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ORIGINAL ARTICLE

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UV-C light as an alternative for disinfecting orthodontic pliers

Luz UV-C como alternativa para desinfecção de alicates ortodônticos

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ABSTRACT

Objective: Orthodontists use orthodontic pliers continuously, and these tools have a strong potential for nosocomial infections. This study aimed to compare the efficiency of three methods for disinfecting orthodontic pliers. **Material and Methods:** The active tips of 26 orthodontic pliers (distal end cutters and Weingart pliers) were contaminated with *S. aureus, E. coli,* and *C. albicans* microorganisms, viruses, and spores. The microbial control methods were 70% alcohol disinfection, glass bead sterilization (250 °C dry heat), and ultraviolet light irradiation (250 nm UV-C) for 30 and 60 seconds. The number of colony-forming units (CFU) and plaque-forming units (PFU) was quantified and compared for each microorganism after incubation in culture plates. **Results:** All tips of the pliers in the groups that received ultraviolet light or were subjected to glass bead sterilization showed a significantly lower number of spores, bacteria, and fungi than their respective control samples ($p<0.001$). Physical disinfection with UV-C light may represent a reliable alternative compared to other chemical and physical methods due to the increase in microorganisms resistant to chemical products and the emission of harmful by-products after chemical treatment. **Conclusion:** The tested microbial control methods were effective in the disinfection of orthodontic pliers, making ultraviolet-C light a promising alternative to eliminate microorganisms from pliers.

KEYWORDS

Biosafety; Contamination; Disinfection; Microorganisms; Ultraviolet light.

RESUMO

Objetivo: Os ortodontistas usam alicates ortodônticos continuamente, e essas ferramentas têm um forte potencial para infecções nosocomiais. Este estudo teve como objetivo comparar a eficiência de três métodos de desinfecção de alicates ortodônticos. **Material e Métodos:** As pontas ativas de 26 alicates ortodônticos (cortadores distais e alicates Weingart) foram contaminadas com microrganismos, vírus e esporos *S. aureus, E. coli e C. albicans.* Os métodos de controle microbiano foram desinfecção com álcool 70%, esterilização com esferas de vidro (250 °C calor seco) e irradiação com luz ultravioleta (250 nm UV-C) por 30 e 60 segundos. O número de unidades formadoras de colônias (UFC) e unidades formadoras de placas (UFP) foi quantificado e comparado para cada microrganismo após incubação em placas de cultura. **Resultados:** Todas as pontas do alicate dos grupos que receberam luz ultravioleta ou foram submetidos à esterilização com esferas de vidro apresentaram número significativamente menor de esporos, bactérias e fungos do que suas respectivas amostras controle ($p<0,001$). A desinfecção física com luz UV-C pode representar uma alternativa confiável em comparação com outros métodos químicos e físicos devido ao aumento de microrganismos resistentes a produtos químicos e à emissão de subprodutos nocivos após o tratamento químico. **Conclusão:** Os métodos de controle microbiano testados foram eficazes na desinfecção de alicates ortodônticos, tornando a luz ultravioleta-C uma alternativa promissora para eliminar os microrganismos dos alicates.

PALAVRAS-CHAVE

Biossegurança; Contaminação; Desinfecção; Microganismos; Luz ultravioleta.

INTRODUCTION

Healthcare-associated infections remain a high public health concern. Also known as "nosocomial" or "hospital" infections, they occur in patients during assistance in a hospital or other health units [1,2].

Microorganisms that cause healthcare-associated infections belong to different groups, such as Gram-negative bacteria (*Acinetobacter baumannii, Pseudomonas aeruginosa,* and *Escherichia coli)*, Gram-positive bacteria (*Staphylococcus aureus*), spore-producing bacteria (*Bacillus spp.*), yeasts (*Candida albicans*), and viruses (bacteriophage) [3].

Biosafety is highly significant for dental practice [4] and needs to be more judicious, especially at current times due to the wide SARS-CoV-2 transmission [5,6].

Current orthodontic practice must extensively reassess and restructure this infection control to prevent cross-contamination [4,7], requiring effective sterilization and disinfection methods. Sterilization completely removes microorganisms, including viruses, bacteria, fungi, and spores. Disinfection is a short-term process that reduces microbial contamination and does not remove all vegetative spores. High-level disinfection destroys all microorganisms except spores. Intermediate-level disinfection extinguishes most microorganisms, including the tuberculosis bacillus, but not all viruses and spores. Finally, low-level disinfection is when chemical agents eliminate a few microorganisms [8-10].

However, diseases may be transmitted in a dental environment from (1) patients to dentists, (2) dentists to patients, (3) one patient to another, and (4) the dental office to the community. When communicable diseases from saliva or blood contamination increase, dentists are responsible for minimizing risks, following strict aseptic principles in the dental clinical environment, which includes the dental chair, laboratory equipment, light cables, suction tips, high- and low-rotation pens, curing units, sinks, laptops, pens, loupes, and keys, among others [11].

The infection control methods used in dental offices are steam autoclaving (121 °C - 20 minutes), dry-heat oven (180 °C - 60 minutes), glass bead sterilization (218-240 °C - 10 to 60 seconds), exposure to gaseous agents (ethylene oxide), and disinfection by chemical agent immersion. The Brazilian Health Regulatory Agency (ANVISA) recommends steam autoclaving for sterilizing orthodontic pliers because it completely removes microorganisms. However, this method has some disadvantages, such as long instrument exposure and cooling times, high cost, and, given the metallic composition, large hinge areas and sharp edges that must be cleaned and dried before sterilization to minimize damage and corrosion [9].

