Imaging and Radiation Research (2021) Volume 4 Issue 1
doi:10.24294/irr.v4i1.1730

ORIGINAL RESEARCH ARTICLE

Bacteria on surfaces contacted during intraoral radiographic examinations

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ABSTRACT

Objective: To determine the presence of bacteria by means of microbiological analysis on the surfaces contacted by the operator during the taking and processing of intraoral radiographs at different times of the day in the Oral Radiology Service of the UPCH. **Materials and methods:** Nine surfaces of the oral radiology service were sampled. The samples were taken at two times by the same investigator; at the beginning and the end of the activities in the service, the surfaces were swabbed with Trypticase Soy Broth (TSB). The samples were inoculated and incubated in three culture media (Plate Count Agar, Lamb's Blood Agar and Cetrimide Agar). Then the respective Colony Forming Unit (CFU) count was performed and Gram staining was also performed. **Results:** A high concentration of bacteria (4180 CFU/mL) and fungi was found in the oral radiology service. Gram-positive cocci were the most frequently found microorganisms and gram-negative bacilli were less frequently found. **Conclusions:** There is a high contamination of bacteria in the oral radiology service. When the activities are completed, the number of bacteria decreases, but the variety of bacteria increases.

Keywords: Infection Control; Disinfection; Dental Radiography; Microbiology

ARTICLE INFO

Received: 3 January 2021
Accepted: 5 February 2021
Available online: 12 February 2021

1. Introduction

Generally, radiographic procedures are not invasive, however there is always a risk of contamination due to the procedure being performed inside the oral cavity where microorganisms exist[1–3]. The application of means and measures for biosafety is applicable to all actions performed by the health professional, however in the application of radiographic techniques we can frequently observe that these biosafety principles are neither responsible nor correctly applied[1–4]. Although the American Dental Association published a biosafety guide in the practice of oral radiology[2,3], unfortunately there is no mechanism to supervise the application of these means and measures[3,6–10].

The objective of this research was to determine the number and types of bacteria according to Gram staining, existing on the surfaces that are contacted by the operator during intraoral radiographic examinations in the Oral and Maxillofacial Radiology Service of the Central Stomatology Clinic of the Universidad Peruana Cayetano Heredia on a day in November 2010.

2. Material and methods

A total of nine surfaces contacted by the operator during the taking of intraoral radiographs in the Oral and Maxillofacial Radiology Service of the Dental Clinic were evaluated (**Figures 1, 2** and **3**). The sur-

Figure 1. Radiographic plate dispenser and sample taking.

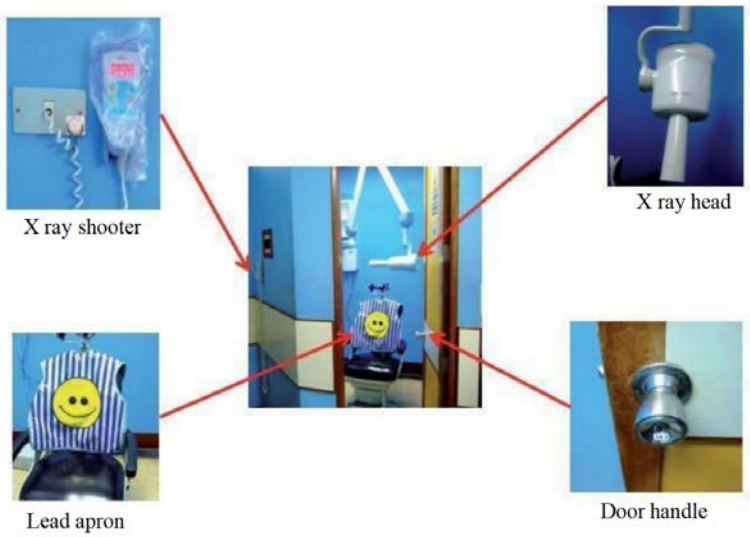


Figure 2. Selected contact surfaces in the intraoral X ray room.

