

# Sterilization and Disinfection of Extracted Human Teeth for Institutional Use

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## Abstract

**Objective:** The study was done to determine the various effective methods of sterilization in extracted human teeth.

**Materials & methods:** 80 non carious molar teeth were extracted due to periodontal disease and used to determine the effectiveness of different disinfection, sterilization method of using bacillus subtilis, a spore bearing thermo resistant bacteria frequently used to test sterilizers. The teeth were collected and stored in normal buffered saline. 10 teeth were placed in each of 8 groups. 7 groups with 10 teeth were immersed in 5% Naocl, 2.5 % Naocl,, 10 % formalin, 3 % H<sub>2</sub>O<sub>2</sub>, 2% gluteraldehyde, 3% hypochlorite solution and autoclaving at 121<sup>0</sup>C, 16 lbs pressure for 20 minutes respectively and last was immersed with normal saline as control ,subsequently incubated for 7 days at 370c in blood agar, mackonkey agar, nutrient agar before and after treatment for 48 hrs. The plates were examined before treatment and after treatment for growth of Bacteria.

**Result:** It was found that autoclaving at 121<sup>0</sup>C for 20 minutes at 16 lbs pressure, 10 % formalin, 3 % sodium hypochlorite solution was 100% effective.

**Conclusion:** It was concluded that formalin, hypochorite, autoclaving was the choice of sterilization of human extracted teeth for institutional use.

**Key words:** Extracted human teeth, autoclaving, formalin, hypochlorite, sterilization, culture media.

## Introduction

Extracted teeth were used routinely in dental schools to teach and to build the technical and preclinical skills of the students in the beginning of the course and before the entry to clinical environment to deliver dental care to the patients. Some endodontic procedures can be taught using models, plastic blocks, and typodont tooth models. However there are instances where no substitute were available for extracted teeth for examination, preparation or research.<sup>1</sup> Human extracted teeth were used in dental institutes to teach the clinical methods for patient's treatment . More over these teeth were used for various purposes such as preparing ground section for histological study and for learning endodontic procedures such as cavity preparation , root canal treatment, developing and testing various restorative materials, for crown preparation, and inlay<sup>2</sup> Now a days typodonts are used as educational tool for students, allow them to practice certain dental procedures before performing on live patients. In India due to less income group, extracted human teeth were used which serves as a low budget substitute to typodont. There were instances typodont can't replace extracted teeth.<sup>3</sup> Teeth extracted for various purposes were the possible cause for cross contamination to various laboratory equipments and workers . So, extracted teeth must be sterilized to prevent spread of infection. Various methods of sterilization were used for instruments that comes in direct contact with blood stream to avoid infections such as human immunodeficiency Virus, Hepatitis B virus ( HBV) and Hepatitis C virus(HCV). It can be achieved by autoclave, hot air oven, gluteraldehyde or

formaldehyde solutions or physical agents (radiation). The effectiveness of any method of sterilization depends on time, contact, temperature with steam sterilization, high pressure, number of microorganism present, amount and type of organic material that protects the microorganisms. Extracted human teeth were difficult to sterilize because of the structure as they might be spoiled or changed by sterilization methods.<sup>4,5</sup> In recent years, infection control guidelines in dental institutions has been revised due to the possibility of cross contamination from extracted teeth.<sup>3</sup> Directives by the American Dental Association (ADA) and Centre for Disease Control (CDC) called for removal of any organism capable of transmitting disease from non disposable items used in patient care. The materials used might have come in contact with blood or saliva. These body fluids were associated with extracted human teeth that are routinely used in dentistry to develop technique and skills.<sup>4</sup> Various method have been tried for disinfection / sterilization of extracted human teeth with variable results. The centre for Disease control (CDC) recommended storing extracted teeth in 1:10 household Bleach<sup>4</sup>. However Tate and white showed household bleach was poor disinfectant for this purpose.<sup>5</sup> Ethylene oxide can also used as sterilizer agent but its efficacy has been found 20 % - 30% on bacillus subtilis spores in extracted teeth.<sup>6</sup> Various new method of sterilization have been introduced with minimal effect on the tooth structure. Y radiation sterilizes without high temperature, pressure, chemicals or gases. They have no effect on teeth.<sup>7</sup> Sodium hypochlorite subsonic agitation caused more effective reduction in bacterial load in the root canal as compared to sodium hypochlorite alone.. As in institutional set up in which extracted teeth were used in preclinics for number of procedures, it was necessary to find best method of sterilizing extracted teeth.

### Objective

The main purpose of this study was to determine the effectiveness of various disinfection methods of extracted human teeth in Institute of Dental Sciences, Bareilly.

### Materials and Method

80 freshly extracted human teeth were obtained from the Department of oral and Maxillofacial surgery of Institute of Dental Science, Bareilly and bacteriological process was carried out in the Department of Microbiology, Rohilkhand Medical College and Hospital, Bareilly. The teeth were kept in sterile normal saline in bottle till tested and divided in to eight groups containing 10 teeth in each including control.

