

# Gingival bleeding on probing: relationship to change in periodontal pocket depth and effect of sodium hypochlorite oral rinse

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**Background and Objective:** This study evaluated the potential of gingival bleeding on probing to serve as a predictor of future periodontal breakdown. It also assessed the ability of 0.25% sodium hypochlorite twice-a-week oral rinse to convert periodontal pockets showing bleeding on probing to nonbleeding sites.

**Material and Methods:** The study was performed as a randomized, single-blinded, clinical trial in parallel groups. Seven periodontitis patients rinsed twice-weekly for 3 mo with 15 mL of a fresh solution of 0.25% sodium hypochlorite, and five periodontitis patients rinsed with water. The 12 study patients received no subgingival or supragingival scaling. Clorox® Regular-Bleach was the source of sodium hypochlorite. At baseline and 3-mo visits, gingival bleeding was assessed within 30 s after probing to full pocket depth using an approximate force of 0.75 N.

**Results:** A total of 470 (38%) of 1230 periodontal pockets in the bleach-rinse group revealed bleeding on probing at the initial visit but not at the 3-mo visit; only 71 (9%) of 828 pockets in the control group became bleeding-negative during the study ( $p < 0.001$ ). Bleeding on probing in 4- to 7-mm-deep pockets decreased by 53% in the bleach-rinse group but increased by 6% in the water-rinse group ( $p < 0.001$ ). Ninety-seven pockets showed depth increases of  $\geq 2$  mm after 3 mo: 60 (62%) of those pockets exhibited bleeding on probing at both the initial and the 3-mo visits; 24 (25%) bled at only one of the two visits; and 13 (13%) never demonstrated gingival bleeding ( $p < 0.001$ ).

**Conclusions:** Persistent gingival bleeding on probing was associated with an increased risk for periodontal breakdown, and the absence of gingival bleeding seemed to be a useful, although not perfect, indicator of disease stability. Twice-weekly oral rinsing with dilute bleach (0.25% sodium hypochlorite) produced a significant reduction in bleeding on probing, even in deep unscaled pockets. Sodium hypochlorite constitutes a valuable antiseptic in periodontal self-care.

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Periodontitis remains a diagnostic and therapeutic challenge. Dental professionals lack accurate biomarkers to predict ongoing or future progression of periodontal disease, and practitioners often resort to surgery to prevent an anticipated scenario of continued attachment loss. Successful treatment of aggressive/active periodontitis usually requires surgical intervention and/or adjunctive systemic antibiotic therapy (1). On the other hand, as many as 85% of patients with chronic periodontitis may remain disease-stable for 5–6 years after only a one-time scaling (2) or no treatment at all (3), and may derive relatively little benefit from surgical treatment. The availability of a reliable test to assess risk for periodontal disease activity would help to optimize treatment of individual patients and specific periodontal sites.

Gingival bleeding on probing comprises one of the more promising diagnostic predictability tests in periodontics. Claffey *et al.* (4) found loss of probing attachment in 41% of periodontal sites that bled on probing at 75% of recall visits during a 3.5-year observation period. Lang *et al.* (5) reported that periodontal sites which bled on probing at four consecutive maintenance visits showed a 30% risk of losing attachment, whereas sites with bleeding on probing at one out of four consecutive recall visits had only a 3% risk of breakdown, and periodontitis patients who demonstrated gingival bleeding in fewer than 10% of sites in the dentition were at low risk for progressive disease. A meta-analysis showed that persistent bleeding on probing after treatment was associated with progression of periodontitis with an odds ratio of 2.8 (6). Charalampakis *et al.* (7) found that bleeding on probing yielded a better prediction of progressive periodontal disease if combined with quantified bacterial markers. Also, repeated mucosal bleeding on probing around implants may serve as a risk indicator of peri-implantitis (8), and the absence of mucosal bleeding may be an indicator of low risk for implant failure as a result of infection (9).

