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Brief Report

The practice of disinfection of high-speed handpieces with 70% w/v alcohol: An evaluation.

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A high-speed handpiece is used in several dental procedures and the official recommendation for safe decontamination consists of rinsing with water, washing with detergent, mechanical friction, and sterilization; however, many professionals only apply 70% w/v alcohol without also cleaning the tool between patients. We performed an analysis of high-speed handpieces reprocessed only with 70% w/v alcohol and found that the methods used in clinical practice are not safe.

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The risk of infection due to noncompliance with decontamination procedures of devices used in the health field is broadly recognized. Dental tools are potential sources of microbial dissemination among patients. They host and carry a universe of microorganisms, which greatly justifies safe management processing.[1-5]

A high-speed handpiece (HSH) is used in several dental procedures, such as dental prophylaxis, preparing cavities for restoration, odontosections, and osteotomies.[6,7] This technologically innovative tool not only has undeniable benefits in its application, but also has challenges in its decontamination due to its complexity. The official recommendation by scientific societies for safe HSH decontamination is rinsing with water, washing with detergent, and applying mechanical friction, followed by autoclave sterilization.[8,9]

Currently, many professionals only apply 70% w/v alcohol without any other cleaning of the tool between patients.[6,7,10] Dentists claim practicality and insufficient inventory to justify their practice of such methods.[6,7,10]

The use of 70% w/v alcohol in the disinfection process, as opposed to sterilization, is expected to eliminate only microorganisms with lower lethal power, because there is no destruction of bacterial spores.[2,11] Alcohol is widely used as an intermediate surface-level disinfectant with both low cost and low toxicity. It is considered a nonspecific antimicrobial due to the multiplicity of its mechanism with toxic effects on microbial cells.[2]

There is no reliable data linking dental infections to deficient reprocessing of HSHs. Given the small number of infection notifications[12] and the disagreement among official recommendations and the practices observed in reprocessing HSH, this study aims to settle this doubt.

METHODS

The samples were collected from a dental practice in which the decontamination of HSHs was the use of alcohol without cleaning previous. The dental practice under study performs an average of 100 procedures per day.

To analyze the microbial load present on the external surface of HSHs, a carrier was chosen and validated to carry the microorganisms. This process was conducted because HSHs cannot be directly immersed into culture medium or liquid to extract the rinsed product. A sterilized hydrophilic cotton gauze (7.5 × 7.5 cm, 8 layers) (Cremer SA, Blumenau – SC, Brasil) was used because it has greater carrying capacity when compared with cotton swabs.

Validation of the capacity for microbial release from the gauze was performed by means of agitation and sonication. The gauze was previously contaminated with 1 mL suspension of *Serratia marcescens* (ATCC 14756), 10^6 CFU/mL, followed by colony count by means of

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serial dilution after 24 hours and 48 hours kept at 37°C ± 2°C in a bacteriologic incubator.

For the analysis of extraction capacity of the microbial load presented in HSH using the gauze, triplicated HSH samples were contaminated with 1 mL suspension containing 10^6 CFU *S marcescens* (ATCC 14756). One piece of gauze embedded in 5 mL saline solution at 0.9% (0.9% SS) was rubbed on the HSH body, deposited in 300 mL 0.9% SS, and later sonicated and agitated. A total of 2 mL was removed from the rinse for serial dilution and later colony count.

The amount of times the gauze should be rubbed to ensure the maximum carrying of microorganisms from the HSH surface after disinfection was also evaluated. The HSHs were then contaminated with the same *S Marcescens* suspension and after, disinfection with 70% w/v alcohol was performed, using different contact times (30, 60, and 90 seconds). Three gauzes embedded in 0.9% SS were then consecutively rubbed on the HSHs and deposited in flasks containing 300 mL 0.9% SS. After agitation and sonication, aliquots were collected to perform serial dilution and, later, colony count.

Decontamination of the water system in dental units

To prevent HSH contamination with microorganisms originating from the water system, validation of the decontamination of the water system used for cooling HSHs was performed. This procedure was applied before each sample was collected.

To disinfect the reservoir and the hose connecting the HSH, the water reservoirs were substituted by others containing a hydrogen peroxide-based disinfectant—Dentosept P Solution from Sirona (Sirona, Bensheim, Germany)— diluted at the 1:100 mL rate, resulting in the final concentration of 0.014%.[13] With the HSH connected to the hose, the switch of the water line was activated for 2 minutes, with continuous disinfectant flow.[13]

After the contact time of the disinfectant with the water line, the original reservoir was supplied with sterilized distilled water and attached to the equipment. The switch was activated for 2 more minutes for complete removal of the disinfectant and residue.

