

Are our dental units safe: Is faecal contamination a possibility? Dental Unit Waterline Contamination

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ABSTRACT:

Background: Aim of the study was to assess bacterial contamination of water and biofilm samples of dental unit waterlines that used no decontamination measures.

METHODS: Water and biofilm samples were collected from 27 dental unit water lines using borewell water and 20 using distilled water. Samples were cultured and number of colony forming units counted. The presence/absence of *Escherichia coli* was assessed.

RESULTS: Important findings were (a) Dental units using distilled water were also contaminated, though non significantly. (b) Colony forming units were significantly higher in dental unit waterlines using borewell water (c) the proportion of water samples positive for *E coli* was non-significantly higher in the dental unit waterlines using borewell water. A Substantial proportion of water samples from both dental unit waterlines had colony forming units above the American Dental Association standards with a higher proportion in bore well water violating these standards.

CONCLUSION: Dental units which do not use decontamination measures show substantial microbial contamination with possibility of even contamination with *Escherichia coli*. Contrary to general perception that dental units using distilled water are safe, fecal contamination is a possibility in these too. Contamination is more in units using borewell water than in those using distilled water.

Key words: dental unit waterlines; colony forming units; *E.coli*; bore well water; distilled water.

INTRODUCTION

Contamination of Dental Unit Water Lines (DUWLs) and its consequent risk to patients has been controversial for a long time now. The source of water used in the dental units may vary in different settings ranging from municipal water to sterile water to storage tank water. The water then passes through narrow bore dental tubings. Long extended periods of stagnation of water within these tubings are responsible for formation of biofilms on the inner wall of these tubings¹ which in turn may contribute to further contamination of water in the dental unit waterlines. Contribution of the water used in the dental units to contamination has not been well investigated. Coliform bacteria are considered to be the principal indicators of bacteriological quality of water and are also the commonest organisms responsible for water borne diseases in India.

The aim of the present study was to assess and compare the bacterial contamination in input water (bore well water and distilled water before they were run through the DUWLs), output water (after it was run through the DUWLs) and biofilm samples of dental unit waterlines using bore well water and those using distilled water. Presence/absence of E.coli was also assessed in the same.

MATERIAL AND METHODS

20ml of output water samples from 27 DUWLs using bore well water (BW) and 20 DUWLs using distilled water (DW) were collected from a hospital setting. None of the dental units used measures of decontamination. The samples were collected in sterile disposable containers and immediately filtered using 5mm thick, 0.45 micron porous cellulose nitrate membrane. Using the pour plate method, the filtrate was cultured on nutrient agar. The culture plates were incubated for 48hrs at 37°C. After 48hrs, the culture plates were sent for bacterial evaluation. Colony forming units (CFUs) were counted from nutrient rich agar plates using manual colony counting device. Smear was made from the colonies grown on nutrient rich agar plates and gram's staining was done and the type of organisms was observed. 20 ml of input water (bore well water and distilled water) samples were also collected and evaluated as above.

Biofilm samples were collected aseptically from a 2 cm long fragment of the tubing from the booster end of dental unit. The tubings were cut longitudinally and the samples were collected from the internal walls using a sterile cotton swab. Immediately the collected samples were dipped in 2ml of saline in a sterile test tube (for 2 mins) and plugged with cotton to prevent contamination. The samples were then filtered and cultured as above. The number of CFUs was counted using manual colony counting device. M- Endo agar was used specifically for culture of E.coli from water and biofilm samples.

The difference in the number of CFUs was analysed using independent sample t-test after log-transformation. The presence/absence of E coli was compared using chi-square test.

The number of CFUs in the DUWLs was then compared with the ADA recommended standard for dental unit output water of a microbial load of less than 200 CFU/ml.

RESULTS

The mean (SD) log CFUs of output water and

biofilm samples was significantly more in DUWLs using bore well water [5.17 (0.53)] (Fig 1) and [5.37 (0.83)] (Fig 2) respectively than in DUWLs using distilled water [4.71 (0.91); p=0.036] (Fig 3) and [4.94 (0.83); p=0.047] (Fig 4) respectively (Table 1&2; Graph 1&2). Gram negative organisms were isolated most commonly. While the distilled water used as input water was sterile (Fig 5)(0 CFUs), the bore well input water (Fig 6) had on an average 78CFUs. No E.coli was seen in input water (Fig 7&8). With respect to the output water, there was no significant difference in the proportion of DUWLs which were positive for E. coli (40.7% in those using Bore well water (Fig 9) vs. 25% in Distilled water (Fig 10); p=0.26) (Table 3& Graph 3). Same for true for E.coli in the biofilm samples (37% in those using Bore well water (Fig 9) vs. 20% in Distilled water (Fig 10); p=0.28) (Table 4& Graph 4)

The number of DUWLs using BW and DW that maintained the ADA standards for Dental Unit Output Water Quality was 17/27 and 15/20 respectively (Table 5& Graph 5). No statistical significance was observed.