Other types of diagnostic tests to predict periodontal breakdown have also been evaluated. A high score of 4 in the Community Periodontal Index of Treatment Needs screening system yielded a low positive-predictive value for progressive periodontitis, but low sextant scores of 0–2 provided a presumptive identification of nonprogressive sites (10). The absence of a radiographic lamina dura in angular periodontal defects (11) and in peri-implantitis lesions (12) was an indicator of progressive disease, although with low positive predictability, but periodontal sites with a radiographically intact crestal lamina dura exhibited literally no risk of disease progression for at least 2 years (13). The lack of periodontal disease progression at Ramfjord's six index teeth was suggestive of a low risk of progressive disease in the entire dentition (14). A clinically based periodontal risk calculator failed, largely because of false-positive high-risk scores, to reliably predict progression of periodontal disease during a 3-year post-treatment period (15). Periodontopathic bacteria in subgingival plaque (7,16–18) or saliva (19), or gingival crevice fluid biomarkers (20,21), may also serve as indicators of periodontal disease status. The modest to moderate positive-predictive value of diagnostic tests for progressive periodontal disease are the result of a low incidence of disease-active periodontitis and a relatively high number of false-positive assessments (22). Diagnostic tests may be improved by combining several independent variables from different aspects of the periodontal disease process (7,23), but such an approach remains to be defined and validated in periodontics.

As persistent bleeding on probing provides one of the most robust predictors of periodontal breakdown, and because little information exists on the potential of periodontal self-care to resolve gingival bleeding, this study examined the ability of 0.25% sodium hypochlorite oral rinse, used twice-weekly for 3 mo, to convert gingival bleeding sites to nonbleeding sites in periodontitis patients. The study subjects received no subgingival

or supragingival scaling before or during the study in order to stress-test the boundary of effectiveness of sodium hypochlorite oral rinse.

## Material and methods

The details of the material and methods are outlined elsewhere and are only briefly summarized below (24). The study included 12 periodontitis patients, who completed a controlled clinical trial with sodium hypochlorite oral rinse. The study patients had a mean age of 41 years and an average of 28.6 teeth. Each patient exhibited at least four separate teeth with a pocket depth of  $\geq 6$  mm. The patients were medically healthy and required no emergency dental care. Excluded from the study were individuals who were unable to comply with the research protocol, smoked  $> 10$  cigarettes daily or had received periodontal therapy or systemic antibiotics during the 6 mo before entering the study. The University of Southern California Health Sciences Campus Institutional Review Board approved the study (# HS-10-00509). All patients understood and signed informed consent and HIPAA documents before enrolling in the study.

All study patients received a comprehensive clinical examination and conventional oral hygiene instruction at baseline, but no subgingival or supragingival scaling. Standard periodontal therapy was performed at the conclusion of the study. By random assignment, seven study participants rinsed with sodium hypochlorite (test group) and five rinsed with water (control group). The source of sodium hypochlorite was Clorox® Regular-Bleach (The Clorox Company, Oakland, CA, USA), which contains 6% sodium hypochlorite and is registered as a bactericide, virucide and fungicide agent with the United States Environmental Protection Agency (CAS number 5813-100). Participants in the sodium hypochlorite rinse group were provided with Clorox Regular-Bleach and instructed to mix 5 mL (one teaspoonful) of bleach with 120 mL (one half-glass) of tap water to yield a sodium hypochlorite

concentration of 0.25%. A fresh bleach solution was to be made up at each time of rinsing. The study patients were requested to rinse their mouth for 30 s every Wednesday and Sunday for 3 mo, with either 15 mL of a fresh solution of 0.25% sodium hypochlorite or 15 mL of water, and were given a rinse log to record the exact date and time of rinsing.

A calibrated blinded examiner determined pocket depth and gingival bleeding within 30 s after probing to full pocket depth using a Marquis CP-12 probe (Hu-Friedy Mfg. Co., Chicago, IL, USA) and a probing force of approximately 0.75 N (76 gram-force). Mid-facial, mid-lingual, mesiofacial, distofacial, mesiolingual and distolingual surfaces were examined, and a total of 1230 pockets in the bleach-rinse group and 828 pockets in the water-rinse group were studied.

The statistical analysis was performed using the SPSS-19.0 software program (SPSS Inc., Chicago, IL, USA). The individual pockets were treated as independent statistical units, based on the nonspecific and wide-ranging antimicrobial action of sodium hypochlorite and the observations that pockets with a large range of depths responded positively to the bleach treatment and that residual bleeding on probing sites showed no tendency to cluster in particular patients or around specific teeth. Based on the percentage of periodontal pockets with bleeding on probing and pocket depth changes, the chi-square test was used to identify significant differences between the bleach-rinse and the water-rinse groups. The Spearman's rank correlation coefficient test assessed the relationship between the frequency of bleeding on probing and increased pocket depth. A *p*-value of  $\leq 0.05$  was considered statistically significant.

