

Hydrogen Peroxide Versus Sodium Hypochlorite: All a Matter of pH?



Erik Trautmann, DMD,*
Thomas Attin, DMD,* Dirk Mohn,
PhD,*† and Matthias Zehnder,
DMD, PhD*

ABSTRACT

Introduction: Hydrogen peroxide (H_2O_2) and sodium hypochlorite (NaOCl) solutions are similar in that they contain oxidizing agents with a bleaching effect. NaOCl solutions are stable at a high pH, at which they also exert increased cleansing/tearolysis. On the other hand, H_2O_2 solutions are natively acidic, yet gain bleaching power on organic stains when alkalinized. It was investigated whether alkalinizing a H_2O_2 solution would also let it dissolve soft tissue or increase its bleaching power on blood-stained dentin. **Methods:** The stability of alkalinized H_2O_2 solutions was assessed by iodometric titration. Soft tissue dissolution was investigated on porcine palatal mucosa. The bleaching effect (ΔL^*) after 60 minutes of exposure was monitored in blood-stained human dentin using a calibrated spectrophotometer. To compare similar molarities, 2.5% H_2O_2 solutions were used here, and 5.0% NaOCl was used as the positive control, whereas nonbuffered saline solution served as the negative control.

Results: Adding alkali (NaOH) to the H_2O_2 solutions rendered them unstable in a dose-dependent manner. A H_2O_2 solution of pH 11.1 was chosen for the main experiments (tissue dissolution and bleaching effect) and compared with a native counterpart (pH = 4.7). Alkalinizing the H_2O_2 solution had no discernible effect on its soft tissue dissolution or bleaching power ($P = .75$ compared with the native H_2O_2 solution). The NaOCl solution of similar molar concentration had a considerably ($P < .001$) higher tissue dissolving and bleaching effect under current conditions. **Conclusions:** The proteolytic/bleaching effects of NaOCl solutions are unique and cannot be achieved by altering the pH of peroxide solutions. (*J Endod* 2021;47:297–302.)

KEY WORDS

Bleaching; dentin; hydrogen peroxide; tissue dissolution

The use of strong oxidizing agents as root canal irrigants has a long tradition in dentistry. Solutions of sodium hypochlorite (NaOCl) and hydrogen peroxide (H_2O_2) have been used in different parts of the world to irrigate and disinfect root canals^{1,2}. Peroxide-containing pastes are still marketed globally as aids in root canal instrumentation and disinfection³. What hypochlorite and peroxide-containing products have in common is that they are bleaching agents and, thus, in theory, have the potential to remove discolorations from teeth during root canal treatment. Both chemicals are used in industrial bleaching procedures^{4,5} as well as in different types of sanitizers⁶. Despite their similarity in application, these agents differ in one major aspect—their stability at different pH levels in aqueous solution. NaOCl solutions are stable at high pH and are thus sold in this form^{7,8}. On the other hand, peroxide-containing formulations are stable in acidic form and have to be stabilized if the pH is to be elevated⁹.

In the context of endodontics, there is one reason why NaOCl solutions have become the main irrigants: their unique ability to dissolve necrotic soft tissue^{10,11} and biofilm matrix components¹². At their native pH, peroxide solutions do not display these cleansing effects, which are highly desirable in endodontics^{11,12}. A recent study showed that a NaOCl solution (pH = 12) at a lower concentration (molar) than a H_2O_2 solution (pH = 4.5) dissolved all visible components from human whole blood, whereas the H_2O_2 solution did not. Consequently, the NaOCl had a higher bleaching power in human blood-stained dentin than H_2O_2 ¹³.

It is known from NaOCl solutions that they lose their dissolving efficacy on necrotic soft tissues once their pH is lowered¹⁴ and gain extra power in this context if alkali is added to the solution⁷. These pH effects have not been studied in peroxide solutions at concentrations that are relevant in endodontics. It is

SIGNIFICANCE

This research highlights the uniqueness of sodium hypochlorite when it comes to its proteolytic effect, which also results in higher bleaching power in blood-stained dentin compared with hydrogen peroxide, even if the latter was alkalinized.

