# Influence of surgical instrument identifiers on microbial contamination of scalpel handles after cleaning and sterilization

## *Influência dos identificadores de instrumental cirúrgico na contaminação microbiana de cabos para lâminas de bisturi após limpeza e esterilização*

## *Influencia de los identificadores de instrumental quirúrgicos en la contaminación microbiana de mangos para cuchillas de bisturí después de la limpieza y esterilización*

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**ABSTRACT: Objective:** To evaluate the influence of different instrument identifiers on the microbial growth of critical devices after cleaning and sterilization steps. **Method:** Fifteen No. 3 scalpel handles were used as test specimens, divided into 3 groups (n=5), with each group consisting of an instrument without an identifier and four other instruments with the following identifiers: laser engraving, silicone ring, vinyl identifier with permanent adhesive and personalized adhesive label. After being submerged in the surgical aspirate, the instruments were processed in accordance with the good practice requirements of Resolution No. 15/2012 from the National Health Surveillance Agency (Anvisa). Microbial growth was evaluated at different times qualitatively, by the presence or absence of growth, and quantitatively for positive cases, by counting viable colony-forming units (log CFU/scalpel handle). **Results:** At some point, there was microbial growth on the instruments regardless of the type of identifier. However, there was no continuous and progressive contamination after repeating the cleaning and sterilization steps. **Conclusion:** The type of dental instrument identifier does not interfere with microbial growth as long as cleaning and sterilization standards are respected.

**Keywords:** Dental instruments. Sterilization. Microbiology.

**RESUMO: Objetivo:** Avaliar a influência dos diferentes identificadores de instrumental no crescimento microbiano de dispositivos críticos após etapas de limpeza e esterilização. **Método:** Foram utilizados 15 cabos nº 3 para lâminas de bisturi como corpo de prova, divididos em 3 grupos (n=5), sendo cada grupo composto de um instrumental sem identificador e outros quatro instrumentais com os seguintes identificadores: gravação a laser, anel de silicone, identificador vinílico com adesivo permanente e etiqueta adesiva personalizada. Após serem submersos no aspirado cirúrgico, os instrumentais foram processados de acordo com os requisitos de boas práticas da Resolução n. 15/2012 da Agência Nacional de Vigilância Sanitária (Anvisa). O crescimento microbiano foi avaliado em diferentes momentos de forma qualitativa, pela presença ou ausência de crescimento, e de forma quantitativa para os casos positivos, por meio de contagem de unidades formadoras de colônia viáveis (log UFC/cabo de bisturi). **Resultados:** Em algum momento, houve crescimento microbiano nos instrumentais independentemente do tipo de identificador. No entanto, não houve contaminação contínua e progressiva após repetição das etapas de limpeza e esterilização. **Conclusão:** O tipo de identificador de instrumental odontológico não interfere no crescimento microbiano desde que sejam respeitadas as normas de limpeza e esterilização.

**Palavras-chave:** Instrumentos odontológicos. Esterilização. Microbiologia.

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**RESUMEN: Objetivo:** Evaluar la influencia de los diferentes identificadores de instrumental en el crecimiento microbiano de dispositivos críticos después de las etapas de limpieza y esterilización. **Método:** Se utilizaron 15 mangos #3 para cuchillas de bisturí como cuerpo de prueba, divididos en 3 grupos (n=5), cada grupo compuesto por un instrumental sin identificador y otros cuatro instrumentales con los siguientes identificadores: grabado a láser, anillo de silicona, identificador vinílico con adhesivo permanente y etiqueta adhesiva personalizada. Después de ser sumergidos en el aspirado quirúrgico, los instrumentales fueron procesados de acuerdo con los requisitos de buenas prácticas de la Resolución nº 15/2012 de la Agencia Nacional de Vigilancia Sanitaria (Anvisa). El crecimiento microbiano se evaluó en diferentes momentos de forma cualitativa, por la presencia o ausencia de crecimiento, y de forma cuantitativa para los casos positivos, mediante el recuento de unidades formadoras de colonias viables (log UFC/mango de bisturí). **Resultados:** En algún momento, hubo crecimiento microbiano en los instrumentales independientemente del tipo de identificador. Sin embargo, no hubo contaminación continua y progresiva después de la repetición de las etapas de limpieza y esterilización. **Conclusión:** El tipo de identificador de instrumental odontológico no interfiere en el crecimiento microbiano siempre que se respeten las normas de limpieza y esterilización.