## Results

The post-study interview and the patient rinse log revealed that the study participants fully understood the instructions for mixing the bleach rinse solution and complied with the

Table 1. Bleeding on probing (BOP) in relation to periodontal pocket depth in patients receiving 0.25% sodium hypochlorite rinse (test) or water rinse (control)

Item	Study time point	Probing pocket depth (mm)											
		1	2	3	4	5	6	7	8	9	10	11	12
Test	Baseline	0/17 (0)	72/239 (30.3)	256/547 (46.8)	130/203 (64.0)	76/81 (93.8)	86/97 (88.7)	19/20 (95.0)	5/5 (100)	22/22 (100)	0/0	0/0	0/0
	3 mo	0/1 (0)	23/357 (6.4)	70/526 (13.3)	37/163 (22.7)	28/62 (45.2)	54/107 (50.5)	5/11 (45.5)	0/1 (0)	0/2 (0)	0/0	0/0	0/0
<i>p</i> -value		0.999	0.001	0.001	0.001	0.001	0.001	0.001	0.167	0.004	–	–	–
Control	Baseline	0/0	4/86 (4.7)	90/265 (34.0)	41/80 (51.3)	53/87 (60.9)	113/130 (86.9)	46/53 (86.8)	22/25 (88.0)	66/82 (80.5)	6/6 (100)	3/3 (100)	11/11 (100)
	3 mo	0/1 (0)	6/316 (1.9)	112/253 (44.3)	20/46 (43.5)	34/50 (68.0)	118/134 (88.1)	31/34 (91.2)	17/18 (94.4)	67/87 (77.0)	1/1 (100)	0/0	6/8 (75.0)
<i>p</i> -value		–	0.231	0.016	0.401	0.407	0.780	0.532	0.628	0.581	0.999	–	0.164

Values are given as no. of bleeding pockets/no. of total pockets at the specific pocket depth (percentage of bleeding pockets at the specific pocket depth) at baseline and at 3 mo, in control and test groups.

Table 2. Periodontal pocket depth changes in relation to changes in bleeding on probing

Pocket depth changes from Visit 1 to Visit 2	No. (%) of pockets with bleeding on probing at Visit 1 but not at Visit 2	No. (%) of pockets with bleeding on probing at Visit 1 and at Visit 2	No. (%) of pockets with no bleeding on probing at Visit 1 and at Visit 2	No. (%) of pockets with no bleeding on probing at Visit 1 but bleeding on probing at Visit 2
Pocket depth reduction of $\geq 2$ mm	Test: 57 (55.9% <sup>a</sup> ) Control: 25 (16.6%) Pooled: 82 (32.4%)	Test: 30 (29.4%) Control: 88 (58.3%) Pooled: 118 (46.6%)	Test: 15 (14.7%) Control: 28 (18.5%) Pooled: 43 (17.0%)	Test: 0 (0%) Control: 10 (6.6%) Pooled: 10 (4.0%)
<i>p</i> -value	0.001	0.001	0.451	0.014
No pocket depth changes (< 2 mm)	Test: 400 (36.5% <sup>a</sup> ) Control: 45 (7.3%) Pooled: 445 (26.1%)	Test: 157 (14.3%) Control: 249 (40.6%) Pooled: 406 (23.8%)	Test: 517 (47.2%) Control: 281 (45.8%) Pooled: 798 (46.7%)	Test: 21 (1.9%) Control: 38 (6.2%) Pooled: 59 (3.5%)
<i>p</i> -value	0.001	0.001	0.887	0.279
Pocket depth increase of $\geq 2$ mm	Test: 13 (39.4% <sup>a</sup> ) Control: 1 (1.6%) Pooled: 14 (14.4%)	Test: 10 (30.3%) Control: 50 (78.1%) Pooled: 60 (61.9%)	Test: 6 (18.2%) Control: 7 (10.9%) Pooled: 13 (13.4%)	Test: 4 (12.1%) Control: 6 (9.4%) Pooled: 10 (10.3%)
<i>p</i> -value	0.001	0.001	0.160	0.489

<sup>a</sup>Percentage of total pockets in the particular Table row. 'Visit 1' denotes baseline and 'Visit 2' denotes termination of the study at 3 mo. Test patients rinsed with 0.25% sodium hypochlorite and control patients rinsed with water.

twice-weekly rinsing guidelines. No adverse events were identified in any of the participants, except for minor complaints about the taste of bleach.