From the *Clinic of Conservative and Preventive Dentistry, Center of Dental Medicine, University of Zurich, Zurich, Switzerland; and †Institute for Chemical and Bioengineering, Department of Chemistry and Applied Biosciences, ETH Zurich, Switzerland

Address requests for reprints to Dr Matthias Zehnder, Division of Endodontics, Clinic of Conservative and Preventive Dentistry, Center of Dental Medicine, University of Zurich, Plattenstrasse 11, CH-8032 Zurich, Switzerland.
E-mail address: matthias.zehnder@zzm.uzh.ch
0099-2399

Copyright © 2020 The Authors. Published by Elsevier Inc. on behalf of American Association of Endodontists. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).
<https://doi.org/10.1016/j.joen.2020.10.016>

known that the bleaching power of H₂O₂ on wine and tobacco colors increases with pH¹⁵. In industrial bleaching, the pH of H₂O₂ solutions is elevated because it is more effective at pH levels above 10. The dissociation of H₂O₂ occurs at pH levels between 10 and 12, and the perhydroxyl anion (HO₂⁻) becomes the predominant ion above a pH of 11. The perhydroxyl anion is thought to be responsible for this increase in bleaching power and especially stain removal^{9,16}. However, it is not known whether an increased pH will also render a H₂O₂ solution proteolytic and, thus, could add an effect that is not seen at native pH levels.

In the present study, we created a 2.5% H₂O₂ solution with a pH >11 and then compared its soft tissue dissolution capacity and bleaching effects on blood-stained dentin to those of a native counterpart at a pH of 4.8 and a NaOCl solution of similar molarity (5%).

MATERIALS AND METHODS

Ethics

The porcine palatal mucosa used here was obtained from the local slaughterhouse (SBZ Schlachtbetriebe, Zurich, Switzerland). Human whole blood was excess material used for blood agar plates in microbiology. The teeth in this study were caries-free third molars extracted for reasons not related to the current work. All patients gave informed written consent that these teeth could be used to perform anonymized *in vitro* tests. None of these human-derived materials could be traced back to their donors. Under local law, the current protocol was exempt from the necessity to obtain individual ethics approval¹⁷.

Solutions and Stability Assessment

The solutions used in the current study were made from concentrated stock solutions of NaOCl (PanReac Applichem, Darmstadt, Germany) and H₂O₂ (Sigma-Aldrich, St Louis, MO). Concentrations were verified by iodometric titration¹⁸. The pH of these solutions was measured using a calibrated pH electrode (827 pH lab; Deutsche Metrohm, Filderstadt, Germany).

In a first set of experiments, the impact of pH on the available H₂O₂ was assessed over the course of 1 hour using iodometric titration. H₂O₂ concentration and pH were measured immediately after mixing 5.0% H₂O₂ 1:1 (wt/wt) with deionized water, 0.1 mol/L NaOH, or 0.5 mol/L NaOH. Measurements were repeated at 15, 30, and 60 minutes. These experiments were performed 4 times on different experimental days, with all 3 solutions

being assessed simultaneously at room temperature (25°C).

Based on these results, a fresh 1:1 mixture of 0.5 mol/L NaOH with 5.0% H₂O₂ was used as an alkalinized 2.5% H₂O₂ solution for the following experiments. This alkalinized H₂O₂ solution had an initial pH of 11.1, whereas the native solution had a pH of 4.7. Nonbuffered physiological saline solution (0.9% NaCl; B Braun Melsungen, Berlin, Germany) and 5.0% NaOCl (pH = 12.2) were used as controls with a minimal and maximal proteolytic effect, respectively. The concentrations of NaOCl and H₂O₂ used here were chosen under the assumption of almost complete dissociation (OCl⁻ and OOH⁻), when a 5% NaOCl (K_{Diss} = 2.9 × 10⁻⁸ at 25°C) solution corresponds to a 2.3% H₂O₂ (K_{Diss} = 2.4 × 10⁻¹² at 25°C) solution in molarity. This is the case at a highly alkaline pH.

Soft Tissue Dissolution Assay

A simple yet reliable weight loss assay¹⁹ was used to test the soft tissue dissolution efficacy of test and control solutions. Twenty-four frozen specimens of similar volume and surface area were obtained from porcine palatal mucosa with a round punch (8 mm) applied at the crest of the rugae. Specimens were thawed at room temperature in a humid environment. They were blotted dry and then weighed using a precision balance (AT 261; Mettler Instrumente AG, Nänikon-Uster, Switzerland). The specimens were then immersed without agitation in 10 mL test or control solutions for 20 minutes in cylindrical 40-mL polypropylene containers with a plastic lid (Semadeni, Ostermundigen, Switzerland). Subsequently, specimens were immersed in 0.1 mol/L sodium thiosulfate (Na₂S₂O₃) to stop the oxidizing action of H₂O₂ or NaOCl, blotted dry, and weighed again. Tests were repeated 5 times (*n* = 6) on different experimental days. In each run, 1 specimen per group was assessed to avoid bias.

