Effect of 12-month weekly professional oral hygiene care on the composition of the oral flora in dentate, dependent elderly residents: A prospective study

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Objective: To study the effect of weekly professional oral hygiene care on the proportion of micro-organisms associated with good oral health, caries, and periodontal and soft tissue diseases in oral biofilms in dentate, dependent elderly residents.

Background: Assisted oral hygiene care reduces the plaque score and number of micro-organisms in the oral biofilms in elderly residents. Less is known about the effect on the quality/composition of the remaining oral flora.

Materials and methods: Participants comprised 33 residents in the study and 35 in the control group. Dental status (≥10 natural teeth and no removable dentures to be included), plaque score, salivary secretion rate and prescription medicines were recorded. Duplicate samples, collected from supragingival plaque and tongue, were analysed using cultivation technique. Differences between and within groups were analysed using one-way and two-way ANOVA, respectively.

Results: At the baseline, the number of teeth in the participants (mean age, 83.7 ± 7.4 years) was 22.0 ± 4.5. The number of prescription medicines was 9.4 ± 4.5. Seventy-six per cent had low salivary secretion rate. Fifty per cent had “visible thick” supragingival plaque. At the 12-month registration, “no visible” or “visible but thin” plaque was recorded in 92% in the study group. The proportions of bacteria associated with good oral health and periodontal diseases were decreased over time, while the frequency and proportions of micro-organisms associated with caries and soft tissue infection were unaffected or increased.

Conclusion: The results indicate that assisted oral hygiene care alone is not sufficient to regain an oral microbial flora associated with good oral health in dentate, dependent elderly residents.

KEYWORDS dependant elders, minor salivary secretion, oral microbial flora, professional oral hygiene care

1 | INTRODUCTION

The importance of a natural and healthy dentition at an older age is recognised not only for a good nutritional status but also for good general functions and health. Also, the elders consider the maintenance of natural teeth and oral health to be of great importance. The vast majority of nurses at homes for dependent elders are of the opinion that assistance with oral hygiene care is important and included in their duties. Despite
a basic, positive attitude in both caretakers and providers, several studies reveal poor oral health with dental plaque accumulation, tooth decay and the need of periodontal treatment in dependent residents.5,6

One of the two major reasons for tooth loss is dental caries. In addition, caries lesions are a common cause of oral pain while drinking and eating. In dependent elderly people, root caries is the most prevalent form of caries lesions.7 Caries lesions are associated with high proportions of acidogenic and aciduric bacteria, such as mutans streptococci, lactobacilli and Actinomyces, in the supragingival plaque.8,9 At a decreased salivary secretion rate, which is a common disorder in dependent elders,10 the proportion of the acidogenic and aciduric micro-organisms is increased in the dental biofilm.11-13

There are results indicating that an oral acidic milieu, in combination with diseases affecting general health, will contribute to the frequent colonisation of opportunistic micro-organisms reported in frail elders.10-14 Opportunistic micro-organisms are involved in both oral mucosal infections and aspiration pneumonia in institutionalised elders.15,16 In specimens collected from respiratory secretion in elders ≥75 years of age with pneumonia and admitted to intensive care, more opportunistic microbial species frequently occurring in samples collected from the oral cavity in dependent elderly residents were identified.17 Second to urinary infection, aspiration pneumonia is the most common infection in dependent elderly residents and has a high mortality rate.16

The second of the two main reasons for tooth loss is periodontitis. Gingivitis, the most common form of periodontal disease, will be the effect when bacteria are allowed to accumulate on the teeth. In the anaerobic milieu created at plaque accumulation, bacteria such as Fusobacterium nucleatum and Prevotella intermedia will thrive.18 Gingivitis is a reversible disease, and the inflammation will normally subside after a week with proper oral hygiene procedures. Porphyromonas gingivalis, having the advantage of the components released at the inflammatory reaction, and Aggregatibacter actinomycetemcomitans are both associated with destruction of the tooth-supporting tissues, periodontitis.19

The results obtained in intervention studies, in which dependent elderly residents received regular oral care by a dental hygienist for ≥5 months, indicate a reduction in the total number of micro-organisms and streptococci, a reduced frequency of black-pigmented Bacteroides species, and reduced frequencies and numbers of opportunistic micro-organisms.20 Less is known about the effect on bacteria with acidogenic and aciduric potential, and the quality/composition of the oral flora, with regular long-term professional oral health care in dentate, dependent elderly residents.

