## **Original Article**

# Awareness and Knowledge of Undergraduate Dental Students about Sterilization/Disinfection Methods of Extracted Human Teeth

# Deogade SC, Mantri SS, Saxena S<sup>1</sup>, Sumathi K

Departments of Prosthodontics and Crown and Bridge and <sup>1</sup>Public Health Dentistry, Hitkarini Dental College and Hospital, Jabalpur, Madhya Pradesh, India

Address for correspondence: Dr. Deogade SC, Flat No: 502, Block-D, Apsara Apartment, South Civil Lines, Pachpedi Road, Jabalpur - 482 001, Madhya Pradesh, India. E-mail: dr. deogade@yahoo.co.in

# Abstract

Background: Dental undergraduate students work on extracted human teeth in preclinical practical's to learn technical skills before entering the clinics and delivering dental care to the patients. Aim: The aim of the present investigation was to assess the awareness and knowledge toward sterilization/disinfection methods of extracted human teeth in a selected group of Indian dental students. Materials and Methods: In this descriptive cross-sectional study, the participants consisted of 2<sup>nd</sup>-, 3<sup>rd</sup>-, 4<sup>th</sup>-, and 5<sup>th</sup>-year dental students. Data were collected by questionnaires and analyzed by Mann-Whitney U-test and Kruskal-Wallis test using SPSS software version 16 for Windows (SPSS Inc., Chicago, IL, USA). Results: In this study, 235 dental students participated in the study. The average awareness and knowledge score was 7.27 (1.92). Based on the opinion of 57% (134/235) students, hydrogen peroxide was selected as the suitable material for sterilization and 24.6% (58/235) students believed that autoclave sterilization is a good way for the purpose. **Conclusion:** The results of this investigation indicated that awareness and knowledge of undergraduate dental students in relation to sterilization/disinfection methods of extracted human teeth were good. However, deficiencies were observed in relation to teaching the material and methods suitable for sterilization.

Keywords: Awareness, Dental student, Extracted human teeth, Knowledge

# Introduction

Infection control is the most crucial and critical issue of dentistry including subbranches that are related to the health of dental clinicians, the assistant dental personnel, and the patients. Dental undergraduate students have to learn various technical and preclinical skills before entering the clinics and delivering dental care to the patients. To fulfill this curriculum, the students generally practice on extracted human teeth that simulate different dental procedures performed while delivering treatment care to the patients.<sup>[1]</sup> The other methods to follow this include artificial plastic blocks or teeth on manikins

Access this article online		
Quick Response Code:	Website: www.amhsr.org	
	DOI: 10.4103/amhsr.amhsr_85_16	

and models. However, these instructional tools cannot replace the natural human teeth in practical examinations, education, and research work.<sup>[1,2]</sup> Furthermore; these tools are only used when access to extracted human teeth is restricted or impossible. These extracted teeth have been noticed as a resource for infection. Some researchers, hence, advocated evaluating the effects of disinfection/sterilization on human extracted teeth.<sup>[2,3]</sup>

American Dental Association (ADA) and Centers for Disease Control (CDC) recommend the thorough removal

This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 3.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

For reprints contact: reprints@medknow.com

How to cite this article: Deogade SC, Mantri SS, Saxena S, Sumathi K. Awareness and knowledge of undergraduate dental students about sterilization/disinfection methods of extracted human teeth. Ann Med Health Sci Res 2016;6:348-55.

of microorganisms capable of transmitting disease from non-disposable materials used in treating patients. These also include the materials used in simulated preclinical education that might have contaminated with blood or saliva. These body fluids are attached with the teeth after its extraction causing a source of infection. These extracted teeth may lead to cross-contamination to dental undergraduate students who routinely work on these teeth to improve their clinical skills and techniques.<sup>[2-5]</sup> Kumar et al<sup>[2]</sup> have brought in notice that hepatitis B virus (HBV), human immunodeficiency virus (HIV), and many other blood-borne bacterial pathogens may be present in the pulp and radicular and periradicular tissues of extracted human teeth. In the preclinical practical's, the tooth preparation procedures on extracted teeth are carried out as a routine educational exercise. This procedure is generally done without using a liquid coolant which leads to a greater risk of exposure to pathogenic bacteria in the preclinics. As a result of this, the risk of contagion spread through aerosols and accidental penetrating wounds that might happen during handling of dental instruments such as low-speed micromotor and diamond burs, is increased doublefold.<sup>[4]</sup>

