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# Plaque inhibitory effect of a 0.05% cetyl-pyridinium chloride mouth-rinse in a 4-day non-brushing model

**Abstract:** *Objectives:* Results from clinical studies evaluating the efficacy of the adjunctive use of cetyl-pyridinium chloride (CPC) containing oral hygiene products have shown wide variability, probably due to differences in formulations. The objective of this study was to determine the inhibitory plaque effect of a 0.05% CPC mouth rinse in *de novo* plaque formation in a 4-day non-brushing experimental model. *Materials and methods:* The study was designed as a short-term double-blind randomized cross-over experimental model aimed to compare three products: a negative control (similar to the test product, without active ingredients), a positive control (with 0.12% chlorhexidine and CPC) and the test product (with 0.05% CPC) in terms of plaque index, gingival inflammation and microbiological variables. *Results:* Plaque levels after 4 days were 2.88 for the positive control, 3.86 for the negative control and 3.60 for the test. Differences among groups on day 4 were statistically significant ($P < 0.001$). Gingival index showed comparable values at baseline ($P = 0.745$), and significant increases were observed, with the exception of the positive control. Total colony forming units showed comparable values at baseline ($P = 0.125$) and significant increases were observed only in the negative control. *Conclusions:* The tested 0.05% CPC mouth-rinse is capable of inhibiting plaque formation.

**Key words:** cetyl-piridinium chloride; chlorhexidine; mouth-rinse; plaque

## Introduction

The prevalence of plaque-associated gingivitis is high. It has been estimated to affect 82% of the children and adolescent populations and nearly half of the adult population (1). The importance of gingivitis is that it can precede periodontitis, although not all gingivitis will lead to the development of periodontitis. Epidemiological studies show that periodontitis has lower prevalence than gingivitis, ranging from 30 to 50%, although this variability may depend on the case definition used to describe the destructive forms of periodontal diseases (1). In addition, recent studies demonstrate a significant association between periodontitis and systemic diseases (2), so the importance of preventing gingivitis and periodontitis is further highlighted.

The prevention of gingivitis is based on supragingival plaque control, which may also help to prevent periodontitis in susceptible subjects. The maintenance of good oral hygiene is then of paramount importance, in spite of epidemiological studies demonstrating that most part of the population does not perform adequate mechanical hygiene control (3). This

fact has led to the rise in interest in the development of oral hygiene products based on chemical plaque control, especially those effective antimicrobial mouth-rinses that may prevent gingivitis development by making an impact on the supragingival and subgingival colonization of teeth by oral bacteria (3).

Oral hygiene products for chemical plaque control must be scientifically evaluated to prove their efficacy. However, as suggested by Addy *et al.* (4), studies attempting to assess the effects of mouth-rinses on plaque formation are hampered not only by the number of components in the formulation, but also by the mechanical action of the toothbrush delivery method. Short-term study methods, measured in hours, largely overcome this tooth brushing effect and are relevant, as by demonstrating significant reductions in plaque formation after such shorts periods (16–94 h) in absence of oral hygiene, they are able to show the true chemical antiplaque effect of the tested product (3).

Among the antimicrobial agents most commonly used, cetyl-pyridinium chloride (CPC) is a monocationic member of the quaternary ammonium family, which is adsorbed easily on oral surfaces but has a limited substantivity (5). Cetyl-pyridinium chloride is a cationic surface-active agent with a broad antimicrobial spectrum capable of killing Gram-positive pathogens and yeast in particular. It is suggested that interaction with bacteria occurs by the disruption of membrane function, leakage of cytoplasmic material and ultimately the collapse of the intracellular equilibrium (6). As required by the guidelines of the American Dental Association (7), it has a minor effect on the composition of the normal oral microbiota. Data on the adjunctive effect of CPC reported at a recent systematic review (8) demonstrated a statistically significant improvement in plaque and gingival indexes. Cetyl-pyridinium products have demonstrated effectiveness and safety as a plaque inhibitory agent in a range of concentrations between 0.045 and 0.1%, but there is a great variety in the results depending on the formulations (9). Based on the different effects of different CPC formulations, it seems justified to evaluate the clinical and microbiological effects of new CPC formulations, such as the one investigated in the present research.

