

## ORIGINAL ARTICLE

# Disinfection Of Gypsum Casts: Comparing Microwave Irradiation And Chemical Disinfection-An In Vivo Study

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## ABSTRACT

**Introduction:** Disinfection procedures cannot only be limited to disinfecting the impressions, disinfection also needs to be carried out at intermittent steps during exchange between the laboratory and the dental clinic. The aim of this study was to test whether microwave oven irradiation can disinfect gypsum casts satisfactorily and whether it would be as effective as a validated method of chemical disinfection of impressions.

**Materials and methods:** Three successive impressions of the maxillary arch were made for each volunteer. The Impressions were randomly divided into three groups. For each volunteer one impression was poured in type III gypsum without disinfection to serve as a control. The second impression was chemically disinfected by immersing it in 0.535% sodium hypochlorite for 10 minutes and poured in type III gypsum. The third impression was poured without disinfection and the cast was irradiated in a microwave oven at 2,450 MHz and 800 W for 5 min. All casts were incubated aerobically in glucose broth at 37°C for 18 hours.

**Results:** Untreated gypsum casts showed cfu/ml counts with a median log value of 10<sup>5</sup>. While microwave irradiated casts showed cfu/ml counts with a median log value of 0. Casts poured from chemically disinfected impressions demonstrated cfu/ml counts with a median log value of 10<sup>4</sup>

**Conclusion:** Under the given conditions, it can be concluded that microwave irradiation (800W) not only reduces the microbial load on gypsum casts but is more effective than chemical disinfection of impressions in doing so.

**Keywords:** Microwave irradiation, disinfection of gypsum casts, chemical disinfection, cross contamination

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## INTRODUCTION

The recognition of the potential for transmission of numerous infectious microorganisms during dental procedures has led to an increased concern for infection control in dental practice. The main sources of cross infection between dentists and patients are impression trays, impression materials and poured stone casts.<sup>1-3</sup> The most common dental procedure that may cause cross-infection, especially between patients and dental laboratory personnel, is the making of impressions. Among various impression materials irreversible hydrocolloid impressions have produced a relatively higher level of contamination.<sup>4-7</sup> Previous studies have shown that majority of the impressions arriving at a dental laboratory are contaminated with bacteria and other microorganisms,<sup>8-11</sup> irrespective of whether they had been exposed to a disinfection procedure or merely rinsed with tap water.<sup>11,12</sup>

American Dental Association has advised all dental workers to disinfect patient's impression trays.<sup>13,14</sup>

The conventional solution to the problem of cross contamination through dental impressions has been chemical disinfection.<sup>15</sup> However the disinfection of impression materials hinders possible cross-contamination only at the time the cast is poured. A cast from a properly disinfected impression may subsequently be contaminated by a technician or clinician.<sup>16</sup> Also, the prosthesis will become contaminated by the patient after trial and adjustment in the mouth and will recontaminate the cast on repositioning. Microwave irradiation might provide an effective, quick, easy, inexpensive and versatile tool for inactivation of microbial organisms on gypsum casts, which can be performed by dentists, assistants and technicians alike.<sup>17</sup>

A PubMed, Medline search from 1990 to 2014 for published articles citing the use of microwave irradiation as a means of disinfection of gypsum casts revealed only few relevant studies.<sup>18-22</sup> The present study aims to use a household microwave with relatively lower levels of irradiation to disinfect casts from patient derived irreversible hydrocolloid impressions. It also provides a direct comparison of the microbial load reduction between chemical disinfection of impressions and microwave irradiation of casts.

*Null Hypothesis:* The null hypothesis was that there is no difference in the reduction of the microbial load of a gypsum cast obtained from an irreversible hydrocolloid impression disinfected using 0.535% sodium hypochlorite and directly subjecting the cast to microwave irradiation at 800W for 5 min.

## MATERIAL AND METHODS

Ethical committee clearance from the Goa University Ethics Committee and informed consent from the volunteers was taken before starting the procedure. Three successive impressions of the upper arch were made for each of the ten volunteers with an irreversible hydrocolloid. All procedures were carried out in the Department of Microbiology, Goa Medical College and Hospital in an attempt to avoid contamination that may occur during transport of these impressions. The Impressions were randomly divided into three groups and subjected to three different chair side procedures.