There are three categories of dental instruments (critical, semi-critical, and non-critical) according to the risk of infection, and sterilization need is determined between use and contamination levels [12]. Orthodontic pliers are semi-critical instruments.

The most common disinfectants are formaldehyde, glutaraldehyde, peracetic acid, potassium peroxymonosulfate complexes, phenols, alcohols, iodine compounds, chlorate compounds, quaternary ammonium salts, and chlorhexidine [4,13-18]. However, several studies have shown that orthodontists still prefer chemical disinfection, and only glutaraldehyde can be considered a high-level disinfectant for semi-critical materials, despite being a tissue irritant and potentially causing allergic reactions to handlers, which is a disadvantage for daily use [19].

Isopropyl alcohol (70%) is an intermediate-level disinfectant used for disinfecting surfaces and instruments. Alcohol precipitates nucleic acids, denatures proteins, and dissolves fats, thus performing antimicrobial action. It is a fast-acting bactericidal, slightly irritant, inexpensive, non-toxic, and colorless, and does not leave residues [13]. However, alcohol has disadvantages, such as the absence of sporicidal activity; decreased activity in the presence of organic matter; fast volatilization with lower antimicrobial activity in dry blood, saliva, and other organic matter; non-acceptance by the ADA (American Dental Association) as a fixed surface and instrument disinfectant, potentially corroding metallic instruments; inactivity against hydrophilic viruses; no residual action; and it is an intermediate-level disinfectant [10,19]. Orthodontists use it extensively in clinical practice to disinfect orthodontic pliers because these instruments are expensive and unfeasible to own several, especially because orthodontic

care presents short appointments and simple, relatively fast, and inexpensive procedures to justify the process of microorganism elimination.

Glass bead sterilization can be used as a fast and convenient method to sterilize directly contaminated instruments. This method includes a metal recipient and glass beads with a diameter between 1.0 and 1.5 mm, at a temperature of 218-240 °C, for three to five seconds. Dutra et al. [16] showed disinfection potential efficacy of this method with only 10 seconds of exposure to pre-heated glass beads. Glass bead sterilization has shown bactericidal and viricidal effects, including the hepatitis B virus, when used for five seconds at 233 °C. Authors such as Rane et al. [20] and Sinha [10] report that an interval of three, seven, and 12 minutes is sufficient for sterilization. Manufacturers recommend using this method for 10 to 15 seconds but no more than 60 seconds because the potential for iatrogenic contact burns should be considered during application. Therefore, a cooling time of around two minutes should be allowed before using the sterilized instruments. However, it is a reliable method that can be used routinely in clinical practice [10,16,20-23] but may present a high corrosion index in metallic orthodontic instruments, mainly damaging pliers by cutting.

Ultraviolet light has been used as a disinfection method, showing a broad action spectrum against several microorganisms, such as bacteria, fungi, and viruses, destroying pathogens with antimicrobial resistance [24]. The most common ultraviolet bands are UV-C, UV-B, and UV-A, with spectral bands of 200–280 nm, 280–315 nm, and 315–380 nm, respectively. UV-C has the most potent antimicrobial/antiviral properties because it inactivates microorganisms by damaging DNA with photon absorption [25]. It can disinfect clinical environments, hospital and radiology rooms, ICUs, electronic equipment, hospital instruments, and PPEs [26-32]. The cycle time required for disinfection is relatively short. Authors report that 30 seconds are sufficient to disinfect a 35-cm-high template, as long as the UV-C source is close to the object to be disinfected. However, UV-C efficacy remarkably decreases as the distance from the lamps increases [33].

The present study aimed to compare the efficacy of three disinfection methods (UV-C light, glass bead sterilization, and 70% alcohol) in the active tips of contaminated orthodontic pliers using the microbiological method.

MATERIALS AND METHODS

This study was submitted to and approved by the ethics committee of Faculdade São Leopoldo Mandic, and it was exempt for not including patients or animals (2021-0294).

Tested microbial species and materials

The microbiological study was performed at the Microbiology Department of Faculdade São Leopoldo Mandic to assess the efficacy of three disinfection methods on the active tips of orthodontic pliers. The microorganisms used in the present study were *S. aureus* (ATCC 25923), *E coli* (ATCC 25922), *C. albicans* (ATCC 10231), a non-enveloped virus (bacteriophage), and *Geobacillus stearothermophilus* spores (ATCC 7953).

Sample selection

The study sample included 26 pliers (13 distal end cutters and 13 Weingart pliers). These pliers were selected because their active tips were different, and they were the most used in orthodontic care. All pliers were from the same brand; had the same characteristics and metal constitution/alloy; did not present grooves, wear, or fractures; were in excellent condition; and were sterilized in an autoclave for 30 minutes at 121 °C before contamination with microorganisms.

Preparation of the microbial inoculum and contamination of pliers

The *S. aureus* and *E. coli* strains were sown in BHI agar (Brain Heart Infusion KASVI, K25-1048, Spain) at 37 °C for 24 hours. For achieving the inoculum to contaminate the pliers, colonies were collected from the plate with a platinum strap and then homogenized in 5 ml of saline solution until reaching standard 1 in the McFarland scale, corresponding to a final concentration of $3x10^8$ CFU/ml.

C. albicans was sown in Sabouraud agar at 37 °C for 24 hours. For achieving the inoculum to contaminate the pliers, colonies were collected from the plate with a platinum strap and then homogenized in 5 ml of saline solution until reaching standard 1 in the McFarland scale,

corresponding to a final concentration of $3x10^8$ CFU/ml.

