

Disinfection Efficiency of Irreversible Hydrocolloid Impressions Using Different Concentrations of Sodium Hypochlorite: A Pilot Study

Maryam Memarian, DDS, MS; Mohamad Reza Fazeli, PharmD, PhD;
Hossein Jamalifar, MSc; Ahmad Azimnejad, DDS



Abstract

Aim: Dental impressions are potential sources of bacterial contamination which could eventually lead to transmissible infectious diseases through the blood or saliva. Sodium hypochlorite is an effective disinfectant recommended by the American Dental Association (ADA) in a 1:10 dilution for a ten minute immersion to disinfect irreversible hydrocolloid impressions. As the ADA protocol is sometimes neglected in busy practice settings, this pilot study was designed to determine an efficient and effective protocol for disinfection of irreversible hydrocolloid impressions.

Methods and Materials: Various concentrations of sodium hypochlorite and disinfection times were challenged against irreversible hydrocolloid impressions contaminated with six Gram-positive and Gram-negative bacteria.

Results: A two minute immersion time in a 0.6% solution of sodium hypochlorite protocol was found to prevent bacterial growth on the impressions.

Conclusion: Disinfection of irreversible hydrocolloid impressions in a 0.6% solution of sodium hypochlorite for two minutes was as effective as the ADA's protocol of using a 0.5% sodium hypochlorite solution for ten minutes to destroy the test bacteria.

Keywords: Infection control, sodium hypochlorite, irreversible hydrocolloid, disinfection

Citation: Memarian M, Fazeli MR, Jamalifar H, Azimnejad A. Disinfection Efficiency of Irreversible Hydrocolloid Impressions Using Different Concentrations of Sodium Hypochlorite: A Pilot Study. J Contemp Dent Pract 2007 May;(8)4:027-034.

Introduction

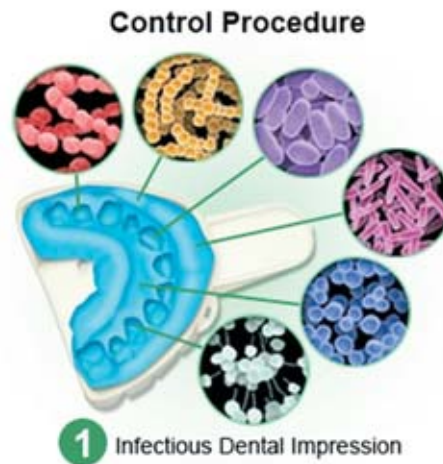
The risk of infections transmitted by saliva and blood is considered a potential occupational hazard in dentistry.^{1,2} Dental impressions can act as a vehicle for the transmission of infectious agents to dental personnel or the resultant dental casts.^{3,4} Irreversible hydrocolloid impressions have produced a significantly higher level of contamination.⁵ Effective infection control procedures should be established to break the chain of transmission of infection and reduce cross-contamination including the disinfection of all impressions prior to delivery to dental laboratories.⁶

The American Dental Association (ADA) recommends a ten-minute immersion in a 1:10 dilution (0.525%) of sodium hypochlorite solution for disinfection of irreversible hydrocolloid impressions.⁷ There are several studies related to the efficacy of different concentrations of sodium hypochlorite. Minagi et al.⁸ have recommended a 60-minute immersion of an impression in sodium hypochlorite (10,000 ppm of available chlorine). Tebrock et al.⁹ have reported total growth inhibition of *Bacillus subtilis* on stone dies poured from contaminated irreversible hydrocolloid impressions immersed in a range of concentrations of sodium hypochlorite for ten minutes. Jennings et al.¹⁰ have reported reduced colony forming units (CFUs) of *Pseudomonas aeruginosa* and *Candida albicans* when contaminated impression materials were disinfected with a 0.0125% solution of sodium hypochlorite for 30 seconds. Impressions contaminated with *Streptococcus sorbines* showed no growth of bacteria after a ten minute immersion in 0.5% sodium hypochlorite.