The aim of this study was to evaluate the effect of weekly professional oral hygiene care on the proportion of micro-organisms associated with good oral health, caries, periodontal diseases and opportunistic infections in oral biofilms, in dentate, dependent elderly residents.

2 | MATERIALS AND METHODS

2.1 | Ethics statement

The study was approved by the Ethics Committee at the University of Gothenburg, Sweden (Permit Number 039/00).
2.4 | Collection of clinical data and samples for microbial analysis

Clinical examinations, measurements of labial minor gland salivary secretion rate and microbial samplings were performed between 9 and 12 AM (by author KL. K) and were carried out in the residents’ private rooms. All microbial sampling was performed in duplicate, 1 week apart. The clinical examination was performed at the first of the two duplicate sampling occasions. Plaque registration followed by the collection of supragingival plaque for microbial analysis was performed at four sites, in that order. The sites selected were interproximally the upper right first and second molars, the lower left first and second molars, the upper right second incisor and the canine and the lower left second incisor and the canine. If one or more of these teeth were missing, the closest available site was selected. The measurement of the labial minor gland salivary secretion rate, followed by microbial sampling from the tongue, was performed in that order and at the end of the registration and sampling sessions.

In both the study and control groups, clinical data, data on prescription medicines, the labial minor gland salivary secretion rate and microbial samples were collected at the baseline and at the end of the 12-month study period. In the study group, additional plaque registration and sampling for microbial analysis were performed after 3, 6 and 9 months from the baseline.

2.5 | Clinical examination

The clinical examination was carried out by the bedside and in the light of an adjustable headlight, using a dental mirror and a dental probe. The number of natural teeth, clinically visible caries lesions and plaque were recorded. Plaque was recorded as no visible plaque (score 0), visible but thin plaque (score 1) or visible thick plaque (score 2). A mean of the plaque scores recorded at the four interproximal sites selected for registration was calculated.

2.6 | Prescription medicines

Data on prescription medicines were collected from the residents’ medical records.

2.7 | Labial minor gland salivary secretion rate

For measurement of the labial minor gland salivary secretion rate, performed as described by Eliasson et al.,22 the area of the lower labial mucosa was gently dried with a cotton pad. A pre-cut piece of a standard filter paper was then placed for 15 seconds near the midline approximately 3 mm from the outer border of the mucosa. The volume of liquid absorbed by the filter paper was measured using a calibrated Periotron® (8000, ProFlow™ Inc., Amityville, NY, USA). At each examination, four samples were collected from each participant and an individual mean was calculated.

2.8 | Microbial sampling and analysis

Microbial samples were collected from the supragingival plaque and the dorsum of the tongue. An experienced laboratory assistant blinded to which group of participants, study or control group, and to which of the examination occasions the samples were collected from, analysed the samples.

The supragingival plaque samples were collected using sterile toothpicks (TePe Birch; TePe) and pooled. The tongue samples were collected using sterile tweezers, cotton pellets and plastic spatulas. The spatula, with a circular hole 1.5 cm diameter, was placed on the back part of the dorsum. A cotton pellet, immersed in sampling fluid, was swept over the area inside the hole as described previously. The two samples were each transferred to 3.5 mL of transport medium VMGA III® and processed within 4 hours.

The analyses of the samples were performed as previously described using enriched blood agar plates and selective agar plates. The total number of micro-organisms growing under anaerobic conditions, the total number and proportion of streptococci and the number and proportion of Streptococcus sanguinis/oralis and Streptococcus salivarius, both associated with good oral health,24 mutans streptococci, lactobacilli and Actinomyces spp., associated with dental caries,8,9 F. nucleatum, and P. intermedia/nigrescens, associated with gingival inflammation,18 P. gingivalis and A. actinomycetemcomitans, associated with periodontitis. Candida albicans, Staphylococcus aureus and enteric rods, associated with mucosal and respiratory infections,15 were calculated. The detection limit was 100 colony-forming units for all species except A. actinomycetemcomitans, where the detection limit was 10 colony-forming units. If possible, the number of micro-organisms was calculated from their number on a plate giving 30-300 colonies. A mean of the results from the duplicate samplings was calculated.