As for spores, a biological indicator disc (Attest, 3M, USA) for monitoring steam sterilization cycles was homogenized in 30 ml of sterile saline solution in a vortex tube agitator for 30 seconds to obtain a concentration of $3x10^4$ CFU/ml.

Bacteriophages were cultivated with *E. coli* to prepare the viral inoculum, and after replication, the purified bacteriophage solution was subjected to plaque-forming unit counts to determine the initial concentration of viral particles. A 30-ml saline solution was prepared with $3x10^8$ PFU/ml.

When the suspensions were ready, the activated tips of the pliers were immersed for 15 minutes in an inoculum solution. Then, they were removed and left to dry for 20 minutes under sterile gauze in a laminar flow chamber for 10 minutes on each side of the pliers.

Analyzed disinfection methods

Immediately after drying, the pliers underwent the following disinfection procedures: ultraviolet-C light radiation for 30 and 60 seconds, glass bead sterilization (STERI 350, Sweden) at a temperature of 218-240 °C for 30 and 60 seconds, and 70% isopropyl alcohol (Dell Cosméticos Ltda., Ibaté, SP, Brazil) by the rubbing method (gauze soaked in 1 ml of 70% alcohol) in three interpolated phases with a natural drying period of 10 minutes, as recommended by ANVISA RDC #15, 1503/2012.

This study developed a device composed of a stainless-steel box (for reflecting light) of 14.5 cm in width, 23.5 cm in height, and 25 cm in length to use the UV-C light. Also, two 8-watt ultraviolet lamps (200-240 nm) were placed at the bottom of the box (base) and another on the top (lid) (Figure 1). Stainless steel was the selected box material because of its reflection ability.

Microbiological analysis

The active surfaces of the pliers contaminated with spores, bacteria, and fungi were collected with a sterile swab soaked in sterile saline solution and rubbed on the active surfaces of the pliers. Next, this swab was sown in specific agar with the scattering technique. After seeding, the plates were incubated in a bacteriological oven at 37 °C for 24 hours. The colony-forming units (CFU) were counted with the help of a colony counter.

For the virus, the swab tip was cut with sterile scissors and placed in an Eppendorf microtube with 1 ml of sterile saline solution. The microtube was homogenized for 30 seconds, and three aliquots of 10 μL were dripped on the overlayer of Trypticase Soy Broth (TSB) 0.7% agar-agar (previously contaminated with *E. coli* at the concentration of $1.5x10^8$ CFU/ml) in a Petri dish with Tryptic Soy Agar (TSA). The plates were incubated at 37 °C for 24 hours for PFU counts.

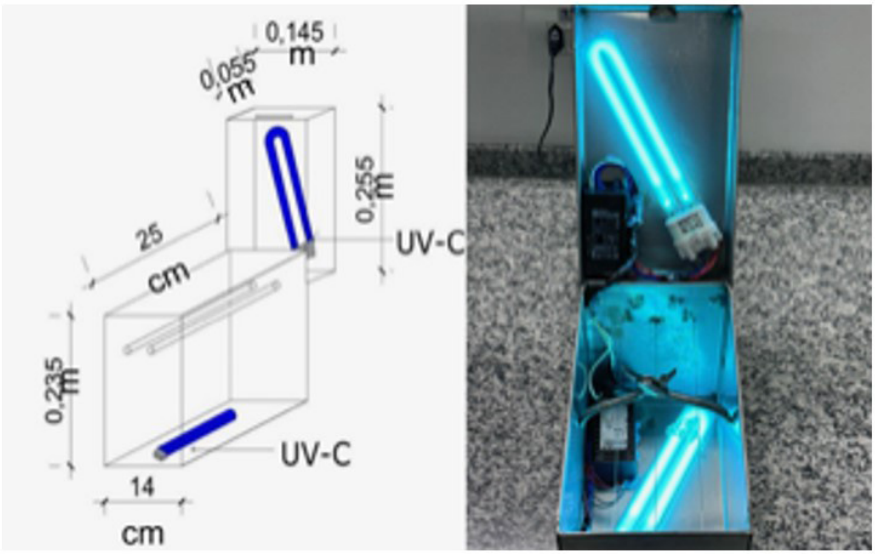

Figure 1 - UV-C light box design.

A single examiner performed the entire procedure and a second examiner randomly assessed the samples to prevent an outcome bias.

Statistical analysis

Considering the dichotomous nature (presence and absence) in the groups subjected to 70% alcohol, first, the ultraviolet light and glass bead values were also dichotomized into the presence and absence of spores, *E. co*li + S. aure*us,* and *Candida*, and subjected to Fisher's exact or G test to allow their comparison with the other groups presenting quantitative data. Next, t-tests for one sample verified the potential difference among the sterilization/disinfection method data with ultraviolet light and glass beads compared to their respective controls. Mann-Whitney tests were applied because the quantitative data (ultraviolet light and glass beads) did not adhere to a normal distribution and homoscedasticity for comparing pliers, methods, and times. In turn, Kruskal-Wallis and Dunn tests jointly compared the groups composed of the two pliers, two disinfection methods, and two times. The SPSS 23 (SPSS INC., Chicago, IL, USA) and BioEstat 5.0 (Fundação Mamirauá, Belém, PA, Brazil) performed the statistical calculations at a 5% significance level.