**Palabras clave:** Instrumentos dentales. Esterilización. Microbiología.

## INTRODUCTION

The close contact of the dentist with the patient's mouth, the high number of consultations, the use of instruments that produce aerosol and the variety of oral microbiota are situations that increase the risk of cross-infection in dentistry[1], and that was proven during the of COVID-19 pandemic[2]. In addition to the existence of these direct routes of infection spread, the existence of indirect routes must also be pointed out, such as what occurs after the use of contaminated materials and instruments[3]

Scalpel handles are classified as critical medical devices, that is, products used in invasive procedures with penetration of skin and adjacent mucous membranes, subepithelial tissues and vascular system, capable of causing a high risk of cross-infection, and must be subjected to sterilization, after cleaning and other processing steps[4]. For critical devices, the accepted probability of survival is one microorganism for every $10^6$ units processed[5]. Considering that the absolute sterility of a product (100% death) does not exist, it is extremely important that all stages of its processing are respected, as most of these instruments have narrow lumens, corrugated surfaces and multiple joints that facilitate the retention of organic matter[6].

As concern about the processing of health products increases, new challenges are emerging, which include the increased use of dental instrument identifiers to facilitate the identification, separation and organization of materials in sterilization centers of public and private services that work with a large number of daily appointments. Radio frequency markings and the use of Data Matrix codes are modern technologies that still have a very high cost[7]. Therefore, many professionals opt for simpler and cheaper methods, such as laser engraving, silicone rings, vinyl labels with permanent adhesive and personalized adhesive labels.

## OBJECTIVE

To evaluate the influence of different surgical instrument identifiers on the microbial contamination of scalpel handles after being subjected to cleaning and sterilization steps.

## METHOD

A longitudinal study was conducted that evaluated microbial contamination on No. 3 handles number for scalpel blades (Quinelato®, Brazil), identified with: A (silicone ring), B (personalized adhesive label), C (vinyl identifier with permanent adhesive), D (laser engraving) and E (without identification), the last group being considered the negative control. To this end, the instruments were previously identified and divided into three groups that represented biological replicas (Figure 1).

The choice of scalpel handles was due to the grooves they have on their surface, capable of facilitating the retention of organic matter, and their dimensions that allowed their inclusion inside the test tubes. Initially, all scalpel handles were subjected to the processing recommended by Resolution No. 15/2012 of the National Health Surveillance Agency (Anvisa)[8] and subjected to microbiological analysis

to prove the absence of microbial contamination before carrying out the challenges. Next, several challenges were carried out on all 15 instruments after being submerged for 30 min in surgical aspirate (blood, saliva and sterile saline), collected with the aid of a Nevoni aspirator vacuum pump, model 5005 (NSR®, Brazil), connected to the suction system of the dental equipment of the Maxillofacial Surgery Clinic of the Faculty of Dentistry of the Federal University of Juiz de Fora. The flowchart in Figure 2 illustrates the challenges and microbiological analyses carried out after the 1st, 6th, 11th, 16th, 21st and 26th surgery.

The processing of scalpel handles involved pre-cleaning the instrument by immersing it in a solution containing water and neutral detergent Deter Rio (Rioquímica®, Brazil) for 5 min, followed by removing visible dirt by scrubbing. Subsequently, the instruments were rinsed with running water and transported to the ultrasonic washer (Cristófoli®, Brazil) containing Riozyme enzymatic detergent (Rioquímica®, Brazil), leaving in the tank for 4 min and 30 s, according to the manufacturer's recommendations. After rinsing again in running water, the instruments were dried with a disposable compress before being individually packaged in surgical grade (Clean up biotechnology®, Brazil), sealed and sent to the autoclave (Sercon®, Brazil) at the Material and Sterilization Center (CME) of the Faculty of Dentistry.