The weight loss was calculated in the percent of the original tissue weight. The pH values of the test and control solutions before and after exposure to the soft tissue were determined using a calibrated pH electrode (827 pH lab, Deutsche Metrohm). These experiments were also performed at room temperature (25°C).

Bleaching of Blood-stained Human Dentin

For these experiments, a recently developed model was used¹³. In brief, dentin cylinders were obtained from the roof of the pulp chamber of human third molars, which had been stored at 5°C in a 0.1% thymol solution.

Personnel handling the teeth applied all necessary precautions for infection control. Dentin cylinders of a 6.6-mm diameter and 3.5 mm in height were cut using a low-speed sawing apparatus (IsoMetTM LS; Buehler, Esslingen, Germany) equipped with a diamond-coated wheel saw (Struers, Ballerup, Denmark) for horizontal cuts and a water-cooled diamond core drill (Proxxon, Trier, Germany) for vertical cuts. Dentin specimens were embedded in methacrylate (Paladur; Kulzer, Hanau, Germany) to form a cylinder of an 8.8-mm diameter with the coronal dentin surface exposed. This surface was polished (Tegramin 30, Struers) using silicon carbide grinding paper (Prüfag, Schlieren, Switzerland) up to 4000 grit. Subsequently, a hollow cylinder with a diameter of 5 mm was prepared in the middle of each specimen using a water-cooled diamond core drill so that the dentin had a remaining thickness of 1 mm and the lateral walls of the resulting holding chamber a height of 4 mm. Specimens were soaked in 17% EDTA (Kantonsapotheke, Zurich, Switzerland) for 30 seconds to open the dentinal tubules and remove the smear layer and then washed in deionized water.

To stain the dentin specimens, human whole blood was used containing citrate phosphate dextrose as an anticoagulant and a stabilizer consisting of sodium chloride, adenine, glucose monohydrate, and mannitol²⁰. Fifty microliters of blood per specimen were pipetted into the holding chamber. Specimens were centrifuged for 2 minutes at 1200g (Z320; Hermle, Gosheim, Germany) in 15-mL Corning tubes (Corning Inc, Corning, NY) with their polished dentin surface (assessment side) facing down. Subsequently, the remaining blood was removed from the holding chamber. The specimens were then stored in 100% humidity for 3 weeks at 37°C in an incubator (NCU-Line IL 23; VWR International, Leuven, Belgium). After that period, a stable discoloration is reached, as has been assessed previously¹³.

To irrigate the specimens, 50 µL irrigating solution was pipetted into the holding chamber (Fig. 1A). Irrigants were replaced (ie, removed and 50 µL fresh solution applied) every 10 minutes. After a total irrigation time of 60 minutes at room temperature (25°C), the solutions were removed. Spectrophotometric measurements were performed before and after irrigation with test and control solutions. A timer was used so that the irrigation times were exactly the same for all specimens. Experiments were repeated 9 times (*n* = 10) on separate days. Color changes on the polished dentin surfaces (Fig. 1B) were assessed using a black/white-calibrated spectrophotometer (CM-2600d; Konica