Several authors <sup>[4,6]</sup> have discussed the problems associated with the use of extracted human teeth as these teeth are grossly contaminated and difficult to sterilize because of their structure, and furthermore these teeth might be damaged or altered by sterilization procedures used for it. Kumar et al<sup>[2]</sup> studied the knowledge of dental students about infectious risk of extracted teeth used in preclinical practical's and concluded that about 90% of students know that extracted teeth are sources of infection but only 75% of them performed a disinfection method to eliminate contamination from these teeth. They also found that most of the students used boiling water and sodium hypochlorite solution to sterilize these extracted teeth. Tate and White<sup>[7]</sup> revealed from their study that formaldehyde is the only antiseptic solution that can achieve an effective antimicrobial concentration within the pulp space. White and Hays<sup>[8]</sup> tested ethylene oxide to sterilize extracted human teeth and demonstrated its inefficacy against Bacillus subtilis spores placed in the chamber of extracted human molar teeth. White et al<sup>[9]</sup> studied the effect of gamma radiation while sterilizing teeth and found that these radiations sterilize teeth and endodontic filling materials without altering the structure and function of dentin. The authors also recommended a dose of 173 k-rad with the help of a cesium radiation source for sterilization purpose. Dominici et al<sup>[1]</sup> suggested the use of autoclaving for 40 min at 240°F and 20 psi or soaking in 10% formalin for 1 week to achieve 100% efficacy in preventing microbial growth. Pantera and Schuster<sup>[4]</sup> showed that Rockal solution (benzalkonium chloride) for 24 h and 3 weeks was inefficient to eliminate microorganisms from extracted teeth. Attam et al<sup>[10]</sup> compared the effect of autoclaving and 10% formalin storage on extracted teeth and found that the chemical materials such as 2.6% sodium hypochlorite, 3% hydrogen peroxide, and boiling water are not suitable and effective for disinfecting/sterilizing extracted human teeth.

Since there are limited studies about the awareness and knowledge of undergraduate dental students in relation to sterilization/disinfection methods of extracted human teeth used in preclinical practices, this study was designed to evaluate these parameters among a group of dental students of Hitkarini Dental College and Hospital in Madhya Pradesh (India).

## **Materials and Methods**

This descriptive cross-sectional study was carried out on 2<sup>nd</sup>-, 3<sup>rd</sup>-, 4<sup>th</sup>-, and 5<sup>th</sup>-year dental students at Hitkarini Dental College and Hospital, Rani Durgawati University, Jabalpur, Madhya Pradesh, India. It was planned to use census sampling method for this investigation. Census sampling is the method of data collection from every member of the population.<sup>[11]</sup> In the present study, questionnaires were distributed among all 2<sup>nd</sup>-, 3<sup>rd</sup>-, 4th-, and 5th-year undergraduate students. A self-administered questionnaire was prepared based on previous studies. <sup>[1,2,4]</sup> This questionnaire included the demographic data and personal information of the participants along with the questions related to their awareness and knowledge about sterilization/disinfection of extracted human teeth. Nature of the study was explained with a patient information sheet, and an informed consent of all the participants was obtained. The approval was obtained from the Ethical Committee of the Rani Durgawati University, after which, this investigation was carried out from July 2014 to September 2014.

To examine the validity of the designed questionnaire, it was produced before ten specialist dental practitioners to advise their level of agreement to the question statements using a five-point rating scale (extremely appropriate, appropriate, no idea, inappropriate, and extremely inappropriate). On this basis, some of the test questions were modified to improve clarity of understanding, and a discussion was arranged for each patient to validate the questions and apply the necessary modifications to validate the questionnaire.

The validity of entire questionnaire and each question was 79% and 75%-89%, respectively. These values of validity were acceptable. Cronbach's alpha was used to assess the reliability of the questionnaire and collecting the feedback from 15 dental students to the same questionnaire within a 15-day period. Cronbach's coefficient for the reliability was 0.87, which was suitable for an acceptable study. After the analysis and discussion, the final questionnaire consisted of 11 questions in relation to awareness and knowledge in addition to questions about demographic data and personal information. The questionnaires were distributed among 2nd-, 3rd-, 4th-, and 5<sup>th</sup>-year students by the investigator. The goal of the study was explained to the participants, and they were left alone to fill the questionnaire anonymously. To score the awareness and knowledge questions, each correct response was given a score of 2; each wrong one was given a score of 0 score and no answer was given a score of 1.

Shapiro–Wilk test showed that data did not follow normal distribution; nonparametric tests, namely Mann–Whitney U-test and Kruskal–Wallis test were used for further data analysis. P < 0.05 was considered statistically significant. Data analysis was done using Statistical Package for the Social Sciences (SPSS Inc., Chicago, IL, USA) version 16 for windows.

