

Comparison of disinfection protocol of irreversible hydrocolloid (alginate) impressions through plastic and metallic trays

Comparação do protocolo de desinfecção de moldes com hidrocolóide irreversível (alginate) em moldeiras plásticas e metálicas

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ABSTRACT

Introduction: Many studies have evaluated the disinfection of irreversible hydrocolloid impressions through different disinfecting agents. However, impression trays can be source of cross-infection requiring disinfection. This study aimed to determine which would be the most suitable tray (metallic or plastic), available in dental market, and the ideal time to achieve disinfection by using 1% sodium hypochlorite poured into the alginate impression. **Material and method:** Thirty dental impressions from the patients aged from 7-12 years and treated in the Discipline of Orthodontics of the institution were divided into two groups according to the impression tray type: 15 impressions through plastic tray (Morelli) and 15 impressions through metallic tray (Tecnodont). The material collection was performed before and after the application of 1% sodium hypochlorite for 3, 5 and 10 min. After the incubation period of 48 h at 37 °C, the microorganism colonies were counted on the plates presenting from 30 to 300 colonies to determine the colony-forming unit (CFU) per mL. CFU/mL results were transformed into logarithm and submitted to statistical analysis by applying ANOVA and Tukey test ($p \leq 0.05$). **Results:** Greater CFU percentage reduction occurred in alginate after three min, in both tray types. Concerning to tray types, it could be observed that the plastic tray showed 100% of reduction after 5 min while the metallic tray exhibited 81.49% of reduction after 3 min. **Conclusion:** 1 – The plastic tray showed the most effective disinfection after 5 min, with 100%

RESUMO

Introdução: Muitos são os trabalhos que avaliam a desinfecção das moldagens com hidrocolóide irreversível e com diferentes agentes desinfetantes, mas as moldeiras também podem ser vetores de infecção cruzada e é necessária a sua desinfecção. A proposta do trabalho foi determinar qual a melhor moldeira (metálica ou plástica), disponíveis no mercado para a utilização do cirurgião dentista, e o tempo ideal para se obter a desinfecção utilizando o hipoclorito de sódio a 1% vertido na moldagem de com alginate. **Material e método:** Foram obtidas 30 moldagens de pacientes em tratamento na Disciplina de Ortodontia, do Curso de Odontologia, do ICT-UNESP-SJC, com idades entre 7 e 12 anos, divididas em dois grupos de acordo com o tipo de moldeira empregada: 15 moldagens com moldeiras de plástico (Morelli) e 15 com moldeiras de metal (Tecnodont). A coleta de material foi realizada antes e após a aplicação do hipoclorito de sódio a 1% durante 3, 5 e 10 min. Após período de incubação de 48 h a 37 °C foram contadas colônias de microorganismos nas placas que apresentaram de 30 a 300 colônias para determinação de Unidades Formadoras de Colônias (UFC) por mL. Os resultados em UFC/mL foram transformados em logaritmo e submetidos à análise estatística Anova e teste de Tukey ($p \leq 0,05$). **Resultados:** No alginate ocorreu uma maior redução percentual de UFC após 3 min, em ambas as moldeiras. Em relação a maior redução nas moldeiras, pudemos observar que a moldeira plástica ocorreu 100% de redução após 5 min e 81,49% de redução na moldeira metálica após 3 min. **Conclusão:** 1 - A moldeira plástica apresentou desinfecção mais eficiente, após 5 min, com redução de 100% de UFC; 2 - O tempo mais eficaz de desinfecção da moldagem com hipoclorito

of CFU reduction; 2 – The most effective time of disinfection with 1% sodium hypochlorite poured into the impression was 5 min, for both tray types.

KEYWORDS

Dental impression materials; Disinfection; Sodium hypochlorite; Alginates.

de sódio a 1% vertido na moldagem foi de 5 min, para moldeira de plástico e de metal.

PALAVRAS-CHAVE

Materiais para moldagem odontológica; Desinfecção; Hipoclorito de Sódio; Alginatos.

INTRODUCTION

Irreversible hydrocolloid, so-called alginate, is a material indicated for impressions demanding less details and accuracy, while other materials as polysulfide, polyether, and condensation silicone are more indicated when more accurate dental casts are necessary to construct indirect restorations [1]. Notwithstanding, alginate has been preferentially used because of relatively low cost and easy handling [2].

Currently in Dentistry, there is an increasingly concern about the transmission of infectious agents to the professionals involved in the construction of prostheses and orthodontic appliances. The risk of acquiring infectious diseases is very high because of the direct contact of the dentist with the oral cavity, which contains saliva and blood that have been considered potential transmission sources of some microorganisms.