Table 1 details bleeding on probing in relation to periodontal pocket depth. Both the bleach-rinse group with 1230 pockets and the water-rinse group with 828 pockets showed positive correlations at baseline between frequency of gingival bleeding and increased pocket depth ( $p < 0.001$ ). No periodontal site of 1 mm depth bled on probing at any study time point. At baseline, bleeding on probing occurred in 37% of 2- to 3-mm-deep pockets and in 88% of pockets with a depth of  $\geq 6$  mm. At 3 mo, bleeding on probing in pockets of 4–7 mm depth showed a 53% decrease in the bleach-rinse group but a 6% increase in the water-rinse group ( $p < 0.001$ ). At baseline, 27 pockets in the bleach-rinse group had a probing depth of  $\geq 8$  mm and all bled on probing. At the 3-mo visit, the bleach-rinse group revealed the presence of only three pockets of  $\geq 8$  mm, and none showed bleeding. In comparison, 127 pockets in the control group had probing depths of  $\geq 8$  mm at baseline and 108 (85%) bled on probing. At 3 mo, 114 control pockets of  $\geq 8$  mm depth were present and 91 (80%) bled on probing. The difference between the bleach-rinse and the

water-rinse groups in decrease of percentage of  $\geq 8$  mm pockets and bleeding sites was statistically significant ( $p < 0.001$ ).

Table 2 describes how changes in bleeding on probing affected periodontal pocket depth. A total of 470 (38%) periodontal pockets in the bleach-rinse group revealed bleeding on probing at the initial visit but not at the 3-mo visit; only 71 (9%) pockets in the control group became bleeding-negative during the study ( $p < 0.001$ ). Conversely, 197 (16%) periodontal pockets in the bleach-rinse group, but as many as 387 (47%) pockets in the control group, showed persistent bleeding throughout the study ( $p < 0.001$ ). Ninety-seven pockets (5% of the total pockets in the study) increased in probing depth by  $\geq 2$  mm: 60 (62%) of those pockets exhibited bleeding on probing at both the initial and the 3-mo visits; 24 (25%) bled at only one of the two visits; and 13 (13%) never demonstrated gingival bleeding ( $p < 0.001$ ). A total of 25 pockets in the bleach-rinse group (2% of test pockets) developed bleeding on probing during the study; none of those pockets showed a depth reduction of  $\geq 2$  mm, and four (0.3%) pockets experienced a depth increase of  $\geq 2$  mm. No statistically significant changes were found in gingival recession, furcation involvement or tooth mobility, within or between the bleach-

rinse and control groups, during the 3-mo study.

## Discussion

This study sought to assess the proficiency of bleeding on probing to predict progression of periodontal disease and the capability of sodium hypochlorite oral rinse to resolve bleeding on probing in periodontal pockets of different depths. Bleeding on probing is a potentially attractive risk indicator for periodontal breakdown because it is a relatively simple, quick, all-or-none visual testing method that is not greatly dependent on special skills or exceptional tactile sensitivity on the part of a clinical observer. Also, probing of the periodontal pocket depth is already a standard assessment technique in dental practice, requiring no extra training for experienced operators. However, to avoid underestimating gingival bleeding, we probed to the base of periodontal pockets and not just around the gingival margin (25) and used a probing force of approximately 0.75 N instead of the conventional 0.50 N (26). Lang *et al.* (27) showed that an increase in probing force from 0.50 to 0.75 N doubled the percentage of periodontal pockets that bled on probing.

Bleeding on probing occurred in 55% of untreated periodontal pockets in our study compared with 71% (28),

63% (29) and 57% (30) pockets in previous studies. We found that untreated, 3-mm-deep pockets exhibited a frequency of bleeding on probing of 43%, whereas Farina *et al.* (31) reported a frequency of bleeding on probing of 18%. Bleeding on probing scores are elevated with deep pockets and heavy plaque accumulation, and are reduced by cigarette smoking, which may explain some of the differences in the findings across studies (28,31,32). Rather than merely determine an overall correlation between bleeding on probing and progression of periodontal disease, we aimed to quantify, in more detail, the relationship between bleeding on probing and periodontal disease activity. Of the 97 pockets (5% of the total pockets in the study) that showed increased probing depth of  $\geq 2$  mm, disease progression was detected 2.5-fold more frequently in pockets that bled on probing at both the initial and the 3-mo visits (60 pockets) than in pockets that bled on probing at only one of the two visits (24 pockets). Disease progression occurred in only 13 (1.5%) of the 854 pockets that never demonstrated bleeding on probing. The increased rate of periodontal breakdown in periodontal sites having repeat bleeding on probing and the low occurrence of false-negative readings show the benefits of maintaining the periodontium in a nonbleeding state.