## **Results**

A total of 240 questionnaires were distributed among dental undergraduates, and only 235 students returned valid questionnaires. This represented 97.9% (235/240) response rate. This response rate is adequate for analysis and therefore 97.9% response rate was considered as being very good for analysis. Of 235 respondents, 75.7% (178/235) were female students and 24.2% (57/235) were male students. Distributions of participants by sex and education year are shown in Figures 1 and 2, respectively. The mean age of the respondents was 22.2 (1.65) years (a range of 19-27 years). The overall mean score for awareness and knowledge was 7.2 (1.9) (7.1 [2.2] and 7.3 [1.8] for males and females, respectively, range = 0-10) [Table 1]. No significant relationship was observed between male and female respondents for awareness and knowledge scores (P = 0.69). However, Kruskal-Wallis test showed a significant difference between the awareness and knowledge scores of different education years [Table 2]. When Mann–Whitney U-test was applied for pair-wise comparison, it showed a significant difference between 2<sup>nd</sup> and 4<sup>th</sup> Year, 2<sup>nd</sup> and 5<sup>th</sup> Year, 3<sup>rd</sup> and 4<sup>th</sup> Year, 3<sup>rd</sup> and 5<sup>th</sup> Year, and 4<sup>th</sup> and 5<sup>th</sup> Year (P < 0.001).

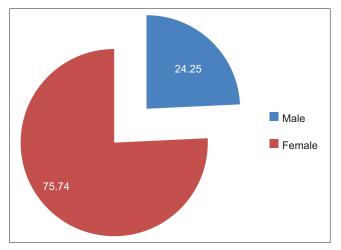


Figure 1: Gender-wise distribution of participants (in percentage)

Difference between  $2^{nd}$  and  $3^{rd}$  year was statistically not significant.

The results showed that 90.2% (212/235) of the respondents were aware that extracted human teeth can be a source of infection, whereas only 1.7% (4/235) of them had no idea about it. Most of the participants were not aware that these extracted teeth can be a source of HBV (26.0% [61/235]), hepatitis C virus (HCV) (29.8% [70/235]), and HIV (38.3% [90/235]) infections [Table 3]. The results revealed that 48% (113/235) of the students had no previous formal training in sterilization of extracted teeth and other students were trained about sterilization in the Department of Endodontics. About 35.3% (83/235) of the students responded that they were not asked to sterilize these teeth. Almost 89.3% (210/235) of the students had worked on extracted human teeth and 93.6% (220/235) of the participants thought that they need education regarding disinfection of these teeth. Regarding the question about respondents' opinion on the most appropriate methods for disinfection or sterilization of extracted human teeth, 57.0% (134/235) of the students chose hydrogen peroxide and 24.6% (58/235) chose autoclave [Table 4]. About 73.2% (172/235) of the students chose one method, 14.5% (34/235) students chose two methods, 10.2% (24/235) students chose three methods, and 2.1% (5/235) students chose more than three methods for disinfection/sterilization of extracted teeth [Table 5]. The most commonly used method was hydrogen peroxide and autoclave. The results showed that 91.5% (215/235) of

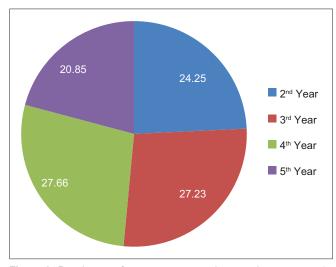


Figure 2: Distribution of participants according to education year (in percentage)

Table 1: Gender-wise comparison of awareness and knowledge scores					
Gender	ender A			Awareness and knowledge scores	
	Minimum	Maximum	Mean (SD)	Mann-Whitney U-test value	Р
Male	1	10	7.12 (2.23)	4900.500	0.695 (NS)
Female	0	10	7.31 (1.81)		
Over all	0	10	7.27 (1.92)		

SD: Standard deviation, NS: Not significant

#### Deogade, et al.: Behavior of dental students toward sterilization of extracted teeth

Education	Awareness and knowledge scores					
year	Minimum	Maximum	Mean (SD)	Kruskal-Wallis test	Mann-Whitney <i>U</i> -test for pairwise comparison (significant differences	
2 <sup>nd</sup> year	1	10	6.16 (1.72)	74.46	2 <sup>nd</sup> year and 4 <sup>th</sup> year	
3 <sup>rd</sup> year	2	10	6.59 (1.59)	<i>P</i> <0.001 (S)	2 <sup>nd</sup> year and 5 <sup>th</sup> year	
4 <sup>th</sup> year	0	10	7.58 (1.63)		3 <sup>rd</sup> year and 4 <sup>th</sup> year	
5 <sup>th</sup> year	5	10	9.02 (1.51)		3 <sup>rd</sup> year and 5 <sup>th</sup> year	
Over all	0	10	7.27 (1.92)		4 <sup>th</sup> year and 5 <sup>th</sup> year	