RESULTS

First, the mean values of the groups evaluated in a bar diagram of the relative frequency (%) of distal end cutters and Weingart pliers with tips containing spores, *E. coli* + *S. aureus* and Candida, according to the method and time of sterilization/disinfection (Figure 2).

When converting the quantitative data of ultraviolet light and glass beads into dichotomous answers (present and absent) to compare them with the similar data achieved for 70% alcohol, the study did not find a statistically significant difference between distal end cutters and Weingart pliers for spores, *E. coli* + *S. aureus* and *Candida*.

This finding applies to the individual comparison (p-values in each row in the last column of Table I) of groups that received 70% alcohol, ultraviolet light (30 and 60 seconds), or glass bead sterilization (30 and 60 seconds) and the joint contrast of all groups (p-values identified as "global comparison between the two pliers" in Table I).

The proportion of distal end cutters presenting tips with spores was not significantly affected by the sterilization/disinfection method,

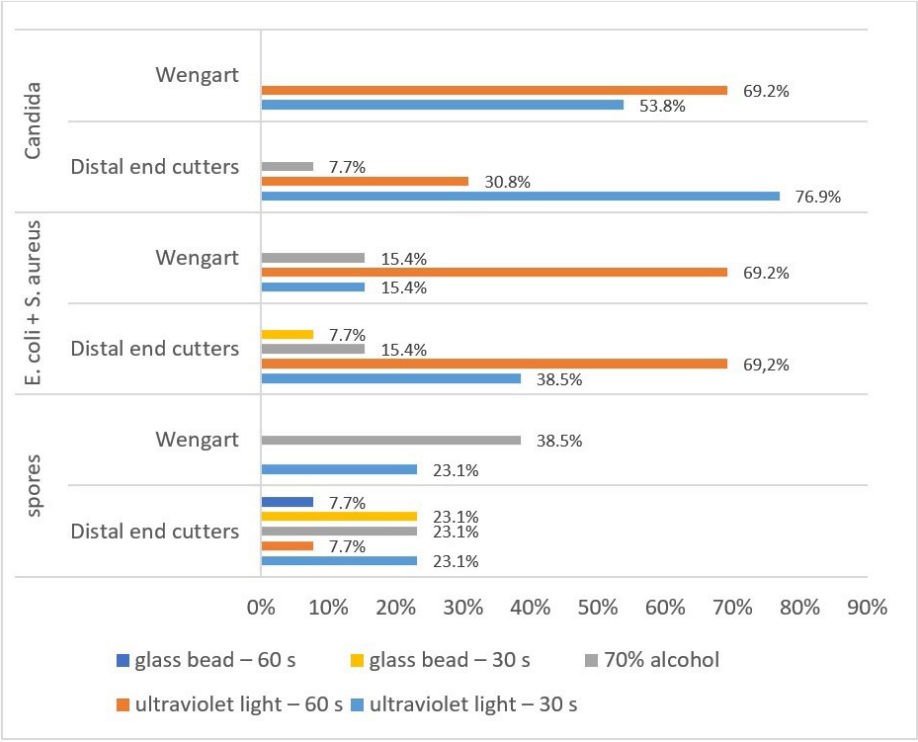

Figure 2 - Bar diagram of the relative frequency (%) in distal end cutters and Weingart pliers with tips containing with spores, *E. coli* + *S. aureus* and Candida, according to the method and time of sterilization/disinfection. Source: Produced by the authors.

Table I - Absolute (n) and relative (%) frequencies of distal end cutters and Weingart pliers with tips containing spores, *E. coli + S. aureus,* and *Candida*, according to the sterilization/disinfection method and time (CFU/ml)

	Method Distal end cutter	Pliers Weingart		p-value (comparison between pliers for each of the methods)
Spores	Ultraviolet light - 30 seconds	3 (23.1%)	3 (23.1%)	1.000*
	Ultraviolet light - 60 seconds	1 (7.7%)	0 (0.0%)	1.000*
	70% alcohol	3 (23.1%)	5 (38.5%)	0.673*
	Glass beads - 30 seconds	3 (23.1%)	0 (0.0%)	0.220*
	Glass beads - 60 seconds	1 (7.7%)	0 (0.0%)	1.000*
	p-value (global comparison between methods for each of the pliers)	0.580**	**0.002****	—
	p-value (global comparison between the two pliers)		0.138**	—
	Method Distal end cutter	**Pliers Weingart**		**p-value (comparison between pliers for each of the methods)**
E. coli and *S. aureus*	Ultraviolet light - 30 seconds	5 (38.5%)	2 (15.4%)	0.378*
	Ultraviolet light - 60 seconds	9 (69.2%)	9 (6.2%)	1.000*
	70% alcohol	2 (15.4%)	2 (15.4%)	1.000*
	Glass beads - 30 seconds	1 (7.7%)	0 (0.0%)	1.000*
	Glass beads - 60 seconds	0 (0.0%)	0 (0.0%)	1.000*
	p-value (global comparison between methods for each of the pliers)	**< 0.001****	**< 0.001****	—
	p-value (global comparison between the two pliers)		0.703**	—
	Method Distal end cutter	**Pliers Weingart**		**p-value (comparison between pliers for each of the methods)**
Candida	Ultraviolet light - 30 seconds	10 (76.9%)	7 (53.8%)	0.411*
	Ultraviolet light - 60 seconds	4 (30.8%)	9 (69.2%)	0.115*
	70% alcohol	1 (7.7%)	0 (0.0%)	1.000*
	Glass beads - 30 seconds	0 (0.0%)	0 (0.0%)	1.000*
	Glass beads - 60 seconds	0 (0.0%)	0 (0.0%)	1.000*
	p-value (global comparison between methods for each of the pliers)	**< 0.001****	**< 0.001****	—
	p-value (global comparison between the two pliers)		0.425**	—

Legend: p-values ≤ 0.05 indicate a statistically significant difference by Fisher's exact (*) or G (**) test.
Source: Produced by the authors.

as seen in the p-value in the penultimate row of results for spores in Table I. However, the tips of Weingart pliers showed significantly more samples with spores using 70% alcohol than ultraviolet light for 30 seconds. Glass bead sterilization for 30 or 60 seconds and ultraviolet light applied for longer (60 seconds) did not show spores (penultimate column and row called "global comparison between methods for each of the pliers" in Table 1).