Considering the possibility busy dental practices and laboratories may ignore the ten minute immersion recommended by the ADA, the aim of this study was to determine whether shorter immersion times of various concentrations of sodium hypochlorite would be effective in the disinfection of irreversible hydrocolloid impressions.

Methods and Materials

Household bleach (Golrang, Karaj, Iran) containing 5.25% sodium hypochlorite was used as a disinfectant agent on Iralgin (Golchai,



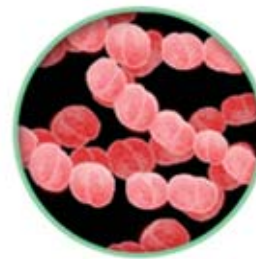
Tehran, Iran) irreversible hydrocolloid dental impressions. The microorganisms were obtained from the Persian Type Culture Collection (PTCC) and clinically isolated by morphological and biochemical tests. They included *Streptococcus sanguis* (PTCC 1449), *Streptococcus pyogenes*, *Streptococcus agalactiae*, *Staphylococcus epidermidis* (ATCC 12228), *Staphylococcus aureus* (PTCC 1112, ATCC 6538), and *Pseudomonas aeruginosa* (PTCC 1074, ATCC 9027).



Streptococcus Sanguis



Staphylococcus Aureus



Streptococcus Pyogenes



Staphylococcus Epidermidis



Streptococcus Agalactiae



Pseudomonas Aeruginosa

Test tubes containing 1 ml of soy bean casein digest broth (SCDB) were inoculated with 1 ml of bacterial suspension (10^8 cfu/ml of individual microorganisms), then 0.5 ml of the desired concentration of freshly prepared sodium hypochlorite was added. The tubes were incubated for 24 hours at 37°C before they were plated on soy bean casein digest agar (SCDA) plates. Plates were incubated for 24 hours at 37°C for determination of the Minimum Inhibitory Concentrations (MICs) using the macrodilution method.

Using this protocol, four experiments were conducted to ultimately determine the efficacy of disinfection on irreversible hydrocolloid. Experiments I to III were designed to narrow the range of concentrations of sodium hypochlorite solution capable of destroying test organisms within controlled time limits. Once the concentration and time of exposure parameters were identified, the fourth experiment was designed to actually determine the concentration and exposure time required for the disinfection of irreversible hydrocolloid impressions under controlled conditions.

The experiments are described as follows:

- **Experiment I:** To initiate the determination of the range of sodium hypochlorite concentrations required to achieve disinfection of the test microorganisms within 20 minutes,

seven different concentrations (0.05%, 0.1%, 0.25%, 0.5%, 0.75%, 1%, and 2%) were used.

- **Experiment II:** Once the results were obtained in Experiment I, the 0.05% concentration was eliminated from further consideration. Then the antimicrobial efficacy of sodium hypochlorite concentrations between 0.1% and 1% (0.15%, 0.2%, 0.25%, 0.5%, 0.75%, and 1%) were investigated against the challenged microorganisms using an exposure time of ten minutes.
- **Experiment III:** Once the results were obtained in Experiment II, the concentrations below 0.5% were eliminated from consideration in an attempt to decrease the time of disinfection. Therefore, sodium hypochlorite concentrations greater than 0.5% (0.6%, 0.7%, 0.8%, 0.9%, and 1%) were used to disinfect microorganisms. The time of disinfection achieved for concentrations of 0.8% or more of sodium hypochlorite was five minutes or less (4, 3, and 2 minutes, respectively). The disinfection times obtained for concentrations of 0.6% and 0.7% were less than 10 minutes (9, 8, 7, 6, 5, 4, 3, and 2 minutes).
- **Experiment IV:** As a result of the first three experiments, 0.6% and 0.7% sodium hypochlorite solutions were used to disinfect the contaminated irreversible hydrocolloid impressions for 4, 3, and 2 minutes.



Thirty-six impressions were made using a sterile typodont and sterile impression trays. Irreversible hydrocolloid was hand mixed with sterile water according to the manufacturer's recommendations.

The third molar regions of all impressions were cut and removed from trays. The impressions of the right molars were immersed in individual microbial suspensions of 10^8 cfu/ml for one minute and transferred to sterile tubes containing 15 ml of nutrient broth (NB) media to create a control.