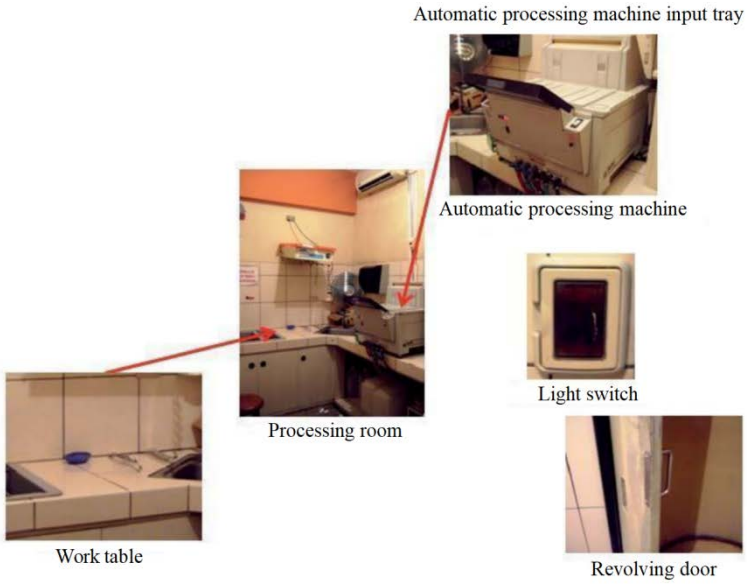
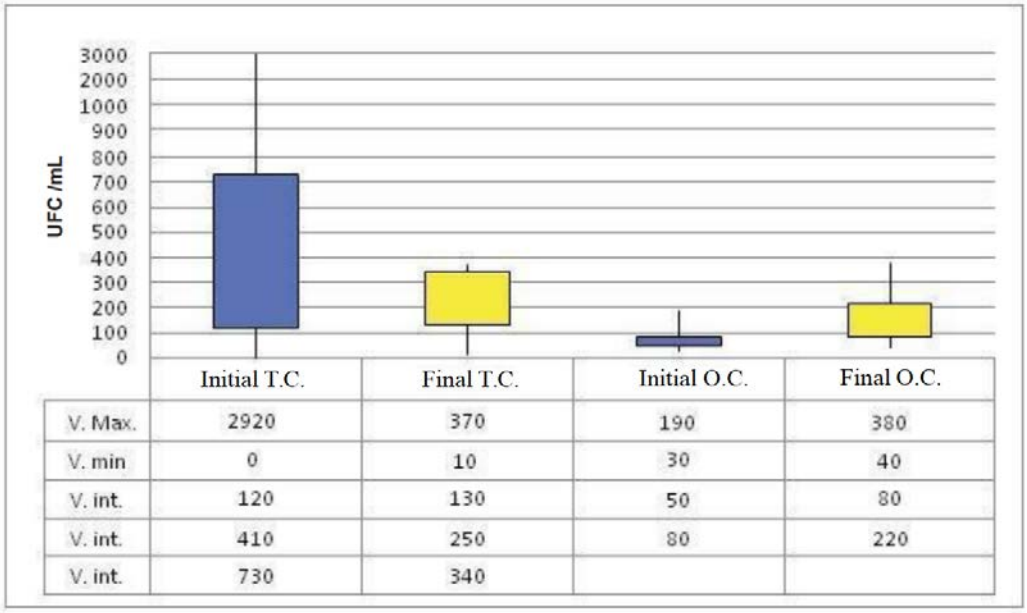


Figure 3. Contact surfaces in the darkroom.



	Initial T.C.	Final T.C.	Initial O.C.	Final O.C.
V. Max.	2920	370	190	380
V. min	0	10	30	40
V. int.	120	130	50	80
V. int.	410	250	80	220
V. int.	730	340		

Figure 4. Quantity of microorganisms (CFU/mL) found on contacted surfaces in the radiographic acquisition room and darkroom, at the beginning and end of the activities. Initial T.C.: Imaging room at the beginning of the activities, Final T.C.: Imaging room at the end of the activities, Initial O.C.: Dark room at the beginning of the activities, Final O.C.: Dark room at the end of the activities.

faces selected were: 5 surfaces of the intraoral radiographic acquisition room (new periapical radiographic plate wrapper, door knob of the acquisition room, X-ray trigger, X-ray head and external surface of the leaded apron) and 4 surfaces of the dark room (surface of the work table, handle of the dark room revolving door, surface of the X-ray inlet tray in the developing machine and light switch).

