the washing out patient's mouth. The unexpected increase of the number of airborne actinomycetes after DUWL disinfection could be probably explained by the absence of large amounts of streptococci which earlier inhibited their growth.

This study is the first in which the species composition of airborne bacteria, recovered during dental treatment procedures with the use of a volumetric method, is presented. Thus, our results could be compared only with those obtained by the authors using inaccurate sedimentation method for study the air microflora [1, 15, 17]. Al Maghlouth *et al.* [1] reported that among airborne bacteria isolated during dental treatments there prevailed *Staphylococcus epidermidis* (37.1%) followed by *Micrococcus* spp. (32.6%) and "diphteroids" (28.2%). It is striking that these authors did not detect streptococci which in the present study formed nearly 80% of total airborne bacteria. In contrast, Osorio *et al.* [15] and Rautemaa *et al.* [17] reported

Table 4. Species and genera of bacteria isolated from air of dental unit operation areas after DUWL disinfection.

Gram-negative bacteria
Brevundimonas vesicularis (8); ***Empedobacter brevis*** (7, 11); ***Pantoea agglomerans*** (6); ***Ralstonia pickettii*** (5, 7, 10, 16, 25); ***Sphingomonas paucimobilis*** (5, 9)

Staphylococci and other catalase-positive cocci
Dermacoccus nishinomiyaensis (13); *Kocuria rosea/erythromyxa* (1, 4, 5, 6, 7, 8, 9, 13, 21, 22, 23, 24); *Kytococcus sedentarius* (1, 3, 4, 5, 7, 11, 13, 14, 18); *Macrococcus carouselicus* (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 13, 14, 15, 17, 18, 19, 20, 21, 22, 23, 24); ***Micrococcus luteus*** (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25); ***Micrococcus lylae*** (1, 4, 5, 6, 11, 19, 20); ***Micrococcus* spp.** (1, 3, 4, 5, 6, 8, 9, 11, 12, 15, 16, 18, 19, 20, 21, 22, 24); *Staphylococcus arlettae* (1, 2, 3, 4, 5, 6, 7, 8, 11, 12, 13, 14, 15, 16, 17, 18, 19, 21, 22, 23, 24, 25); *Staphylococcus gallinarum* (5, 6, 7, 8, 12, 19, 20, 21); ***Staphylococcus haemolyticus*** (1, 3, 4, 5, 7, 8, 10, 11, 13, 14, 16, 17, 20, 21, 22, 23, 25); ***Staphylococcus hominis*** (5, 9, 14, 18); ***Stomatococcus mucilaginosus*** (9, 22); Unidentified catalase-positive cocci (1, 2, 6, 8, 10, 12, 15, 17, 21, 24, 25)

Streptococci and other catalase-negative cocci
Aerococcus viridans (15, 24); ***Lactococcus lactis ss lactis*** (9); *Leuconostoc fallax* (1, 3, 4, 5, 7, 8, 9, 10, 11, 14, 15, 16, 18, 20, 21, 22, 23, 24); *Pediococcus pentosaceus* (1, 2, 8, 15, 16, 17, 24, 25); ***Streptococcus acidominimus*** (13); ***Streptococcus mutans/ratti*** (7); ***Streptococcus sanguis*** (3); ***Streptococcus* spp.** (2, 3, 7, 10, 13, 14, 16, 17, 18, 21, 22, 23, 25)

Endospore-forming bacilli
***Bacillus* spp.** (1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 18, 19, 20, 21, 22, 23, 24, 25)

Corynebacteria and related organisms
Aureobacterium flavescens (4, 5, 6, 11, 16, 19, 20, 22); *Brevibacterium* spp. (3, 8, 18); ***Corynebacterium urealyticum*** (16); ***Corynebacterium* spp.** (1, 2, 6, 7, 8, 10, 12, 14, 17, 21, 23); ***Leifsonia aquatica*** (13)

Actinomycetes
Actinomyces naeslundii (3, 18); ***Streptomyces* spp.** (2, 4, 7, 8, 11, 15)

Boldface: species potentially pathogenic. Shaded: species indigenous for oral cavity. Underlined: species isolated also from DUWL before disinfection [20, 21]. Numbers in parentheses indicate dental units at which species was isolated.

on the prevalence of streptococci and staphylococci, but did not give the exact percentages of both groups among total airborne bacteria.

A total of 50 species or genera of bacteria were identified in the present study in air samples collected during dental treatment. Of these, at least 40 species or genera, to the best of our knowledge, have not been reported until recently, from the air of dental surgeries.

In conclusion, the high pollution of dental operation areas with bacteria found in the present work and a large proportion of species considered as potentially pathogenic (72%) indicates a need for the use of preventive measures protecting dental staff and patients from the airborne infection related to dental treatment. Apart from the DUWL disinfection, the efficiency of which has been tested in this study, these measures should include: wearing a surgical mask and safety glasses by the dentist, a pre-procedural rinse with antiseptic mouthwash by the patient before treatment, the use of a high-volume evacuator for all procedures, the use of a rubber dam, maintenance of a high-efficiency ventilation system, and the use of air filters and ultraviolet lamps [7, 8, 9, 10, 11, 16, 17].

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