The impressions of the left molars were immersed in individual microbial suspensions of 10^8 cfu/ml for four minutes. The contaminated specimens were then immersed in 0.7% and 0.6% of sodium hypochlorite for 4, 3, and 2 minutes based on the findings of the preceding experiments.

Each specimen was washed with 50 ml of sterile distilled water and transferred to sterile tubes containing 15 ml of NB media.

In addition, aliquots of 0.5 ml of water were used to wash each specimen and were transferred to tubes containing 15 ml of NB media which served as controls. All tubes were incubated for 24 hours at 37°C. One loop of each tube was streaked on SCDA culture plates and incubated for 24 hours at 37°C for MICs determination.

Results

Experiment I

Of the seven different concentrations (0.05%, 0.1%, 0.25%, 0.5%, 0.75%, 1%, and 2%) used, microorganisms were disinfected with 0.1% and more concentrations of sodium hypochlorite in 20 minutes (Table 1).

Experiment II

When the antimicrobial efficacy of sodium hypochlorite concentrations between 0.1% and 1% (0.15%, 0.2%, 0.25%, 0.5%, 0.75%, and 1%) were investigated against the challenged microorganisms, a ten-minute immersion in 0.5% sodium hypochlorite was found to prevent bacterial growth, which was supportive of the ADA recommendation.⁷ In addition, immersion in a 0.75% solution of sodium hypochlorite for five minutes could also destroy microorganisms (Table 2).

Experiment III

When sodium hypochlorite concentrations greater than 0.5% (0.6%, 0.7%, 0.8%, 0.9%, and 1%) were used to disinfect the test microorganisms, disinfection was achieved for concentrations of 0.8% or more of sodium hypochlorite in five

Table 1. Growth inhibition of challenged bacteria following disinfection with different concentrations of sodium hypochlorite for 20 minutes.

Microorganisms	Concentrations of Sodium Hypochlorite (%)						
	2.0	1.0	0.75	0.5	0.25	0.1	0.05
<i>Streptococcus sanguis</i>	-	-	-	-	-	-	-
<i>Streptococcus pyogenes</i>	-	-	-	-	-	-	-
<i>Streptococcus agalactiae</i>	-	-	-	-	-	-	-
<i>Staphylococcus aureus</i>	-	-	-	-	-	-	+
<i>Staphylococcus epidermidis</i>	-	-	-	-	-	-	+
<i>Pseudomonas aeruginosa</i>	-	-	-	-	-	-	+

Legend: - No bacterial growth, + Bacterial growth

Table 2. Growth inhibition of challenged bacteria following disinfection with different concentrations of sodium hypochlorite for 5, 10, and 15 minutes.

Microorganisms	Concentrations of Sodium Hypochlorite (%)						Time (minutes)
	1	0.75	0.5	0.25	0.2	0.15	
<i>Streptococcus sanguis</i>	-	-	-	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15
<i>Streptococcus pyogenes</i>	-	-	+	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15
<i>Streptococcus agalactiae</i>	-	-	-	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15
<i>Staphylococcus aureus</i>	-	-	-	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15
<i>Staphylococcus epidermidis</i>	-	-	-	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15
<i>Pseudomonas aeruginosa</i>	-	-	+	+	+	+	5
	-	-	-	+	+	+	10
	-	-	-	+	+	+	15

Legend: - No bacterial growth, + Bacterial growth

minutes or less (4, 3, and 2 minutes, respectively). The disinfection times obtained for concentrations of 0.6% and 0.7% were less than ten minutes (9, 8, 7, 6, 5, 4, 3, and 2 minutes). Therefore, sodium hypochlorite concentrations of 0.6% and higher can disinfect microorganisms in less than ten minutes.

Experiment IV

As a result of the first three experiments, 0.6% and 0.7% sodium hypochlorite solutions were used to disinfect the contaminated irreversible hydrocolloid impressions for 4, 3, and 2 minutes. No bacterial growth was observed in any disinfected irreversible hydrocolloid impressions or in the wash-out solutions while contaminated impressions showed visible bacterial growth.

