

An *in vitro* Study of the Contamination Remaining on the Surface of Used Healing Abutment and Impression Copings After Cleaning and Sterilizing

**Dr. Shally Rana¹, Dr. Dron Lakhani², Dr. Dushyant Soni³,
Dr. Rajshree Bhandari⁴, Dr. Amrit Assi⁵, Dr. Kshitija Hatkar⁶,
Dr. Rushita Dodia⁷**

¹Post Graduate Student, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

²Professor, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

³HOD & Professor, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

⁴Reader, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

⁵Senior lecturer, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

⁶Post Graduate Student, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

⁷Post Graduate Student, Department of Prosthodontics and Crown and Bridge and Implantology,
Vyas Dental College and Hospital, Jodhpur, Rajasthan

Corresponding Author: Dr. Shally Rana.

DOI: <https://doi.org/10.52403/ijhsr.20251002>

ABSTRACT

In daily dental practice, cleaning and sterilization of reusable components like healing abutments (HAs) and implant impression copings (IICs) are critical for preventing cross-infection and ensuring successful clinical outcomes. Although often designated as single-use, these titanium components are frequently reused after sterilization for economic reasons. Contamination from blood, saliva, epithelial tissue, and bacterial biofilms can persist even after standard sterilization, posing a risk for infection and implant failure. Proper decontamination is challenging due to strong protein adhesion on titanium surfaces. Phloxine B, a protein-staining dye, can detect residual contamination, with stereomicroscopy offering three-dimensional visualization of stained surfaces. This study aims to assess the effectiveness of different sterilization protocols in removing organic residues from reused HAs and IICs, highlighting the importance of meticulous cleaning to maintain hygiene and implant success. In this, thirty used healing abutments (HAs) and thirty used implant impression copings (IICs) were collected and evaluated in their "as received" condition. Samples were stained with Phloxine B and divided into three cleaning protocols: (1) Distilled water + Cidex OPA; (2) Distilled water + 5.25% NaOCl with 15-min ultrasonic cleaning; (3) Distilled water + NaOCl + ultrasonic cleaning followed by autoclaving. After treatment, each sample was examined under a stereomicroscope at 2x magnification. HAs were imaged at five key sites (B, C, S, D, O), and IICs at three (E, S) to assess residual contamination.

Microscopic analysis showed that Distilled water + Cidex OPA was ineffective in reducing contamination on healing abutments and impression copings. Significant reduction was observed with Distilled water + NaOCl, especially on external surfaces. The combination of NaOCl with autoclaving showed the most effective decontamination across all regions of both components.

This study revealed that standard cleaning and sterilization methods fail to completely eliminate contaminants from healing abutments and impression copings, with 99% showing residual proteins.

KEYWORDS: Healing abutment, Impression copings, Sterilization, Phloxine B

INTRODUCTION

In routine dental practice, the cleaning and sterilization of various instruments and components are essential to prevent infections and ensure optimal clinical outcomes. While many devices are supplied for single-use in sterilized packaging, several metal components require cleaning before sterilization and are often reused.

Although healing abutments and implant impression copings are typically designed for single-use, many clinicians clean and sterilize them for reuse due to economic reasons. Studies show that titanium HAs can be effectively sterilized, and certain sterilization methods can even enhance soft tissue cell adhesion to the titanium surface. However, recent research¹ suggests that these components may not be as sterile as previously assumed, raising concerns about the safety of reusing them.

During the procedures, clinicians often observe contamination on the surfaces of healing abutments and implant impression copings.³ This contamination can originate from bacterial plaques, epithelial attachments that tear during abutment removal, blood, food debris, and saliva. Failure to properly clean, disinfect, or sterilize instruments contaminated with pathogens can lead to cross-contamination and infections. Additionally, proteins on the surface of implants can resist standard sterilization methods, posing an infection risk. Extra caution is necessary when dealing with blood, saliva, and contaminated instruments to prevent cross-infection. Contamination of implants during placement is inevitable due to contact with

saliva, a fluid containing over 700 species of bacteria. Early implant failure, which accounts for 1% to 2% of all implant failures, is strongly associated with bacterial contamination, which can negatively affect osseointegration, especially in augmented areas.