Impression one: Rinsed with 250 ml distilled water for 15 seconds<sup>23</sup>

Impression two: Immersed in 0.535% sodium hypochlorite for 10 minutes as per ADA recommendations for chemical disinfection of irreversible hydrocolloid impressions.<sup>24</sup>

Impression three: Rinsed with 250 ml distilled water for 15 seconds and poured without disinfection. The cast obtained was irradiated in a microwave oven at 2,450 MHz and 800 W for 5 min. To ensure that the casts were adequately irradiated on all surfaces, they were first exposed for 2.5 minutes and subsequently turned upside down and irradiated again for the same amount of time. All casts were poured in type III gypsum and allowed to set for one hour.

Sample selection: Inclusion criteria included all the male subjects within the age group of 25- 30 years with intact maxillary and mandibular dentition and good oral hygiene. Exclusion criteria: Presence of any systemic disease that can alter normal oral flora. Presence of dental caries or periodontal disease. History of smoking or tobacco use.

## Bacteriologic Procedures

All casts were incubated aerobically in glucose broth at 37°C for 18 hours. After serial dilutions of 1:2, 1:4 and 1:8 this broth was plated on blood agar using a wire loop. These inoculated plates were incubated aerobically at 37°C for 24 hours and assessed for bacterial growth by counting the colony forming units per millilitre (cfu/ml) of the culture.

*Determining number of colony forming units/ millilitre - Cfu/ml = (number of colonies visible under the magnifying glass) × (dilution factor)<sup>25</sup>*

Dilution factor: Broth after 1:8 dilution was used for plating therefore 10<sup>8</sup> is the dilution factor, also the internal diameter of the wire loop was 4mm , therefore volume of nutrient broth on loop = 0.01ml= 10<sup>-2</sup>

For eg. Cfu/ml= 54(number of colonies visible under the magnifying glass) × 10<sup>8</sup> × 10<sup>-2</sup>

Cfu/ml=54 × 10<sup>-2</sup> × 10<sup>8</sup> = 5.4 × 10<sup>5</sup>

## Asepsis protocols

All procedures were carried out in a fumigated room in the Department of Microbiology. Standard barrier technique was used; impression

trays were autoclaved, spatulas and mixing bowls disinfected with 70% ethanol. Distilled water was used for all procedures. Irreversible hydrocolloid and gypsum were dispensed from vacuum sealed packets.

## STATISTICAL ANALYSIS

Statistical analysis was performed using a specialized software S.P.S.S. (Statistical Package for Social Sciences version 20). The non-parametric Kruskal Wallis test and Mann Whitney U test were used for pairwise comparison to determine if a statistically significant difference existed between the three groups.

## RESULTS

Untreated gypsum casts showed cfu/ml counts with a median log value of  $10^5$ . Microwave irradiated casts showed cfu/ml counts with a median log value of 0. Casts poured from chemically disinfected impressions demonstrated cfu/ml counts with a median log value of  $10^4$  (Table I). The Kruskal Wallis test gave a p value of less than 0.05 that indicates there is a significant difference between the three groups (Table II). Further Mann-Whitney test was used for pairwise comparison at an error rate of 0.05 (Table III). A p value of less than 0.05 and Z value -3.297 indicates a significant difference between chemical method of disinfection and microwave irradiation.

Thus the null hypothesis was rejected.

## DISCUSSION

The conventional solution to the problem of cross contamination through dental impressions has been chemical disinfection.<sup>12,15</sup> Some studies claim that washing the impression materials with tap water removes 40% of the bacteria even though other studies indicate it has the capacity to remove 90% of the microorganisms.<sup>26</sup> The efficacy of chemical disinfectants has been the subject of several studies.<sup>23,24,26,27</sup> The most common chemical disinfectants used by dental professionals are alcohols, aldehydes, phenols, chlorine combinations, biguanides and ammonium.<sup>27</sup>

Sample No	Untreated CfU/MI	Chemical Method CfU/MI	Microwave Irradiation CfU/MI
1	$1.24 \times 10^4$	$8 \times 10^3$	0
2	$4 \times 10^5$	$4.4 \times 10^4$	0
3	$6 \times 10^5$	$8 \times 10^3$	0
4	$4 \times 10^5$	$1.2 \times 10^5$	$8 \times 10^3$
5	$5.4 \times 10^5$	$8 \times 10^4$	0
6	$3.5 \times 10^5$	$2.8 \times 10^4$	0
7	$4.8 \times 10^5$	0	0
8	$6.9 \times 10^5$	$4.4 \times 10^4$	$4 \times 10^3$
9	$4 \times 10^5$	$2 \times 10^5$	0
10	$4.8 \times 10^5$	$4 \times 10^3$	0
Median	$4.4 \times 10^5$	$3.6 \times 10^4$	0

