Control of bacterial contamination of dental unit water using sodium hypochlorite

Controle da contaminação bacteriana da água de equipos odontológicos com o uso de hipoclorito de sódio

Control de la contaminación bacteriana del agua del equipo dental con hipoclorito de sodio

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ABSTRACT

Objective: To evaluate bacterial load of water using and not using sodium hypochlorite aiming at bacterial control of dental unit water. **Method:** Dental unit reservoirs from a dental clinic from School of Dentistry of Ribeirão Preto – USP were filled with public-supply water with and without sodium hypochlorite solution at 0.0003%, and water samples were collected and evaluated by Petrifilm™ AC (Aerobic Count) system from 2014 to 2017. **Results:** No bacterial contamination was observed in the water samples using sodium hypochlorite solution. However, bacterial contamination was detected in water from reservoirs, air-water syringes and high-speed outputs (without handpieces) without the disinfectant. **Conclusion:** The use of sodium hypochlorite solution controlled bacterial load of all the dental unit water samples as established by Brazilian legislation (<500CFU/mL).
Descriptors: Biofilms; Dental Equipment; Water Microbiology.

RESUMO

Objetivo: Avaliar a carga bacteriana da água com e sem o uso de hipoclorito de sódio visando o controle bacteriano da água de equipos odontológicos. **Método:** Os reservatórios dos equipos odontológicos de uma clínica odontológica da Faculdade de Odontologia de Ribeirão Preto – USP foram preenchidos com água de abastecimento público com e sem solução de hipoclorito de sódio a 0,0003%, e as amostras de água coletadas e avaliadas pelo sistema Petrifilm™ AC (Aerobic Count) de 2014 a 2017. **Resultados:** Nenhuma contaminação bacteriana foi observada nas amostras de água com uso de solução de hipoclorito de sódio. Entretanto, a contaminação bacteriana foi detectada na água de reservatórios, seringas tríplices e saídas de alta rotação (sem as peças de mão) sem o desinfetante. **Conclusão:** O uso da solução de hipoclorito de sódio controlou a carga bacteriana de todas as amostras de água dos equipos odontológicos conforme estabelecido pela legislação brasileira (<500UFC/mL).
Descritores: Biofilmes; Equipamentos Odontológicos; Microbiologia da Água.

RESUMÉN

Objetivo: evaluar la carga bacteriana de agua con y sin el uso de hipoclorito de sodio para el control bacteriano del agua de los equipos dentales. **Método:** Los depósitos de los equipos dentales de una clínica dental de la Facultad de Odontología Ribeirão Preto-USP se llenaron de agua pública con y sin solución de hipoclorito de sodio al 0,0003%, y las muestras de agua recolectadas y evaluadas por Petrifilm™ AC (Aerobic Count) de 2014 a 2017. **Resultados**: No se observó contaminación bacteriana en muestras de agua con solución de hipoclorito de sodio. Sin embargo, se detectó contaminación bacteriana en el agua de depósitos, jeringas triples y salidas de alta velocidad (sin piezas de mano) sin desinfectante. **Conclusión**: El uso de una solución de hipoclorito de sodio controló la carga bacteriana de todas las muestras de agua del equipo dental según lo establecido por la legislación brasileña (<500UFC/mL).
Descriptores: Biopelículas; Equipo dental; Microbiología del agua.

INTRODUCTION

The mouth, with dental biofilm and saliva, contains more than 700 microorganism species, since the cultivable ones do not reach half this quantity.[1] Furthermore, it is the largest source of contamination in dentistry, followed by water from dental unit waterline, in consequence of biofilm presence.[2-3]

The objective of the control infection is to reduce or eliminate the exposure of patients and dental staff members to microorganisms.[4] This way, the biosafety in dentistry can be based on BSDAP (Barrier, Sterilization, Disinfection, Antisepsis and Preservative) system, in which each one of the letters are initials of an important term or procedure for controlling contamination/infection.[2]

The water that arrives to the unit contains, in most cases, small quantity of microorganisms because it is the same one consumed by the population. In Brazil, according to ordinance No. 2,914 of December 12, 2011 from the Ministry of Health the limit is 500 colony-forming units per milliliter (CFU/mL) of water.[5]

However, the literature reports the contamination of dental unit water in an alarming level[6-7] since the first report of this contamination was made by Blake.[8] Moreover, Kelstrup and collaborators[9] were the pioneers in demonstrating the growth of microorganism colonies adhered to dental unit waterline walls, that later would be denominated biofilm.