2.9 | Statistical methods

To the best of this study’s knowledge, relevant data for a proper power analysis were not available in literature. Therefore, the number of participants included was based on the results in our previous studies. In the studies on oral microbial flora in groups with hyposalivation due to Sjögren’s syndrome, radiation therapy to the head and neck region, medication and their controls, the number of participants in each group was approximately 20.11-13,25,26 In our studies on elderly nursing home residents, the dropout rate was about one-third over a 1-year period.10,27

To normalise the microbial data, the numbers were transformed logarithmically. Zero counts were treated as one colony-forming unit/mL. Mean and median values and standard deviations were calculated. One-way ANOVA was used for the analysis of differences between the study and control groups. Two-way ANOVA was used for statistical analysis of differences at different time points within the two groups. Results were regarded statistically significant at P-values <.05. Owing to the multiple influence aspect, isolated significances should be interpreted with some caution.
3 | RESULTS

Of the 33 participants included in the study group, 26 participated at the 12-month registration. Of the dropouts, three were deceased and four declined to participate due to general health deterioration. Of the 35 participants included in the control group, 30 participated at the 12-month registration. All five dropouts were deceased. Baseline clinical and microbial results in the 12 dropouts were not significantly different from those participating during the whole 12-month study period.

3.1 | Clinical results

Results regarding oral clinical status and prescription medications at the baseline and the 12-month examination are shown in Table 1.

3.1.1 | Baseline

At the baseline, no significant differences between the study and control groups could be revealed regarding age, number of prescription medicines or oral clinical data. Of all 68 participants included in the study, 93% had ≥4 prescription medicines and 76% had a labial minor gland secretion rate ≤4 μL/cm²/min. No statistically significant correlation between the number of prescription medicines used and the labial minor gland secretion rate, or in the labial secretion rate between the group of men and group of women, could be revealed. Seventy-one percent had ≥20 teeth and 50% had the maximum plaque score 2 (visible thick plaque).

3.1.2 | 12-month examination

At the 12-month examination, there were still no significant differences detected between the two groups for age, number of prescription medicines, number of teeth and the labial minor gland secretion rate (Table 1).

Regarding the supragingival plaque score at the 12-month registration, there was a substantial decrease in the study group, both in comparison with their baseline score and with the score in the control group (P<.0001 for both). Further analysis indicated that the decrease in plaque score occurred from the 3-month to the 6-month registration. At the 12-month registration, 92% in the study group and 13% in the control group had a plaque score ≤1. No differences in the plaque score could be revealed between the 12 participants in the study group who had been using the electric toothbrush, the eight participants who had been using both the electric and a manual toothbrush and the six participants who had been using a manual toothbrush only.

3.2 | Microbial results

Results of the microbial analysis, at the baseline and the 12-month examination, of the supragingival plaque samples, are shown in Table 2. The tongue samples are shown in Table 3. The frequency of the opportunistic micro-organisms in samples collected from the supragingival plaque and the dorsum of the tongue, in the study group at every 3-month follow-up occasion, is given in Table 4.

3.2.1 | Baseline

The differences detected at the baseline between the study and control groups in the micro-organisms analysed in the supragingival plaque included a lower proportion of lactobacilli (P=.03), a tendency to a lower number of Actinomyces (P=.05), a higher frequency and number, as well as proportion of P. intermedia/nigrescens (P=.006, P=.01 and P=.02, respectively), and a lower frequency of S. aureus (P=.007) in the study group.

No statistically significant differences between the two groups could be revealed in samples collected from the tongue.

3.2.2 | 12-month examination

Total viable count

At the 12-month examination, there was a tendency towards a lower total count of micro-organisms in the supragingival plaque samples (P=.06) and a significantly lower total microbial count in the tongue samples (P=.04) in the study group, compared to that in the control group. The decrease in the tongue samples (P=.03) occurred from the 3-month to 6-month registration.

| TABLE 1 | Mean, standard deviation and median (in parentheses) values of age, prescription medications and results of the oral clinical examination in the study and control groups, at baseline and at the 12-month registration |
|----------|-----------------------------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
|          | Women | Men | Age | Number of medicines | Number of teeth | Labial minor gland secretion (μL/cm²/min) | Plaque score |
| Study group |       |     |     |                      |                  |                                  |             |
| Baseline   | 21    | 12  | 83 ± 8 (84) | 9 ± 4 (9) | 21 ± 5 (22) | 3.20 ± 2.45 (2.98) | 1.7 ± 0.5 (2.0) |
| 12 months  | 16    | 10  | 82 ± 8 (84) | 8 ± 4 (8) | 21 ± 5 (22) | 2.57 ± 1.34 (2.53) | 0.7 ± 0.5 (1.0)* |
| Control group |     |     |     |                      |                  |                                  |             |
| Baseline   | 23    | 12  | 84 ± 7 (84) | 10 ± 5 (10) | 22 ± 4 (24) | 3.42 ± 2.65 (2.83) | 1.6 ± 0.4 (1.8) |
| 12 months  | 20    | 10  | 85 ± 7 (85) | 10 ± 5 (10) | 23 ± 3 (24) | 2.77 ± 1.68 (2.71) | 1.6 ± 0.6 (1.5) |