Data collection

A sample size of 100 units (experimental group) was established, ensuring a 95% confidence interval and 9.8% absolute precision in the worst case, which happens when estimating a contamination incidence of 50%. The calculations were performed using the sampling book package from R software (R Foundation for Statistical Computing, Vienna, Austria).

The experimental group analyzed the microbial load on the HSH surface, without previous treatment, after disinfection with 5 mL 70% w/v alcohol. A gauze embedded in 5 mL 0.9% SS was rubbed on the HSH external surface to carry possible microorganisms.

Half of the samples (n = 50) were analyzed by the membrane filtering indirect method (method I). The gauzes were placed inside a previously sterilized container with 300 mL 0.9% SS. The container was submitted to 3 sessions of 5-second ultrasound (Ultrasonic Rinser USC-2800, Enge Solutions; Enge Solutions and Fanem, São Paulo, Brasil) and an additional 10 minutes of orbital agitation (Kline model 255; Fanem) at 160 rotations per minute to obtain the maximum dislocation of the possible microbial load collected by the gauze. In a biological protection cabin, the rinse was filtered in 3 equal parts, through membranes with 0.45 µm porosity (Sterifil Holder; Millipore Industria e Comercio LTDA, Barueri, Sao Paulo, Brazil) and they were sown in specific culture medium for recovering aerobic and anaerobic microorganisms (Blood agar, *Mitis salivaris* & Anaeroinsol; BD Difco, Spark, MD).

The other 50 samples were analyzed by direct immersion of the gauze in the culture medium (method II). After the gauze was rubbed on the HSH's external surface, it was placed in a test tube containing 40 mL Fluid Thioglycollate Medium (BD Difco). The tube containing the gauze was agitated for 30 seconds in a vortex agitator to provide microbial detachment and greater contact of the gauze with the culture medium.

Positive and negative control groups were also formed, with 10 samples each. The samples in the negative control group were collected after standard cleaning and vapor sterilization by means of an autoclave cycle at 134°C for 5 minutes. The procedure to recover the microbial load followed that used in method I.

The samples of the positive control group were collected after the dentist attended to a patient without any previous disinfection treatment. After using the HSH, the same technique used in method I (quantitative) was used for microbial recovery.

All samples were incubated in a bacteriologic incubator at 37°C ± 2°C, with daily reading of the samples. In the presence of microbial growth in culture media, the samples were forwarded to quantification and/or identification of genus and species of the microorganisms. This procedure was in accordance with the routine at the Laboratory of Microbiology of the Hospital Infection Service at Irmandade Santa Casa de Misericórdia de São Paulo.

RESULTS

In the validation study of the method used to release microorganisms from the gauze used as a carrier, the average microbial recovery was 4.09×10^5 CFU and the microbial load carried by the gauze was 2.28×10^5 CFU. The application of 70% w/v alcohol to an HSH for 90 seconds presented the best contact time of the germicide in the reduction of the initial load, requiring only 1 gauze for microbial carry. The validation studies performed showed satisfactory microbial release from the gauze, as well as its effectiveness at extracting the microorganisms present on an HSH.

The samples analyzed by method I (filtration by membrane) presented positive growth in 27 out of 50 samples (54%). The total positive growth presented by method II (direct sowing of the gauze) was in 12 out of 50 samples (24%). The microorganisms found in both methods are described in Table 1.

The samples in the positive control group presented satisfactory results regarding the presence of microorganisms on HSHs (Table 2).

All 10 samples in the negative control group presented an absence of microbial growth.

DISCUSSION

The results of the present investigation rebuke the practice of disinfecting HSHs by friction with 70% alcohol without previous cleaning. This statement is substantiated by the survival of microorganisms that do not correspond to the fungal and bactericide action expected from the alcohol in the condition of intermediary disinfection that should eliminate mycobacteria, vegetative bacteria, fungi, and viruses.[14]

The microbial load present in the mouths of healthy human individuals was high, ranging from 10^6-10^9 CFU/mL (or organisms per gram) in saliva, 10^5 CFU/mL in the gingival sulcus and 10^{11} organisms per gram on the dental plate.[15,16] Considering this bioburden, the reduction obtained in the study conditions was impressive, reducing to 10^0-10^2 CFU, but this did not eliminate microorganisms supposedly susceptible to the disinfectant used.