Therefore, the main objective was to determine if a 0.05% CPC mouth-rinse is capable of inhibiting plaque accumulation in a formulation with zinc lactate, permethol and provitamin B5, as compared with a placebo mouth-rinse, in a 4-day non-brushing model in healthy volunteers. As a secondary objective, the microbiological impact was also assessed and the gingival index was evaluated as a control variable.

## Materials and methods

### Participants

Volunteers, with ages ranging between 20 and 30 years were selected by two researchers among students from the Faculty of Odontology at the University Complutense of Madrid, after demonstrating good oral hygiene and gingival health and being willing to adhere to the study protocol. Subjects were excluded if referring any relevant systemic disease, presenting an untreated oral condition, having moderate-to-severe gingivitis (defined as Lobene gingival index ≥1.75 in the Ramfjord teeth) or periodontitis, or if they had been taking antibiotics or using antimicrobials 1 month before the beginning of the study.

All eligible volunteers were informed about the objectives and the protocol of the study by the researchers and agreed to participate by signing an informed consent, approved by the institutional ethics committee (protocol 194-2007), in accordance with the ethical principles that have their origin in the Declaration of Helsinki and are consistent with good clinical practice.

### Experimental design

The study was designed as a short-term double-blind, randomized, cross-over and plaque regrowth (non-brushing) experimental study. This study design was proposed by Addy *et al.* (4) and aims to detect the effect of antimicrobial products on new dental plaque formation in the absence of mechanical oral hygiene. It consisted of the use of the tested products by the same subject during a period of 4 days when all mechanical oral hygiene practices were ceased. After this period, the volunteers were examined and outcome measurements registered. To avoid the carry-over effects, after each test period the subject entered a washout time of at least 1 week, a longer period than the one used by other authors (10–13), during which mechanical oral hygiene was resumed and then, another 4-day test interval commenced with a new assigned product. The study had a randomized double-blind design as neither the volunteer nor the investigators were aware of the composition of the products, identified by codes kept by the study promoter and only opened when the study was finished. Three formulations were compared: a negative control (a placebo similar to the test product but without active ingredients), a positive control (Perio-Aid tratamiento®, Dentaid, Cerdanyola, Barcelona, Spain, which included 0.12% CHX and 0.05% CPC, as active ingredients) and the test product (Vitis Encías®, Dentaid) formulated with 0.05% CPC as the main active ingredient and also containing zinc lactate, permethol and provitamine B5. The product assignment was carried out following a bloc randomization order through a computer-generated sequence, associated with the number of the selected subjects when entering the study. The researchers were unaware of the product allocation.

### Outcome variables

Measurement of plaque index (PlI) on day 4 of each experimental period was considered the main outcome variable. New plaque formation was measured by the Turesky modification of the Quigley-Hein index, in six sites per tooth (14) after disclosure of dental plaque with erythrosine (Plac Control®, Dentaid). The Lobene index (15) was used to score gingival inflammation (GI) on days 0 and 4 and considered as a

secondary variable as it may be affected by plaque accumulation and it was also used to assure the healthy gingival conditions at the start of every study period. Both indexes were evaluated on all teeth present, excluding the third molars.

Subgingival microbiological samples were taken at the beginning and at the end of each experimental period.

The volunteer's compliance and occurrence of adverse effects were evaluated at the end of each study interval through a questionnaire and by the measurement of the product remaining in the returned bottles.

Two trained and calibrated examiners registered all the variables. Table 1 shows the chronogram of one experimental week of the study.

#### Interventions

On Monday morning of each experimental week, subjects received professional prophylaxis and tooth polishing until all plaque, stain or calculus were removed. Then, the gingival index was scored and, finally, subgingival microbiological samples were taken.

Subjects were then asked to stop mechanical oral hygiene until Friday afternoon and to rinse with 15 ml of the assigned product for 30 s twice daily (after breakfast and dinner).

On Friday morning, plaque index, gingival index, adverse effects and compliance were evaluated, in the enumerated order, and subgingival samples were taken after gingival index assessment. Subjects were then allowed to resume their usual oral hygiene practices. After each experimental week, there was a washout period of at least 1 week.