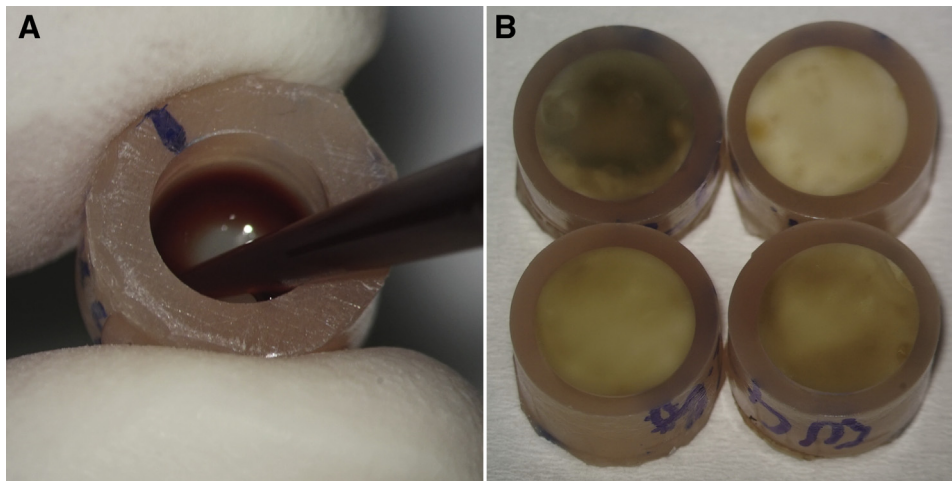


FIGURE 1 – Human dentin specimens that were used as a model of blood-stained dentin. (A) Human whole blood was pipetted into the treatment side of the specimens. Subsequently, the blood was centrifuged into the dentinal tubules and left there for 3 weeks at 37°C. These blood-stained dentin specimens were then treated with test and control solutions for 60 minutes, and (B) the color change was measured on the assessment side after 60 minutes of treatment with the following solutions: (upper right) nonbuffered saline solution and 5% NaOCl; (lower left) native 2.5% H₂O₂ (pH = 4.7), or (lower right) alkalized H₂O₂ (pH = 11.1).

Minolta, Tokyo, Japan) connected to an external computer running the analysis software (Spectra Magic NX, version 2.8, Konica Minolta). Specimens were positioned on the device using a customized aluminum holder so that they could be reassessed in the same position. Measurements were performed in the CIEL*A*B* color space in the reflectance mode, where the L* value indicates the white to black, a* the green to red, and b* the blue to yellow hue. However, in the current study, only the change in L* values were considered (ΔL^*) because this difference represents the bleaching effect¹³. Images were taken with a field of view of 3 mm (Target Mask A147, Konica Minolta) under simulated natural light illumination (D65).

Sample Size and Data Analysis

Changes in H₂O₂ concentration with added alkali over time are presented using descriptive

statistics (line graphs representing mean values). All data were distributed evenly, and, consequently, they are presented as means and standard deviations. The exception are pH data, which are logarithmic by nature, and are presented as medians and ranges. The sample sizes for the 2 experiments involving biological samples (soft tissue and blood-stained dentin) were calculated based on the mean values and standard deviations of the first triplicates with a power of 80% and a type I error rate of 5%. The mean values between different irrigants were compared using 1-way analysis of variance followed by the Tukey honestly significant difference test. The alpha-type error was set to 5% ($P < .05$).

RESULTS

There was a dose-dependent negative effect on H₂O₂ stability when alkali was added

(Fig. 2). After 60 minutes, the 2.5% H₂O₂ solution with an initial pH of 11.1 had a content of $1.9\% \pm 0.1\%$ and a pH of 11.3. This solution, which was always created from a fresh mixture of 5.0% native H₂O₂ and 0.5 mol/L NaOH, was used as the alkalized peroxide solution in the subsequent experiments.

The results of the soft tissue dissolution experiments revealed that the alkalized H₂O₂ solution did not gain any discernable proteolytic effect (Table 1). This was contrasted by the distinct soft tissue dissolution efficacy observed with the 5% NaOCl solution ($P < .001$ compared with all other groups). These observations were confirmed in the blood-stained human dentin specimens; no increase in the bleaching effect was observed by the alkalized 2.5% H₂O₂ solution compared with the native (acidic) counterpart ($P = .75$). By far, the strongest bleaching effect was attained by the 5% NaOCl

Repeated Chemical Assessment of 1:1 (wt/wt) Mixtures of 5 % H₂O₂ with:

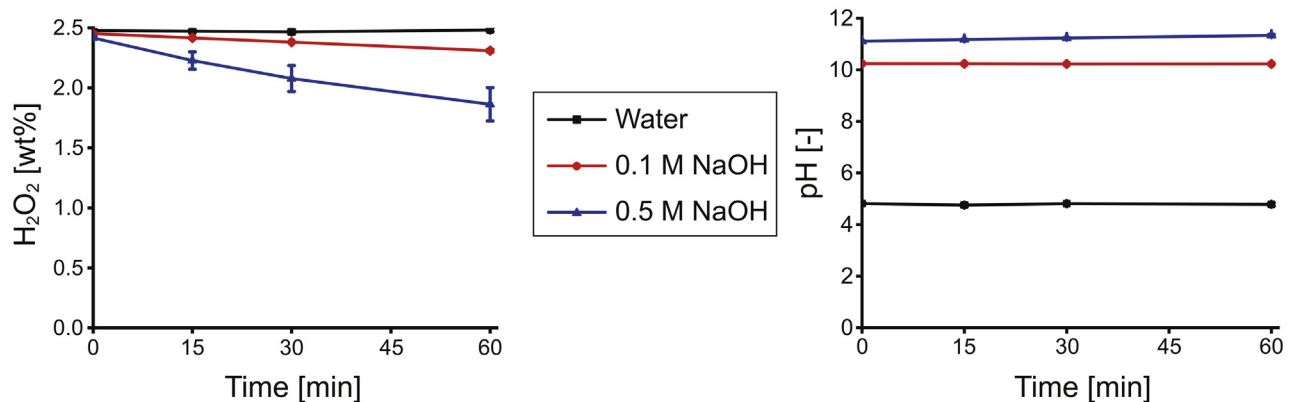


FIGURE 2 – Line graphs depicting the change of H₂O₂ concentration and pH when alkali (NaOH) was added to the solution.

TABLE 1 - The Weight of Standardized Soft Tissue Specimens ($n = 6$) in the Percent of Initial Weight (Means \pm Standard Deviations) after 20 Minutes of Immersion in Test and Control Solutions and pH Values (Medians [Ranges]) of these Solutions before and after the Experiment

Solution	Final weight (%)	Initial pH	Final pH
Saline (0.9% NaCl)	102 \pm 2 ^A	5.6 (5.6–5.6)	6.6 (6.6–6.6)
2.5% native H ₂ O ₂	100 \pm 2 ^A	4.7 (4.7–4.8)	5.7 (5.7–5.8)
2.5% alkalized H ₂ O ₂	104 \pm 2 ^A	11.1 (11.1–11.2)	11.2 (11.1–11.2)
5.0% NaOCl	69 \pm 8 ^B	12.2 (12.1–12.4)	12.3 (12.2–12.4)

H₂O₂, hydrogen peroxide; NaOCl, sodium hypochlorite.

Different superscript letters indicate that there was a significant difference between data sets pertaining to tissue weight loss at the .001 level (analysis of variance, Tukey honestly significant difference test).

solution (Figs. 1B and 3). This difference was statistically highly significant ($P < .001$).

DISCUSSION

This study showed that alkalinizing a H₂O₂ solution does not add a proteolytic effect to it, and, therefore, no soft tissue dissolution capacity or increased bleaching effect on blood-stained dentin was observed.

Many things that we know about chemicals with a bleaching effect we know from industrial applications and not in the context of dentistry. For NaOCl solutions, it is known that their proteolytic/cleansing effect is best at extreme pH values (ie, at a very high or very low pH)²¹. However, NaOCl solutions are only stable at a high pH. The opposite is the case for peroxide solutions. They are stable in an acidic environment; yet, their bleaching effect is also increased at a higher pH^{9,16}. Both

NaOCl and H₂O₂ are oxidizing agents. Based on the reported finding that H₂O₂ solution gains bleaching efficacy against organic stains at a higher pH¹⁵, we speculated that a high pH could also add a proteolytic effect to these solutions, which, under current conditions, could not be confirmed. NaOCl is classified as a halogen-releasing agent, whereas H₂O₂ is a peroxygen²². Even though these compounds are among the oldest disinfectants that are still in use, their exact mode of action is not understood completely^{9,22}. Interestingly, and perhaps surprisingly, NaOCl, also known as “bleach,” has a much stronger bleaching effect on blood-stained dentin than a H₂O₂ solution of comparable concentration. This fact has not received the attention it probably deserves. A bleaching effect on blood-stained roots is a desirable effect, which may be added to the other unique features that make NaOCl indispensable in root canal irrigation²³.

However, the use of hypochlorite-releasing agents (eg, in the form of calcium hypochlorite powder in suspension) as internal bleaching agents would probably not be advisable because a sustained exposure of root dentin to these collagen-depleting agents would affect mechanical dentin properties²⁴.

As a limitation of this study, it needs to be acknowledged that the experiments were performed at room temperature (25°C) rather than body temperature. Chemical reactions are driven by concentration, pH, and temperature. However, no essential differences in the hierarchy of the outcomes presented here should be expected. This was a laboratory and not a clinical study, and room temperature is laboratory standard.