SD: Standard deviation, S: Significant

Table 3: Responses of participants to awareness and knowledge questions (percentage)			
Questions	Yes (%)	No (%)	No idea (%)
Is preclinical practical procedure practiced on mounted extracted human teeth?	229 (97.5)	6 (2.5)	0
Can extracted human teeth be a source of infection?	212 (90.2)	19 (8.1)	4 (1.7)
Can these teeth be a source of HBV transmission?	124 (52.8)	61 (26.0)	50 (21.3)
Can these teeth be a source of HCV transmission?	85 (36.2)	70 (29.8)	80 (34.0)
Can these teeth be a source of HIV transmission?	111 (47.2)	90 (38.3)	34 (14.5)
Is there a necessity to disinfect/sterilize extracted teeth before working on them?	215 (91.5)	16 (6.8)	4 (1.7)
Is there a necessity to use mask while working on extracted human teeth?	224 (95.3)	8 (3.4)	3 (1.3)
Is there a necessity to use safety glasses while working on extracted human teeth?	180 (76.6)	40 (17.0)	15 (6.4)
Is there a necessity to use gloves while working on extracted human teeth?	231 (98.3)	3 (1.3)	1 (0.4)
Should students be taught about different methods of disinfection for extracted human teeth? HBV: Hepatitis B virus, HIV: Human immunodeficiency virus, HCV: Hepatitis C virus	220 (93.6)	9 (3.8)	6 (2.6)

Table 4: Responses of participants regarding sterilization/ disinfection methods of extracted human teeth according to gender (percentage)

Method	Male (%)	Female (%)	Total (%)
Hydrogen peroxide	24 (42.10)	110 (61.80)	134 (57.02)
Formalin	9 (15.79)	41 (23.03)	50 (21.28)
Alcohol	2 (3.51)	10 (5.62)	12 (5.11)
Chlorhexidine	2 (3.51)	6 (3.37)	8 (3.40)
Boiling water	9 (15.79)	11 (6.18)	20 (8.51)
Sodium hypochlorite	8 (14.03)	12 (6.74)	20 (8.51)
Saline	1 (1.75)	7 (3.93)	8 (3.40)
Glutaraldehyde	1 (1.75)	12 (6.74)	13 (5.53)
Autoclave	15 (26.31)	43 (24.16)	58 (24.68)
Oven	0	1 (0.56)	1 (0.42)
No idea	6 (10.53)	2 (1.12)	8 (3.40)

Table 5: Number of sterilization/disinfection methodspreferred by the participants (percentage) (n=235)

Number of methods	Number of participants (%)
One	172 (73.2)
Two	34 (14.5)
Three	24 (10.2)
Four	5 (2.1)

the participants wished to disinfect extracted teeth before working on them and 95.3% (224/235) of the participants responded to use mask, whereas working on these teeth, 98.3% (231/235) responded for gloves and 76.6% (180/235) responded for safety glasses.

# **Discussion**

It has already been discussed that all body fluids and tissues must be considered as sources of infection for HIV, HBV, and HCV or other blood-borne pathogenic microbes. In preclinical practical education in dentistry, different procedures are performed on extracted human teeth. These teeth are always in direct contamination with body fluids and are therefore considered as dangerous sources for cross-contamination.<sup>[7,12]</sup> Infectious disease transmission has long been a concern in the practice of medicine and dentistry. The infection sources such as saliva, blood, and body fluids which are present in clinical settings may contaminate and exist in the extracted stored teeth which are used further for preclinical education.<sup>[1]</sup> The Occupational Safety and Health Administration considers extracted human teeth in research and teaching purposes as potential infection sources of blood-borne pathogenic microbes.<sup>[13]</sup>