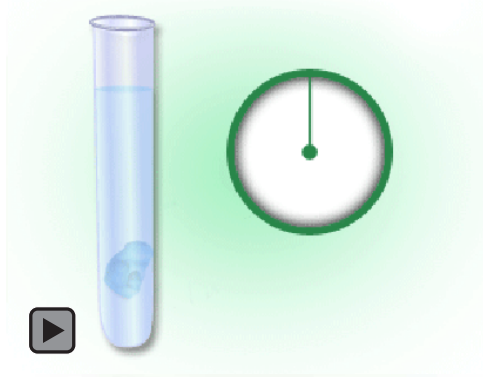
Discussion

As indicated by the present study, immersion in concentrations of 0.6% and 0.7% sodium hypochlorite for two minutes is sufficient to disinfect irreversible hydrocolloid impressions and could prevent bacterial growth.

Several studies have dealt with the efficacy of sodium hypochlorite disinfection including McNeil et al.¹¹ who have reported both the Polio virus and *Streptococcus sanguis* on irreversible hydrocolloid could be inactivated by a 7.5-minute immersion in a 0.1% hypochlorite solution. Schwartz et al.¹² have also reported a 4-log₁₀ (99.99%) reduction of bacteria on irreversible hydrocolloid impressions when the contaminated materials were immersed in a 0.5% sodium hypochlorite solution for ten minutes. Westerholm et al.¹³ have found more than a 4-log₁₀ reduction of *M. phlei* on irreversible hydrocolloid impressions after only a one minute exposure to full-strength (5.25%) sodium hypochlorite spray. Best et al.¹⁴ have reported only a 3.2-log₁₀ reduction of *M. tuberculosis* when a 1% sodium hypochlorite was used for one minute.

Different disinfection procedures and immersion times may affect the dimensional accuracy or surface detail of an irreversible hydrocolloid impression.¹ Ruggeberg et al.⁷ have reported dimensional change and loss of reproduction

0.6% and 0.7% Sodium Hypochlorite Solution



of detail in Microstone[®] (Whip Mix Corporation, Louisville, KY, USA) casts using irreversible hydrocolloid immersed in a 0.5% sodium hypochlorite solution for ten minutes. Tullner et al.¹⁵ have noted partial dissolution of irreversible hydrocolloid materials when immersed in a 1% sodium hypochlorite solution for 15 minutes. Herrera and Merchant¹⁶ have found no significant effect on the dimensional accuracy of Vel-Mix[®] (KerrLab, Orange, CA, USA) casts after a

30 minute immersion of full-arch irreversible hydrocolloid impressions in 0.5% and 1% sodium hypochlorite solution.

The results of the present pilot study tend to suggest immersion in a sodium hypochlorite solution for ten minutes or less could be practiced in clinical settings and dental laboratories. Reducing the immersion time can also minimize changes in physical properties like dimensional stability and surface integrity.

Conclusion

Disinfection of irreversible hydrocolloid impressions in a 0.6% solution of sodium hypochlorite for two minutes was sufficient to prevent bacterial growth and achieve disinfection of irreversible hydrocolloid impressions. This was as effective as the ADA's protocol of 0.5% sodium hypochlorite in ten minutes for the tested bacteria.

However, this was only a pilot study. Further investigation is needed to determine if these results translate to the disinfection of complete mouth impressions in an actual clinical trial.