The American Dental Association (ADA)[10] recommended that the whole community, both scientific and industrial, directed efforts so that in the year of 2000 water would not present contamination greater than 200CFU/mL, and

that it would develop faster, more practical and lower-cost techniques to effectuate the microbial count as well as implement measures to prevent and control biofilm formation in waterline.

Aiming at the reduction of bacterial contamination of dental unit water and, consequently, the improvement of aspects related to biosafety: contamination/infection control in dentistry, the objective of this study was to evaluate the bacterial load of water from taps and dental units (reservoirs, air-water syringes and high-speed outputs) using and not using sodium hypochlorite at 0.0003%.

METHODS

Water samples were collected from 12 taps and 14 dental units (reservoirs, air-water syringes and high-speed outputs without handpieces) from the Special Patients Clinic at School of Dentistry of Ribeirão Preto USP, using and not using sodium hypochlorite solution for reducing the bacterial contamination of dental unit water in two stages.

In the study's first stage, 14 dental unit water samples that contained sodium hypochlorite at 0.0003% (0.15mL of sodium hypochlorite at 1% − 3 drops − in 500mL of water) in reservoir were collected. In the second stage, 14 reservoirs of dental units were filled only with tap water for analysis of bacterial load, without sodium hypochlorite interference. It is worth noting that a specific tap was established to supply all the reservoirs.

10mL water samples were collected in sterile test tubes (25x150mm). The collection of water samples from air-water syringes, high-

speed outputs (without handpieces) and taps were performed after flush or water drainage for about 30s.[10-11]

Residual chlorine from water samples was neutralized by adding 0.05mL of aqueous solution of sodium thiosulphate at 2%.[12]

For total aerobic bacteria count Petrifilm™ AC (Aerobic Count) system was used, presenting a prepared culture medium with standard nutrient agar, a gelling agent soluble in cold water, and triphenyltetrazolium chloride (TTC).[12]

The experiment was carried out in Class II Type A1 Biological Safety Cabinet. According to the manufacturer's instructions, the seeding of 1mL aliquots of *in natura* water samples were carried out after upper film suspension of Petrifilm™ AC through slow and careful deposit on central part of the bottom film of codified plates. The upper film was dropped on the sample, avoiding air bubble formation, and next the plastic diffuser on the center of plate was pressed for around 10s. After removing the diffuser, the Petrifilm™ AC plates were remained untouched for at least 1min for gel solidification. Following this, the plates were stored in containers with pieces of cotton wetted with water (humid chamber) and in horizontal position. The Petrifilm™ AC plates were incubated at 37°C for 48h.

Reading of Petrifilm™ AC plates was performed with the help of a stereomicroscope under reflected light and the numbers of colony-forming units of total aerobic bacteria were expressed per milliliter of water sample (CFU/mL), according to the microbiological parameter from ordinance No. 2,914 of December 12, 2011 from the Ministry of Health that stipulates the limit at 500CFU/mL of water.[5]

RESULTS

The water bacterial load using and not using sodium hypochlorite at 0.0003% was evaluated aiming at contamination control of dental unit water. It is highlighted that the water samples were collected from 14 dental units (reservoirs, air-water syringes and high-speed outputs), and the results are expressed in Table 1.