In the present investigation, the awareness and knowledge of dental undergraduate students were evaluated in relation to the use of extracted human teeth and methods that they preferred appropriate to disinfect them. Dental undergraduate students had an acceptable level of knowledge in relation to disinfection methods for extracted human teeth which is consistent with the findings of Kumar *et al.*<sup>[2]</sup> Hashemipour *et al.*<sup>[14]</sup> reported that more than half of the students in their study did not had any education about sterilization of extracted teeth and were not asked to disinfect these teeth while working in the laboratory. However, in the study conducted by Kumar *et al.*<sup>[2]</sup>

about 87.5% of the dental students in Indian dental school were forced to sterilize the extracted teeth. In the present investigation, about 96.1% of students were aware regarding sterilization of the extracted teeth. This difference can be attributed to difference in education in the past 10 years about disinfection/sterilization. Furthermore, the other reason may be that Indian education is more concerned about the infection of extracted teeth due to the higher prevalence of different diseases in this country during these 10 years. Extracted human teeth are a source of various transmitting infections; hence, disinfection of these teeth becomes essential to prevent dissemination of diseases. Different sterilization/disinfection methods can be practiced, which include sodium chloramines, formalin, sodium hypochlorite, alcohol, glutaraldehyde, autoclaving, normal saline, freezing, 1:10 household bleach, ethylene oxide sterilization, and gamma radiation.<sup>[2,15]</sup> In the present study, students chose hydrogen peroxide and autoclave as the best options to sterilize extracted teeth although most of the students used boiling water and storing in formalin and sodium hypochlorite to sterilize these teeth. This finding was almost consistent with Kumar *et al*<sup>[2]</sup> as in their study most of the students used sodium hypochlorite as the first option. Most of the students preferred only one method of sterilization/disinfection for extracted human teeth. The possible reason for this is that since the 3<sup>rd</sup>-, 4<sup>th</sup>-, and 5<sup>th</sup>-year BDS students have already passed the microbiology and pathology examinations, they had fair knowledge about blood-borne pathogens, most of them considering extracted teeth a possible source of infection, capable of provoking cross-infections in educational settings.

Sterilization/disinfection methods should not alter the physical qualities of dentin and enamel structures and hence that the operating characteristics of the shear bonding and the sense of touch will be the same as clinical environments. This method must also be able to disinfect the harmful bacteria within the root canals.<sup>[7]</sup> Very few studies are available regarding the methods practiced for disinfection and sterilization of extracted teeth. The important factor that should be considered while performing educational work on extracted teeth is that the time duration since the extraction can alter the properties of these teeth while they are still a rich source for different transmitting infections.<sup>[16,17]</sup> Tate and White<sup>[7]</sup> suggested the formaldehyde solution as the most powerful antiseptic that can achieve an effective antimicrobial concentration within the pulp space. They reported that the 10% formalin is the only disinfectant which penetrates the pulp chamber efficiently. George et al<sup>[18]</sup> studied the effect of formalin storage on the apical microleakage of obturated canals and found that the rate of apical microleakage in the case group stored in formalin was much less than that in the control group. They also found that this rate decreases for the extracted teeth in formalin in comparison to nonfixed specimens and this was significant. This finding is consistent with the findings of other studies.<sup>[8,19-22]</sup>

Some workers<sup>[23,24]</sup> doubted the application of formalin for educational purpose as it releases dangerous and carcinogenic materials. White and Hays<sup>[8]</sup> reported the failure of ethylene oxide against B. subtilis spores placed in the chamber of extracted human molars. In their study, they found that 64% of the teeth exposed to cold ethylene oxide treatment and 80% of the teeth exposed to the warm treatment still contained viable spore. This proved that ethylene oxide is not an effective antimicrobial agent in eradicating infections from extracted human teeth. White et al<sup>[9]</sup> compared different methods of sterilization/disinfection of extracted teeth. They used gamma radiation and compared with autoclaving, ethylene oxide, and dry heat and found that gamma radiation sterilizes teeth and endodontic filling materials without altering the structure and function of dentin. They recommended a dose of 173 k-rad for complete sterilization with the help of a cesium radiation source. The authors found undetectable changes with gamma radiation and some detectable changes with all the other methods in the spectra. Dominici et al<sup>[1]</sup> reported that only autoclaving for 40 min at 240°F and 20 psi or soaking in 10% formalin for 1 week was 100% effective in preventing microbial growth. Furthermore, Kumar *et al*<sup>[2]</sup> found that autoclaving at 121°C, 15 lbs psi for 30 min, and immersion in 10% formalin for seven is effective in disinfecting/sterilizing extracted human teeth and chemicals such as 2.6% sodium hypochlorite, 3% hydrogen peroxide, and boiling in water are not effective in disinfecting teeth. While performing spectroscopic studies, White et al<sup>[9]</sup> observed that autoclave does not lead to color changes in the teeth, but it increases the rate of light attraction by dentin. They also found that autoclaving induces some changes in the dentin mineral and organic material.