Table-1: cfu/ml for the three groups

Groups	N	Median Number of Microorganisms	Mean Rank	
Untreated	10	$4.4 \times 10^5$	24.90	Chi-Square=22.89 Df= 2 Asymp. Sig=.000
Chemical Disinfection	10	$3.6 \times 10^4$	15.25	
Microwave Irradiation	10	0	6.35	
Total	30			

Table-2: Kruskal-Wallis Test

Mann-Whitney U Value	Z Value	Asymp. Sig (2-tailed) value
8.500	-3.297	.001

Table-3: Mann-Whitney U (Chemical method vs. Microwave irradiated)

Numerous studies have shown sodium hypochlorite in various concentrations to be most effective in disinfecting irreversible hydrocolloid impressions.<sup>1,24</sup>

However, chemical disinfectants used for disinfection of impressions have a number of problems associated with their use. They are time consuming and expensive to perform in dental practice. Almost all chemical disinfectants are potentially harmful to the health of the user and to the environment. Furthermore they are not always readily compatible with irreversible hydrocolloid<sup>15,28-30</sup> which is one of the most frequently used impression materials.<sup>16</sup> Some studies have detected bacterial contamination even after disinfection had been performed.<sup>30</sup>

Thus if elimination of possible cross contamination is considered a requirement then disinfection measures should be applied through all phases of dental treatment to both cast and

prosthesis<sup>18</sup> In practice, conventional autoclaving of the dental cast could easily damage the surface of the dental stone and immersion of the cast in a chemical disinfectant could lead to dissolution of a significant amount of gypsum to cause measurable reduction in dimensions of the cast and decrease in the compressive strength of the dental stone. Thus microwave oven disinfection might provide a convenient solution.

The results of this study shows that microwave irradiated casts exhibit a median log value of 0 cfu/ml. This is consistent with findings of previous studies<sup>[18-22]</sup> The limitations of previous studies<sup>18-19</sup> were that in the in vitro aspect of these studies impressions were made of either a resin model or a sterile metal model further these impressions were contaminated with bacterial suspensions. In another in vitro study<sup>20</sup> impressions were made of a standard silicone mold, casts were poured and then these casts were contaminated with bacterial suspensions. These studies do not take into consideration the bacterial load present in plaque that is routinely found on the surfaces of teeth. This study proves that the disinfecting potential of microwave irradiation in an in-vivo situation is consistent with the findings of the disinfecting potential of microwave irradiation in the previous in-vitro studies. The present study proves that even relatively lower levels of microwave irradiation (800W) within the range of most mid-range house hold microwaves (650-800) is as effective in disinfecting gypsum casts as higher values of irradiation used in the previous study.<sup>18</sup> Results show that casts poured from chemically disinfected impressions demonstrated cfu/ml counts with a median log value of  $10^4$  and untreated gypsum casts showed cfu/ml counts with a median log value of  $10^5$ . Hence in this study chemical disinfection fails to meet the minimum acceptable reduction of a 4-log<sub>10</sub> (99.99%) of bacteria for effective disinfection of irreversible hydrocolloid impressions.<sup>1-2</sup> The clinical importance of this study is obvious. Microwave irradiation provides an effective, quick, easy and inexpensive versatile tool for inactivation of microbial organisms on gypsum casts, which can be performed by dentists, assistants and technicians alike.<sup>17</sup>

Although there was some concern that cracks or porosity in the surface might occur when gypsum casts were exposed to irradiation of a very high wattage (1,450W) but the wattage used in this study was well within this limit.

This study shows that chemical disinfection of impression materials also has the potential to hinder possible cross-contamination but only at the time the cast is poured.

## CONCLUSION

Within the limitations of this study it can be concluded that five minutes of microwave irradiation at 800 W is most effective in significantly reducing the microbial load of dental casts. Routine use of microwave radiation for disinfection of casts could be recommended and used between procedures to prevent cross contamination in the dental clinic.

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