References

1. Muller-Bolla M, Lupi-Pegurier L, Velly AM, Bolla M. A survey of disinfection of irreversible hydrocolloid and silicone impressions in European Union dental schools: Epidemiologic study. *Int J Prosthodont.* 2004; 17(2): 165-71.
2. Samaranayake LP, Hanjan M, Jennings KJ. Carriage of oral flora on irreversible hydrocolloid and elastomeric impression materials. *J Prosthet Dent.* 1991; 65 (2): 244-9.
3. Beyerle MP, Hensley DM, Bradley DV Jr, Schwartz RS, Hilton TJ. Immersion disinfection of irreversible hydrocolloid impressions with sodium hypochlorite. Part I: Microbiology. *Int J Prosthodont.* 1994; 7 (3): 234-8.
4. Leung RL, Schonfeld ES. Gypsum cast as a potential source of microbial cross-contamination. *J Prosthet Dent.* 1983; 49(2): 210-11.
5. Al-Omari WM, Gly Jones JC, Hart P. A microbiological investigation following the disinfection of alginate and addition cured silicone rubber impression materials. *Eur J Prosthodont Res Dent.* 1998; 6(3): 97-101.
6. Kohn WG, Collins AS, Cleveland JL, Harte JA, Eklund KJ, Malvitz DM. Centers for Disease Control and Prevention (CDC). Guidelines for infection control in dental health-care settings--2003. *MMWR Recomm Rep.* 2003; 52(RR-17):1-61.
7. Vandewalle KS, Charlton DG, Schwartz RS, Reagan SE, Koeppen RG. Immersion disinfection of irreversible hydrocolloid impressions with sodium hypochlorite. Part II: Effect on gypsum. *Int J Prosthodont.* 1994; 7(4): 315-22.
8. Minagi S, Fukushima K, Maeda N, Satomi K, Ohkawa S, Akagawa Y, Miyake Y, Suginaka H, Tsuru H. Disinfection method for impression materials: freedom from fear of hepatitis B and acquired immunodeficiency syndrome. *J Prosthet Dent.* 1986; 56(4):451-4.
9. Tebrock OC, Engelmeier RL, Mayfield TG, Adams HJ. Managing dental impressions and casts of patients with communicable diseases. *Gen Dent.* 1989; 37 (6): 490-5.
10. Jennings KJ, Samaranayake LP. The persistence of microorganisms on impression materials following disinfection. *Int J Prosthodont.* 1991; 4 (4): 382-7.
11. McNeill MR, Coulter WA, Hussey DL. Disinfection of irreversible hydrocolloid impressions: A comparative study. *Int J Prosthodont.* 1992; 5 (6): 563-7.
12. Schwartz RS, Hensley DH, Bradley DV Jr. Immersion disinfection of irreversible hydrocolloid impression in PH adjusted sodium hypochlorite. Part I: Microbiology. *Int J Prosthodont.* 1996; 9(3): 217-22.
13. Westerholm HS, Bradly DV, Schwartz RS. Efficacy of various spray disinfection on irreversible hydrocolloid impressions. *Int J Prosthodont.* 1992; 5 (1): 47-54.
14. Best M, Sattar SA, Springthorpe VS, Kennedy ME. Efficacies of selected disinfectants against *Mycobacterium tuberculosis*. *J Clin Microbiol.* 1990; 28 (10): 2234-9.
15. Tullner JB, Commette JA, Moon PC. Linear dimensional changes in dental impressions after immersion in disinfectant solutions. *J Prosthet Dent.* 1988; 60 (6): 725-8.
16. Herrera SP, Merchant VA. Dimensional stability of dental impressions after immersion disinfection. *J Am Dent Assoc.* 1986; 113 (3): 419-22.

About the Authors

Maryam Memarian, DDS, MS

Dr. Memarian is an Assistant Professor in the Department of Removable Prosthodontics of the Faculty of Dentistry at Tehran University of Medical Sciences in Tehran, Iran.

e-mail: memarian@tums.ac.ir

Mohamad Reza Fazeli, PharmD, PhD

Dr. Fazeli is an Assistant Professor in the Department of Pharmaceutical and Biotechnology in the School of Pharmacy at Tehran University of Medical Sciences in Tehran, Iran.

Hossein Jamalfar, BSc

Mr. Jamalfar is an Instructor in the Department of Pharmaceutical and Biotechnology of the School of Pharmacy at Tehran University of Medical Sciences in Tehran, Iran.

Ahmad Azimnejad, DDS

Dr. Azimnejad is a general dentist in the Department of Removable Prosthodontics of the Faculty of Dentistry at Tehran University of Medical Sciences in Tehran, Iran.