Some authors<sup>[20,21,25]</sup> investigated the effect of sterilization methods on the physical properties and perceived cutting characteristics of extracted teeth. Chandler<sup>[20]</sup> investigated the effect of autoclaving and found that it produced significant softening of bovine enamel. He has also reported that the changes in microhardness recorded in bovine enamel were similar to those produced by some experimental cariogenic substrates.<sup>[20]</sup>Soares et al<sup>[21]</sup> carried a Raman Spectroscopy analysis and showed that gamma irradiation caused no significant changes in enamel hardness. They also showed that the mineral and organic contents of dentin were more affected due to autoclaving compared while the teeth stored in thymol. Autoclaving has been recommended as the best method of sterilization for materials (such as natural teeth after extraction) exposed to body fluids by ADA and CDC.<sup>[26]</sup> Moreover, autoclaving extracted teeth with amalgam restorations may release mercury vapors in air through autoclave exhaust. It may also lead to residual mercury contamination of the autoclave. However, the physical properties of the teeth can be damaged of altered by the autoclave sterilization.<sup>[25,27]</sup> Berutti et  $al^{[12]}$  questioned autoclaving as an option for sterilization of extracted human teeth as the thermal cycling may cause

fracture of teeth with amalgam restorations due to differences in their coefficient of thermal expansion. Pantera and Schuster<sup>[4]</sup> showed that Rockal solution (benzalkonium chloride) for 24 h and 3 weeks and sodium chloride (5.25%) for 5 min failed to eliminate microorganisms in teeth. However, they found that autoclaving for 40 min at a pressure of 15 psi and a temperature of 121°C destroyed all bacterial species. Salem-Milani *et al*<sup>[28]</sup> showed that autoclaving is not an appropriate sterilization method in studies related to dentin microhardness. They found a significant reduction in microhardness of dentin after autoclaving, hence suggested a 2-week immersion in formalin for sterilization of extracted teeth.

Attam *et al*<sup>[10]</sup> performed a microleakage evaluation and showed that the chemical solutions such as sodium hypochlorite (2.6%), hydrogen peroxide (3%), and boiling water are unsuitable and ineffective for disinfection/sterilization of extracted human teeth. The teeth used for educational and research purposes should be disinfected/sterilized with sodium hypochlorite or liquid chemical germicides.<sup>[26]</sup> However, while using sodium hypochlorite as a disinfectant solution, the surface zone deproteinization of human enamel has been reported which may lead to the porosity of enamel.<sup>[29,30]</sup>

Recently, a safer alternative (Gigasept PA) to formalin has been advised for the sterilization of human teeth destined for the use in preclinical and clinical training.<sup>[31]</sup> Gigasept PA was the only disinfectant that sterilized 100% of the extracted human teeth samples. Gigasept PA should be considered a safer and effective alternative to formalin for the sterilization of extracted teeth destined for teaching purposes. Gigasept PA is a high-level hospital disinfectant used for medical instruments, not easily available. Inspite of the evidence demonstrated by numerous studies, students still use hydrogen peroxide, sodium hypochlorite, normal saline, etc., because of their availability in the dental teaching school.<sup>[32]</sup>

This undertaken investigation showed that there is a significant difference between the year education and total mean score for knowledge. Since the participated students are touched with extracted human teeth just at the second years of education, it is evident that the number of year in their education has a positive influence on their awareness and knowledge. Concerning the teaching of infection control practice, the two things such as cleaning with boiling water and sodium hypochlorite or hydrogen peroxide are touched only. These practices do not have any effect on students' awareness and knowledge. In the present investigation, the students had good awareness and knowledge in relation to sterilization/disinfection of extracted human teeth although it seems that more teaching, training, and education are needed. This research work showed that the students under investigation are well aware about sterilization of these teeth but less aware of using the necessary protective barriers such as masks, safety glasses, and gloves while practicing preclinical work on extracted teeth. Infection control measures to protect students and teaching faculty staff are not restricted to disinfection/sterilization

of extracted teeth. Along with sterilization of instrumentation, the use of other protective accessories such as gloves, eye protection, and masks should also be given prime considerations in the preclinical laboratory.<sup>[12]</sup> It has been shown that bacterial colonies grew on plates placed in the area of the dentists' nose and mouth while carrying dental procedures with an air-turbine hand-piece.[13] This investigation revealed that the students have a good level of knowledge about disinfection of extracted teeth although some of them did not favor to disinfect or sterilize these teeth. Hence, we conclude that the methods in which most students practice to sterilize extracted teeth are not effective practically, and more attention should be given to teach and train them an appropriate method to sterilize the teeth while they perform practical work in preclinical laboratory. Instructions regarding handling of extracted teeth need to be given to students when they enter BDS curriculum. It is always safe to follow the guidelines issued by the CDC while handling extracted teeth.[33]