Bleeding on probing can decrease substantially after mechanical debridement but, as also shown in the control group of this study, not after conventional oral hygiene alone (28,29,33). Scaling and root planing of 4- to 7-mm-deep pockets has been reported to reduce bleeding on probing scores by approximately 50%, with reductions of 6–64% occurring after 1 mo and reductions of 12–80% occurring after 3 mo (33). However, our study showed that twice-weekly oral rinsing with 0.25% sodium hypochlorite for 3 mo, in the absence of subgingival scaling, also resulted in a reduction in bleeding on probing, of 53%, in pockets of 4–7 mm depth. Kalkwarf *et al.* (32) found that bleeding on probing returned in 35–60% of pockets  $\geq 5$  mm deep during the first

3 mo following nonsurgical or surgical therapy. However, our study revealed that 3 mo of twice-weekly rinsing with sodium hypochlorite/dilute bleach gave rise to a 49% decrease in gingival bleeding in pockets  $\geq 5$  mm deep. The finding that bleeding on probing was resolved in sites with deep probing depths, exposed to sodium hypochlorite oral rinsing, which has not been reported previously, lends substantial credence to the use of sodium hypochlorite in periodontal maintenance care. Further studies are needed to determine if periodontitis patients may experience even less disease progression if rinsing with dilute bleach is combined with scaling and root planing.

The marked decrease in gingival bleeding occurred despite oral rinse only penetrates 0.1–0.2 mm into the subgingival area (34). The improved gingival health after rinsing with sodium hypochlorite was most probably a result of the very low level of supragingival plaque, causing a decrease in the subgingival counts of pathogenic bacteria (35–37). Lobene *et al.* (38) and De Nardo *et al.* (39) found anti-plaque and anti-gingivitis effects of sodium hypochlorite oral rinse similar to those reported here. An attractive feature of the present rinsing protocol is the minimal extra time (30 s, twice weekly) that is imposed on patients' usual oral hygiene effort.

Sodium hypochlorite is a powerful bactericidal and antiviral agent (40), which can even neutralize hard-to-destroy pathogenic prions (41). The antimicrobial action of sodium hypochlorite relies on oxidation or uncoupling of electrons from proteins and nucleotides, and the resulting wide-range damage of infectious agents virtually precludes the development of resistance. Properties of sodium hypochlorite other than plaque removal may also have contributed to the reduction of gingival bleeding (42). Sodium hypochlorite at low concentration was recently shown to interfere with the ability of nuclear factor- $\kappa$ B cellular signaling pathways to activate proinflammatory gene programmes (43). Also, as sodium hypochlorite and essentially all other components of Clorox® Regular-

Bleach occur naturally in the human body (24), dilute bleach oral rinse does not give rise to allergic reactions or other types of disease.

In summary, bleeding on probing tended to persist in periodontal sites with progressive disease, as measured by an increase in probing pocket depth of  $\geq 2$  mm, and to remain absent or discontinued in disease-stable or healing sites. Continued absence or cessation of bleeding on probing appeared to be a useful, but not perfect, indicator of stable periodontal conditions. Twice-weekly oral rinsing with sodium hypochlorite/dilute bleach, coupled with conventional oral hygiene, controlled bleeding on probing more efficiently than conventional oral hygiene alone, and provided a resolution of bleeding on probing even in pockets of considerable depth. The reduction in bleeding on probing in this study, beyond those noted in previous oral rinse studies, was probably the result of substantial antimicrobial, anti-inflammatory and tooth-substantive properties of sodium hypochlorite. Dilute bleach used as an oral rinse offers convenience for patients and deserves serious consideration in periodontal healthcare.