#### Microbiological methods

Microbiological samples were taken using two sterile standard-sized paper points (Maillefer, Ballaigues, Switzerland) that were inserted consecutively in each selected site, two posterior and two anterior buccal sites, after the removal of supragingival plaque (16). Before the insertion of the paper points, sites were isolated with cotton rolls to avoid saliva contamination and the area was dried with the syringe from the dental chair. Paper points were kept in place for 10 s and then they were pooled in a screw top vial containing 1.5 ml of reduced transport fluid (17). Samples were transferred to the laboratory within 2 h where they were homogenized by vortex vibration for 30 s (18) and sequentially diluted in Phosphate-buffered solution. Samples were cultivated on agar-blood medium (enriched with haemine and menadione) and incubated for 15 days in jars with an anaerobic atmosphere; and on selective medium *Dentaid*-1, samples were incubated for 3–5 days in 5% carbon dioxide (19). Bacterial species identification was carried out by the assessment of the colony morphology and confirmed by the application of biochemical standard tests. In addition to the conventional evaluation of the plates, the possible overgrowth of opportunistic species, both in blood agar and in selective plates, was investigated. The main microbiological outcome variables included total anaerobic counts and the presence, counts and proportions of different bacterial species, including opportunistic species to detect possible undesired microbiological adverse effects.

#### Statistical analysis

The mean plaque index, standard deviation and 95% confidence intervals (CI) on day 4 were calculated per subject and per treatment. First, it was checked whether this outcome variable was normally distributed and if there were no statistically significant differences between the variances (*F*-test). The analysis of variance was used to compare the different products, using the treatment as factor and patient, sequence of treatment and baseline gingival index as cofactors. The multiple range test was used as the *post hoc* test.

For gingival index and log of total counts of colony forming units (CFU) per ml, baseline and final values were calculated for each group and distributions were checked for normality. ANCOVA was used for intergroup comparison at baseline, at 4 days and in changes, with product as factor and patient, order and baseline values as cofactors. For intragroup comparison, a paired *t*-test was used.

Due to the lack of previous studies with the same product, a proper sample-size calculation could not be made. Based on similar studies, a sample size of 15 patients was considered adequate. After the study, a power calculation was performed, showing an 85% of power to detect a difference of 0.50 in plaque index between the test and the negative control, considering a standard deviation of 0.66 and a clinically relevant difference a 15% of reduction.

Table 1. **Chronogram of an experimental week**

| Day | Monday | Tuesday | Wednesday | Thursday | Friday |
|---|---|---|---|---|---|
| Morning | Prophylaxis<br>Gingival index<br>Micro samples<br>OH cessation | Use 2 | Use 4 | Use 6 | Use 8<br>Plaque index<br>Gingival index<br>Micro samples<br>Compliance<br>Adverse effects |
| Night | Use 1 | Use 3 | Use 5 | Use 7 | |

OH, mechanical oral hygiene procedures; Micro, microbiological.

## Results

### Experimental population and calibration of the examiners

Figure 1 shows a flow chart of the participants who were enrolled in the study. The final sample was composed of six men and nine women aged from 22 to 29 years (mean age 26.6). All subjects attended all scheduled visits and complied with product usage in, at least, seven of the eight programmed uses.

Only one of the 45 microbiological samples had to be discarded due to technical problems (a baseline sample of patient using the test product). The final sample of that study period was not included in the result, but the other samples from the same patient were included. Thus, an intention-to-treat analysis was used for clinical variables and a per protocol analysis was selected for microbiological variables.

Calibration between examiners was carried out for both GI and PlI at baseline and at the end of the study, and kappa values were calculated. Regarding the intra-examiner calibration, examiner 1 scored 0.68 for GI and 0.65 for PlI, and examiner 2 scored 0.42 and 0.41 respectively. For the inter-examiner calibration, after the study, a value of 0.59 was calculated for GI (85% of agreement), and 0.68 for PlI (80%). The overall agreement was therefore considered good.

### Clinical outcomes

On day 4, plaque scores were 3.86, 2.88 and 3.60 for negative control, positive control and test groups, respectively, as shown in Table 2a. A statistically significant effect of the treatment group was observed ($P = 0.0003$, ANCOVA), and no effect of the co-variables patient, order of use and baseline gingival index. The *post hoc* test showed statistically significant differences among all the groups ($P < 0.05$), as shown in Table 2b.