Proper decontamination and sterilization are vital for preventing infections and ensuring successful clinical outcomes.⁴ While it is clear that healing abutments and impression copings should be replaced, organic material may still remain on their surfaces even after sterilization. Cleaning these surfaces thoroughly remains challenging due to the strong attachment of proteins and other molecules to titanium surfaces. To detect protein residue, Phloxine B dye is used, and the stained surfaces are examined under a stereomicroscope².

This study aims to evaluate the residual contamination on used healing abutments and implant impression copings after cleaning and sterilizing them with different sterilization protocols.

MATERIAL AND METHOD

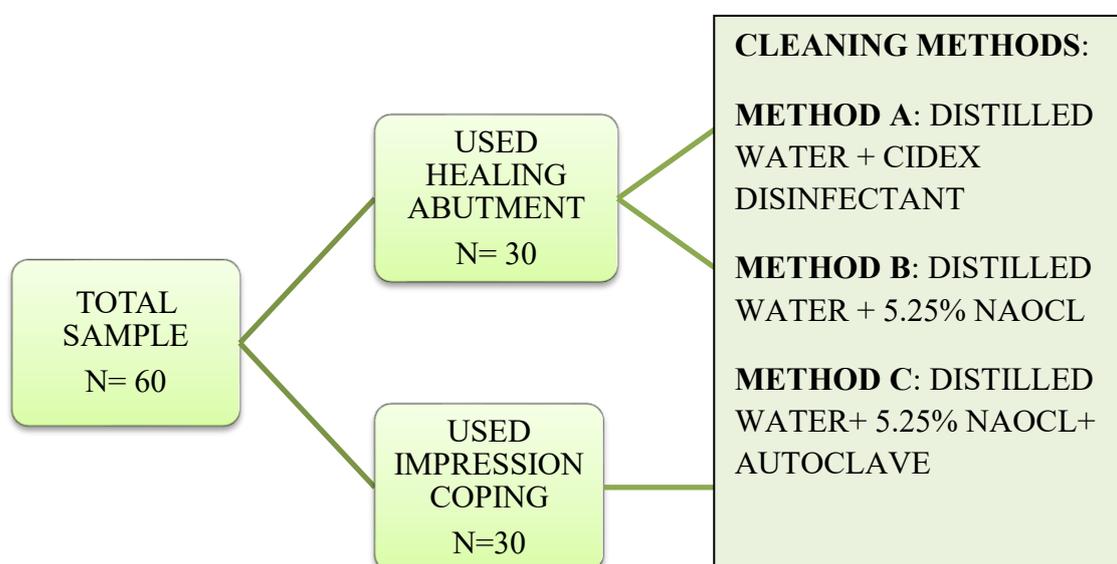
A total of 30 used healing abutments (HAs) and 30 used implant impression copings (IICs) were collected from the Department of Prosthodontics, Vyas dental college and hospital. Re-use of HAs and IICs was confirmed occasionally. Collected sample were used irrespective of the brand and size. After collection, the HAs and IICs designated as the test group were the “as received” group. These were initially inspected with unaided vision to determine if any debris or other damage or

contamination could be detected. For staining, each HA and IIC was placed in an individual plastic bag with 2 ml of Phloxine B stain and sealed. Now, 10 samples of each HA and IIC were cleaned with three different methods.

1. Distilled water + cidex OPA Disinfectant
 - The contaminated HAs and IIC were firstly cleaned with distilled water followed by swiping the surface with cidex opa and were then air dried.
2. Distilled water + NAOCL
 - The contaminated HAs and IIC were firstly cleaned with distilled water

followed by placing them in an individual plastic bag containing 1 ml 5.25% sodium hypochlorite solution and ultrasonicated for 15 min, and then air dry.

3. Distilled water + NAOCL + AU
 - The contaminated HAs and IIC were firstly cleaned with distilled water, followed by placing them in an individual plastic bag containing 1 ml 5.25% sodium hypochlorite solution and ultrasonicated for 15 min and subsequent steam autoclaving for 30 min at 121° with a 15 min minimum air dry.



Evaluation of the number and site of contamination of the healing abutments -

The HAs and IIC were photographed using a stereomicroscope at a magnification of 2x. For each healing abutment three images were captured: two of the body of healing abutments rotated at 180 degrees, one from the occlusal and one from the top (screwdriver engagement site). And have examined these different sites:

- The main body (B)
- Connection (C) to implant site
- Screw (S) thread shank
- Screw driver (D) engagement site
- Occlusal (O)

Evaluation of the number and site of contamination of the impression copings-

For each Impression coping two images were captured: two of the external surfaces rotated at 180 degrees and one screw thread. And have examined these different sites:

- External surface (E)
- Screw thread (S)

A control group consisting of three unused healing abutments and three unused impression copings were separately cleaned with the three cleaning methods mentioned above, followed by staining to evaluate the cleaning process and its effect based on Phloxine B staining.