*Difference between groups at the 12-month examination and within the study group between the baseline and the 12-month examination, P<.0001 for both.
TABLE 2  Supragingival plaque. Total viable microbial count (log 10) and proportions of streptococci, lactobacilli, Actinomyces, F. nucleatum, P. intermedia/nigrescens, P. gingivalis, A. actinomycetemcomitans, C. albicans, S. aureus and enteric rods. Mean, standard deviation and frequency (in square brackets) are given.

<table>
<thead>
<tr>
<th></th>
<th>Study group</th>
<th>Control group</th>
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<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>12 months</td>
</tr>
<tr>
<td>Total viable count</td>
<td>7.47 ± 0.37 (7.42) [100]</td>
<td>6.95 ± 0.56 (6.95) [100]</td>
</tr>
<tr>
<td>Proportion</td>
<td></td>
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<tr>
<td>Streptococci</td>
<td>25.5 ± 20.7 (20.4) [100]</td>
<td>64.9 ± 54.9 (54.2) [100]</td>
</tr>
<tr>
<td>S. sanguinis/oralis⁴</td>
<td>15.9 ± 14.3 (12.5) [91]</td>
<td>4.1 ± 6.9 (1.6) [81]</td>
</tr>
<tr>
<td>S. salivarius</td>
<td>5.6 ± 12.0 (0.9) [79]</td>
<td>0.7 ± 0.9 (1.2) [65]</td>
</tr>
<tr>
<td>Mutans streptococci³</td>
<td>14.6 ± 16.2 (7.6) [97]</td>
<td>15.6 ± 22.3 (2.5) [77]</td>
</tr>
<tr>
<td>Lactobacilli</td>
<td>0.9 ± 1.7 (0.1) [88]</td>
<td>2.6 ± 3.4 (0.9) [89]</td>
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<tr>
<td>Actinomyces</td>
<td>1.1 ± 1.5 (0.3) [100]</td>
<td>0.5 ± 0.8 (0.2) [92]</td>
</tr>
<tr>
<td>F. nucleatum</td>
<td>1.2 ± 1.7 (0.7) [97]</td>
<td>0.9 ± 2.2 (0.1) [73³]</td>
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<tr>
<td>P. intermedia/nigrescens</td>
<td>2.9 ± 5.1 (0.8) [79⁹]</td>
<td>1.2 ± 1.6 (0.6) [69]</td>
</tr>
<tr>
<td>P. gingivalis</td>
<td>0.1 ± 0.2 (0.0) [12]</td>
<td>0.4 ± 1.8 (0.0) [4]</td>
</tr>
<tr>
<td>Actinomycetemcomitans</td>
<td>0.0 ± 0.0 (0.0) [0]</td>
<td>0.0 ± 0.0 (0.0) [0]</td>
</tr>
<tr>
<td>C. albicans</td>
<td>0.04 ± 0.11 (0.00) [55]</td>
<td>0.15 ± 0.28 (0.00) [54]</td>
</tr>
<tr>
<td>S. aureus</td>
<td>0.00 ± 0.01 (0.00) [24⁴]</td>
<td>0.01 ± 0.01 (0.00) [35]</td>
</tr>
<tr>
<td>Enteric rods</td>
<td>0.00 ± 0.00 (0.00) [12]</td>
<td>0.98 ± 4.04 (0.00) [19]</td>
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</table>

⁴Difference between the study and control groups, P=.01.
³Difference within the study group from the baseline to the 12-month examination, P<.0001.
⁵Difference between the study and control groups, P=.03.
³Difference within the study group from the baseline to the 12-month examination, P=.007.
⁶Difference between the study and control groups, P=.02.
⁷Difference between the study and control groups, P=.006.
⁸Difference between the study and control groups, P=.007.
Proportion of total number of Streptococci.

