

Bacterial Contamination of Dental Unit Water Lines (DUWL) in Baghdad City

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Abstract: This study was designed to detect the microbial contamination in dental unit water line DUWL of several dental clinic in Baghdad city . Fifteen dental clinic were surveyed in current study; water samples were collected from the air/water syringe, high speed handpiece and washing water from each dental clinic. The samples were examined for bacterial contamination and colony forming unit (CFU) for each water sources in dental chair (air/water syringe, high speed handpiece and washing water) were determined. Bacterial flora were isolated with the filtration method. Recovery rate of bacterial growth was 40.4% (20 sample of 45). The average of concentration of total bacteria was 90 cfu/ 100ml (high speed hand pieces), 60 cfu/ 100ml (air –water syringe and 50 cfu ml (washing water) . Bacteria were identified with gram staining followed by the biochemical methods , further confirmation was done by using API 20E (bioMérieux, France) . The following bacteria were identified: *Pseudomonas aeruginosa*, *Aeromonas hydrophila*, *Klebsiella* spp., *Klebsiella pneumoniae*, *Staphylococcus* spp., *Staphylococcus aureus*, *Escherichia* spp., *Escherichia coli*, *Bacillus* spp., *Enterobacter* spp., *Streptococcus* spp. The prevailing bacteria were *Pseudomonas aeruginosa* (21%); while *Aeromonas hydrophila* (19.2%) were the next most frequently occurring bacteria. The quantitative bacterial analysis of water samples collected from the dental clinic for handpiece and air/water syringe indicated of some the dental clinic under study delivered water that could meet the accepted standard of American Dental Association (ADA) dental unit water quality (<200 CFU) but other clinic gave bacterial concentration reached excessive values, the bacterial flora were composed of the bacteria characteristic for water distribution systems, opportunistic pathogens and oral flora.

Keywords: Dental Unit Water Lines , Biofilm , Bacterial Contamination, water syringe

I. Introduction

Dental unit waterlines

In Dental unit, the system of unit waterlines (DUWL) is made of thin, plastic tubes delivering water to the different hand pieces. In the case of an opened water system, the source of water is a municipal water supply and in a closed water system – water from a reservoir belonging to a unit. That lead to concern attention on potential occupational hazards in the dental office take into account increasing numbers of dental patients considered as immunocompromised persons for example: elderly people, smokers, people with alcoholism, organ transplant and blood transfusion recipients, AIDS and cancer patients, people with diabetes, people with autoimmune disease and people with chronic organic disorders [1]. Dental unit waterlines (DUWL) are environment for the growth of biofilms of aerobic, mesophilic, and heterotrophic microorganisms [2]. High counts of bacteria in this system of pipeline are well documented and have been described in many studies in the reports of potential opportunistic pathogens such as *Streptococcus* spp., *Enterococci* spp., *Pseudomonas aeruginosa*, *Legionella* spp., and other gram-negative rods [3,4,5] , that caused pneumonia, other respiratory infections, or wound infections in immunocompromised people. Workers in Dental clinic have been shown to have altered nasal flora, with colonization of *Pseudomonas* spp. similar to those found in their dental units [6,7] The microorganisms capable of growing and forming biofilms on surfaces of DUWLs may also growth and form biofilms on heart valves, causing endocarditis. [8]

The assessment of quality of dental unit water is more important since patients and dental staff are regularly exposed to water and aerosols generated from the dental unit, therefore the aim of the study was to assess microbial contamination of water in dental unit waterlines of dental clinics in Baghdad city .

II. Material and methods

Samples collection

Forty five water samples were taken from 15 dental units (from air/water syringe and high speed handpieces and mouth washing water) at Baghdad city as illustrated in table (1), 100 ml of water samples were collected aseptically in sterile containers, 15 water samples were taken from the tap water as control group. The samples were carried out to the laboratory in special aseptic cool box and analyzed within 24h.[9,10].

Table 1 : Distribution of water samples were taken from dental chair.

Samples types	NO.
air/water syringe	15
High speed handpiece	15
Mouth washing water	15
Tap water (control)	15
total	60

Filtration and culturing of water samples

Water were tested for the presence of heterotrophic bacteria by filtering 100ml volume through 0.45µm nitrocellulose filter (Sartorius) and incubated on R2 agar 37 °C for 5 day, and the number of colony forming units (CFU) was determined.

Isolation and Identification of colonies

Bacterial isolates were differentiated based on the morphological characteristics of colonies (diameter, color, surface, pigmentation, edges and elevation), as well as the Gram stain of bacterial cells. For further confirmation, API-test kits (BioMérieux, France) were used.

III. Results and Discussion

Isolation of bacterial isolates

Forty five samples were collected from 15 dental clinic (from handpiece, air/water syringe and mouth washing water) also 15 tap water as control. Recovery rate of bacterial isolates was 40.4% (20 samples) with the average of CFU/ml was illustrated in table 2.