All photographs were examined by one examiner. Recordings of the number of protein-stained sites was recorded for each abutment and coping aspect for both “as received” group and the control group. The presence of residual contamination was evaluated.

RESULT

Representative photographs of the specimens are seen in Figures (1-8). Although it was also possible to see contamination with the unaided vision on some of the abutments and copings, many appeared to be free of residual contamination. The visualized results revealed a reddish-orange color in the areas of residual protein and amino acid contamination under the stereomicroscope.

Microscopic analysis of the contamination before and after decontamination procedure in Healing Abutments:

- No significant difference was observed in the degree of contamination before

and after the decontamination procedure in the first group i.e, in Distilled water and Cidex opa disinfectant group. (figure 1)

- The percentage of contaminated surface was significantly decreased in the connection, body, and occlusal regions of healing abutments in the Distilled water and 5.25% NaOCL group. No significant difference in the screw driver engagement region in this group. (figure 2)
- The percentage of contaminated surface was significantly reduced in all regions: body, screw, connection, and screw driver engagement and occlusal of healing abutment in the Distilled water + 5.25% NaOCL + autoclave. (figure 3)

The residual contamination after the decontamination procedure is listed in Table 1. The various degrees of contamination before and after decontamination procedures within the groups are shown in figure1-4. The unused healing abutment showed no stained at any site.

TABLE 1: NUMBER AND SITE OF HEALING ABUTMENT CONTAMINATION OF “AS RECEIVED” GROUP

Cleaning method	Contaminated healing abutments before treatment (n)	Contaminated healing abutments after treatment (n)	Healing abutment regions	Contaminated abutment surfaces Before n/N (%)	Contaminated abutment surfaces After n/N (%)	P value
Distilled water+ Cidex	10	10	Body (B)	18/20 (90%)	18/20 (90%)	1.000
			Connection (C)	16/20 (80%)	16/20 (80%)	1.000
			Screw thread (S)	20/20 (100%)	20/20 (100%)	1.000
			Occlusal (O)	10/10 (100%)	10/10 (100%)	1.000
			Screwdriver engagement (D)	10/10 (100%)	10/10 (100%)	1.000
Distilled water+ NaOCL	10	10	Body (B)	14/20 (70%)	6/20 (30%)	<0.001
			Connection (C)	16/20 (80%)	12/20 (60%)	0.013
			Screw thread (S)	18/20 (90%)	18/20 (90%)	1.000
			Occlusal (O)	8/10 (80%)	4/10 (40%)	<0.001
			Screwdriver engagement (D)	10/10 (100%)	8/10 (80%)	0.032
Distilled water+ NaOCL+	10	5	Body (B)	12/20 (60%)	2/20 (10%)	<0.001
			Connection (C)	12/20 (60%)	8/20 (40%)	<0.001

autoclave			Screw thread (S)	18/20 (90%)	14/20 (70%)	0.012
			Occlusal (O)	8/10 (80%)	2/10 (20%)	<0.001
			Screwdriver engagement (D)	10/10 (100%)	5/10 (50%)	0.015

REPRESENTATIVE MICROSCOPE IMAGES OF HEALING ABUTMENT BEFORE CONTAMINATION, STAINED WITH PHLOXINE B (a1, a2, a3), AND AFTER DECONTAMINATION PROTOCOLS (b1, b2, b3):