The careful use of mechanical barriers (gloves, caps, aprons, protective glasses) and the concern of working in aseptic medium not only should occur during the handling of instruments and devices, but also caution should be given during the handling of the impressions because they have the capacity of storing pathogenic microorganisms, such as: *Streptococcus mutans*, *Salmonella choleraesuis*, *Bacillus subtilis*, *Mycobacterium bovis*, and act as medium to disseminate diseases among patients, dentists, dental assistants, and dental technicians [3].

After obtaining the impression, it is essential that its disinfection is executed before

dental stone is poured, since microorganism transmission from contaminated impressions to dental casts has been proved [4-9]. If the disinfection procedure is not carried out, the dental casts obtained from the contaminated impression and sent to either dental prosthesis or orthodontic laboratory could transmit pathogenic microorganisms. Notwithstanding, the dental trays are also contaminated with oral fluids, so that they can be vectors of cross-infection and their disinfection is mandatory to enable the safe handling of dental impressions.

Researches have been conducted aiming to determine which would be the most suitable disinfection method to be routinely applied to obtain orthodontic study/working cast and to avoid cross-infection [6-8,10]. According to Philips et al. [11], to avoid dimensional alterations of the orthodontic dental cast, both the disinfection and the stone pouring into the alginate impression should be fast.

The disinfection method through immersion of the alginate may result in soaking and altering the physical dimensions of the impressions, while the spraying method could not totally covered the contaminated area [12]. Impression disinfection becomes an indispensable procedure at dental offices because of the increase in infectious diseases. The alginate during the impression procedure contacts with patient's saliva, dental biofilm, and blood and may easily transmit viral diseases (herpes, hepatitis, and AIDS) to the dentist, dental staff, and dental technician who will manipulate these casts. Researches have concluded that the impression disinfection is highly necessary [6,7,12,13].

Osório et al. [13] verified that 100% of alginate impressions not submitted to disinfection exhibited positive results in the analysis of culture medium turbidity and microscopic analysis of bacterial presence. It is important that the disinfection agent does show satisfactory antimicrobial potential and does not degrade the physical properties of the impression material and resulting dental casts.

Bergman, Mauad and Olsson [14] recommended that all impressions should be washed in running water to remove saliva and blood, then to be disinfected, and sent to dental laboratory, safely and avoiding the risk of cross-infection.

Cucci et al. [15] recommended that alginate impression should be stored in 100% relative humidity environment during the period elapsed from the impression to dental stone pouring.

Because most of impression materials do not support high temperatures without undergoing distortion, one should opt for chemical agents promoting proper sterilization and disinfection [11]. The disinfection agents should still have low toxicity, easy handling, and low cost.

Casemiro et al. [12], evaluated the level of contamination of the following six alginate commercial brands: Jeltrate - 389 UFC/g; Jeltrate Plus - 516 UFC/g; Jeltrate Chromatic - 135 UFC/g; Hydrogum - 1,455 UFC/g; Kromopan - 840 UFC/g; and Greengel - 59 UFC/g (two containing chlorhexidine) and identified the contamination present in these materials with viable microorganisms. The authors observed the presence of bacteria (*Staphylococcus epidermidis*, *Bacillus subtilis*, *Bacillus* sp., *Bacillus coagulans*, *Bacillus licheniformis*, *Bacillus cereus*, *Micrococcus luteus*, and *Nocardia* sp.); filamentous fungi (*Aspergillus niger*, *Aspergillus flavus*, *Rhizopus* sp., *Neurospora* sp.); and yeasts (*Candida* sp.). The authors concluded that all studied alginate types showed viable bacteria, fungi,

and yeasts. Notwithstanding, the materials containing chlorhexidine exhibited the lowest levels of contamination. The contamination detected in the materials pointed out the necessity for adopting measures to improve the control of microbiological quality. The use of contaminated materials inside the mouth does not meet the basic principles of cross-infection control and may put at risk impaired or immunocompromised patients. Methods of sterilization such as gamma radiation should be performed at the ending of production. Notwithstanding, gamma radiation method requires the previous knowledge of the microbial load, both qualitatively and quantitatively, aiming to determine the doses to be applied. These authors concluded that alginate Jeltrate and Greengel obtained an effective result to reduce cross contamination caused by the impressions.

Esteves et al. [3] evaluated the antimicrobial efficacy of alginates considered as self-disinfecting (containing 1% sodium hypochlorite and 2% chlorhexidine) compared with conventional alginates. The microorganisms used for the experimental trials were *Streptococcus mutans* and *Staphylococcus aureus*. The authors concluded that the antimicrobial action of self-disinfecting alginates were more effective for *Streptococcus mutans* and ineffective for *Staphylococcus aureus*. 2% chlorhexidine and 1% sodium hypochlorite were effective for both microorganisms.