H₂O₂ was indirectly measured in the current study via a so-called coupled reaction. At first, the available H₂O₂ reacted with an iodide solution to form iodine, which was then titrated with a defined sodium thiosulfate solution. Starch served as the indicator. The consumed amount of thiosulfate solution to reach the colorless end point of the titration indicates the amount of H₂O₂ present. In a simulated clinical approach, Zollinger et al¹³ used a higher concentration of H₂O₂ (5%) and a lower concentration of NaOCl (2.5%) than was done here. In the current study, we adjusted the molarities of the solutions under investigation so that the outcomes are more comparable. Because the alkalinized peroxide solution under investigation was not completely stable for the duration of the experiments, we deemed 5% NaOCl (which corresponds to 2.3% H₂O₂) and an initial strength of 2.5% H₂O₂ the correct concentrations to tackle the research question under investigation. The results that were obtained are clear-cut; alkalinizing H₂O₂ did not possess any desirable effect under current conditions. This could not necessarily have been predicted based on the available literature when H₂O₂ at a higher pH is assumed to be more efficient due to the perhydroxyl anion action. The only article that assessed H₂O₂ pH in the context of bleaching dental tissues showed an increased effect of alkalinized solutions against wine and tobacco colors¹⁵. Two particular differences to the aforementioned study are the use of standard concentrations for root canal treatments, which are lower than concentrations used for bleaching, and the approach to bleach tissue rather than to decolorize mere liquids. H₂O₂ concentrations in this study are also similar to concentrations used in pulp bleaching, but various other factors, such as time, temperature, and material accessibility, are different and not in favor for the system that

Bleaching Effect After 60 min of Treatment

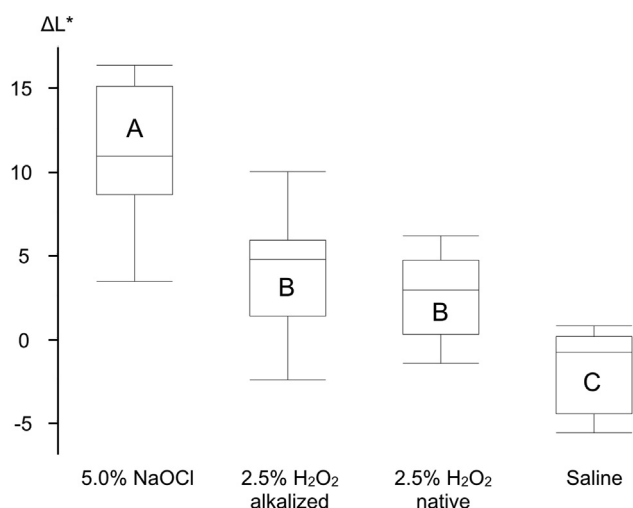


FIGURE 3 – Box plots (medians and interquartile ranges, whiskers: upper and lower data point values) depicting the bleaching effect (ΔL^*) of the solutions under investigation. Note that the physiological saline solution that was used as a negative control caused a slight darkening of the specimens, which can be explained by wetting. Data sets that did not differ significantly at the 5% level (1-way analysis of variance, Tukey honestly significant difference test) are marked by identical letters ($n = 10$).

was applied here¹⁶. Bleaching in dentistry with a peroxide system uses rather higher concentrations, as applied in Torres et al¹⁵ and, thus, may compensate for the lack of time, temperature, and other industrial modifications. However, it should be cautioned that the pH of the alkalized H₂O₂ solution used here was still 1 unit under that of the NaOCl solution, and, thus, an extra effect of the alkali contained in the NaOCl cannot be excluded. NaOCl solutions are naturally alkaline because they are produced electrochemically from cooled NaOH (Hooker process)⁸. However, NaOH per se has no effect on soft tissue dissolution^{7,25}.

The current results suggest that there are systematic differences between the actions of active chlorine and the oxidative species generated by H₂O₂ in an aqueous solution that are not merely a function of pH. How these differences can be explained awaits further clarification. Furthermore, the unique bleaching power of NaOCl solutions should be further assessed in tooth discolorations that are not related to blood/protein.

CONCLUSIONS

This study showed that alkalizing a H₂O₂ solution rendered it slightly unstable; yet, it did

not equip it with any discernible dissolving effect on necrotic soft tissue or an increased bleaching power on blood-stained dentin. This study highlights the uniqueness of NaOCl solutions in the context of endodontic treatment needs; their bleaching effect on blood-stained dentin has not gained the attention it probably deserves.