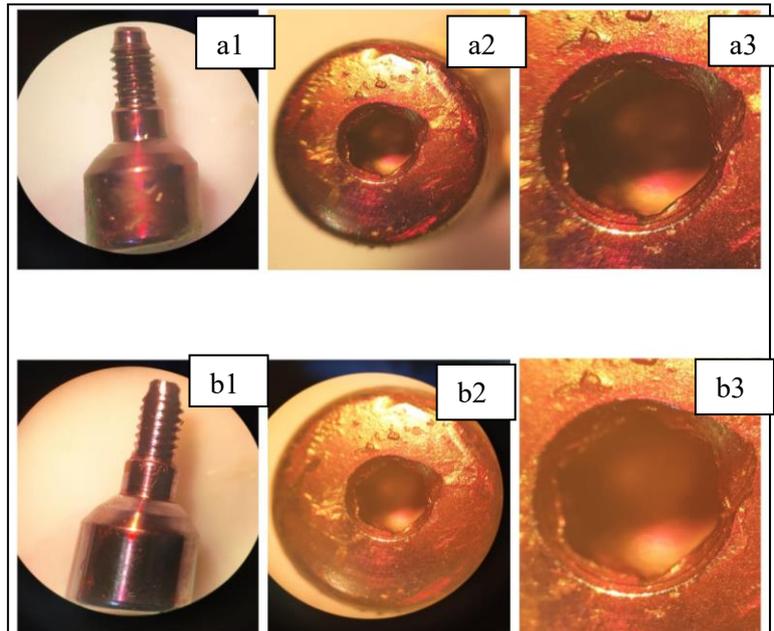


Figure 1: Distilled water + cidex showing stained debris in all regions of healing abutment

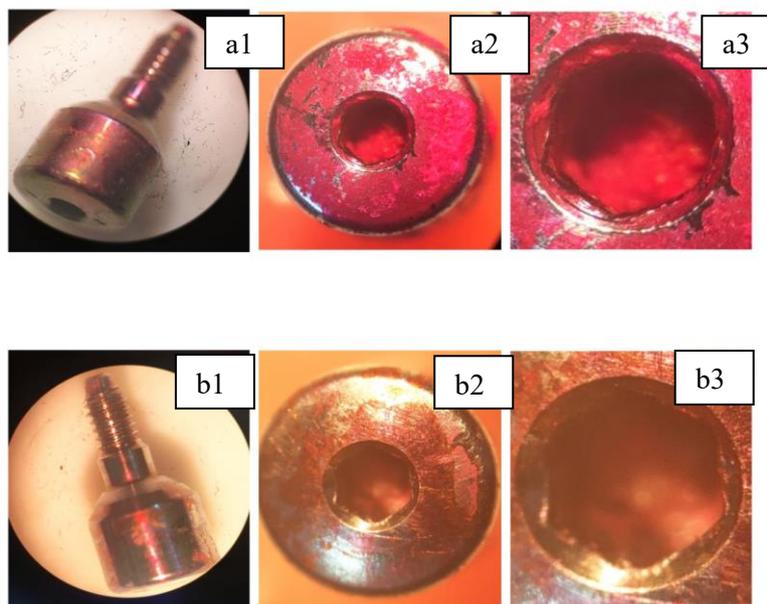


Figure 2: Distilled water + 5.25% NaOCL demonstrating unstained debris on the occlusal region of a healing abutment

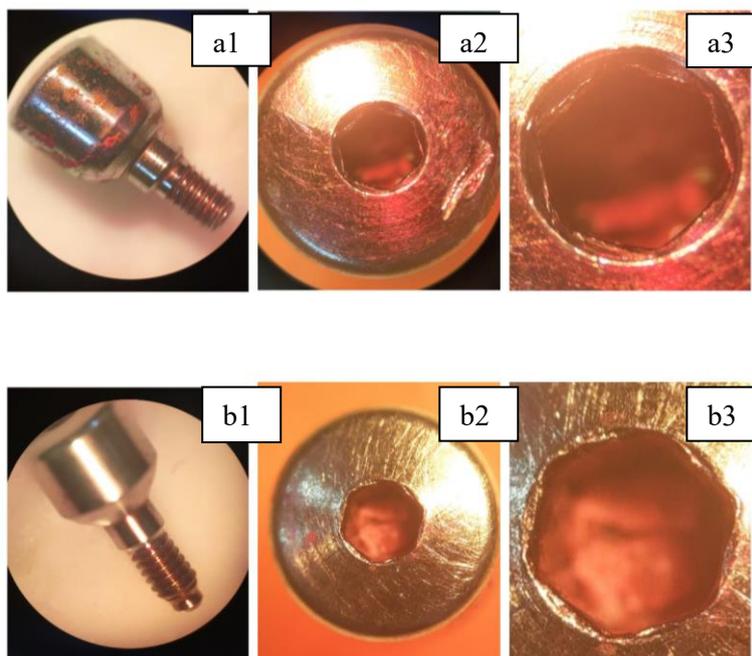


Figure 3: Distilled water + 5.25% NaOCL + Autoclave illustrating no visible stained areas on the body and occlusal region on the healing abutment