As for *E. coli* and *S. aureus*, the proportion of samples with bacteria was significantly higher when applying ultraviolet light for 60 seconds for both tested pliers. Using this same light for 30 seconds caused a significantly higher proportion of distal end cutter tip samples with bacteria than those treated with 70% alcohol. The *E. coli + S. aureus* proportion was equal in the samples subjected to ultraviolet light for 30 seconds and 70% alcohol. The tips of Weingart pliers did not show bacteria in the groups sterilized with glass beads for either 30 or 60 seconds, but distal end cutters did not show spores when applying the beads for 60 seconds (Figure 2 and Table I).

Regarding the data of the presence/absence of *C. albicans*, the proportion of tips of distal end cutters with this fungus was significantly higher when using ultraviolet light as a sterilization/disinfection method for 30 seconds than 60 seconds. In turn, the latter showed more samples with *C. albicans* than the group treated with 70% alcohol. Glass bead sterilization did not show *Candida* in the tips of distal end cutters and Weingart pliers. However, the tips of the latter instruments sterilized with 70% alcohol also did not show fungi, which appeared in 69.2% and 53.8% of samples subjected to ultraviolet light for 60 and 30 seconds, respectively (Figure 2 and Table I).

When focusing on groups with quantitative original data of spores, *E. coli*, *S. aureus*, and *C. albicans*, meaning the groups that received ultraviolet light or glass bead sterilization, the analyses showed a significantly lower number of spores, bacteria, and fungi in the tips of all pliers and all methods than their respective controls ($p < 0.001$) (Table II).

Regarding the quantification data, the application time of 30 or 60 seconds did not

Table II - Quantification (CFU/ml and PFU/ml) of spores, *E. coli + S. aureus, Candida*, and viruses in the tips of distal end cutters and Weingart pliers, according to the sterilization/disinfection method and time

	Method	Distal end cutters			Weingart pliers		
		30 seconds	60 seconds	p-value*	30 seconds	60 seconds	p-value*
Spores	Ultraviolet light	0.23 (0.44) 0.00	0.08 (0.28) 0.00	0.505	0.38 (0.87) 0.00	0.00 (0.00) 0.00	0.317
	Glass beads	0.23 (0.44) 0.00	0.08 (0.28) 0.00	0.505	0.00 (0.00) 0.00	0.00 (0.00) 0.00	1.000
	p-value**	1.000	1.000	—	0.317	1.000	—
	Control			p-valueΨ			p-valueΨ
	Ultraviolet light	4	2	< 0.001	3	6	< 0.001
	Glass beads	1	1	< 0.001	1	1	< 0.001
	Method	**Distal end cutters**			**Weingart pliers**		
		30 seconds	**60 seconds**	**p-value***	**30 seconds**	**60 seconds**	**p-value***
E. coli/S. aureus	Ultraviolet light	2.85 (3.89) 0.00	6.31 (7.30) 5.00	0.191	0.69 (1.80) 0.00	6.46 (11.10) 2.00	**0.021**
	Glass beads	0.08 (0.28) 0.00	0.00 (0.00) 0.00	0.739	0.00 (0.00) 0.00	0.00 (0.00) 0.00	1.000
	p-value**	0.144	**0.003**	—	0.505	**0.003**	—
	Control			p-valueΨ			p-valueΨ
	Ultraviolet light	300	300	< 0.001	300	136	< 0.001
	Glass beads	277	277	< 0.001	44	44	< 0.001
	Method	**Distal end cutters**			**Weingart pliers**		
		30 seconds	**60 seconds**	**p-value***	**30 seconds**	**60 seconds**	**p-value***
Candida	Ultraviolet light	5.38 (7.61) 3.00	1.62 (3.43) 0.00	**0.003**	1.62 (2.06) 1.00	2.38 (3.59) 1.00	0.663
	Glass beads	0.00 (0.00) 0.00	0.00 (0.00) 0.00	1.000	0.00 (0.00) 0.00	0.00 (0.00) 0.00	1.000
	p-value**	**< 0.001**	0.182	—	**0.020**	**0.003**	—
	Control			p-valueΨ			p-valueΨ
	Ultraviolet light	300	300	< 0.001	300	980	< 0.001
	Glass beads	300	300	< 0.001	99	99	< 0.001
	Method	**Distal end cutters**			**Weingart pliers**		
		30 seconds	**60 seconds**	**p-value***	**30 seconds**	**60 seconds**	**p-value***
Viruses	Ultraviolet light	0.08 (0.15) 0.00	1.08 (1.38) 0.33	**0.017**	0.28 (0.47) 0.00	0.28 (0.36) 0.33	0.778
	Glass beads	—	—	—	—	—	—
	p-value**	—	—	—	—	—	—
	Control			p-valueΨ			p-valueΨ
	Ultraviolet light	16.8 (9.0)		< 0.001	2.8 (1.6)	< 0.001	
	Glass beads	—	—	—	—	—	—