Total streptococci
The proportion of streptococci in the supragingival plaque was higher in the study group compared to that in the control group at the 12-month follow-up (P=.01). The increase in the proportion of streptococci in the supragingival plaque samples in the study group (P=.001) occurred from the baseline to the 3-month registration. The proportion of streptococci in supragingival plaque in the study group increased from 26% ± 21% at the baseline to 65% ± 55% at the 12-month examination. No corresponding differences were detected in the samples collected from the tongue.

Bacteria associated with good oral health
At the 12-month examination, the number of S. sanguinis/oralis in the supragingival plaque samples was lower in the study group compared to that in the control group (P=.02). Within the study group, both the number of S. sanguinis/oralis and their proportion of the total number of streptococci in the supragingival plaque decreased (P=.008 and P<.0001, respectively). The decrease in number of S. sanguinis/oralis occurred from the 6-month to the 12-month registration and in proportion from the 6-month to the 9-month registration. No corresponding differences for S. sanguinis/oralis were detected in samples collected from the tongue.

For S. salivarius, no significant differences were detected between the study and control groups regarding the number, proportion or frequency in the supragingival plaque or tongue samples during the study period.

Acidogenic and aciduric bacteria
At the 12-month examination, there were no significant differences between the study and the control groups for mutans streptococci, lactobacilli and Actinomyces in the supragingival plaque and for mutans streptococci and lactobacilli in samples collected from the tongue. The lower proportion of lactobacilli in the supragingival plaque in the study group, as compared to that in the control group at the baseline, was thus levelled out. The proportion of lactobacilli in the supragingival plaque increased (P=.007) gradually over time.

Bacteria associated with periodontal diseases
The frequency of F. nucleatum in the study group was lower in both the supragingival plaque and the tongue samples compared to that in the control group at the 12-month examination (P=.02 and P<.0001, respectively). In the supragingival plaque, the number of F. nucleatum in the study group was lower compared to that in the control group (P=.0004). For the tongue samples, both the number and proportion...
of *F. nucleatum* were lower in the study group compared to that in the control group (*P* < .0001 and .03, respectively), with a gradual decrease over the study period.

For *P. intermedia/nigrescens*, which showed a higher frequency, number and proportion in the supragingival plaque in the study group at the baseline, there were no statistically significant differences at the 12-month examination between the study and control groups, neither in the supragingival plaque nor in samples collected from the tongue.

*P. gingivalis* was a rare finding and detected in the study group only. At the baseline examination, *P. gingivalis* was detected in both the supragingival plaque and the samples collected from the tongue in three participants and in one participant in samples collected from the tongue only. In 1 participant, *P. gingivalis* was detected in the supragingival plaque from the 3-month examination and throughout the study period.

*Aggregatibacter actinomycetemcomitans* was not detected in any of the participants included in the study.

Opportunistic micro-organisms

No statistically significant differences regarding frequency, number or proportion of *C. albicans*, *S. aureus* and enteric rods could be revealed between the study and control groups at the 12-month examination. *C. albicans* was the most frequently detected and showed only small variations over time, between 50% and 57% in the supragingival plaque and between 27% and 42% in the tongue samples.

### DISCUSSION

In the present study, weekly professional oral hygiene care in dentate, dependent elderly residents resulted in a decreased plaque score and