Figure 4: Microscopic images of healing abutment in control group stained with Phloxine B illustrating no visible stained area on the surface

Microscopic analysis of the contamination before and after decontamination procedure in Implant Impression Copings:

- No significant difference was observed in the degree of contamination before and after the decontamination procedure in the first group i.e., in Distilled water and Cidex opa disinfectant group. (Figure5)
- The percentage of contaminated surface was significantly decreased in the surface of the impression coping in the Distilled water and 5.25% NaOCL

- group. No significant difference in the screw region in this group. (Figure 6)
 - The percentage of contaminated surface was significantly reduced in all regions i.e., the external surface and screw thread region in the Distilled water + 5.25% NaOCL + autoclave. (Figure 7)
- The residual contamination after the decontamination procedure is listed in Table 2. The various degrees of contamination before and after decontamination procedures within the groups are shown in figures (5-7). The unused Impression coping showed no stained at any site. (figure8).

TABLE 2: NUMBER AND SITE OF IMPRESSION COPINGS CONTAMINATION “AS RECEIVED” GROUP

Cleaning method	Contaminated impression copings before treatment (n)	Contaminated impression copings after treatment (n)	Impression copings regions	Contaminated coping surfaces Before n/N (%)	Contaminated coping surfaces After n/N (%)	P value
Distilled water + Cidex	10	10	External surface (E)	20/20 (100%)	20/20 (100%)	1.000
			Screw thread (S)	20/20 (100%)	20/20 (100%)	1.000
Distilled water + NaOCL	10	10	External surface (E)	18/20 (90%)	16/20 (80%)	0.012
			Screw thread (S)	20/20 (100%)	20/20 (100%)	1.000
Distilled water + NaOCL+ autoclave	10	8	External surface (E)	12/20 (60%)	5/20 (25%)	<0.001
			Screw thread (S)	18/20 (90%)	13/20 (65%)	0.032

REPRESENTATIVE MICROSCOPE IMAGES OF IMPRESSION COPING BEFORE CONTAMINATION, STAINED WITH PHLOXINE B (a1, a2), AND AFTER DECONTAMINATION PROTOCOLS (b1, b2):

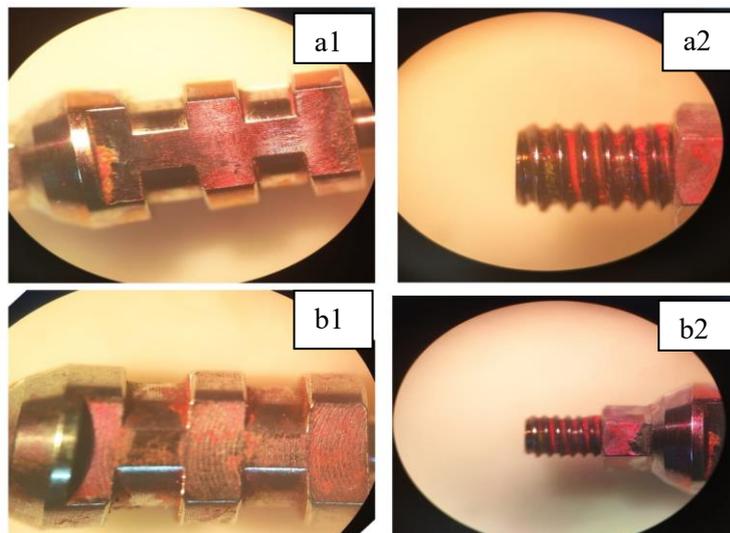


Figure 5: Distilled water + cidex showing stained debris in all regions of impression coping