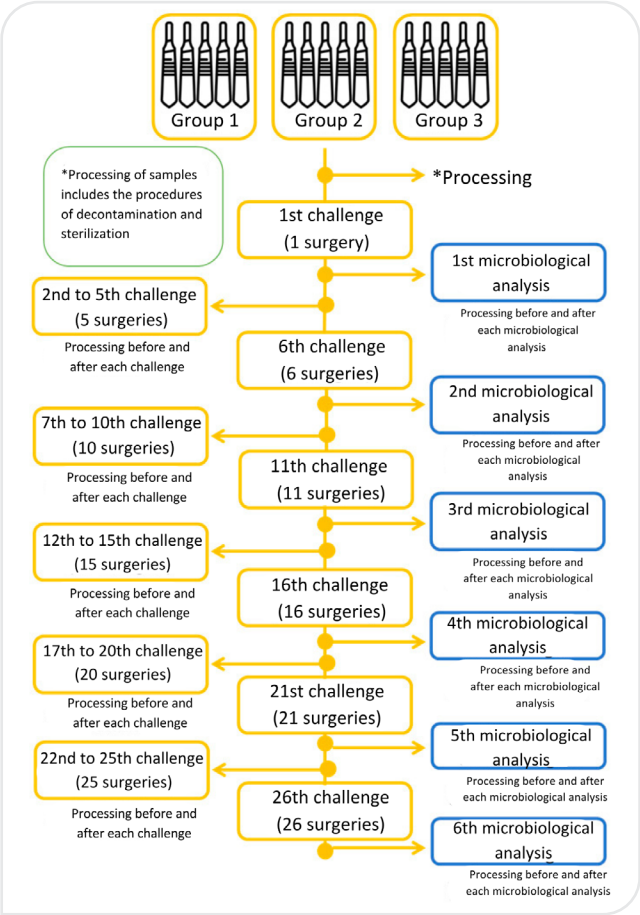


**Figure 2.** Study flowchart.

**Figure 1.** Instruments properly identified and grouped.

Sterilization monitoring was carried out with the aid of class 5 Clean test chemical integrators (Clean up biotechnology®, Brazil) in each packaging and physical indicators obtained from the equipment displays. Clean test biological indicators (Clean up biotechnology®, Brazil) were also used daily, in a package located at the point of greatest challenge, defined during thermal studies in the performance qualification of the sterilization equipment. After sterilization, the instruments were stored in a closed, clean and dry cabinet until they were sent to the Center for Microbiology Studies at the same university. After completing the microbiological test, the scalpel handles were handed over to the clinical collaborator to be reprocessed as previously described, without her having any information about the laboratory stage.

For microbiological analyses, the laboratory collaborator received the scalpel handles, inspected the integrity of the surgical grade, opened the package and, with the aid of sterile hemostatic forceps, submerged the instruments individually in test tubes containing 20 mL of brain heart infusion (BHI) broth, next to a group of test tubes containing only the BHI broth used to control the sterility of the culture medium. All tubes were incubated in an aerobic atmosphere at ±36°C and microbial growth was assessed by the presence of turbidity/biofilm formation on the instruments, over periods of 24 h, 48 h, 7 days and 14 days. These time intervals aimed to observe the growth of fast-growing species, seen in a period of 24 to 48 h, and those that are fastidious, with growth occurring in up to 14 days.

For the test tubes that showed microbial growth, homogenization of the BHI broth and serial dilution (up to $10^{-7}$) in 0.85% saline was performed. Then, a 100-µL aliquot of each dilution was plated on sterile BHI agar, providing three technical replicates for each dilution. The plates were incubated in a microaerophilic atmosphere at ±36°C for 24 h and, subsequently, the colony forming units (CFU) were counted, considering the reading standard between 30 and 300 CFU. To measure contamination in log CFU, the following equation was used: log CFU/scalpel handle = log (20 x average number of CFU x 10 x $10^{\text{inverse of dilution}}$).

The data were tabulated using Microsoft Excel 2019 (Microsoft Corporation, USA) and subjected to descriptive and inferential statistical analysis using SPSS 21.0. Fisher's exact test was used to evaluate the association between microbial growth and the type of identifier at different times. To evaluate the association of microbial growth and the type of identifier over time, the Cochran Q test was used. Regarding the evaluation of microbial growth using CFU/scalpel log, the Shapiro-Wilk test was performed to verify the normality of the data, followed by the Mann-Whitney test, for comparison between groups (A and E) for 24 h of incubation, and the Kruskal-Wallis test, for comparison between all groups for 48 h of incubation. A significance level of 5% was considered ($p<0.05$).

## RESULTS

The first microbiological analysis, that is, the one carried out before the instruments were exposed to surgical aspirate, did not detect bacterial contamination in any of the scalpel handles. However, it was possible to identify some bacterial contamination after the first challenge, present with all types of identifiers, including scalpel handles that did not have identifiers. During the challenges, there was no continuity of microbial contamination in any of the five groups (Table 1).