| TABLE 3 | Tongue samples. Total viable microbial count (log 10) and proportions of streptococci, lactobacilli, *F. nucleatum*, *P. intermedia/nigrescens*, *P. gingivalis*, *C. albicans*, *S. aureus* and enteric rods. Mean, standard deviation, median (in parenthesis) and detection frequency (in square brackets) are given |
|---------|------------------|------------------|
| Study group | Control group | Study group | Control group |
| **Baseline** | **12 months** | **Baseline** | **12 months** |
| **Total viable count** | 7.05 ± 0.62 (7.05) [100] | 6.5 ± 0.9 (6.4) [100] | 6.94 ± 0.59 (6.79) [100] | 6.89 ± 0.65 (7.01) [100] |
| **Proportion** | | | | |
| **Streptococci** | 35.2 ± 36.9 (23.6) [100] | 51.6 ± 37.3 (46.8) [100] | 37.6 ± 27.3 (35.9) [100] | 47.0 ± 33.4 (38.8) [100] |
| **S. sanguinis/oralis** | 2.1 ± 6.1 (0.0) [42] | 0.4 ± 1.1 (0.0) [19] | 2.2 ± 5.9 (0.0) [36] | 0.9 ± 2.4 (0.0) [27] |
| **S. salivarius** | 31.0 ± 25.7 (28.9) [97] | 22.7 ± 17.3 (24.8) [89] | 23.7 ± 22.7 (18.9) [90] | 23.4 ± 20.0 (19.1) [97] |
| **Mutans streptococci** | 6.7 ± 14.5 (0.6) [94] | 4.6 ± 12.6 (0.2) [77] | 5.7 ± 11.3 (0.3) [94] | 1.5 ± 3.6 (0.1) [70] |
| **Lactobacilli** | 0.3 ± 0.5 (0.1) [84.8] | 2.3 ± 5.4 (0.3) [85] | 0.9 ± 2.0 (0.0) [86] | 1.9 ± 8.4 (0.1) [87] |
| **F. nucleatum** | 1.8 ± 5.8 (0.4) [97] | 0.3 ± 0.5 (0.1) [42] | 2.4 ± 4.3 (0.4) [89] | 0.8 ± 1.1 (0.3) [90] |
| **P. intermedia/nigrescens** | 1.4 ± 6.1 (0.0) [58] | 0.5 ± 1.9 (0.0) [39] | 0.3 ± 0.6 (0.0) [46] | 0.4 ± 0.7 (0.0) [52] |
| **P. gingivalis** | 0.1 ± 0.1 (0.0) [8] | 0.2 ± 1.0 (0.0) [4] | 0.0 ± 0.0 (0.0) [0] | 0.0 ± 0.0 (0.0) [0] |
| **C. albicans** | 0.01 ± 0.02 (0.00) [42] | 0.03 ± 0.08 (0.00) [31] | 0.01 ± 0.02 (0.00) [31] | 0.01 ± 0.04 (0.00) [33] |
| **S. aureus** | 0.00 ± 0.02 (0.00) [42] | 0.01 ± 0.04 (0.00) [19] | 0.01 ± 0.04 (0.00) [34] | 0.02 ± 0.07 (0.00) [20] |
| **Enteric rods** | 0.00 ± 0.00 (0.00) [9] | 0.00 ± 0.01 (0.00) [8] | 0.01 ± 0.04 (0.00) [17] | 0.08 ± 0.32 (0.00) [10] |

<table>
<thead>
<tr>
<th>TABLE 4</th>
<th>Frequency of opportunistic micro-organisms in supragingival plaque and the dorsum of the tongue in the study group</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Month</strong></td>
<td><strong>Frequency (%)</strong></td>
</tr>
<tr>
<td><strong>After baseline</strong></td>
<td><strong>N</strong></td>
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<tr>
<td><strong>Supragingival plaque</strong></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>33</td>
</tr>
<tr>
<td>3</td>
<td>31</td>
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<tr>
<td>6</td>
<td>30</td>
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<tr>
<td>9</td>
<td>30</td>
</tr>
<tr>
<td>12</td>
<td>26</td>
</tr>
<tr>
<td><strong>Dorsum of the tongue</strong></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>33</td>
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<tr>
<td>3</td>
<td>31</td>
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<td>6</td>
<td>30</td>
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<td>9</td>
<td>30</td>
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<td>12</td>
<td>26</td>
</tr>
</tbody>
</table>

N, number of participants sampled.

The number and proportion of *P. intermedia/nigrescens* in the supragingival plaque were decreased in the study group (*P* = .002 and .02, respectively). In the control group, the number was increased (*P* = .02) and the proportion showed a tendency to be increased (*P* = .07) in the supragingival plaque.

*P. gingivalis* was a rare finding and detected in the study group only. At the baseline examination, *P. gingivalis* was detected in both the supragingival plaque and the samples collected from the tongue in three participants and in one participant in samples collected from the tongue only. In 1 participant, *P. gingivalis* was detected in the supragingival plaque from the 3-month examination and throughout the study period.

*Aggregatibacter actinomycetemcomitans* was not detected in any of the participants included in the study.

**Opportunistic micro-organisms**

No statistically significant differences regarding frequency, number or proportion of *C. albicans*, *S. aureus* and enteric rods could be revealed between the study and control groups at the 12-month examination. *C. albicans* was the most frequently detected and showed only small variations over time, between 50% and 57% in the supragingival plaque and between 27% and 42% in the tongue samples.
a decrease in bacteria associated with periodontal diseases. The proportion of bacteria with acidogenic and aciduric potential, as well as the frequency of micro-organisms associated with oral mucosal infections and aspiration pneumonia, was about unaffected. Further, there was a decrease over time in the proportion of bacteria associated with good oral health.