Researches have shown that the use of, for example, either 1% sodium hypochlorite or 2% chlorhexidine, at time intervals ranging from 10 to 30 min, is an excellent approach to promote the inactivation of pathogenic microorganisms inside the impressions and the disinfection technique could be either immersion or spray [6,7,8,10,13].

This study aimed to determine which would be the most suitable tray (metallic or plastic), available in dental market, and the ideal time to achieve disinfection by using 1% sodium hypochlorite poured into the alginate impression.

MATERIAL AND METHODS

This present study was submitted and approved by the Institutional Review Board under protocol number #309.520.

Thirty impressions were obtained from patients aged from 7-12 years and treated in the Discipline of Orthodontics of the Course of Dentistry of the institution. The impressions were divided into two groups according to the tray type: 15 impressions obtained through plastic trays (Morelli, Sorocaba, São Paulo, Brazil) and 15 impressions obtained through metallic stainless steel metallic tray (Tenax Tecnodent, São Paulo, São Paulo, Brazil).

Both plastic and metallic trays were autoclaved individually and the impression material proportion was performed according to the manufacturer's instruction.

The impressions were executed in the Clinics of Pediatric Dentistry with irreversible hydrocolloid (Jeltrate II, Dentsply, Petrópolis, Rio de Janeiro, Brazil), during the morning period and sent inside plastic boxes to the microbiology laboratory.

Both the impression and the tray were disinfected with 1% sodium hypochlorite solution (Asfer, São Caetano do Sul, São Paulo, Brazil), poured into the impression-tray assembly. The material collection was carried out before and after the application of sodium hypochlorite solution for 3, 5 and 10 min, according to chart 1.

The material collection was executed with the aid of sterile swab (Absorve, Cral, São

Paulo, SP, Brazil) on one half of the impression for each phase of the study. The *swab* was immediately placed inside tubes containing 3 mL of phosphate buffer solution (PBS) and activated for 3 min inside vortex mixer. Next, the swab was discarded and the tubes centrifuged at 1300 x g for 10 min. The supernatant was discarded and the deposit resuspended in 2.5 mL of PBS and activated for 1 min. Following, serial dilutions were performed and 0.1 mL of each dilution was plated on the surface of Brain Heart Infusion (BHI) agar plates (Himedia, Mumbai, India) supplemented with 5% of sheep blood and incubated at 37 °C for 48 h.

After the collection of the material at phase 1, the impressions were washed by pouring 1% sodium hypochlorite solution. Because of the different tray sizes and different alginate amount, sodium hypochlorite volume was not standardized, but the amount required was enough for covering all alginate and tray areas. Elapsed the disinfection time period of 3, 5, and 10 min, the impressions were washed with sterile distilled water in sufficient amount to cover all alginate and tray areas. Next, the material was collected with the aid of another sterile swab on the contralateral side of phase 1 collection and plated as aforementioned.

Elapsed the incubation period of 48 h at 37 °C, microorganism colonies were counted on the plates exhibiting from 30 to 300 colonies to determine the colony-forming units (CFU) per mL. CFU/mL results were transformed into logarithm and submitted to statistical analysis by applying ANOVA and Tukey test ($p \leq 0.05$).

RESULTS

Two-way ANOVA was used to evaluate which would be the best tray type (metallic or plastic), the tray type that would show the easiest sterilization procedure, and the ideal time to achieve disinfection before dental stone pouring. Tukey test was applied to evaluate the differences among disinfection time periods of the studied samples. Graph Pad Prism software was used to demonstrate the CFU percentage reduction.

Chart 1 - Sample distribution according to the tray type and disinfection time periods.

Tray types (n = 30)	Disinfection time periods	
		3 min (n = 5)
Plastic tray (n = 15)	5 min (n = 5)	
	10 min (n = 5)	
	3 min (n = 5)	
Metallic tray (n = 15)	5 min (n = 5)	
	10 min (n = 5)	