Based on the results of the present investigation, educating dental students in the right method of handling (collection, storage, and disinfection) of extracted human teeth is very much important which must begin in the 1<sup>st</sup> year of BDS course. The students need to be acquainted with the latest published research data regarding this aspect. Since the use of extracted human teeth start from preclinical exercise, students need to practice extreme caution while handling them. Even after use, these tissues can be a source of infection and adequate precautions need to be followed not only while handling them but also to dispose them after use. It is recommended to follow the following guidelines issued by the CDC while handling extracted human teeth:<sup>[26]</sup>

- Extracted teeth used for teaching purpose in dentistry should be considered infective and classified as clinical specimens as they contain blood
- All persons who collect, transport, or manage extracted teeth should handle them with the similar precautions as a sample for biopsy
- Before extracted teeth are manipulated in dental educational training, the teeth first would be cleared of adherent patient material by scrubbing with detergent and water or using an ultrasonic cleaner
- Teeth should then be stored and immersed in a fresh solution of sodium hypochlorite (household bleach 1:10 with tap water) or any liquid chemical germicide for clinical specimen fixation
- Persons handling extracted teeth should wear gloves. Gloves should be disposed of properly, and hands washed after completion of work activities
- Additional personal protective equipment, for example, face shield or surgical mask and protective eye wear should be worn if mucous membrane makes contact with debris or spatter is expected when the specimen is handled, cleaned, or manipulated
- Work surfaces and instruments should be cleaned and decontaminated with a suitable liquid sterilizer after completion of work activities.

There seems future scope for the present study as that has been proved that autoclaving reduces the microhardness of enamel or dentin,<sup>[28]</sup> further study can be conducted on this aspect. Microhardness of dentin or enamel is believed to be dependent on the amount of mineral content in their composition, and its determination usually provides indirect evidence of mineral loss or gain in dental hard tissues. The exact mechanism of this reducing dentin microhardness by autoclaving needs to be found in future studies. CDC protocol is a useful guideline for sterilization of extracted teeth for educational or research purposes. However, its application should be limited to studies that investigate the mechanical or physical properties of dental substrates which are not influenced by sterilization procedures.

## Conclusion

The results of this investigation indicated that awareness and knowledge of undergraduate dental students about sterilization/disinfection methods of extracted human teeth were at a good level. However, a proper teaching, training, and education about the methods and materials suitable for sterilization/disinfection are still required for them. Along with instrument sterilization, the other safety measures such as the use of gloves, masks, and safety eye glasses should be stressed while practicing preclinical work on these teeth.

### Acknowledgments

We would like to acknowledge Dr. Chandrashekar KT, MDS, Principal, Hitkarini Dental College and Hospital, Jabalpur, Madhya Pradesh, India.

#### Financial support and sponsorship

Nil.

## **Conflicts of interest**

There are no conflicts of interest.

## References

- Dominici JT, Eleazer PD, Clark SJ, Staat RH, Scheetz JP. Disinfection/sterilization of extracted teeth for dental student use. J Dent Educ 2001;65:1278-0.
- Kumar M, Sequeira PS, Peter S, Bhat GK. Sterilisation of extracted human teeth for educational use. Indian J Med Microbiol 2005;23:256-8.
- 3. DeWald JP. The use of extracted teeth for *in vitro*bonding studies: A review of infection control considerations. Dent Mater 1997;13:74-81.
- 4. Pantera EA Jr., Schuster GS. Sterilization of extracted human teeth. J Dent Educ 1990;54:283-5.
- 5. Schulein TM. Infection control for extracted teeth in the teaching laboratory. J Dent Educ 1994;58:411-3.
- Shaffer SE, Barkmeier WW, Gwinnett AJ. Effect of disinfection/sterilization on *in-vitro* enamel bonding. J Dent Educ 1985;49:658-9.
- 7. Tate WH, White RR. Disinfection of human teeth for educational purposes. J Dent Educ 1991;55:583-5.

