



## Evaluation of Sterilization Efficacy of Four Different Sterilization Methods Used for Tried-In Orthodontic Bands

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### Abstract

**Objective:** To evaluate the efficacy of four different methods of sterilization for tried in orthodontic bands after initial cleaning procedures.

**Materials and Methods:** Eighty molar bands were tried-in 20 patients. After try-in, 72 bands were equally divided into four groups; Autoclave, Glass-bead sterilizer, UV cabinet and Formalin chamber. After initial cleaning and drying, bands in each group were sterilized by respective method and incubated in separate test tubes containing Brain Heart Infusion broth. The remaining eight bands were neither cleaned nor sterilized. These served as a positive control and directly incubated in BHI broth. The BHI broth incubated at 37°C for 48 hours after the inoculation of bands and checked for turbidity.

**Results:** This study showed 100% sterilization with autoclave, 83.3% with Glass bead and 94.4% with Formalin chamber and UV tube sterilization.

**Conclusion:** This study recommended the use of Autoclave, formalin chamber and UV sterilization for tried-in orthodontic bands.

**Keywords:** Autoclave; Formalin Chamber; Glass Bead Sterilizer; Orthodontic Band; Sterilization

### Abbreviations

HBV: Hepatitis B Virus; HCV: Hepatitis C Virus; HIV: Human Immunodeficiency Virus; BHI: Brain Heart Infusion

### Introduction

The oral cavity has a huge variety of microorganisms that forms a complex environment with diverse and often pathogenic microbiota [1]. Some of the potentially transmissible pathogens are HBV, HCV, herpes simplex and HIV viruses, Mycobacterium tuberculosis, different Staphylococcal and Streptococcal strains and other microorganisms responsible for upper respiratory tract infections [2]. Not all individuals with infectious diseases can be identified before a procedure is performed; therefore, all patients, indiscriminately, should be considered potentially infectious, and consequently, standard precautions should be taken in all procedures with all patients [3].

In an orthodontic clinical practice the preformed molar bands are commonly used as it is time saving procedure. However, unlike directly bonded orthodontic attachments; where one size fits all teeth, preformed molar bands have to be carefully selected according to the size of the tooth to which they are to be cemented. Therefore, one may need to carry out several molar band trials in the mouth before the appropriate size is selected. The chances of the cross contamination with these bands are very high as the design of these bands presents a significant potential for contamination by saliva, plaque and even blood.

Lowe., *et al.* mentioned that there was a high level of residual contamination in spite of performing decontamination of Siqveland matrix bands in the dental surgery [4,5]. The presence of residual restorative materials and dental cements may compromise the subsequent decontamination process.

Benson and Douglas have shown that ultrasonic cleaning for 15 min reduces, but does not totally eliminate, detectable salivary proteins (amylase) from tried in bands [6].

Fulford MR, Ireland AJ, and Main BG suggested that the decontamination of orthodontic bands contaminated with oral secretions is safely achieved using an enzymatic cleaning agent and a bench top steam sterilizer [7]. As such, the re-use of previously tried-in molar bands should not constitute a cross-infection hazard.

Gerald E Smith proposed the use of glass bead sterilizer for sterilization of orthodontic bands and recommended its 1 minute use at 226°C [8]. Saugat Ray, *et al.* demonstrated that glass bead sterilization is equally effective as bench top autoclave for sterilization of orthodontic bands [9].

Hohlt Miller, *et al.* determined the effectiveness of three methods; standard steam, chemical vapour or dry heat sterilizing cycles for the decontamination of orthodontic instruments and bands contaminated with blood or saliva and bacterial spores [10]. He stated that the residual spores on the instruments and bands after ultrasonic cleaning and rinsing had indeed been killed in all cases and demonstrated that the three types of sterilizations were equally effective.

The aim of present study was to evaluate the efficacy of four different methods of sterilization for decontamination of tried in orthodontic bands after initial cleaning procedures.

## Materials and Methods

80 autoclaved stainless steel first molar bands were selected for the study. They were tried in the mouths of 20 such patients requiring fixed mechanotherapy in both the arches and having all four 1st molars.

All selected patients had to undergo complete oral prophylaxis followed by insertion of separator modules. Following the adequate separation, molar bands were tried-in with all four 1st molars of selected patients. Thus and so, every patient contributed 4 bands. These bands were randomly distributed in four study groups in such a way that each patient had to contribute 1 band for each group. Two bands from each group were randomly selected and included in positive control groups (n=8). These eight bands were neither cleaned nor sterilized. Remaining 72 bands in four study groups (n=18) were cleaned in ultrasonic cleansing bath for 180 sec, and then dried by oil and moisture free compressed air.