*Comparisons between times for the same sterilization/disinfection method and pliers (comparisons within each row). **Comparisons between sterilization/disinfection methods for the same time and pliers (comparisons within each column). Ψp-value for the comparison with respective control groups.
Legend: Mean and standard deviation values (in parentheses) in the first row of each group and median under them. p-values ≤ 0.05 indicate a statistically significant difference by Mann-Whitney tests.

significantly interfere with spore counts in the tips of any of the two pliers and sterilization/disinfection methods (ultraviolet light and glass beads). This lack of difference between counts from the application time (30 versus 60 seconds) was repeated for *E. coli, S. aureus*, and *C. albicans* quantification, but with two exceptions. First, the tips of Weingart pliers that received ultraviolet light for 60 seconds showed a significantly higher number of *E. coli* and *S. aureus* than the 30 second application time. Second, the tips of distal end cutters that received ultraviolet light for 30 seconds showed a significantly higher number of *C. albicans* than the 60-second application time (Figure 2). As for viruses in which only ultraviolet light was investigated, the time affected counting significantly only for the tips of distal end cutters, and application for 60 seconds produced a significantly higher viral load than 30 seconds how observed in Table II.

The comparison of ultraviolet light and glass bead methods for spores did not show a significant difference, regardless of pliers and time (30 or 60 seconds). However, bacteria presented a significantly higher number of *E. coli* and *S. aureus* when using ultraviolet light than glass beads in the tips of both pliers, but only for the 60 second application time. Moreover, the number of fungi in the tips of pliers was significantly higher for the ultraviolet light application than for glass beads. There was no significant difference in *C. albicans* counts between ultraviolet light and glass beads only when subjecting distal end cutters to these methods for 60 seconds.

DISCUSSION

This study addressed the need for a new disinfection method, comparing it with the ones most commonly used by orthodontists. UV-C light was compared with 70% alcohol and glass bead sterilization.

The oral cavity is a natural habitat for numerous biological agents (microorganisms). This ecological niche may represent a reservoir of opportunistic and pathogenic microorganisms and a risk of cross-contamination and infection, potentially causing systemic infections [4,7].

The microorganisms tested in this study are often used for controlling and monitoring the action of disinfectants in specific culture media [3]. During orthodontic care, pathogens may be transposed through a direct interaction of contaminated instruments or materials [5,6]. However, orthodontists often neglect the sterilization method, treating it as something that may reduce profitability and efficacy in the dental office due to the need for investing in several orthodontic pliers and the time demand, respectively. This situation makes orthodontists consider disinfection an alternative to sterilization, which is a common mistake [19].

Orthodontic pliers present high contamination rates, so microorganism dissemination through these tools must not be neglected [12]. The present study did not show statistically significant differences in pliers contaminated with spores, *E. coli + S. aureus,* and *C. albicans*, but the CFU percentage decreased for microorganisms, thus promoting disinfection instead of sterilization.

The tests to verify the sufficiency of 70% alcohol rub, glass bead sterilization, and exposure to UV-C light after the clinical use of these pliers showed a decrease in microorganisms, as expected. However, all pliers maintained some degree of contamination, remaining potential infectants from the biological standpoint.

The 70% alcohol is an intermediate-level disinfectant efficient in disinfecting semi-critical items, showing consistent disinfection in this study. However, the ease of use, low cost, and virtually non-existent toxicity associated with the false impression of infection control make several clinicians to use it in their instruments between patient appointments [10,13,17,19].

The glass bead method applied for 30 and 60 seconds did not show microorganisms, corroborating a previous study, which is similar to the results of Kangane et al. [21], who found that 30 seconds of exposure would be sufficient to disinfect the tips of orthodontic pliers. Therefore, it would take approximately 20 minutes to warm the beads to 250 °C, increasing the time for instrument use and representing a fast and convenient method for high-level disinfection. However, it presents a high potential for iatrogenic contact burns and a high corrosion index and requires a cooling time of two minutes before using the sterilized instruments.

Although there was no statistically significant difference among the tested methods, contamination with *S. aureus* and *E. coli* was

more frequent in the UV-C light method for 30 or 60 seconds, corroborating authors who deemed the assessment of irradiation and time of exposure to UV-C light essential for disinfection with this method [24,25,30,33]. A study by El Haddad et al. [27] found that two minutes of exposure would be sufficient to reduce by 70% the bacterial load of surfaces in hospital operating rooms, showing that time and irradiation affect disinfection efficiency [28].

The comparison of light with other methods and microorganism varieties involved in health-related infections did not find an adequate disinfection capacity for UV-C light even after 60 seconds of exposure to irradiation. The insufficient disinfection efficacy with this method might be due to its low ability to reach all sides of the active tips of pliers, suggesting an alternative for further studies to develop a movement in pliers for complete irradiation.

However, this study showed that UV-C light disinfects the active tips of orthodontic pliers. This method is not adequate for sterilization but is often used for disinfection, indicating a reliable alternative compared to other physical and chemical methods. The present study shows the antimicrobial effectiveness of UV-C light, but future studies with new UV-C light equipment with greater emission power can be tested in order to reduce the application time.

The impact of glass bead sterilization on viruses was not assessed for the active tips of orthodontic pliers, so the results do not guarantee complete instrument disinfection. It is worth noting that the microbial load used for infecting the instruments is perhaps much higher than that applied in the clinical routine.