The microbial samples were analysed using a cultivation technique to make it possible to calculate proportions of specific microbial species of the total viable count in a sample and to have the possibility to save strains for further studies. To improve the validity of the microbial results, especially for those usually present in small numbers when detected, sampling for microbial analysis was performed in duplicate, 1 week apart.

This study’s decision to include only dentate residents, with ≥10 teeth and no removable dentures, was based on previous studies reporting that tooth loss and edentulism are declining in the elderly population and are reflected in dependent elders living at nursing homes.

A contributing factor to the relatively high number of natural teeth in the participants included in the present study, 22 ± 5 teeth, could be that the elders living in the two nursing homes were recruited from a socio-economic, fairly well-situated area of the city. There are several indications of a positive correlation between increased prosperity and an improved dental status.

In dentated elderly dependent residents, however, oral hygiene has been found to worsen with an increasing number of natural teeth. Willumsen et al. found that having more than 10 natural teeth was a predicting factor for having unacceptable oral hygiene. The finding in the present study of a visible thick plaque at the baseline in 50% of the included residents is in congruence with that recorded in the study by Willumsen et al. and in our previous study.

An additional rational to include only dentate residents, with ≥10 teeth and no removable dentures, was that the effect of interventions on oral hygiene and oral flora can be expected to show significant differences in groups with different dental status. In the study by Grimoud et al., they found the best effect of the intervention in edentulous participants, second in participants with full dentures, followed by those with natural teeth only and worst in dentate participants with removable dentures.

In the present study, caries assessment was limited to the presence of open coronal and root surface caries cavities, readily visible to the naked eye. Open cavities are likely to influence the microbial flora in the dental biofilm. It is the authors’ experience that caries incidence registration in severe functionally impaired residents, in the last period of life, will involve too many difficulties to justify inclusion in the study.

In a review performed by Deery et al. on the effectiveness of electric vs manual toothbrushes, it was concluded that electric toothbrushes with rotation oscillation action, such as the toothbrush used in the present study, are more effective in reducing plaque and gingivitis. The no-additional effect on the plaque score and microbial load in those in the study group using the electric toothbrush for their daily oral hygiene care vs those who did not may be due to the few participants who preferred a manual toothbrush for their daily oral care and the effect of the weekly professional oral hygiene care.

In the present study, it was not until the 6-month examination that an effect of the intervention on the dental plaque score could be noted. A delayed effect of the intervention of an oral hygiene programme in dependent elderly residents was noted also in a previous study.

At the 12-month registration, a plaque score ≤1, indicating “no visible supragingival plaque” or a “visible but thin plaque,” was recorded in 92% in the study group and in 13% in the control group. With the same dental hygienist involved in both the effectuation of the weekly oral hygiene care and performing the plaque score registrations in both the study and the control groups (author KL K), the validity of the plaque registrations could be questioned. An improvement of the oral hygiene in the study group was, however, supported by an increase in the proportion of streptococci in the supragingival plaque, which was noted already at the 3-month examination. The streptococci adhere to the cleaned tooth surface in the first phase of the biofilm development.

The proportion of streptococci in the supragingival plaque increased from 26% ± 21% at the baseline to 65% ± 55% at the 12-month examination (P<.001). The decrease over time of F. nucleatum and P. intermedia/nigrescens in samples collected from both the tongue and supragingival plaque, as compared to that in the control group, further supported an improved oral hygiene during the 12-month study period. F. nucleatum and P. intermedia/nigrescens are both later colonisers and require the anaerobic milieu created by the facultative anaerobic streptococci.

The proportion of S. sanguinis/oralis in the supragingival plaque of the study group was, at the end of the study, decreased (P<.0001) and lower than that in the control group (P=.02). No significant differences for mutans streptococci, Actinomyces and opportunistic micro-organisms between the study and the control groups could be detected. Also, there was no significant difference for lactobacilli between the study and the control groups at the 12-month examination. The proportion of lactobacilli, which was lower in the supragingival plaque in the study group, as compared to that in the control group, at baseline (P=.03), was increased during the study period (P=.007). The results are in accordance with results obtained in our previous studies on participants with primary Sjögren’s syndrome and participants who had received radiation therapy due to cancer of the head and neck region. Both groups had a substantially reduced salivary secretion rate. However, in spite of a plaque score closely similar to their healthy controls, matched according to gender, age and number of teeth, the composition of their oral microbial flora was characterised by bacteria with an acidogenic and acidic potential and a frequent occurrence of opportunistic micro-organisms.