- 8. White RR, Hays GL. Failure of ethylene oxide to sterilize extracted human teeth. Dent Mater 1995;11:231-3.
- 9. White JM, Goodis HE, Marshall SJ, Marshall GW. Sterilization of teeth by gamma radiation. J Dent Res 1994;73:1560-7.
- Attam K, Talwar S, Yadav S, Miglani S. Comparative analysis of the effect of autoclaving and 10% formalin storage on extracted teeth: A microleakage evaluation. J Conserv Dent 2009;12:26-0.
- Kothari CR. Research Methodology-Methods and Techniques. 2<sup>nd</sup>ed. New Delhi: New Age International (P) Limited, Publishers; 2004.
- Berutti E, Marini R, Angeretti A. Penetration ability of different irrigants into dentinal tubules. J Endod 1997;23:725-7.
- Occupational Safety and Health Administration. 29 CFR 1910.1200. Hazard communication. Fed Regist 1994;59:17479.
- Hashemipour MA, Mozafarinia R, Mirzadeh A, Aramon M, Nassab SA. Knowledge, attitudes, and performance of dental students in relation to sterilization/disinfection methods of extracted human teeth. Dent Res J (Isfahan) 2013;10:482-8.
- 15. Humel MM, Oliveira MT, Cavalli VG. Effect of storage and disinfection methods of extracted bovine teeth on bond strength to dentin. Braz J Oral Sci 2007;6:1402-6.
- Watanabe I, Nakabayashi N. Measurement methods for adhesion to dentine: The current status in Japan. J Dent 1994;22:67-72.
- 17. Titley KC, Chernecky R, Rossouw PE, Kulkarni GV. The effect of various storage methods and media on shear-bond strengths of dental composite resin to bovine dentine. Arch Oral Biol 1998;43:305-11.
- George SW, Pichardo MR, Bergeron BE, Jeansonne BG. The effect of formalin storage on the apical microleakage of obturated canals. J Endod 2006;32:869-71.
- Robertson D, Leeb IJ, McKee M, Brewer E. A clearing technique for the study of root canal systems. J Endod 1980;6:421-4.
- 20. Chandler NP. Preparation of dental enamel for use in intraoral cariogenicity experiments. J Dent 1990;18:54-8.
- Soares LE, Brugnera Junior A, Zanin FA, Pacheco MT, Martin AA. Effects of treatment for manipulation of teeth and Er: YAG laser irradiation on dentin: A Raman spectroscopy analysis. Photomed Laser Surg 2007;25:50-7.
- 22. Pashley EL, Tao L, Pashley DH. Sterilization of human teeth: Its effect on permeability and bond strength. Am J Dent 1993;6:189-91.
- 23. Cuny E, Carpenter WM. Extracted teeth: Decontamination, disposal and use. J Calif Dent Assoc 1997;25:801-4.
- Pagniano RP, Scheid RC, Rosen S, Beck FM. Airborne microorganisms collected in a preclinical dental laboratory. J Dent Educ 1985;49:653-5.
- Parsell DE, Stewart BM, Barker JR, Nick TG, Karns L, Johnson RB. The effect of steam sterilization on the physical properties and perceived cutting characteristics of extracted teeth. J Dent Educ 1998;62:260-3.
- Kohn WG, Collins AS, Cleveland JL, Harte JA, Eklund KJ, Malvitz DM. Centers for Disease Control and Prevention (CDC). Guidelines for infection control in dental health-care settings-2003. MMWRRecomm Rep 2003;52:1-61.
- Toro MJ, Lukantsova LL, Williamson M, Hinesley R, Eckert GJ, Dunipace AJ. *In vitro* fluoride dose-response study of sterilized enamel lesions. Caries Res 2000;34:246-53.

- Salem-Milani A, Zand V, Asghari-Jafarabadi M, Zakeri-Milani P, Banifatemeh A. The effect of protocol for disinfection of extracted teeth recommended by center for disease control (CDC) on microhardness of enamel and dentin. J Clin Exp Dent 2015;7:e552-6.
- 29. Robinson C, Hallsworth AS, Shore RC, Kirkham J. Effect of surface zone deproteinisation on the access of mineral ions into subsurface carious lesions of human enamel. Caries Res 1990;24:226-0.
- Bitter NC. A scanning electron microscopy study of the effect of bleaching agents on enamel: A preliminary report. J Prosthet Dent 1992;67:852-5.
- Hope CK, Griffiths DA, Prior DM. Finding an alternative to formalin for sterilization of extracted teeth for teaching purposes. J Dent Educ 2013;77:68-71.
- 32. Amith HV, D'Cruz AM. Attitude and practice on handling of extracted teeth for educational use among the students of a dental college in Southern India. Int J Med Dent 2014;4:73-9.
- 33. Smitha D, Tijare MS, Amith HV, Gujjar KR, Sharma R. Knowledge, attitude and practice regarding handling of extracted human teeth among students of a dental college in Bhopal. J Indian Assoc Public Health Dent 2014;12:276-82.