CONCLUSION

Glass bead sterilization was the most effective of the three disinfection methods in the active tips of orthodontic pliers when used for microorganisms, compared to 70% alcohol and UV-C light.

The 70% alcohol is easy to use, inexpensive, and virtually non-toxic, but it is not an effective method for semi-critical instruments.

UV-C light significantly reduced contamination levels in orthodontic pliers but less than glass bead sterilization. Nonetheless, it represented an alternative method due to the increase in microorganisms resistant to chemical products and the emission of harmful by-products after chemical treatment. Therefore, further studies are required because the simplicity and low resource demand may suggest promising dental clinical applicability.

Author's Contributions

LRL: Conceptualization, Investigation, Resources, Data Curation, Writing – Original Draft Preparation. RSN, AO: Writing – Original Draft Preparation, Writing – Review & Editing. SSS, ASG: Conceptualization, Methodology, Supervision, Project Administration, Writing Review & Editing.

Conflict of Interest

The authors have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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Regulatory Statement

This study was conducted in accordance with national guidelines for the supervision of human and animal research. The study was carried out after approval by the Research Ethics Committee of Faculdade São Leopoldo Mandic. The study was waived because it did not include patients or animals, standard microbiological samples acquired for laboratory studies were used. The approval code for this study is 2021-0294.

REFERENCES

1. Jeong EK, Bae JE, Kim IS. Inactivation of influenza A virus H1N1 by disinfection process. Am J Infect Control. 2010;38(5):354-60. http://dx.doi.org/10.1016/j.ajic.2010.03.003. PMid:20430477.
2. Protano C, Cammalleri V, Romano Spica V, Valeriani F, Vitali M. Hospital environment as a reservoir for cross transmission: cleaning and disinfection procedures. Ann Ig. 2019;31(5):436-48. http://dx.doi.org/10.7416/ai.2019.2305. PMid:31304524.
3. Guridi A, Sevillano E, de la Fuente I, Mateo E, Eraso E, Quindós G. Disinfectant activity of a portable ultraviolet c equipment. Int J Environ Res Public Health. 2019;16(23):4747. http://dx.doi.org/10.3390/ijerph16234747. PMid:31783593.