The no effect on the proportion of acidogenic and acidic bacteria and the frequency of opportunistic micro-organisms may, at least partly, explain the relatively limited effect of weekly professional assistance with the oral hygiene on the mortality rate in aspiration pneumonia among elderly dependent residents.

The great majority of the participants in the present study had a daily intake of six or more prescription medicines, mean number
9.4 ± 4.5. Prescription medications to manage age-related health conditions, for example psychological conditions and cardiovascular disease, of which several will give a reduced salivary secretion rate as a side effect, are widely used in the elderly population. In our previous study on dentate dependent elders, recruited from the same area of the city as those included in the present study, the most frequently used prescribed medications were psychotropics/neuroleptics, in 80% of the participants.\(^\text{30}\) The mean number of medications prescribed per participant, with hyposalivation as a possible side effect, was two. In the study by Shetty et al.,\(^\text{41}\) a statistically significant correlation between the use of xerostomia-inducing medicine and hyposalivation was found. It is thus plausible that the reason for a low salivary secretion rate in the vast majority in the participants in the present study was due to prescription medications.

The study by Perceval et al.\(^\text{42}\) found the unstimulated whole salivary secretion rate in a group of unmedicated ≥80-year-olds to be lower than that in younger age groups, although the mean value was above normal (0.1 mL/min). The stimulated parotid salivary secretion rate in the unmedicated ≥80-year-olds was, by contrast, not significantly different from that in the younger age groups. The labial minor gland salivary secretion rate was measured as described by Eliasson et al.\(^\text{22}\) and shown to be positively correlated to unstimulated and stimulated whole salivary flow rate with no visible effects of increasing age.\(^\text{53}\) Healthy elders, 65-89 years of age, have been found to have a labial minor gland salivary secretion rate >4.0 μL/cm²/min.\(^\text{22}\) In the present study 76% of the participants had a labial minor gland salivary secretion rate <4.0 μL/cm²/min.

At normal conditions, bicarbonate, the principal buffer in saliva,\(^\text{44}\) is positively correlated to the stimulated secretion rate.\(^\text{45}\) In individuals with hyposalivation due to medication, radiation therapy of the head and neck region, primary Sjögren’s syndrome and for unknown reasons, the concentration of bicarbonate was found to be about 50% of that at normal conditions irrespective of the salivary secretion rate.\(^\text{46}\) The concentration of bicarbonate in the saliva was not analysed in the present study but could, in congruence, be expected to be low and to be a major contributing factor to the non-effect on the acidic and acidogenic bacteria and opportunistic microorganisms of the weekly professional oral hygiene care. The results obtained in a recently completed study\(^\text{15}\) support the importance of a normal salivary secretion rate and buffer capacity for an oral microbical flora associated with healthy conditions. In the 3-year follow-up study of individuals after completing radiation treatment of the head and neck region, a "normalisation" of the oral flora was revealed in individuals only who had regained a normal salivary secretion rate (≥1.0 mL/min) and buffering capacity (≥6.0) during the study period. A steady increase in bacteria, with acidogenic and aciduric potential and opportunistic micro-organisms, was observed over time in individuals with a persisting salivary secretion rate and buffering capacity below "normal." Furthermore, in an in vitro study using a mixed culture chemostat system, Bradshaw et al.\(^\text{47}\) found that (compared to a good oral health), a high proportion of acidogenic and aciduric micro-organisms was due to a decrease in pH, caused by microbial metabolism of carbohydrates, rather than the availability of carbohydrates per se. When buffering substances were added, enough to keep the pH neutral, no effect of the addition of glucose was detected. It is thus plausible that, in dentate, dependent elderly residents with low salivary secretion rate, a good oral hygiene level has to be supplemented with strategies improving the buffer capacity to regain an oral microbial flora associated with healthy oral conditions.

5 | CONCLUSION

The results indicate that assisted oral hygiene care alone is not sufficient to regain an oral microbial flora associated with good oral health in dentate, dependent elderly residents with low salivary secretion rate.

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