Bands in the respective study groups underwent the treatment as follows

### Group 1

Bands sterilized in an autoclave.

Bands were picked up aseptically with sterile forceps; wrapped into the sterile gauze piece and autoclaved at 132°C (270 F) at 30 psi for 8 min.

### Group 2

Bands sterilized in Glass Bead sterilizer.

When the glass bead sterilizer was on 'ready mode' with maximum temperature recorded as 220°C, four to five bands were simultaneously kept along the periphery of well of the glass bead sterilizer at 40 mm depth for 90 seconds. Bands were kept at a distance of approximately 5-6 mm from each other.

### Group 3

Bands sterilized in formalin chamber.

Formalin chamber (20x8x10 inch dimension) was saturated with 20 formalin tablets 24 hrs. before the placement of the bands inside it. Bands were kept for 60 min inside it.

### Group 4

Bands sterilized in UV tube sterilizer.

Bands were kept inside the UV tube and sterilized for 3 minutes. (Flash UV Sterilizer sr. no.100/OU/2009/v).

Bands from each group were picked aseptically by sterile forceps and placed inside the sterile container.

BHI broth was prepared according to the manufacturer's instructions. Composition of BHI broth used in this study was: calf brain infusion 200 gm/l, beef heart 250 gm/l, protease peptone 10 gm/l, dextrose 2 gm/l, sodium chloride 5 gm/l, disodium phosphate 2.50 gm/l, final pH 7.4 ± 0.2 (at 25°C).

The test tubes containing broth were sterilized and incubated for 24 hours at 37°C to ensure its sterility. The sterilized bands from four study groups as well as contaminated bands were inoculated in these test tubes under aseptic condition (Figure 1). Thereafter, the test tubes containing bands incubated for 48 hrs at 37°C. One large test tube containing BHI broth was also incubated to serve as a negative control.

The test tubes containing bands were inspected for presence of turbidity. The turbidity indicates the presence of microorganisms and thus the inadequate sterilization.

### Statistical analysis

Fisher's Exact test was performed to calculate the statistical significance among the study groups. The percentage of presence

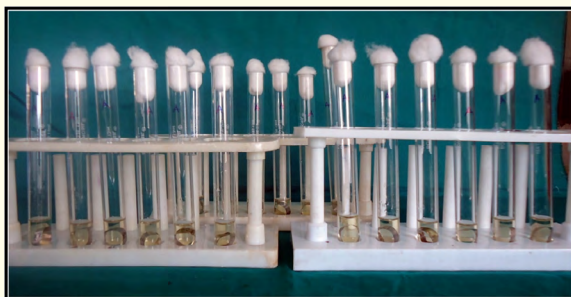


**Figure 1:** Inoculation of the bands in the test tube containing BHI broth.

and absence of turbidity was also calculated. The p-value less than 0.05 was set as statistically significant.

**Results**

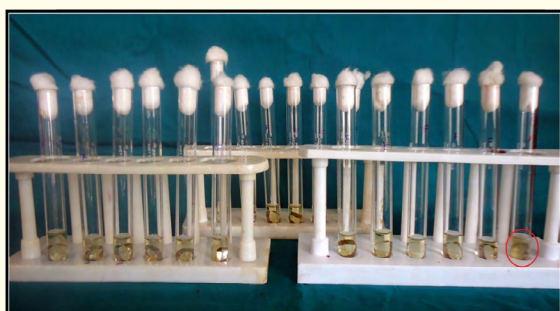
The study showed that the orthodontic bands sterilized by autoclave showed total sterility (Figure 2). The bands subjected to sterilization by glass bead sterilizer showed presence of turbidity in three test tubes. (Figure 3) The orthodontic bands sterilized by formalin chamber (Figure 4) and UV sterilizer (Figure 5) both showed the turbidity in a single test tube. The positive control group has shown the presence of turbidity in all the test tubes (Figure 6). The percentages of sterilization efficacy of four different sterilization methods calculated shown in Table 1. The comparison of group 1 with group 2 showed p-value= 0.23 which was found to be statistically insignificant. The comparison of group 1 and group 2 was done simultaneously with group 3 and 4 as both of these group (i.e. group 3 and 4) showed the same results (turbidity in single test tube). The statistical data for significance among groups showed that the percentage differences of the sterilization found among the study groups are not statistically significant; the statistical results summarized below in Tables 2, 3 and 4.