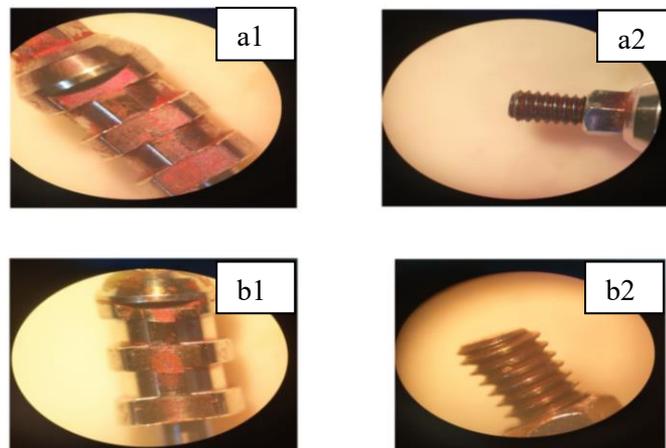


Figure 6: Distilled water + 5.25% NaOCL demonstrating unstained debris on some of the external surface on impression coping

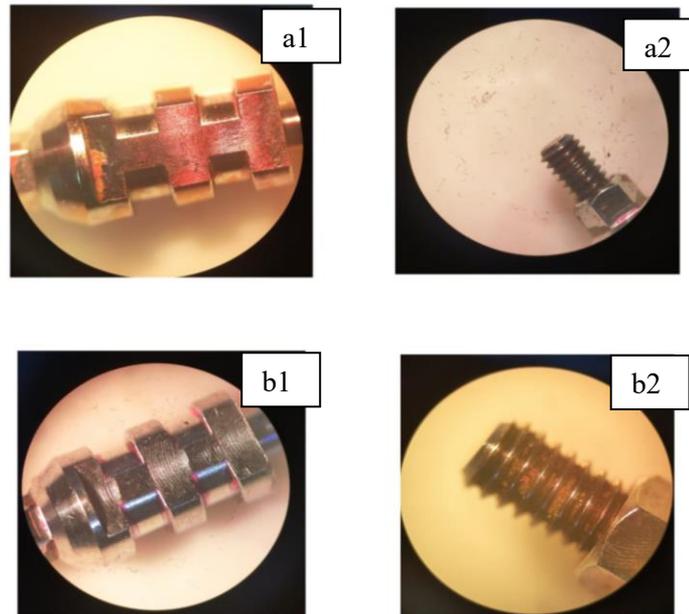


Figure 7: Distilled water + 5.25% NaOCL + Autoclave illustrating no visible stained areas on the surface

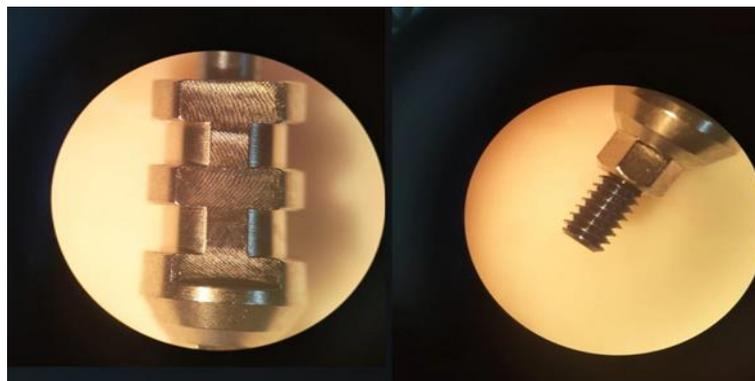


Figure 8: Microscopic images of impression coping in control group stained with Phloxine B illustrating no visible stained area on the surface

DISCUSSION

The study evaluated the effectiveness of different sterilization protocols in cleaning contaminated healing abutments (HAs) and implant impression copings (IICs). Achieving a fully clean surface is crucial, as decontamination methods may alter the surface, affecting soft tissue healing. Healing abutments and impression copings are generally considered single-use, but they are often reused due to economic reasons. Reusing these components is only safe if they can be adequately cleaned and sterilized, which this study suggests may not always be possible. Inadequate cleaning may lead to contamination with proteins and amino acids, which are difficult to remove and can affect healing and implant success.

The study highlights concern about prion proteins, which are resistant to typical sterilization and could potentially transmit infections. Contamination in screw threads and areas like screwdriver holes can affect the mechanical performance of the implant and cause friction that reduces clamping force. This can contribute to implant failure or peri-implant disease.

Various decontamination methods were tested, with combinations of sodium hypochlorite (NaOCl) and enzymatic cleaning (EC) being more effective than using NaOCl or EC alone. However, full cleaning was not always achieved, especially in difficult-to-reach areas. This raises concerns about reusing healing

abutments and impression copings, as they may still harbor contamination.