Fisher's exact test showed that there was no association between microbial growth and the type of identifier at different incubation times, regardless of the moment of microbial challenge. When microbial growth was evaluated for each type of identifier over different incubation times (24 h, 48 h, 7 days and 14 days), the Cochran Q Test showed that there was no difference in the distributions of microbial growth over the period. time (Table 2).

Regarding the CFU count on scalpel handles at different times, an average of 8.22 log CFU/scalpel handle was obtained at 24 h and 7.6 log CFU at 48 h in the first challenge, without there being a significant difference within each incubation time, leading to the interruption of this analysis (Figure 3A). Although there was no significant difference, considering microbial growth at 48 h, there was a trend towards greater microbial density in group E (Figure 3B).

## DISCUSSION

Sterilization with saturated steam under pressure, using an autoclaves, is considered the gold standard in the processing of surgical instruments used in dentistry, as they have excellent penetrability and a relatively short cycle time that allows repeat sterilization and reuse of the instruments in the same day[9]. However, it should be noted

**Table 1.** Quantitative results of microbial growth, negative (-) or positive (+), on identifiers (A, B, C, D and E) when evaluated at different incubation times (24 h, 48 h, 7 days and 14 days), in each of six challenges (after the 1st, 6th, 11th, 16th, 21st and 26th use).

| Microbial challenge | Time | Microbial growth | Identifier % microbial growth (n) | | | | | p value |
|---|---|---|---|---|---|---|---|---|
| | | | A | B | C | D | E | |
| 1 | 24 h | - | 66.7 (2) | 100 (3) | 100 (3) | 100 (3) | 33.3 (1) | 0.407 |
| | | + | 33.3 (1) | 0 (0) | 0 (0) | 0 (0) | 66.7 (2) | |
| | 48 h | - | 33.3 (1) | 66.7 (2) | 66.7 (2) | 66.7 (2) | 33.3 (1) | 1.000 |
| | | + | 66.7 (2) | 33.3 (1) | 33.3 (1) | 33.3 (1) | 66.7 (2) | |
| | 7 days | - | 33.3 (1) | 33.3 (1) | 66.7 (2) | 33.3 (1) | 33.3 (1) | 1.000 |
| | | + | 66.7 (2) | 66.7 (2) | 33.3 (1) | 66.7 (2) | 66.7 (2) | |
| | 14 days | - | 33.3 (1) | 33.3 (1) | 66.7 (2) | 33.3 (1) | 33.3 (1) | 1.000 |
| | | + | 66.7 (2) | 66.7 (2) | 33.3 (1) | 66.7 (2) | 66.7 (2) | |
| | 24 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 48 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| 2 | 7 days | - | 66.7 (2) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 1.000 |
| | | + | 33.3 (1) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 14 days | - | 66.7 (2) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 1.000 |
| | | + | 33.3 (1) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 24 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 48 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| 3 | 7 days | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 14 days | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 24 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 48 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| 4 | 7 days | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 14 days | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 100 (3) | - |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | |
| | 24 h | - | 100 (3) | 100 (3) | 100 (3) | 100 (3) | 66.7 (2) | 1.000 |
| | | + | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 33.3 (1) | |
| | 48 h | - | 100 (3) | 100 (3) | 66.7 (2) | 100 (3) | 66.7 (2) | 1.000 |
| | | + | 0 (0) | 0 (0) | 33.3 (1) | 0 (0) | 33.3 (1) | |
| 5 | 7 days | - | 100 (3) | 100 (3) | 66.7 (2) | 100 (3) | 66.7 (2) | 1.000 |
| | | + | 0 (0) | 0 (0) | 33.3 (1) | 0 (0) | 33.3 (1) | |
| | 14 days | - | 100 (3) | 100 (3) | 33.3 (1) | 100 (3) | 66.7 (2) | 0.407 |
| | | + | 0 (0) | 0 (0) | 66.7 (2) | 0 (0) | 33.3 (1) | |
| | 24 h | - | 100 (3) | 66.7 (2) | 100 (3) | 100 (3) | 100 (3) | 1.000 |
| | | + | 0 (0) | 33.3 (1) | 0 (0) | 0 (0) | 0 (0) | |
| | 48 h | - | 100 (3) | 66.7 (2) | 100 (3) | 66.7 (2) | 100 (3) | 1.000 |
| | | + | 0 (0) | 33.3 (1) | 0 (0) | 33.3 (1) | 0 (0) | |
| 6 | 7 days | - | 100 (3) | 66.7 (2) | 100 (3) | 66.7 (2) | 100 (3) | 1.000 |
| | | + | 0 (0) | 33.3 (1) | 0 (0) | 33.3 (1) | 0 (0) | |
| | 14 days | - | 100 (3) | 66.7 (2) | 100 (3) | 66.7 (2) | 100 (3) | 1.000 |
| | | + | 0 (0) | 33.3 (1) | 0 (0) | 33.3 (1) | 0 (0) | |

that, despite this effectiveness, there are also studies that prove microbial contamination in some of these products after being subjected to sterilization, with emphasis on instruments that have many recesses, such as surgical drills, or that have very small lumens, such as high- and low-speed motors[10-13].