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## References

1. Dentino A, Lee S, Mailhot J, Hefti AF. Principles of periodontology. *Periodontol* 2000 2013;**61**:16–53.
2. Renvert S, Nilvéus R, Dahlén G, Slots J, Egelberg J. 5-year follow up of periodontal intraosseous defects treated by root planing or flap surgery. *J Clin Periodontol* 1990;**17**:356–363.
3. Lindhe J, Haffajee AD, Socransky SS. Progression of periodontal disease in adult subjects in the absence of periodontal therapy. *J Clin Periodontol* 1983;**10**:433–442.
4. Claffey N, Nylund K, Kiger R, Garrett S, Egelberg J. Diagnostic predictability of scores of plaque, bleeding, suppuration and probing depth for probing attachment loss. 3 1/2 years of observation following initial periodon-

- tal therapy. *J Clin Periodontol* 1990;**17**:108–114.
5. Lang NP, Joss A, Tonetti MS. Monitoring disease during supportive periodontal treatment by bleeding on probing. *Periodontol 2000* 1996;**12**:44–48.
  6. Armitage GC. Periodontal diseases: diagnosis. *Ann Periodontol* 1996;**1**:37–215.
  7. Charalampakis G, Dahlén G, Carlén A, Leonhardt A. Bacterial markers vs. clinical markers to predict progression of chronic periodontitis: a 2-yr prospective observational study. *Eur J Oral Sci* 2013;**121**:394–402.
  8. Luterbacher S, Mayfield L, Brägger U, Lang NP. Diagnostic characteristics of clinical and microbiological tests for monitoring periodontal and peri-implant mucosal tissue conditions during supportive periodontal therapy (SPT). *Clin Oral Implants Res* 2000;**11**:521–529.
  9. Jepsen S, Rühling A, Jepsen K, Ohlenbusch B, Albers HK. Progressive peri-implantitis. Incidence and prediction of peri-implant attachment loss. *Clin Oral Implants Res* 1996;**7**:133–142.
  10. Rams TE, Listgarten MA, Slots J. Efficacy of CPITN sextant scores for detection of periodontitis disease activity. *J Clin Periodontol* 1996;**23**:355–361.
  11. Rams TE, Listgarten MA, Slots J. Regards actuels sur les radiographies conventionnelles en parodontie. *J Parodontol* 1994;**13**:179–184. (in French).
  12. Tabanella G, Nowzari H, Slots J. Clinical and microbiological determinants of ailing dental implants. *Clin Implant Dent Relat Res* 2009;**11**:24–36.
  13. Rams TE, Listgarten MA, Slots J. Utility of radiographic crestal lamina dura for predicting periodontitis disease activity. *J Clin Periodontol* 1994;**21**:571–576.
  14. Rams TE, Oler J, Listgarten MA, Slots J. Utility of Ramfjord index teeth to assess periodontal disease progression in longitudinal studies. *J Clin Periodontol* 1993;**20**:147–150.
  15. Rams TE, Listgarten MA, Slots J. Evaluation of a periodontal risk calculator on treated periodontitis patients. *AADR Abstract*, 2014.
  16. Slots J. Low-cost periodontal therapy. *Periodontol 2000* 2012;**60**:110–137.
  17. Rams TE, Listgarten MA, Slots J. Utility of 5 major putative periodontal pathogens and selected clinical parameters to predict periodontal breakdown in patients on maintenance care. *J Clin Periodontol* 1996;**23**:346–354.
  18. Fujise O, Hamachi T, Inoue K, Miura M, Maeda K. Microbiological markers for prediction and assessment of treatment outcome following non-surgical periodontal therapy. *J Periodontol* 2002;**73**:1253–1259.
  19. Saygun I, Nizam N, Keskiner I, Bal V, Kubar A, Açikel C, Serdar M, Slots J. Salivary infectious agents and periodontal disease status. *J Periodontol Res* 2011;**46**:235–239.
  20. Chapple IL, Garner I, Saxby MS, Moscrop H, Matthews JB. Prediction and diagnosis of attachment loss by enhanced chemiluminescent assay of crevicular fluid alkaline phosphatase levels. *J Clin Periodontol* 1999;**26**:190–198.
  21. Ngo LH, Darby IB, Veith PD, Locke AG, Reynolds EC. Mass spectrometric analysis of gingival crevicular fluid biomarkers can predict periodontal disease progression. *J Periodontol Res* 2013;**48**:331–341.