**Group - 1** Teeth (10) were immersed in 10ml of 5 % sodium hypochlorite in bottle for 7 days.

**Group - 2** Teeth(10) were immersed in 10 ml of 2.5 % sodium hypochlorite in bottle for 7 days.

**Group - 3** Teeth(10) were immersed in 10 ml of 10 % formalin in bottle for 7 days.

**Group - 4** 10 teeth were kept in 10 ml of 3 % of Hydrogen peroxide in bottle for 7 days.

**Group - 5** 10 teeth were immersed in 10 ml of 2% Gluteraldehyde in bottle for 7 days.

**Group - 6** 10 teeth were kept in 10 ml of 3% sodium hypochlorite solution in bottle for 7 days.

**Group - 7** 10 teeth were autoclaved at 121°C , 16lbs pressure for 20 minutes duration.

**Group - 8** (control) 10 teeth were kept in 10 ml Normal saline as Control.

A platinum wire loop was flamed in red heat in burner and cooled. A sample was taken before treatment and after treatment from the container group wise. A loop full sample was inoculated in simple, selective, enrichment media like Nutrient agar, Blood agar, Mackonkey agar. The plates were placed at incubator at 37<sup>0</sup>c for 48 hours. After 48 hours of incubation, growth in of different types of colony, its size, consistency, lactose fermentation, haemolysis etc were observed carefully. On the basis of colony morphology, Grams and Biochemical reaction the organisms are identified. The colony count also noted to observe the quantity of microorganism.

### Results

Table 1 shows that 10% formalin, 3% Na hypochlorite solution, Autoclaving 121<sup>0</sup>c, 16 lbs pressure for 20 minute was 100 % effective showing no growth of bacteria where as other disinfectants like 5% sodium hypochlorite, 2.5 % sodium hypochlorite, 2% glutaraldehyde , 3% hydrogen peroxide showed the growth of bacteria was 40%, 50%, 60%, 30% respectively.

### Discussion

Sterilization is the process by which article, surface, medium made free from microorganisms excluding bacterial spores.<sup>8</sup> A extracted teeth serve for educational tool in teaching institute in particular to dental students. It was documented HIV, HBV, HCV and aerobic, anerobic bacteria were present as pathogenic and non pathogenic state in pulp, radicular and periradicular tissue of extracted human teeth.<sup>9,10</sup> Sterilizing agent should be highly effective for because mouth is the gate of infection in human body and eventually the microorganism normally present in oral cavity may be one of the source of systemic organ involvement. HIV,HBV, HCV virus which are related to dental surgery. Sterilization technique is an important for health care delivery system in hospital.

This study documented 10% formalin, 3% sodium hypochlorite solution, autoclaving of extracted teeth are highly effective for sterilization and disinfection of extracted human teeth for institutional use. 10% formalin and autoclaving 121<sup>0</sup>c, 16 lbs for 20 minutes was consistant with the study<sup>11,12</sup>. In the studies of Dominici et al & Simarjeet et al showed that 2.5 % hypochlorite solution is 100% effective<sup>1,12</sup>. But in this study 3 % hypochlorite solution was effective with margin difference of concentration of sodium hypochlorite solution 5%.<sup>1,11 12</sup> Other chemicals, disinfectant used in this study were less effective in comparison with 10%

Type of disinfection/sterilisation	Duration	No of Teeth	Efficacy of different disinfectant solution	Growth of bacteria
3 % sodium hypochlorite solution	7day	10	10 (100%)	No Growth
10 % formalin	7 day	10	10 (100%)	No Growth
Autoclaving 121 <sup>0</sup> C, 16lbs pressure,	20 mins	10	10 (100%)	No Growth.
5 % Naocl	7day	10	04 ( 40%)	Growth +ve
2.5 % Naocl	7day	10	05 (50%)	Growth +ve
2 % Glutarlehyde	7 day	10	06 (60%)	Growth +ve
3 % H2O2	7 day	10	03 (30%)	Growth +ve
Normal Saline (Control)	7 day	10	10 ( 100%)	No Growth

**Table 1 : Efficacy of different disinfectant solution in relation to growth of bacteria on 10 teeth for the duration of 7 days.**

formalin, 3% hypochlorite solution and autoclaving for 20 minutes. The difference of effectiveness of disinfectants was due to poor penetration of the agents in to pulp space or in activation of the disinfectants by the organic substances present in teeth. Formalin was highly effective but it acts as irritant and carcinogenic<sup>10</sup>. Autoclave requires continuous source of heat in the form of electricity, strict adherence to time, pressure. It releases mercury vapors in the air through autoclave exhaust and has residual mercury contamination.<sup>1</sup> It was reported sodium hypochlorite solution may increase the enamel porosity by deproteinisation.<sup>13,14</sup> Formalin, autoclaving and hypochlorite treatments are simple and can be routinely used. They do not alter the 'Feel' and cutting characteristics of the teeth.<sup>15,16</sup>

## Conclusion

Based on this study it was evident that formalin, hypochlorite, autoclaving was the choice of sterilization of human extracted teeth for institutional use and extracted teeth are hazardous and should be handled carefully.

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