Furthermore, it should be noted that most of these instruments are marked with identifiers that can have advantages and disadvantages. Bortolato et al.[14] evaluated the functionality of using marking tapes in the process of assembling surgical boxes, comparing samples of surgical boxes that used the tapes as an identification method with samples that did not have an identifier, obtaining a reduction in box preparation time of the first group.

The study carried out by Samit and Dodson[15] in 1983, was the first to raise the hypothesis of the influence of

**Table 2.** Qualitative results of microbial growth, negative (-) or positive (+), on identifiers (A, B, C, D and E) when evaluated over time (24 h, 48 h, 7 days and 14 days), during the six challenges. Cochran's Q test was performed to evaluate the association of microbial growth and the type of identifier over time.

| Identifier | Microbial growth | Identifier % microbial growth (n) | | | | p value |
|---|---|---|---|---|---|---|
| | | 24 h | 48 h | 7 days | 14 days | |
| A | - | 94.4 (17) | 88.9 (16) | 83.3 (15) | 83.3 (15) | 0.194 |
| | + | 5.6 (1) | 11.1 (2) | 16.7 (3) | 16.7 (3) | |
| B | - | 94.4 (17) | 88.9 (16) | 83.3 (15) | 83.3 (15) | 0.194 |
| | + | 5.6 (1) | 11.1 (2) | 16.7 (3) | 16.7 (3) | |
| C | - | 100 (16) | 87.5 (14) | 87.5 (14) | 81.3 (13) | 0.096 |
| | + | 0 | 12.5 (2) | 12.5 (2) | 18.8 (3) | |
| D | - | 100 (18) | 88.9 (16) | 83.3 (15) | 83.3 (15) | 0.066 |
| | + | 0 | 11.1 (2) | 16.7 (3) | 16.7 (3) | |
| E | - | 83.3 (15) | 83.3 (15) | 83.3 (15) | 83.3 (15) | 1.000 |
| | + | 16.7 (3) | 16.7 (3) | 16.7 (3) | 16.7 (3) | |