The following culture media and broth were prepared before starting the activities: 54 Agar Plate Count (PC), 18 Lamb's Blood Agar (AS), 18 Agar Cetri-Measure, (AC) and 54 tubes with 1 ml of Triptic Soy Broth (TSB). The swabs were wrapped in kraft paper and then dry heat sterilized at a temperature of 170 °C for 60 minutes.

For sample collection, the swab wrapper was opened, the tube containing the broth was uncovered, the swab was introduced into the tube with the broth and then the surfaces contacted by the operator during the taking and processing of intraoral radiographs in the oral radiology service environments were rubbed, then the swab was introduced into the same test tube and then closed with a rubber or cotton cap.

This procedure was performed at the following times: the first swabbing of surfaces was performed at the beginning (8 hours) before starting the service activities and the second sampling was performed at the end (18 hours) of the service activities, after the last radiographic sampling in the same sampling room selected for the study and on the same surfaces. The samples taken were taken to the microbiology laboratory of the Faculty of Science of the UPCH and the samples were inoculated using the following methods: the method of poured plate on plate count agar, they were carried out immediately. Dilutions of 10^{-1} and 10^{-2}, and the streak-plate method (Streak-Plate Technique) on blood agar and cetrimide agar were performed. All culture media were incubated at 37 °C temperature for 24 hours. Only the blood agar medium was incubated under microaerophilic conditions.

After the time elapsed, macroscopic observations were made for the respective count to know the number of microorganisms obtained by swabbing the surface. Then colonies with different morphologies were selected, removed with a seeding loop and Gram staining was performed to determine the morphologies and bacterial types (cocci and bacilli).

3. Results

In Agar PC, he found a large amount of CFU with a white-yellow color at the beginning of the

activities, the number of colonies varied between 0 to 2,920 CFU/mL and the areas of greater contamination in the intraoral radiography room obtained a sub-total of 4,180 CFU/mL and the area of less contamination in the processing room obtained a sub-total of 350 CFU/mL. At the end of the activities, it found less quantities of CFU, which varied between 10 to 390 CFU/mL, likewise the highest concentration was found in the intraoral radiography room with a sub-total of 1,100 CFU/mL and in the dark room a sub-total of 720 CFU/mL was found (**Table 1**, **Figures 1** and **2**).

Then, upon microscopic observation, the samples taken at the beginning of the activities showed gram-positive cocci, gram-negative cocci, gram-positive bacilli and fungi, and at the end of the activities, other microorganisms such as gram-negative bacilli were also found (**Tables 2** and **3**).

In the blood agar, at the beginning of the activities, microscopically Gram-positive cocci were found with α and γ hemolysis, Gram-negative cocci with γ hemolysis, Gram-positive bacilli with α and γ hemolysis, Gram-negative bacilli with α hemolysis. At the end of the activities, the same types of bacteria were found as at the beginning, additionally Gram-positive cocci with α, β and γ hemolysis, Gram-negative cocci with γ hemolysis, Gram-positive bacilli with γ hemolysis, Gram-negative bacilli with γ hemolysis and fungi (**Table 4**). No bacterial growth was observed macroscopically on cetrimide agar.