Baseline GI levels were similar for all groups, ranging 0.44–0.50 ($P = 0.745$). Conversely, after 4 days, values showed a tendency towards significance ($P = 0.062$): 0.74 for the negative control; 0.49 for the positive control; and 0.67 for the test group (see Tables 3a and b). Differences were identified

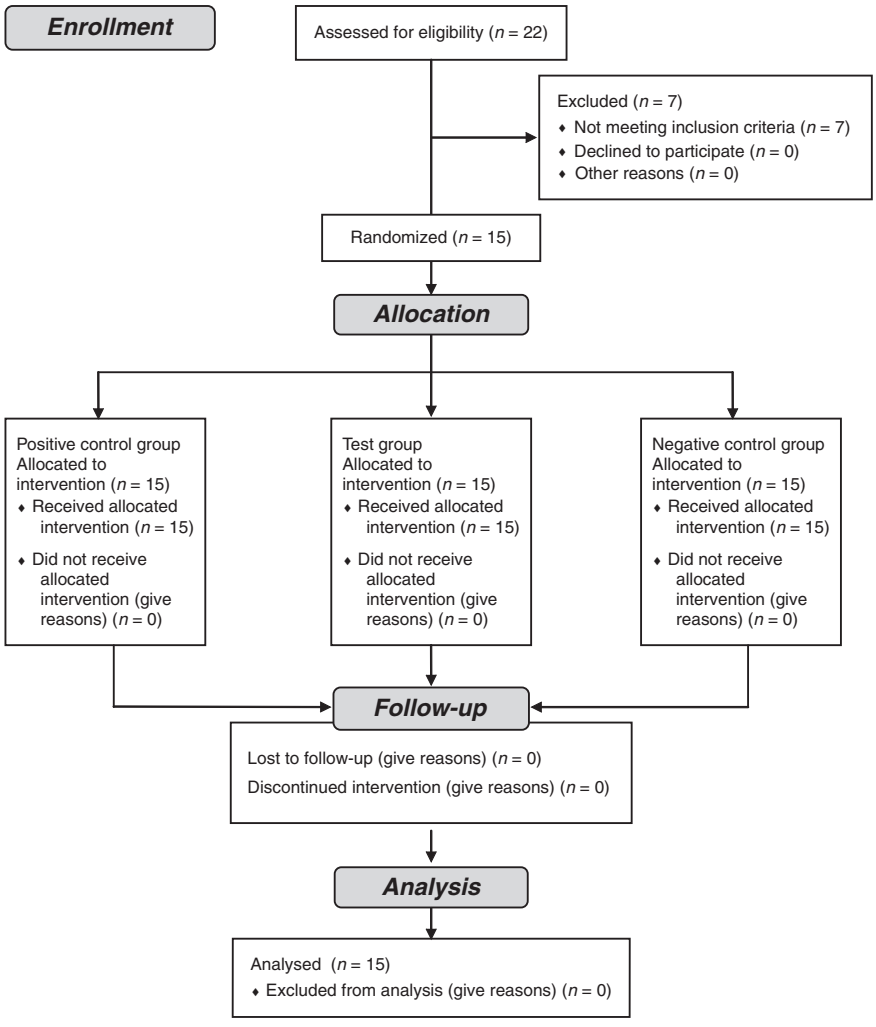


*Fig. 1.* Flow chart.

Table 2. (a) **Mean levels of plaque (with standard deviation and 95% CI) after 4 days. The three groups were compared by means of ANCOVA (with patient, order of sequence and baseline gingival index as covariables). No significant effect of the covariables was observed. (b) Mean differences in plaque levels between groups (with standard deviation and 95% CI) after 4 days. Multiple range test was used as *post hoc* test. A positive value means a higher value of the first group mentioned**