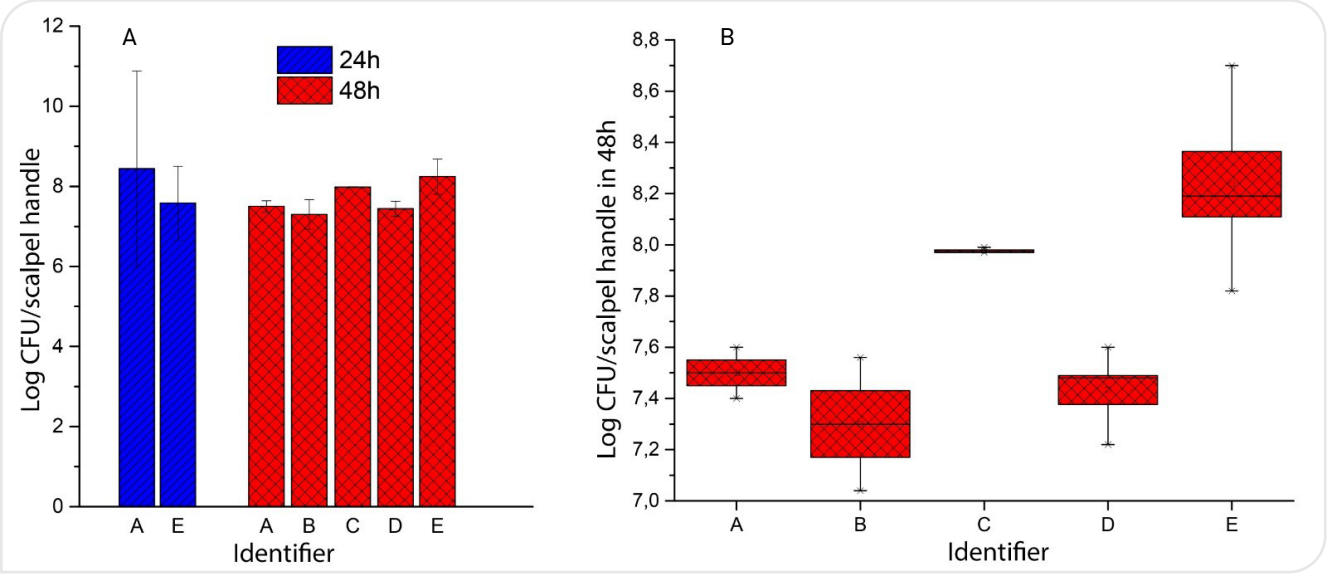


**Figure 3.** (A) Microbial growth in log CFU/scalpel handle for the different identifiers at 24 and 48 h, and (B) distribution of microbial growth at 48 h for the different groups considering the first microbial challenge. Statistical analysis using the Mann-Whitney test, for comparison between the 24-h groups (p=0.667), and the Kruskal-Wallis test, for comparison between the 48-h groups (p=0.053).

identifiers on the microbial retention of instruments. The authors identified the presence of *Staphylococcus epidermidis* in patients undergoing oral surgery within 13 days postoperatively. Later, they isolated the same bacteria on the instrument identification tapes. Finally, they removed the tapes from the instruments and brought an end to the infectious outbreak.

Another complication that may be related to the use of instrument identifiers is the possibility of material detachment during surgery, as reported by Kraayenbrink et al.[16]. It should be noted that any item forgotten in the surgical wound can cause harm to the patient and the professional, as this act is classified as a preventable error[17]. Therefore, to avoid such accidents, it is suggested that manufacturers establish expiration dates for identifiers, as well as removing identifiers that are defective or detached from the instruments.

In 2019, Bruna et al.[18] evaluated microbial contamination on tapes and resins used to identify 140 surgical forceps. After analyzing fragments of instrument identifiers, the authors found bacterial growth on three tape samples. With the aim of reproducing clinical reality as much as possible, the present study proposed the use of the instrument itself as a body of evidence. Although the study found microbial contamination in several instruments, it was not possible to establish a significant correlation with any type of identifier. These findings reinforce the importance of meticulousness in all stages of instrument processing[13].

For cleaning health products that have complex conformations, the use of an ultrasonic washer is advocated, automated equipment that uses the principle of cavitation, in which waves of acoustic energy are propagated in an aqueous solution, breaking the bonds that attach dirt to the surface of the instrument. The study by Pereira et al.[19] compared the effectiveness of cleaning endodontic files using three cleaning methods: gauze with 70% alcohol, detergent and ultrasonic washer. As a result, they achieved a reduction in debris counts in all groups, with the best results found when using the ultrasonic washer.

A limitation of the present study was the difficulty of organizing the logistics of transporting the instruments between the CME of the Faculty of Dentistry and the Center for Microbiology Studies. Considering the work routine and availability of employees, a period of two days was adopted to begin microbiological analyses after sterilization of the instruments. Although this time is much shorter than that recommended by most regulatory bodies, which certify seven days as the limit for using a sterilized product, it is known that most recommended is that the instruments are used as close as possible to the sterilization process, to avoid damage to the packaging, nowadays called the "sterile barrier system". The Brazilian Society of Surgical Center, Anesthetic Recovery and Material and Sterilization Center Nurses states that the loss of sterility of a packaged item is associated with the related event and not with the shelf life, as the theory of abiogenesis must be adopted. It also recommends that health services, through the health products processing committee, should establish the maximum shelf life for sterilized products, based on a plan to assess the integrity of sterile barrier systems[20].

## CONCLUSION

The use of different dental instrument identifiers is not associated with increased microbial contamination, as long as cleaning and sterilization standards for these materials are respected.

## FUNDING

None.

## CONFLICT OF INTERESTS

The authors declare that there is no conflict of interests.

## AUTHORS' CONTRIBUTIONS

LBS: Data curation, Investigation, Writing – original draft, Writing – review & editing. RPB: Conceptualization, Data curation. MSO: Data curation, Investigation, Writing – original draft, Software. MSAP: Writing – review & editing, Software. ACMA: Formal analysis, Methodology, Software, Supervision, Validation, Visualization. MFC: Project administration, Formal analysis, Methodology, Supervision, Validation, Visualization.

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