Table 2: Average of colony forming units(CFU's) of microorganisms obtained from handpiece, air/water syringe, mouth washing water and control (tap water)

Water samples type	CFU/ 100ml
High speed handpiece	90
air/water syringe	60
Washing mouth water	50
control (tap water) .	6



Figure 1: Bacterial isolates on filter paper

Identification of bacterial isolates

The bacterial isolates were identified by gram staining followed by the biochemical methods and were confirmed by API system ,results revealed that a total of 11 different bacterial species obtained from 52 bacterial isolates ,thes species were listed in table 3 .

Table 3: Microorganisms obtained from handpiece and air/water syringe mouth washing water

Bacterial genus or species	NO. isolats (%)
<i>Pseudomonas aeruginosa</i>	11(21%)
<i>Aeromonas. hydrophila</i>	10(19.2%)
<i>Klebsiella spp.</i>	6(11.5%)
<i>Klebsiella pneumoniae</i>	4(7.7%)
<i>Staphylococcua spp.</i>	4(7.7%)
<i>Staphylococcua. aureus</i>	4(7.7%)
<i>Escherichia spp.</i>	3(5.8%)
<i>Escherichia coli</i>	3(5.8%)
<i>Bacillus spp.</i>	3(5.8%)
<i>Enterobacter spp.</i>	2(3.9%)
<i>Streptococcus spp.</i>	2(3.9%)
<i>Total</i>	52(100%)

The results of recent study came in agreement with James *et al.* 2015 [9]. In recent study, *P. aeruginosa* were found to be the prevailing bacteria in the samples obtained from dental clinic . This is in concurrence with a study conducted by Al Hiyasat *et al.* 2007 [11] which showed that *P. aeruginosa* thrive commonly in Dental unit waterlines. A study by Barbeau *et al* reported that *P. aeruginosa* was isolated from 24% of the tested waterlines [12] . In another study conducted by Stampi *et al.* ,there was a notable growth of *P. aeruginosa* was observed in the dental unit waters.

P. aeruginosa is an opportunistic, nosocomial pathogen which affects immunocompromised patients more commonly. *These results may due* to the complex design of the equipment cause in stagnation of water within DUWL and subsequent amplification of contaminating microorganisms,also bacteria and viruses may be aspirated from the oral cavity and cause cacontamination of the handpiece.

The results of recent study show that samples taken from the hand piece showed the maximum CFU's of microorganisms and then water –air syringe , followed by mouth washing water that came in accordance with James *et al.* that may due to the nature of the hand piece and other part of DUWLs that they will develop a biofilm, and water flowing down the biofilm-coated waterlines will contribute to microbial load in the water as it exits the tubing. Frequent periods of water stagnation in DUWLs (related to the rhythm of work during the day, in the evenings, during the nights, weekends and holidays) and the properties of the plastics used in DUWLs construction can promote the attachment and colonization of biofilm-forming microorganisms. Most plastic dental tubing has an inside diameter of 16 mm (1/16 inch) to 8 mm (1/8 inch). This creates a very large ratio of surface area to water volume of narrow bore tubing. [13, 14].

The physics of laminar flow of water passing through the DUWLs results in maximum flow at the centre of the lumen and minimal flow at the periphery, encouraging deposition of organisms onto the surface of the tubing thus promoting further undisturbed bacterial proliferation. In addition, bacteria adhere more readily to hydrophilic polymeric plastic tubing (polyvinyl chloride, polyurethane) than to tubing composed of glass or steel, [15]. The source of bacteria for biofilm in DUWLs may be municipal water piped into the dental unit and, and suck-back of patient saliva into the line due to lack of anti-retraction valves . Bacterial contamination of DUWLs is thought to follow development of biofilms on their inner surface. Frequently, water entering DUWLs is of good microbiological quality, but after shedding of bacteria from the biofilm, it becomes contaminated over the acceptable level [16,17] , biofilm can constantly release bacteria [18] . Aerosols and droplets produced by dental instruments connected to dental unit waterlines (DUWLs) during dental care may contain microorganisms that can be opportunistic pathogens for patients and dentists. Microbial proliferation inside DUWLs is inevitable and is principally associated with biofilm formation. It represents a low but current risk of infection . This becomes quite significant when immunocompromised patients (the elderly, smokers, HIV+ or cancer patients, people with diabetes, alcoholism, etc.) are treated. [19] The dental unit water can also be heavily contaminated with opportunistic pathogens that can pose a major risk for the dental team and the patients [20] The microorganisms included in the dental clinic are usually found in the public water systems and hospital/clinical environment. These microorganisms cause sepsis, pneumonia, periodontitis, oropharyngeal infection and other nosocomial diseases .[21]

Conclusion

The quantitative bacterial examination of water samples collected from the dental clinic for handpiece, air/water syringe and mouth washing water indicated that bacterial concentration in some dental clinic reached excessive values, the bacterial flora were composed of the bacteria characteristic for water distribution systems, opportunistic pathogens and oral flora.

Since the phenomenon of dental unit waterlines contamination must taken more attention.

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*Sanaa R. Oleiwi. "Bacterial Contamination of Dental Unit Water Lines (DUWL) in Baghdad City." IOSR Journal of Dental and Medical Sciences (IOSR-JDMS) 16.8 (2017): 47-50.