| Plaque index | *n* | Mean | Standard deviation | Lower CI | Upper CI | ANCOVA *P*-value |
|---|---|---|---|---|---|---|
| (a) | | | | | | |
| Negative control | 15 | 3.86 | 0.51 | 3.58 | 4.15 | 0.0003 |
| Positive control | 15 | 2.88 | 0.70 | 2.49 | 3.27 | |
| Test | 15 | 3.60 | 0.68 | 3.22 | 3.97 | |
| | | | | | | Multiple range test |
| (b) | | | | | | |
| Negative *versus* positive control | 15 | 0.98 | 0.76 | 0.56 | 1.40 | $P < 0.05$ |
| Negative control *versus* test | 15 | 0.27 | 0.66 | −0.09 | 0.63 | $P < 0.05$ |
| Positive control *versus* test | 15 | −0.71 | 0.84 | −1.18 | −0.24 | $P < 0.05$ |

CI, confidence intervals.

Table 3. **(a) Mean values at baseline, 4 days and changes (final *versus* baseline, positive values mean increase) in gingival index (with standard deviation and 95% CI). The three groups were compared by means of ANCOVA (with patient, order of sequence and baseline gingival index as covariables). A significant effect of baseline gingival index was observed in changes ($P = 0.001$). (b) Mean differences between groups, at baseline, 4 days and changes (final *versus* baseline) in gingival index (with standard deviation and 95% CI). Multiple range test was used as *post hoc* test. A positive value means a higher value of the first group mentioned**

| Gingival index | *n* | Mean | Standard deviation | Lower CI | Upper CI | ANCOVA *P*-value |
|---|---|---|---|---|---|---|
| (a) | | | | | | |
| Baseline | | | | | | |
| Negative control | 15 | 0.46 | 0.28 | 0.31 | 0.62 | 0.745 |
| Positive control | 15 | 0.50 | 0.15 | 0.41 | 0.58 | |
| Test | 15 | 0.44 | 0.16 | 0.35 | 0.53 | |
| Final | | | | | | |
| Negative control | 15 | 0.74 | 0.28 | 0.58 | 0.89 | 0.062 |
| Positive control | 15 | 0.49 | 0.23 | 0.36 | 0.62 | |
| Test | 15 | 0.67 | 0.34 | 0.47 | 0.85 | |
| Changes | | | | | | |
| Negative control | 15 | 0.27 | 0.40 | 0.05 | 0.49 | 0.047 |
| Positive control | 15 | −0.01 | 0.25 | −0.15 | 0.13 | |
| Test | 15 | 0.22 | 0.29 | 0.06 | 0.39 | |
| | | | | | | Multiple range test |
| (b) | | | | | | |
| Baseline | | | | | | |
| Negative *versus* positive control | 15 | −0.03 | 0.32 | −0.21 | 0.14 | NS |
| Negative control *versus* test | 15 | 0.02 | 0.29 | −0.14 | 0.18 | NS |
| Positive control *versus* test | 15 | 0.06 | 0.20 | −0.05 | 0.17 | NS |
| Final | | | | | | |
| Negative *versus* positive control | 15 | 0.25 | 0.23 | 0.12 | 0.37 | $P < 0.05$ |
| Negative control *versus* test | 15 | 0.07 | 0.38 | −0.14 | 0.28 | NS |
| Positive control *versus* test | 15 | −0.17 | 0.26 | −0.32 | −0.03 | $P < 0.05$ |
| Changes | | | | | | |
| Negative *versus* positive control | 15 | 0.28 | 0.39 | 0.07 | 0.49 | NS |
| Negative control *versus* test | 15 | 0.05 | 0.47 | −0.21 | 0.31 | NS |
| Positive control *versus* test | 15 | −0.23 | 0.30 | −0.40 | −0.06 | NS |

CI, confidence intervals.

between the positive control and the other two groups. Regarding changes baseline–final, significant increases were observed for the negative control (0.27, $P = 0.020$) and the test group (0.22, $P = 0.010$), as compared with minor changes in the positive control (−0.01, $P = 0.870$). Significant differences among groups were detected by ANCOVA ($P = 0.047$).