The microorganisms recovered, although of low pathogenic potential, can behave as opportunists capable of attacking a host when environmental and immunological conditions are favorable to the microorganisms and causing infection.[17] This is yet another aspect that reinforces condemnation of the practice observed.

Table 1
Distribution of samples with positive growth by method I (membrane filtration analysis) and by method II (direct sowing of the gauze) according to microorganism and number of colony forming units per sample, São Paulo, Brazil, 2013

Membrane filtration			Method II	
Sample	Microorganism	CFU/sample	Sample	Microorganism
1, 4*, 9, 14, 19, 21, 23, 26, 35*, 48, 50	*Staphylococcus* negative coagulases	< 10	2, 7, 16	*Peptococcus* spp
3, 48	*Staphylococcus* negative coagulases	20-120	14, 15, 24, 27, 37	Gram-positive bacilli
6*, 17, 21*, 22*, 28, 31, 32, 35*, 37, 39, 41, 43, 44, 48*	Gram-positive bacilli	< 5	17	*Staphylococcus* negative coagulases Gram-positive bacilli
5	Gram-positive bacilli	16	20, 30, 32	*Staphylococcus* negative coagulases
2	*Acinetobacter baumannii*	2		-
35*, 45	*Micrococcus* spp	< 10		-
24, 6	*Penicillium* spp	1		-
4	*Candida* spp nonalbicans	2		-

*Samples that showed polymicrobial isolation.

The capability of organic residues protecting microorganisms from the action of disinfectants and sterilizers has been debated[9,18,19] and the results of the present study reinforce these data. Saliva or other residues deposited on HSHs during clinical use possibly protect the microorganisms from the action of the alcohol.

If the HSHs in this investigation had been submitted to cleaning before disinfection, the results could have shown greater safety of the practice, simply by the fact that it initiates the disinfection process from a smaller load of inoculum and the absence of organic residues.

In health care, all possible procedures to control transference of exogenous microorganisms must be redoubled. This research has shown that although the most efficient method for disinfecting is rubbing with alcohol for 90 seconds with 3 repetitions, it is concerning to recognize that in standard practice, smaller time periods are being used.[6]

It is a challenge to classify how significant the HSH equipment is and difficult to define its correct processing. HSHs can be used both as critical equipment (as in implantology) and as semicritical equipment (as in general clinical treatment). In addition, there is the complexity of the equipment and the great diversity of practices being performed. The key to safest processing of HSHs is cleaning followed by sterilization. It is imperative that both inventory and maintenance of HSHs be appropriate.

The method used describes an inability to reach germicide action with 70% w/v alcohol as an intermediate disinfectant. It is important to emphasize that, given the low initial microbial count, the method employed may have underestimated the recovery of microorganisms in the evaluation of the decontamination intervention.

Table 2
Distribution of colony forming units per microbial growth sample in the positive control group, São Paulo, Brazil, 2013

Sample	Microorganism	Colony forming units/sample
1	*Klebsiella oxytoca*	22
	Klebsiella pneumoniae	7
	Staphylococcus negative coagulases	3
	Alcaligenes spp	4
2	*Pseudomonas aeruginosa*	153
	Staphylococcus aureus	9
	Micrococcus spp	32
3	*Penicillium* spp	1
	Staphylococcus negative coagulases	1
4	*Staphylococcus* negative coagulases	5
	Micrococcus spp	15
5	*Staphylococcus* negative coagulases	16
	Corynebacterium spp	1
6	*Candida* spp	8
	Gram positive nonspore-forming bacilli	2
7	*Staphylococcus* negative coagulases	>300
8	*Staphylococcus* negative coagulases	1
9	*Staphylococcus* negative coagulases gram-positive nonspore-forming bacilli	NA*
10	*Staphylococcus* negative coagulases gram-positive nonspore-forming bacilli	NA*

NA, not applied.
*Samples performed by method II (direct sowing of gauze in culture medium).

CONCLUSIONS

The results of this investigation disprove the practice of HSH decontamination by means of rubbing with 70% w/v alcohol for 90 seconds in the absence of previous cleaning. This is substantiated by the survival of microorganisms that do not comply with the expected bactericide and fungicide action of 70% w/v alcohol in the condition of intermediate disinfection.

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