Table 1. Microorganisms (CFU/ml) found on surfaces contacted in intraoral radiographic recordings at the beginning and end of the activities

Area	Surfaces	Start CFU/mL	End CFU/mL
Intraoral radiographic acquisition room	Wrapping of radiographic plate	0	10
	Door handle	410	250
	Switch or trigger	2,920	340
	X-ray tube or head	730	370
	Leaded apron	120	130
Sub-total		4,180	1,100
Darkroom	Work table	80	380
	Revolving door handle	50	80
	Inbox transfer machine	190	220
	Light switch	30	40
Sub-total		350	720
Total		**4,530**	**1,820**

Table 2. Microorganisms according to morphology in Gram staining found on surfaces contacted in intraoral radiographic recordings at the beginning and end of the activities

Area	Surfaces	Start		End	
		Morphology	Gram	Morphology	Gram
Tapping room of radiographs intraoral	Wrapping radiographic plates	------	------	Coconuts in Chains	+
		Coconuts in chains	+	Coconuts in short chains/Bunches of grapes	+
	Door knob	Diploid/Tetra Cocos	+	Coconuts in chains	+
		------		Mushrooms	------
		Coconuts in short chains	+	Pleomorphic curved bacilli	+
	X-ray trigger	Diplococcus	+	Curved Bacilli	-
		Other	------	-----	
		Tetra coconuts	+	Clustered coconuts	+
	X-ray head	Coconuts in chains	-	Curved bacilli	-
		------	------	Straight bacilli	-
		Diplococcus	-	Clustered coconuts	+
	Outer surface	Coconuts in short chains	+	Diplococcus	-
	Leaded apron	Straight bacillus	+	Straight bacillus	-
Darkroom	Work table	Diplococcus	+	Clustered coconuts	+
		Tetra coconuts	+	------	
	Revolving door handle	Coconuts in chains	+	Grouped coconuts	+
		Mushroom	------	Curved bacillus	+
	In-tray developing machine	Tetra coconuts	+	Diplococcus	+
		Mushrooms	------	Coconuts in chains	+
		------	------	Mushrooms	------
	Light switch	Diplococcus	+	Clustered Coconuts	+
		Mushrooms	------	Diplococci in chains	-

Table 3. Types of microorganisms by surface contacted in intraoral radiographic exposures at the beginning and end of the activities

Contact surfaces	Coconut gram +		Coconut gram -		Bacillus gram +		Bacillus gram -		Others	
	I	F	I	F	I	F	I	F	I	F
1	(-)	X	(-)	X	(-)	(-)	(-)	(-)	(-)	(-)
2	X	X	(-)	(-)	(-)	X	(-)	(-)	(-)	X
3	X	X	(-)	(-)	(-)	(-)	(-)	X	X	(-)
4	X	X	X	(-)	(-)	(-)	(-)	X	(-)	(-)
5	X	X	X	X	X	(-)	(-)	X	(-)	(-)
6	X	X	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
7	X	X	(-)	(-)	(-)	X	(-)	(-)	X	(-)
8	X	X	(-)	(-)	(-)	(-)	(-)	(-)	X	X
9	X	X	(-)	X	(-)	(-)	(-)	(-)	X	(-)

I: At the beginning of the activities **F**: At the end of the activities. 1. Wrapping of periapical radiographic plates before taking the radiograph; 2. Door knob of intraoral X-ray taking room; 3. Trigger of intraoral X-ray taking room; 4. X-ray head of intraoral X-ray taking room; 5. External surface of leaded apron; 6. Surface of darkroom working table; 7, Darkroom revolving door handle, 8. X-ray inbox surface on darkroom developing machine, 9. Darkroom light switch.