  22. Slots J, Taichman NS, Oler J, Listgarten MA. Does the analysis of the subgingival flora have value in predicting periodontal breakdown? In: Guggenheim B, ed. *ERGOB proceedings on the conference "Periodontology Today"*. Basel, Switzerland: S. Karger AG, 1988:132–140.
  23. Beltrán-Aguilar ED, Thornton-Evans G, Eke PA, Petersen PE. Recording and surveillance systems for periodontal diseases. *Periodontol 2000* 2012;**60**:40–53.
  24. Galván M, Gonzalez S, Cohen CL, Alonaizan FA, Chen CT-L, Rich SK, Slots J. Periodontal effects of 0.25% sodium hypochlorite twice-weekly oral rinse. A pilot study. *J Periodontol Res* 2013; doi: 10.1111/jre.12151. [Epub ahead of print].
  25. Lie MA, Timmerman MF, van der Velden U, van der Weijden GA. Evaluation of 2 methods to assess gingival bleeding in smokers and non-smokers in natural and experimental gingivitis. *J Clin Periodontol* 1998;**25**:695–700.
  26. Barendregt DS, van der Velden U, Timmerman MF, van der Weijden GA. Comparison of two automated periodontal probes and two probes with a conventional readout in periodontal maintenance patients. *J Clin Periodontol* 2006;**33**:276–282.
  27. Lang NP, Nyman S, Senn C, Joss A. Bleeding on probing as it relates to probing pressure and gingival health. *J Clin Periodontol* 1991;**18**:257–261.
  28. Cercek JF, Kiger RD, Garrett S, Egelberg J. Relative effects of plaque control and instrumentation on the clinical parameters of human periodontal disease. *J Clin Periodontol* 1983;**10**:46–56.
  29. Westfelt E, Rylander H, Dahlén G, Lindhe J. The effect of supragingival plaque control on the progression of advanced periodontal disease. *J Clin Periodontol* 1998;**25**:536–541.
  30. Haffajee AD, Cugini MA, Dibart S, Smith C, Kent RL Jr, Socransky SS. The effect of SRP on the clinical and microbiological parameters of periodontal diseases. *J Clin Periodontol* 1997;**24**:324–334.
  31. Farina R, Tomasi C, Trombelli L. The bleeding site: a multi-level analysis of associated factors. *J Clin Periodontol* 2013;**40**:735–742.
  32. Kalkwarf KL, Kaldahl WB, Patil KD, Molvar MP. Evaluation of gingival bleeding following 4 types of periodontal therapy. *J Clin Periodontol* 1989;**16**:601–608.
  33. Adriaens PA, Adriaens LM. Effects of nonsurgical periodontal therapy on hard and soft tissues. *Periodontol 2000* 2004;**36**:121–145.
  34. Pitcher GR, Newman HN, Strahan JD. Access to subgingival plaque by disclosing agents using mouthrinsing and direct irrigation. *J Clin Periodontol* 1980;**7**:300–308.
  35. Ximénez-Fyvie LA, Haffajee AD, Som S, Thompson M, Torresyap G, Socransky SS. The effect of repeated professional supragingival plaque removal on the composition of the supra- and subgingival microbiota. *J Clin Periodontol* 2000;**27**:637–647.
  36. Hellström MK, Ramberg P, Krok L, Lindhe J. The effect of supragingival plaque control on the subgingival microflora in human periodontitis. *J Clin Periodontol* 1996;**23**:934–940.
  37. Dahlén G, Lindhe J, Sato K, Hanamura H, Okamoto H. The effect of supragingival plaque control on the subgingival microbiota in subjects with periodontal disease. *J Periodontol* 1992;**19**:802–809.
  38. Lobene RR, Soparkar PM, Hein JW, Quigley GA. A study of the effects of antiseptic agents and a pulsating irrigating device on plaque and gingivitis. *J Periodontol* 1972;**43**:564–568.
  39. De Nardo R, Chiappe V, Gómez M, Romanelli H, Slots J. Effect of 0.05% sodium hypochlorite oral rinse on supragingival biofilm and gingival inflammation. *Int Dent J* 2012;**62**:208–212.
  40. McDonnell G, Russell AD. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 1999;**12**:147–179.
  41. Taylor DM. Inactivation of transmissible degenerative encephalopathy agents: a review. *Vet J* 2000;**159**:10–17.
  42. Mainnemaire A, Mégarbane B, Soueidan A, Daniel A, Chapple IL. Hypochlorous acid and taurine-N-monochloramine in periodontal diseases. *J Dent Res* 2004;**83**:823–831.
  43. Leung TH, Zhang LF, Wang J, Ning S, Knox SJ, Kim SK. Topical hypochlorite ameliorates NF- $\kappa$ B-mediated skin diseases in mice. *J Clin Invest* 2013;**123**:5361–5370.