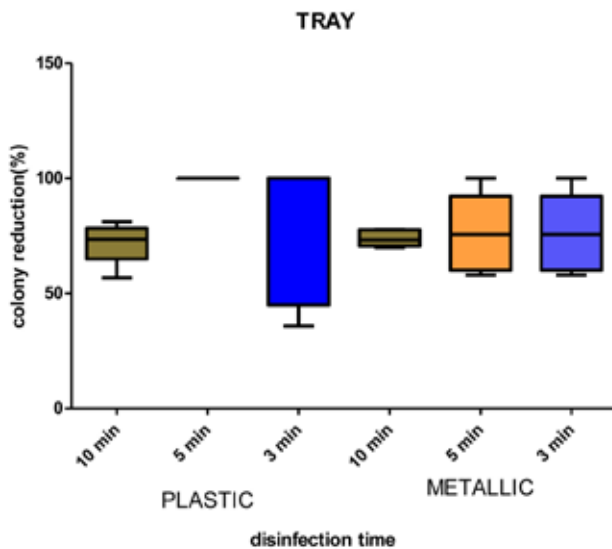
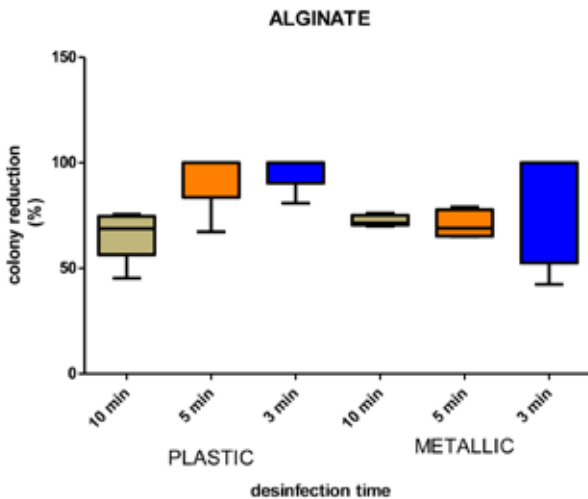
Graph 1 - Colony reduction (%) of alginate employing two tray types after 3, 5 and 10 min of 1% sodium hypochlorite solution.**Graph 2** - Colony reduction (%) of plastic and metallic trays after 3, 5 and 10 min of 1% sodium hypochlorite solution.

Table 2 shows that the difference in CFU percentage reduction among the different disinfection time periods was statistically significant for the assembly alginate+plastic tray when comparing 10 and 5 min and 10 and 3 min.

DISCUSSION

Souza et al. [16], evaluated the disinfection procedure by applying 1% sodium hypochlorite and 2% glutaraldehyde solutions but did not

Table 1 - CFU percentage means on alginate+plastic tray (AP) and alginate+metallic tray (AM) and the tray type (plastic - PT; metallic - MT) after the disinfection time periods of 3, 5 and 10 min.

	AP	AM	PT	MT
3 min	96.16%	80.98%	77.98%	81.49%
5 min	93.47%	71.01%	100%	76.08%
10 min	66.17%	72.54%	72.04%	73.97%

Table 2 - Difference of CFU percentage reduction between disinfection time periods of 10 and 5 min, 10 and 3 min, and 5 and 3 min of 1% sodium hypochlorite solution, considering alginate+plastic (AP), alginate+metallic (AM), plastic tray (PT) and metallic tray (MT).

	10 min and 5 min	10 min and 3 min	5 min and 3 min
AP	-2729 *	-29.98 *	-2.690 ns
AM	1532 ns	-8.436 ns	-9.968 ns
PT	-2795 ns	-5.934 ns	22.02 ns
MT	-2112 ns	-7.518 ns	-5.406 ns

Tukey test ($p \leq 0.05$)

* statistically significance

ns no statistically significance

verify whether it would be difference in their efficacy. These authors observed that both sodium hypochlorite and glutaraldehyde did not caused significant dimensional alterations compared with elastomers, after disinfection for 10 min ($p \leq 0.05$). They affirmed that 1% sodium hypochlorite spray is the most indicated technique because immersion technique tended to erode the metallic trays and used more material amount [7,16,17,18]. In this present study, 1% sodium hypochlorite was poured into the impression for 10 min, corroborating the literature. Notwithstanding, shorter time periods can be used because there were no statistically significant differences when the periods of 3 and 5 min were used (Table 2).

Bergman Maud and Olsson [14], Bergman [19] and Pedrosa [20] concluded that disinfection spray solutions showed better results than those of disinfection immersion solutions regarding to dimensional accuracy of dental casts. However,

in this present study, we used a technique that poured 1% sodium hypochlorite which made disinfection practice easy because it dispenses the use of either sprayer or flask to immerse the impressions, so that the solution can be poured directly from the solution flask/bottle. Because of the methodological differences regarding to the disinfection solution application and the study objectives, comparisons with the literature were not possible. Notwithstanding, we can affirm that the pouring of the disinfection solution on the impressions produced significant reduction in the number of bacterial colonies.

According to Osório et al. [13], alginate impression immersion inside disinfection solutions for 10 min is an effective disinfection method that does not provoke any dimensional alteration in dental cast. The results of this present study corroborate this affirmation, especially for metallic trays, although this study's disinfection method was different. It is worth emphasizing that when using plastic trays, the time period can be reduced for 5 min, as shown by table 2.

CONCLUSION

1 – The plastic tray presented the most effective disinfections after 5 min, with CFU reduction of 100%;

2 – The most effective time period of 1% sodium hypochlorite disinfection poured into the impression was of 5 min, both for plastic and metallic trays.

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