ACKNOWLEDGMENTS

The authors thank Claudia Cucuzza for her help with the laboratory procedures.

The authors deny any conflicts of interest related to this study.

REFERENCES

1. Dutner J, Mines P, Anderson A. Irrigation trends among American Association of Endodontists members: a web-based survey. *J Endod* 2012;38:37–40.
2. Tasic G, Miladinovic M, Kovaevic M, Stojanovic M. Choice of root canal irrigants by serbian dental practitioners. *Vojnosanit Pregl* 2016;73:47–52.
3. Stewart GG, Kapsimalas P, Rappaport H. Edta and urea peroxide for root canal preparation. *J Am Dent Assoc* 1969;78:335–8.
4. Khristova P, Tomkinson J, Dimitrov I, et al. Totally chlorine-free bleaching of flax pulp. *Bioresour Technol* 2002;85:79–85.
5. Wang X, Ni Y, Zhang H, et al. Formation and emission of pccdd/fs in chinese non-wood pulp and paper mills. *Environ Sci Technol* 2012;46:12234–40.
6. Kingsley DH, Vincent EM, Meade GK, et al. Inactivation of human norovirus using chemical sanitizers. *Int J Food Microbiol* 2014;171:94–9.
7. Jungbluth H, Marending M, De-Deus G, et al. Stabilizing sodium hypochlorite at high pH: Effects on soft tissue and dentin. *J Endod* 2011;37:693–6.
8. Jungbluth H, Peters C, Peters O, et al. Physicochemical and pulp tissue dissolution properties of some household bleach brands compared with a dental sodium hypochlorite solution. *J Endod* 2012;38:372–5.
9. McDonnell G. The use of hydrogen peroxide for disinfection and sterilization applications. In: *PATAI'S Chemistry of Functional Groups*. Hoboken: John Wiley & Sons, Ltd; 2009. p. 1–34.
10. Grossman LI, Meiman BW. Solution of pulp tissue by chemical agents. *J Am Dent Assoc* 1941;28:223–5.
11. Naenni N, Thoma K, Zehnder M. Soft tissue dissolution capacity of currently used and potential endodontic irrigants. *J Endod* 2004;30:785–7.
12. Tawakoli PN, Ragnarsson KT, Rechenberg DK, et al. Effect of endodontic irrigants on biofilm matrix polysaccharides. *Int Endod J* 2017;50:153–60.
13. Zollinger A, Attin T, Mohn D, Zehnder M. Effects of endodontic irrigants on blood and blood-stained dentin. *Heliyon* 2019;5:e01794.
14. Aubut V, Pommel L, Verhille B, et al. Biological properties of a neutralized 2.5% sodium hypochlorite solution. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2010;109:e120–5.
15. Torres CR, Crastechini E, Feitosa FA, et al. Influence of pH on the effectiveness of hydrogen peroxide whitening. *Oper Dent* 2014;39:E261–8.
16. Zeronian SH, Inglesby MK. Bleaching of cellulose by hydrogen peroxide. *Cellulose* 1995;2:265–72.
17. Swiss Federal Council. Federal act on research involving human beings. Available at: <https://www.admin.ch/opc/en/classified-compilation/20061313/202001010000/810.30.pdf>. Accessed June 1, 2020.

18. Vogel AI. *A Textbook of Quantitative Inorganic Analysis*. 3rd ed. London, UK: Longmans, Green and Co; 1962. p. 363–5.
19. Zehnder M, Kosicki D, Luder H, et al. Tissue-dissolving capacity and antibacterial effect of buffered and unbuffered hypochlorite solutions. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2002;94:756–62.
20. Routh G, Johnson C. Sag-m blood. *Anaesthesia* 1986;41:1163–4.
21. Fukuzaki S. Mechanisms of actions of sodium hypochlorite in cleaning and disinfection processes. *Biocontrol Sci* 2006;11:147–57.
22. McDonnell G, Russell AD. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 1999;12:147–79.
23. Zehnder M. Root canal irrigants. *J Endod* 2006;32:389–98.
24. Marending M, Luder HU, Brunner TJ, et al. Effect of sodium hypochlorite on human root dentine—mechanical, chemical and structural evaluation. *Int Endod J* 2007;40:786–93.
25. Clarkson RM, Moule AJ, Podlich H, et al. Dissolution of porcine incisor pulps in sodium hypochlorite solutions of varying compositions and concentrations. *Aust Dent J* 2006;51:245–51.