Table 4. **(a) Mean values at baseline, 4 days and changes (final versus baseline, positive values mean increase) in log of total anaerobic colony forming units (with standard deviation and 95% CI). The three groups were compared by means of ANCOVA (with patient, order of sequence and baseline values as covariables). A significant effect of baseline values was observed in changes (*P* = 0.001). (b) Mean differences between groups, at baseline, 4 days and changes (final *versus* baseline) in log of anaerobic total colony forming units (with standard error and 95% CI). Multiple range test was used as *post hoc* test. A positive value means a higher value of the first group mentioned**

| Log of CFU | *n* | Mean | Standard deviation | Lower CI | Upper CI | ANCOVA *P*-value |
|---|---|---|---|---|---|---|
| (a) | | | | | | |
| Baseline | | | | | | |
| Negative control | 15 | 5.59 | 0.53 | 5.29 | 5.88 | 0.125 |
| Positive control | 15 | 5.57 | 0.71 | 5.17 | 5.96 | |
| Test | 14 | 6.04 | 0.75 | 5.60 | 6.48 | |
| Final | | | | | | |
| Negative control | 15 | 6.49 | 0.31 | 6.32 | 6.66 | 0.000 |
| Positive control | 15 | 5.67 | 0.63 | 5.32 | 6.02 | |
| Test | 14 | 6.47 | 0.50 | 6.19 | 6.76 | |
| Changes | | | | | | |
| Negative control | 15 | 0.89 | 0.53 | 0.60 | 1.19 | 0.000 |
| Positive control | 15 | 0.09 | 0.71 | −0.29 | 0.49 | |
| Test | 14 | 0.44 | 0.88 | −0.07 | 0.95 | |
| | | | | | | Multiple range test |
| (b) | | | | | | |
| Baseline | | | | | | |
| Negative *versus* positive control | 15 | 0.02 | 0.85 | −0.45 | 0.49 | NS |
| Negative control *versus* test | 14 | −0.44 | 0.99 | −1.01 | 0.14 | $P < 0.05$ |
| Positive control *versus* test | 14 | 0.42 | 0.75 | −0.85 | 0.01 | $P < 0.05$ |
| Final | | | | | | |
| Negative *versus* positive control | 15 | 0.82 | 0.63 | 0.47 | 1.17 | $P < 0.05$ |
| Negative control *versus* test | 14 | −0.02 | 0.54 | −0.34 | 0.29 | NS |
| Positive control *versus* test | 14 | −0.86 | 0.90 | −1.38 | −0.34 | $P < 0.05$ |
| Changes | | | | | | |
| Negative *versus* positive control | 15 | 0.80 | 0.92 | 0.29 | 1.31 | $P < 0.05$ |
| Negative control *versus* test | 15 | 0.41 | 1.03 | −0.18 | 1.00 | NS |
| Positive control *versus* test | 15 | −0.44 | 0.83 | −0.92 | 0.04 | $P < 0.05$ |

CI, confidence intervals.

#### Microbiological outcomes

Table 4 shows log of total CFU. At baseline, the test group showed significant higher counts. After 4 days, significant differences existed among groups ($P < 0.001$) that corresponded to statistically significant lower values in the positive control as compared with any of the other two groups. With regard to changes baseline–final, a significant increase was observed in the negative control (0.89, $P < 0.001$), versus a non-significant increase in the test group (0.44, $P = 0.080$), and minor changes in the positive control (0.09, $P = 0.060$). Differences in changes among groups were statistically significant ($P < 0.001$) due to the lower increase in counts in the positive control as compared with any of the other two groups.

There were no relevant changes in the presence of the analysed periodontal pathogens, whereas in all groups an increase in *Prevotella intermedia* and *Fusobacterium nucleatum* levels could be observed (data not shown). No overgrowth of opportunistic species was observed.

#### Adverse effects

No relevant adverse effect was reported.