In the study's first stage, all the 14 dental unit water samples supplied with tap water and addition of sodium hypochlorite at 0.0003% did not presented contamination by total aerobic bacteria. In the second stage, among 14 dental unit water samples supplied with tap water without addition of sodium hypochlorite, only 1 (7.1%) of the reservoirs, 2 (14.3%) of the air-water syringes and 3 (21.4%) of the high-speed outputs demonstrated bacterial load. It should be noted that only the dental unit No. 8 demonstrated contamination above that recommended by ADA (<200CFU/mL), but all the samples were below the contamination established by Brazilian legislation (<500CFU/mL) – Table 1.

Table 1: Evaluation of bacterial load (CFU/mL) of dental unit water through Petrifilm™ AC plates. Ribeirão Preto, SP, Brazil, 2017.

Dental Unit No.	RE		AW		HS	
	1st Stage	2nd Stage	1st Stage	2nd Stage	1st Stage	2nd Stage
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	21	0	5
5	0	0	0	0	0	0
6	0	0	0	0	0	0
7	0	0	0	0	0	1
8	0	1	0	0	0	485
9	0	0	0	14	0	0
10	0	0	0	0	0	0
11	0	0	0	0	0	0
12	0	0	0	0	0	0
13	0	0	0	0	0	0
14	0	0	0	0	0	0

Legend: CFU/mL, colony-forming units per milliliter of water; RE, reservoir; AW, air-water syringe; HS, high-speed output; 1st Stage, tap water with addition of sodium hypochlorite at 0.0003%; 2nd Stage, tap water without addition of sodium hypochlorite.

Regarding water samples from taps from clinic used to supply the dental units, 10 were free of total aerobic bacteria and only 2 samples presented 1CFU/mL [taps between the dental units (6 and 7) and (13 and 14)], considering that all presented values below that established by Brazilian legislation (<500CFU/mL) and recommended by ADA (<200CFU/mL).

DISCUSSION

In the year of 2000, American Dental Association (ADA)[10] declared that the bacterial contamination of dental unit water should not exceed 200CFU/mL. Even so, there are reports of dental unit water (air-water syringes and high-speed outputs) with counts of up to

24,700CFU/mL[6] and 1,800,000CFU/mL[7] that pose a risk of infection to patients and professionals present in dentistry environment.

Microorganisms present on waterline biofilm and dental unit water can cause various health problems or even lead to death, mainly, immunocompromised patients and/or smokers, alcoholics, diabetics and carrier of pulmonary, cardiac and renal chronic diseases.[13-15]

In this study, only the water sample from high-speed output from dental unit No. 8 demonstrated contamination above that recommended by ADA (<200CFU/mL), but all the samples were below the contamination established by Brazilian legislation (<500CFU/mL).

Despite the water that supplies the dental units is originating from public water supply, the reservoirs coupled to dental units enable the addition of different types of disinfectants and antiseptics for decontamination of waterlines and, consequently, reduction of bacterial load of water, ensuring potable water for clinical procedures.[16]

One of these disinfectants is sodium hypochlorite, that demonstrated in this study a reduction of bacterial contamination of dental unit water samples, corroborating other authors' reports.[14,17-19]

According to ADA[10], it is recommended to flush for a minimum period of 20 to 30s from air-water syringes and high-speed outputs, at the beginning and end of work days and between patient care, once it can temporarily reduce the microbial contamination of water and remove oral fluids that can be entered via backflow from patient's mouth.[7,20] Additionally, current studies have been demonstrating the need to periodic monitoring of microbial contamination of dental unit water, even of those made up of non-return valves.[21-22]

CONCLUSION

The usage of sodium hypochlorite solution for the improvement of bacterial quality of dental unit water achieved its objective by keeping the bacterial load of all dental units below that established by Brazilian legislation (<500CFU/mL). However, the frequent and prolonged usage of sodium hypochlorite, even in reduced concentration, can possibly cause damages to reservoir and dental unit waterlines, corrosion of metallic surfaces (handpieces) as well as unpleasant taste and smell to water. Thus, future studies are needed to enlighten these questions.

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COLLABORATIONS

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CONFLICTS OF INTEREST

The authors declare that no have conflicts of interest.

AVAILABILITY OF DATA

Available upon request to the authors.

FUDING SOURCE

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