Kyaw TT et al.⁵ observed the combination of NaOCl with subsequent EC can remove soft and hard deposits from the surface of HAs compared to NaOCl alone and EC alone, without altering the surface topography of HAs.

Chew et al.⁷ investigated the use of 0.25% NaOCl support to achieve the effective decontamination of used HAs, including difficult cleaning regions such as screwdriver holes and screw threads.

Kasugai S.⁶ observed the combination of a strong denaturing agent and detergent effectively cleaned contaminated healing abutments, perfect cleaning was not always possible, indicating that the reuse of healing abutments in different patients is not recommended.

In contrast, Almeahmadi⁸ observed residual debris after decontamination with 5.25% NaOCl; it cannot remove the debris-locked area, such as the screwdriver hole and screw thread region of HAs.

In implant impression copings, contamination can be influenced by design, impression technique, and improper disinfection procedures. Areas like screw threads and internal recesses can trap biological material, increasing the risk of disease transmission. Methods for disinfecting these components need further evaluation, especially to ensure all areas are reached.

In conclusion, the study found that different sterilization methods yielded different results, with Method C being the most effective in removing contamination. The findings suggest that reuse of implant components should be reconsidered unless proper cleaning protocols are developed and followed to ensure patient safety.

CONCLUSION

Although this study has its limitations, the results are unmistakable: cleaning and sterilization protocols in dental clinics do not fully eliminate contaminants from healing abutments and impression copings.

Proteins and peptides were still present on 99% of the tested samples. However, a combination of NaOCl followed by autoclaving proved more effective in removing both soft and hard deposits from these surfaces compared to using NaOCl or Cidex alone.

While the idea of reusing or recycling healing abutments may seem financially beneficial, it is essential to carefully weigh the potential cost savings against the significant risks and detrimental effects that incomplete decontamination could have on patient safety and the success of the implant.

Declaration by Authors

Acknowledgement: None

Source of Funding: None

Conflict of Interest: The authors declare no conflict of interest.

REFERENCES

1. Smith A, Lange A, Perrett D, McHugh S, Bagg J. Residual protein levels on reprocessed dental instruments. *Journal of Hospital Infection*. 2005 Nov 1;61(3):237-41.
2. Wadhvani C, Schonnenbaum TR, Audia F, Chung KH. In-vitro study of the contamination remaining on used healing abutments after cleaning and sterilizing in dental practice. *Clinical implant dentistry and related research*. 2016 Dec;18(6):1069-74.
3. Browne V, Flewelling M, Wierenga M, Wilson A, Aprecio R, Richardson P, Angelov N, Johnson N. Sterilization analysis of contaminated healing abutments and impression copings. *Journal of the California Dental Association*. 2012 May 1;40(5):419-21.
4. Fulford MR, Stankiewicz NR. Cleaning methods for dental instruments. *British Dental Journal*. 2023 Jul 28;235(2):105-11.
5. Kyaw TT, Abdou A, Arunjaroen Suk S, Nakata H, Kanazawa M, Pimkhaokham A. Effect of chemical and electrochemical decontamination protocols on single and multiple-used healing abutments: A comparative analysis of contact surface area, micro-gap, micro-leakage, and surface topography. *Clinical Implant Dentistry and Related Research*. 2023 Dec;25(6):1207-15.

6. Kasugai S. Evaluation of residual contamination on healing abutments after cleaning with a protein-denaturing agent and detergent. *Quintessence International*. 2020 Jun;51(6):474.
 7. Chew M, Tompkins G, Tawse-Smith A, Waddell JN, Ma S, Ma S. Reusing Titanium Healing Abutments: Comparison of Two Decontamination Methods. *International Journal of Prosthodontics*. 2018 Nov 1;31(6).
 8. Almeahadi AH. An in vitro analysis of sodium hypochlorite decontamination for the reuse of implant healing abutments. *Journal of Oral Implantology*. 2021 Aug 1;47(4):271-9.
- How to cite this article: Shally Rana, Dron Lakhani, Dushyant Soni, Rajshree Bhandari, Amrit Assi, Kshitija Hatkar et al. An *in vitro* study of the contamination remaining on the surface of used healing abutment and impression copings after cleaning and sterilizing. *Int J Health Sci Res*. 2025; 15(10):10-19. DOI: [10.52403/ijhsr.20251002](https://doi.org/10.52403/ijhsr.20251002)