## Discussion

The results of the study support the bioavailability of the CPC with this formulation (0.05% CPC, zinc lactate, permethol and provitamine B5), as substantiated by the statistically significant difference observed for the plaque index when compared with the negative control group ($P < 0.05$). A recent systematic review found that different CPC formulations may have different clinical effects and some of them may not significantly affect plaque (8). In the present study, the evaluated CPC formulation reduced the plaque index by 7.7% relative to placebo. In comparison with other experimental studies, this result is slightly inferior (10–13, 20, 21), although it should be highlighted that in some of those studies, CPC was used at a higher concentration (0.07%), in combination with other active ingredients, or in a different experimental design, which may

explain these differences. As examples, Versteeg *et al.* (22) proved that 0.07% CPC was capable of reducing plaque formation by 47.4% in comparison with placebo, when using the product three times per day. Moran *et al.* (10) demonstrated that the use of a 0.05% CPC mouth-rinse reduced plaque regrowth by 43.8% in comparison with placebo, but formulated in combination with essential oils. Jenkins *et al.* (11) detected a 19% reduction in plaque, but the washout period was less than 3 days. In addition, in the two last mentioned studies, CPC was formulated with alcohol and that could have had some influence on the obtained results.

CPC is a monocationic antimicrobial agent with demonstrated efficacy as a plaque inhibitor agent in several studies (23–24). There are studies, however, where this efficacy was not demonstrated (25–26). It has been reported that one of the main factors for this heterogeneity is the possible interaction between the active agents and the excipients within the formulation, which may influence the bioavailability of the CPC in a specific product (27). It is also important to realize that the bioavailability of most CPC formulations has not been properly reported. There are other factors that may also explain these differences among the studies, such as the use of different concentrations of CPC (0.07, 0.045 and 0.05%) and the lack of alcohol in some of the evaluated formulations (9).

In 2008, Haps *et al.* published a systematic review in relation to the effects of CPC containing mouth-rinses, when used as adjuncts to either supervised or unsupervised oral hygiene. The results indicated that there is a small but significant additional benefit in reducing plaque accumulation and gingival inflammation with the use of CPC mouth-rinses (8). Study duration was one of the factors evaluated in the quoted study, and the validity of short-term models has been extensively discussed. According to Gunsolley, short-term studies (4 days to 2 weeks) are valid to investigate antiplaque effects (9).

The microbiological results may corroborate the effects of the tested product. A significant increase (0.89, $P < 0.001$) was observed in total bacterial count in the negative control group, whereas the increase in the test group was lower (0.44) and non-statistically significant ($P = 0.080$). Conversely, the positive control with CHX and CPC was even able to decrease the total flora. Both CPC and CHX are known to have antibacterial activity. Moreover, in an *in vitro* experiment, 0.05% CPC and 0.12% CHX were essentially similar in their antibacterial activity (5) even with superior fungicidal properties for 0.05% CPC (28). The microbiological impact of the test mouth rinse was inferior to the positive control, but this is not surprising as the positive control has the added microbiological benefit of 0.12% CHX to the 0.05% CPC. In addition, none of the groups suffered an increase in the opportunistic species demonstrating the safety of the tested product in terms of lacking microbiological adverse effects.

When assessing the control outcome variable, changes in gingival inflammation, no differences were detected at baseline, which favours the validity of the study model, as gingival inflammation may favour plaque accumulation. In addition, the increase of inflammation after 4 days was higher in the negative control (0.27) than in the test group (0.20), whereas minor changes were observed in the positive control.

In the present investigation, a negative and a positive control were used to allow the positioning of the test product between both extremes. The test product showed one-third of the plaque inhibitory effect of the positive control (0.12% CHX with 0.05% CPC). This result was similar to the result reported by Renton-Harper *et al.* (13): a reduction by 40% in relation to the 0.12% CHX was observed. Results obtained by the positive control in our study are in the expected range for the activity of CHX, with significant reductions on the plaque index (25.7%), and the gingival index (96%), when compared with the negative control. Nevertheless, it should be noted that the levels of plaque (mean plaque score on day 4 was 2.86) were higher than those observed for the same product in another study, with a plaque score of 1.5 on day 7 (29).

In conclusion, the results of this study show that the test product was able to reduce plaque formation in a 4-day non-brushing model, this inhibiting capacity being, however, lower than that of a positive control with 0.12% CHX plus 0.05% CPC. Moreover, the test product was well accepted and did not create any adverse effects, either clinical or microbiological.

As the present paper has reported a study model, a home-use study is advised to confirm the results. Longer term home-use studies are needed to evaluate properly the efficacy of this formulation as an adjunct to mechanical plaque control, both on plaque and on gingivitis.

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## Conflict of interest

The authors declare that they have no conflict of interest.

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