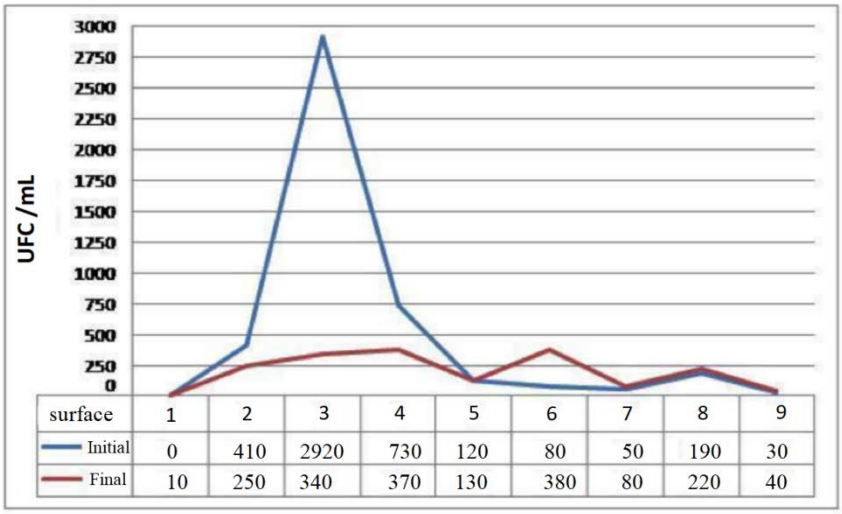


Figure 5. Quantity of microorganisms (CFU/mL) by surfaces contacted in intraoral radiographs at the beginning and end of the activities. 1. Wrap the periapical radiographic plate before taking X-rays; 2. Door knob of intraoral X-ray taking room; 3. Trigger of intraoral X-ray taking room; 4. X-ray head of intraoral X-ray taking room; 5. Outer surface of leaded apron; 6. Surface of darkroom working table; 7. Darkroom revolving door handle; 8. X-ray inbox surface on darkroom developing machine; 9. Darkroom light switch.

4. Discussion

There are few published studies on infection control carried out in the oral radiology service and those that exist do not detail the infection control measures of the services, nor the means used[3,11,12]. When reviewing studies which analyze the effectiveness of the implementation of infection control protocols in the oral radiology service, we can observe the deficiency or failure in the cleaning procedures in the service, since the results show a decrease in the amount of bacteria found at the end, almost half of what was found at the beginning (**Figure 1**); however, the counts are still significant, with a high degree of contamination[3].

In the present study, a value of 6,350 CFU/mL was obtained at 2 points in time as the total cumulative concentration of the study. The concentrations of bacteria at the beginning of the activities of the oral radiology service, obtaining 4,530 CFU/mL and at the finalization of the activities with concentrations of 1,820 CFU/ml (**Table 1** and **Figures 1** and **2**), this decrease could be due to 2 possibilities: a) the bacteria present in the superficies were removed during the procedures of taking radiographs, either taken to the patient's mouth; b) due to the time of the first sample collection at 9:00 am and to the dental assistant, technical personnel or others of the service, who were present and observed the sample collection and it is very likely that they performed better cleaning of the service. This gives us

an explanation why it decreased only in the 3 surfaces (door knob or plate, X-ray head and X-ray trigger), while the rest of the surfaces of the study increased the amount of bacteria at the end of the activities. It also shows that the way of disinfection of our environment does work, but it is still deficient compared to other studies. In many other studies, as in the present one, the environments of oral radiology services present a high degree of contamination[3,11,12]. Silva *et al*. evaluated 7 surfaces during 10 non-consecutive and random days in the oral radiology clinic of the University of the State of Sao Paulo, obtaining a cumulative count of 7,546 CFU/plate in the 48-hour samples, where the highest value was on the third day (1,767 CFU/plate), and the lowest value was on the eighth day (242 CFU/plate). Only on the fourth day of radiography did it obtain a cumulative value of 2,706 CFU/plate[11].

In general, when analyzing the concentrations of bacteria in the radiography room and the dark room, we found that the intraoral radiography room had higher concentrations of microorganisms, especially at the beginning of the activities with 4,180 CFU/mL and the dark room at the beginning of the activities had a lower concentration of microorganisms with a value of 350 CFU/mL. The bacteria present on the surfaces were transferred to the darkroom together with the glove, which would explain why the amount of bacteria in the darkroom increased at the end of the activities.

Silva *et al*. demonstrated the effectiveness of chlorhexidine alcohol solution (70° alcohol with 5% chlorhexidine) as a surface disinfectant applied on the contact surfaces (radiographic plate, the tube head, the dental chair, the leaded apron, the buttons, the protective barrier or leaded apron and the darkroom worktable) in the oral radiology service, reducing the counts of microorganisms from 7,545 to 1,234 CFU/plate[11]. The cumulative concentrations obtained on the surfaces of the darkroom at the beginning of the activities were 350 CFU/mL (**Table 1**), according to the study of Silva *et al*. the samples obtained on the darkroom worktable gave a cumulative count of 710 CFU/plate[11].