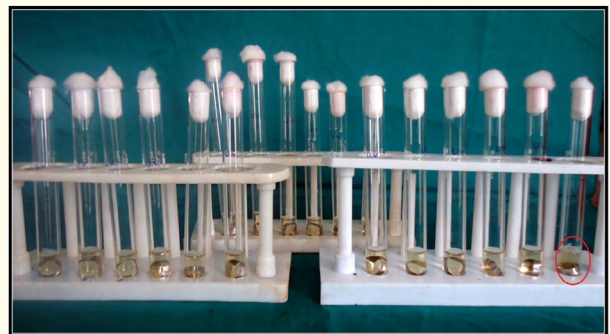
**Figure 2:** Group 1 (Autoclave sterilization).



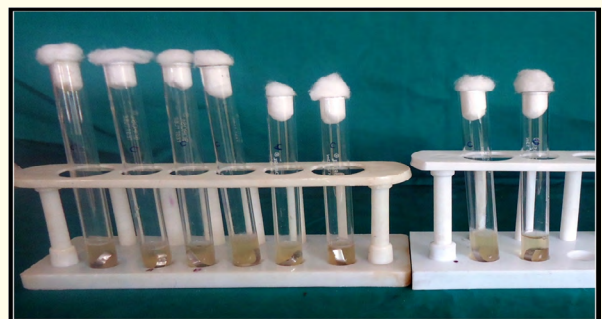
**Figure 3:** Group 2 (Glass bead sterilization)  
[Note: Highlighted test tubes shows presence of turbidity].



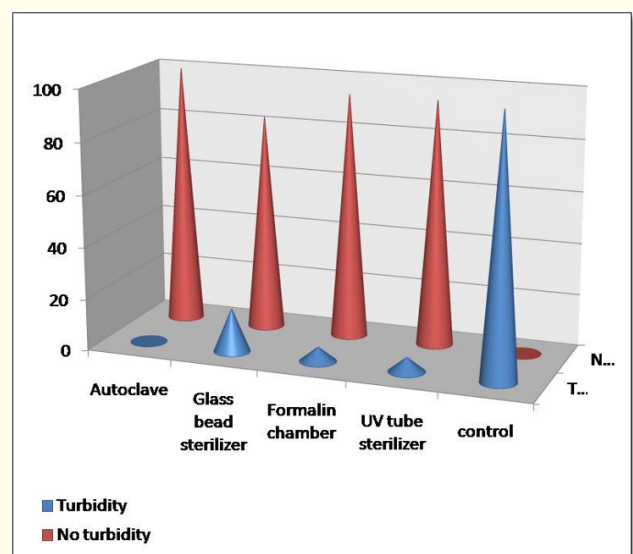
**Figure 4:** Group 3 (Formalin sterilization)  
[Note: Highlighted test tubes shows presence of turbidity].



**Figure 5:** Group 4 (UV sterilization)  
[Note: Highlighted test tubes shows presence of turbidity].



**Figure 6:** Control group  
[Note: all test tube shows presence of turbidity].



**Graph 1:** Showing sterilization efficacy of different groups.  
(Higher the turbidity; lesser the sterilization efficacy).

**Discussion**

Iatrogenic disease is the name given to the doctor-induced illness; one of them may be due to failure to sterilize the instrument causing cross-infection [11]. Matlack’s review of orthodontic offices confirmed the insufficiency of sterilization despite the fact that orthodontic offices were at a high-risk of contracting infections like hepatitis [12-14]. Saliva is one of the modes for non-parenteral spread of hepatitis B [15]. HIV and herpes virus complex are other high-risk cross infections spreading through saliva and blood.

Groups	Turbidity present		Turbidity absent	
	Number	Percentage	Number	Percentage
Group [autoclave sterilization]	-	-	18	100
Group [Glass bead sterilization]	3	16.7	15	83.3
Group 3 [Formalin sterilization]	1	5.5	17	94.4
Group 4 [UV sterilization]	1	5.5	17	94.4
Control	8	100	-	-

**Table 1:** Comparative data showing presence and absence of turbidity in different groups.

Groups	Turbidity Present	Turbidity Absent	Total	p-value
Group 1	0 (0.00)	18 (100)	18	0.23
Group 2	3 (16.7)	15 (83.3)	18	
Total	03	33	36	

**Table 2:** Pair wise comparison between group 1 and group 2.

Groups	Turbidity Present	Turbidity Absent	Total	p-value
Group 2	3 (16.7)	15 (83.3)	18	0.6
Group 3 and Group 4	1 (5.5)	17 (94.4)	18	
Total	04	32	36	