4. Hassan SS, Ghaib NH, Al-Ghurabi BH. Assessment of bacterial contamination of orthodontic arch wire. J. Baghdad Coll. Dent. 2019;31(1):48-51. http://dx.doi.org/10.26477/jbcd.v31i1.2578.
5. Kimbonguila A, Matos L, Petit J, Scher J, Nzikou J-M. Effect of physical treatment on the physicochemical, rheological and functional properties of yam meal of the cultivar "Ngumvu" from *Dioscorea alata* L. of Congo. Int J Recent Sci Res. 2019;10:30693-5. https://dx.doi.org/10.24327/ijrsr.2019.1011.4175.
6. Saltaji H, Sharaf KA. COVID-19 and orthodontics: a call for action. Am J Orthod Dentofacial Orthop. 2020;158(1):12-3. http://dx.doi.org/10.1016/j.ajodo.2020.04.006. PMid:32600749.
7. Ahmad E, Shamim W, Rathore S, Bhatt S, Moiz MA, Bhatt P. Upgrading sterilization in the orthodontics practice: a review. Int J Preventive Clin Dental Res. 2014;1(4):80-4.
8. Brasil. Coordenação de Controle de Infecção Hospitalar. Processamento de artigos e superfícies em estabelecimentos de saúde. Brasília; 1994.
9. Lall R, Sahu A, Jaiswal A, Kite S, Sowmya AR, Sainath MC. Evaluation of various sterilization processes of orthodontic instruments using biological indicators and conventional swab test method: a comparative study. J Contemp Dent Pract. 2018;19(6):698-703. http://dx.doi.org/10.5005/jp-journals-10024-2322. PMid:29959299.
10. Sinha A. Sterilization and disinfection in orthodontics: a literature review. Indian J Public Health Res Dev. 2019;10(11):736. http://dx.doi.org/10.5958/0976-5506.2019.03568.X.
11. Umar D, Basheer B, Husain A, Baroudi K, Ahamed F, Kumar A. Evaluation of bacterial contamination in a clinical environment. J Int Oral Health. 2015;7(1):53-5. PMid:25709369.
12. Azeredo F, Menezes LM, Silva RM, Rizzatto SMD, Garcia GG, Revers K. Microbiological analysis of orthodontic pliers. Dental Press J Orthod. 2011;16(3):103-12. http://dx.doi.org/10.1590/S2176-94512011000300013.
13. Tonetto MR, Silva MAS, Kuga MC, Bandeca MC, Pinzan-Vercelino CRM, Carvalho MRA, et al. Comparison of antimicrobial activity between chemical disinfectants on contaminated orthodontic pliers. J Contemp Dent Pract. 2015;16(8):619-23. http://dx.doi.org/10.5005/jp-journals-10024-1731. PMid:26423496.
14. Laneve E, Raddato B, Dioguardi M, Di Gioia G, Troiano G, Lo Muzio L. Sterilisation in dentistry: a review of the literature. Int J Dent. 2019;2019:6507286. http://dx.doi.org/10.1155/2019/6507286. PMid:30774663.
15. Alsheekhly B, Adnan MM, Al-Zubaydi FS. The effect of multiple glass beads sterilization cycles on cyclic fatigue of AF BLUE S one file. Indian J Forensic Med Toxicol. 2020;14(3):2500-5. http://dx.doi.org/10.37506/ijfmt.v14i3.10812.
16. Dutra SR, Santos VR, Menezes LFS, Drummond AF, Vilaça ÊL, Couto PHA. Esterilização em Ortodontia: eficácia do esterilizador com esferas de vidro. Rev Dent Press Ortodon Ortop Facial. 2008;13(4):60-6. http://dx.doi.org/10.1590/S1415-54192008000400007.
17. Reddy RV, Tanveer K, Sharma KD, Kokkula N, Suresh PL, Sudhakar M. Evaluation of effectiveness of chemical disinfectants in reducing bacterial growth on orthodontic instruments. J Contemp Dent Pract. 2013;14(6):1039-43. http://dx.doi.org/10.5005/jp-journals-10024-1447. PMid:24858747.
18. Wichelhaus A, Bader F, Sander FG, Krieger D, Mertens T. Effektivität der Desinfektion orthodontischer Zangen. J Orofac Orthop. 2006;67(5):316-36. http://dx.doi.org/10.1007/s00056-006-0622-9. PMid:16953352.
19. Venturelli AC, Torres FC, Almeida-Pedrin RR, Almeida RR, Almeida MR, Ferreira FPC. Avaliação microbiológica da contaminação residual em diferentes tipos de alicates ortodônticos após desinfecção com álcool 70%. Rev Dent Press Ortodon Ortop Facial. 2009;14(4):43-52. http://dx.doi.org/10.1590/S1415-54192009000400005.
20. Rane J, Adhikar P, Bakal RL. Molecular imprinting: an emerging technology. Asian J Pharm. 2015;3(11):75-91.
21. Kangane SK, Sawant SK, Patil PS. Instrument sterilizationin the orthodontic clinic: a review. Int J Clin Dental Sci. 2010;1:53-8.14.
22. Ray S, Chopra SS, Mitra R, Jain A. Reliability of glass bead sterilization for tried-in orthodontic bands. J Indian Orthod Soc. 2011;45(4):189-92. http://dx.doi.org/10.1177/0974909820110406.
23. Ronque PPT, Verzosa LG. Effectiveness of the glass-bead sterilizer in ophthalmic instruments. Philipp J Ophthalmol. 2007;32(1):25-7.
24. Botta SB, Teixeira FS, Hanashiro FS, Araújo WWR, Cassoni A. Salvadori MCBSbs. Ultraviolet-C decontamination of a dental clinic setting: required dose and time of UVC light. Braz Dent Sci. 2020;23(2):1-10. http://dx.doi.org/10.14295/bds.2020.v23i2.2275.
25. Nerandzic MM, Cadnum JL, Eckart KE, Donskey CJ. Evaluation of a hand-held far-ultraviolet radiation device for decontamination of Clostridium difficile and other healthcare-associated pathogens. BMC Infect Dis. 2012;12(1):120. http://dx.doi.org/10.1186/1471-2334-12-120. PMid:22591268.
26. Cutler TD, Zimmerman JJ. Ultraviolet irradiation and the mechanisms underlying its inactivation of infectious agents. Anim Health Res Rev. 2011;12(1):15-23. http://dx.doi.org/10.1017/S1466252311000016. PMid:21676338.
27. El Haddad L, Ghantoji SS, Stibich M, Fleming JB, Segal C, Ware KM, et al. Evaluation of a pulsed xenon ultraviolet disinfection system to decrease bacterial contamination in operating rooms. BMC Infect Dis. 2017;17(1):672. http://dx.doi.org/10.1186/s12879-017-2792-z. PMid:29017457.
28. Cadnum JL, Jencson AL, Gestrich SA, Livingston SH, Karaman BA, Benner KJ, et al. A comparison of the efficacy of multiple ultraviolet light room decontamination devices in a radiology procedure room. Infect Control Hosp Epidemiol. 2019;40(2):158-63. http://dx.doi.org/10.1017/ice.2018.296. PMid:30698135.
29. Jinadatha C, Simmons S, Dale C, Ganachari-Mallappa N, Villamaria FC, Goulding N, et al. Disinfecting personal protective equipment with pulsed xenon ultraviolet as a risk mitigation strategy for health care workers. Am J Infect Control. 2015;43(4):412-4. http://dx.doi.org/10.1016/j.ajic.2015.01.013. PMid:25726129.
30. Knox RW, Demons ST, Cunningham CW. A novel method to decontaminate surgical instruments for operational and austere environments. Wilderness Environ Med. 2015;26(4):509-13. http://dx.doi.org/10.1016/j.wem.2015.03.030. PMid:26165581.
31. Penno K, Jandarov RA, Sopirala MM. Effect of automated ultraviolet C–emitting device on decontamination of hospital rooms with and without real-time observation of terminal room disinfection. Am J Infect Control. 2017;45(11):1208-13. http://dx.doi.org/10.1016/j.ajic.2017.06.015. PMid:28757085.
32. Rutala WA, Weber DJ. Disinfectants used for environmental disinfection and new room decontamination technology. Am J Infect Control. 2013;41(5, Suppl):S36-41. http://dx.doi.org/10.1016/j.ajic.2012.11.006. PMid:23622746.
33. Alhmidi H, Cadnum JL, Piedrahita CT, John AR, Donskey CJ. Evaluation of an automated ultraviolet-C light disinfection device and patient hand hygiene for reduction of pathogen transfer from interactive touchscreen computer kiosks. Am J Infect Control. 2018;46(4):464-7. http://dx.doi.org/10.1016/j.ajic.2017.09.032. PMid:29174655.

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