When determining and identifying the types of bacteria in this study, obtained from the samples of the surfaces of the X-ray room and dark room, the microorganisms found were: gram-positive cocci, gram-negative cocci, gram-positive bacilli, gram-negative bacilli, fungus and others; the bacteria most frequently found on the surfaces were gram-positive cocci with aspects of grape clusters with different types of hemolytic properties and the least frequent were gram-negative bacilli, however the figures at the beginning of the activities were slightly higher than at the end of the service activities (**Figures 1** and **2**, **Tables 2**, **3** and **4**). Arredondo *et al*. analyzed 4 surfaces (digital shutter, X-ray tube, the chair and the developing machine) at the University of Chile and observed that the most frequently found bacteria were gram-negative bacilli and gram-positive cocci were less frequently found[12]. According to recent investigations where means and infection control measures have been applied, it was shown that the presence of bacteria decreased in great quantity or the counts were almost null after disinfection of the X-ray and dark room areas of the oral radiology service[8,11,12].

No bacterial growth was found on cetrimide agar, since this agar is exclusively for *Pseudomonas spp.* growth[3]. This procedure was chosen in cetrimide agar due to the antecedents of other studies and publications. Therefore, the results do not mean that the presence of this species is ruled out, but simply that growth was not observed in these tests, suggesting to perform them in another agar medium to obtain better results.

The present study shows high values of Colony Forming Unit counts and a great variety of types of microorganisms on the different surfaces of the oral radiology service, thus showing a high risk of infection and contamination compared to studies carried out in Brazil and Chile[3,11,12]. This suggests that a project should be carried out to identify each species of microorganisms and thus be able to implement and develop the use of adequate means and measures for the disinfection of each of the contact surfaces when taking and processing radiographs in the service.

Table 4. Microorganisms found in the culture of samples of surfaces contacted in intraoral radiographic exposures at the beginning and end of the activities

Area	Surfaces	Start		End	
		Morphology	Gram (hemolysis)	Morphology	Gram (hemolysis)
X-ray examination room	Radiographic plate envelope	Diplococcus	+(γ)	Coconuts in chains	- (γ)
	Door knob	Large bacillus	-(α)	------	------
		Pleomorphic coccus	+ (γ)		
	X-ray trigger	Diplococcus	+ (α)	Diplococcus	+ (γ)
		Diplococcus	- (γ)		
	X-ray head	Diplococcus	+ (γ)	Diploid/Tetra coccus	+ (γ)
		Macrobacillus	+ (γ)		
		Macrobacillus	+ (α)		
	Outer surface of plumb line	Pleomorphic bacillus	+ (γ)	----	----
		Macrobacterium pleomorpha	+ (α)		
		Diploid/tetracoccus	+ (γ)		
Darkroom	Work table	----	----	Diplococcus	+ (β)
				Bacillus in couple	+ (γ)
	Revolving door handle	----	----	Curved Bacilli	- (γ)
				Diploid/Tetra coconuts	+ (γ)
	Processing machine inlet tray	----	----	Coconuts in chains	- (γ)
	Light switch	----	----	Diplococcus	+ (α)
				Mushrooms	

5. Conclusions

In the area of the intraoral X-ray room, there was a higher concentration of bacteria at the beginning of the activities (4,180 CFU/mL) than at the end of the activities (1,100 CFU/mL). Both at the beginning and at the end of the activities there was greater contamination by gram-positive cocci, especially in the X-ray trigger. At the end of the activities there was an increase in the types of bacteria than at the start of the activities. There was a significant decrease in the values of bacterial concentrations when comparing the samples at the beginning and at the end (4,530 CFU/mL and 1,620 CFU/mL) of the activities of the service.

Acknowledgements

Thanks to MSc. Dora Maurtua Torres for her collaboration and support in the microbiology laboratory, Department of Microbiology Faculty of Sciences and Philosophy—UPCH.

Conflict of interest

The authors declare that they have no conflict of interest.

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