**Table 3:** Pair wise comparison between group 2 and group 3, 4.

Groups	Turbidity Present	Turbidity Absent	Total	p-value
Group 1	0 (0.00)	18 (100)	18	1
Group 3 and Group 4	1 (5.5)	17 (94.4)	18	
Total	01	35	36	

**Table 4:** Pair wise comparison between group 1 and group 3, 4

According to the recommendations by British Dental Association, there are three stages of the decontamination process of dental instruments [16]. These are presterilization cleaning, sterilization and storage. Presterilization cleaning can be done as hand cleaning or by ultrasonic cleaners.

Benson and Douglas have shown that ultrasonic cleaning for 15 minutes reduces, but does not eliminate, detectable salivary proteins (amylase) from tried-in bands [6]. They found that 50% of molar bands that were tried for size in the mouth had detectable amylase, albumin or both, even after 15 minutes in an ultrasonic cleaning bath. The volume of detectable amylase significantly

reduced compared with unclean bands; however, the reduction in the volume of albumin was not statistically significant.

Saugat Ray *et.al* validates the fact that autoclave is the gold standard of sterilization but as far as orthodontic bands are concerned glass bead sterilization is equally effective, time saving and reliable method, which can be followed routinely in the busy clinical practice, where rapid turnover is required [9].

In the present study the bands autoclaved after ultrasonic cleaning at 132°C (27°F) at 30 psi for 8 minutes showed no turbidity suggesting no growth of microorganisms after 48 hours of incubation at 37°C.

The glass bead sterilizer uses a metal cup with glass beads of 1.2-1.5 mm in diameter. Larger beads are not effective in transferring heat to due to presence of large air spaces between the beads which reduces the efficiency of the sterilizer when operated at a temperature range of 218°C- 240°C for 3-5 seconds.

In the present study, four to five bands were kept at the periphery and 40 mm deep in the well of the glass bead sterilizer for 90 seconds after the sterilizer is ready with max temperature recorded being 220°C. The bands were kept at a distance of approximately 5-6 mm from each other showed the sterilization efficacy of 83.3%. However, in contemporary orthodontic practice, the prefabricated molar bands have molar tubes or other auxiliaries attached to it, in such cases the glass beads fails to contact the inner surface of the tubes.

The sterilization of bands in present study using formalin chamber had shown sterilization efficacy of 94.4%. It is considered as the safest process as it does not compromise on the cutting efficiency of instruments even though dulling is seen with long-term use. Although advantageous from an orthodontist's perspective, it has not gained popularity due to the presence of a strong odour necessitating separate enclosures with adequate ventilation. Present day chemiclaves meet or exceed OSHA emission standards as they are equipped with a built in chemi-purge and chemi-filter to slash emissions [17]. An added advantage of formalin chamber is its low cost.

The U.V. sterilizer used in present study shows the sterilization efficiency of 94.4% for the sterilization cycle of 3 min. So it has been found to be more time efficient method of sterilization as compared to autoclave or formalin chamber. Although, glass bead sterilizer takes only 90 seconds, its percentage sterilization found to be lower than UV sterilization.

BHI broth is a liquid culture medium used for cultivating wide varieties of bacteria (streptococci, pneumococci, meningococci, etc.) it is a highly nutritive medium. In the present study, any absence of contamination of the broth was confirmed by testing the negative control group where no growth was observed after 48 hours of incubation. The efficacy of the BHI broth was confirmed by the positive control group, which has shown the increase in turbidity of the broth in all the test tubes after 48 hours of incubation.

The present study showed the complete sterilization of the bands sterilized in an autoclave similar to the earlier studies [9]. Though, this study showed lesser efficacy of Glass bead sterilizer, it has not found to be statistically significant. Based on present study the use of formalin chamber and UV tube sterilizer is recommended for sterilization of tried-in orthodontics bands, which showed statistically insignificant difference as compared with the gold standard autoclave sterilization.

### Conclusion

The present study concludes that autoclave is the gold standard of the sterilization but as far as tried-in orthodontic bands are concerned UV tube sterilizer and formalin chamber sterilization are equally effective. The use of glass bead sterilization found to be least effective among the tested groups. This study recommends the use of UV tube sterilization method for the sterilization of tried-in orthodontic bands as it is quick, efficient, and easy to use.

### Conflicts of Interest

Authors deny any conflicts of interest.

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