DISCUSSION

The microbial contamination in dental unit waterlines was first described by Blake in 1963². The contaminated water may be ingested by the patients, it may contact open wounds or may be aerosolized and inhaled by the patient or dental staff. The use of instruments such as an ultrasonic scaler could potentially force the microorganisms into the gingiva raising the possibility of introducing microorganisms into the bloodstream.³ Microbial contamination of DUWLs may originate mainly from the water supplying the dental chair units (DCUs) which itself may contain low levels of microorganisms. Studies have compared microbial contamination in DCUs using tap water and those using distilled water, but such a comparison has been done in DCUs using decontamination measures⁴. Very few studies have compared microbial contamination in DCU's using different kinds of water especially in units which did not use any measures of decontamination⁵. Hence in this study we compared the bacterial contamination in DUWLs using bore well water and those using distilled water which did not use any decontamination measures.

Important findings of this study were (a) the CFUs were significantly higher in output water and biofilms of DUWLs using bore well water and (b) the proportion of output water samples positive for

E. coli was non-significantly higher in the DUWLs using the BW. A substantial proportion of output water samples from both DUWLs had CFUs above the ADA standards with a higher proportion of DUWLs using the bore well water violating these standards.

These results raise concerns about the safety of using BW in the DUWLs. Though the level of contamination in DUWLs using DW was less, it was not significantly lower than in those using BW in the proportion of samples found positive for *E. coli*. This lack of statistical significance could be a type-2 error owing to small sample size. However, it was surprising that the level of contamination even in DUWLs using distilled water was considerable.

This suggests that the input water used in the DUWLs itself may have been contaminated. We assessed the bacterial contamination of BW and DW before they were run through the DUWLs. Both types of input water maintained the drinking water standard and no contamination with *E. coli* were seen in either of them. This is in concordance with findings in others studies where no difference was seen in the microbial contamination in dental unit water systems regardless of whether these systems were main, bottle, or header tank fed or whether the water used was hard, soft, deionized, or distilled⁶. Dental unit waterlines supplied by sterile water also may become colonised¹. Thus the use of water with an initial lower contamination level did not inhibit the increase in contamination in output water/patients mouths or in the environment. This could be attributed to the biofilms formed on the inner walls of dental tubing which could contribute to the microbial contamination of the output water. In our study too, the phenomenon of biofilms contaminating the water may explain the contamination seen in the output water, at least partially. When the biofilms were examined, the number of CFUs in biofilms of DUWLs using bore well water was more than in the biofilms of DUWLs using distilled water. Contamination in biofilms depends on several factors, including (1) the number of patients treated (2) oral status of these patients, (3) the life of the tubings and (4) measures taken towards decontamination. None of the dental units used decontamination measures routinely; we have not assessed the other factors in this study. Hence we cannot rule out the possibility of differences in these factors causing the observed differences in the level of contamination.

Hence, besides using independent reservoirs, it is necessary to develop methods to reduce or

eliminate biofilm coating the internal walls of the DUWLs.⁷

As there was microbial contamination in output water and also in the biofilms of the DUWLs irrespective of whether they were using bore well water or distilled water, the number of CFUs in the DUWLs was compared with the ADA recommended standard for dental unit output water of a microbial load of less than 200 CFU/ml. There were about 15 chairs that did not meet the ADA standard of output water but no statistically significant difference was seen in the number of DUWLs using BW as compared to those using DW.

CONCLUSION

Contamination of dental unit waterlines is considerable with faecal contamination a definite possibility. Though the input water was sterile or had low levels of contamination, there was considerable contamination in the output water implicating biofilms to have some role in contamination of dental unit waterlines. Substantial contamination found in both DUWLs in our study raises concerns about safety of these units. Irrespective of the kind of input water used, decontamination measures targeting water and biofilms need to be used to reduce contamination in dental unit waterlines. There is an urgent need to examine the effect of such measures in reducing the level of contamination of DUWLs.

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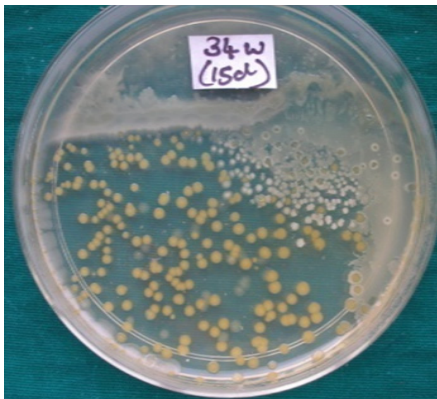


Fig 1: Output bore well water sample: Small cream and yellow, circular, smooth, opaque and opaque colonies

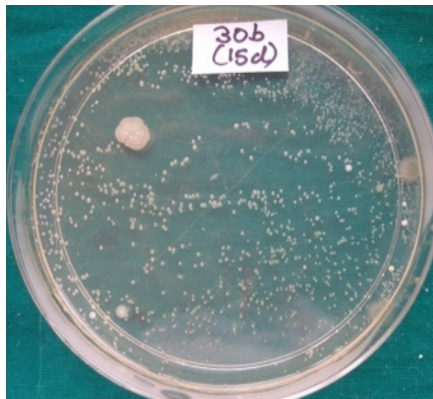


Fig 2: Biofilm samples from tubings of DUWLs using bore well water: Pinpoint, cream, circular and opaque colonies seen

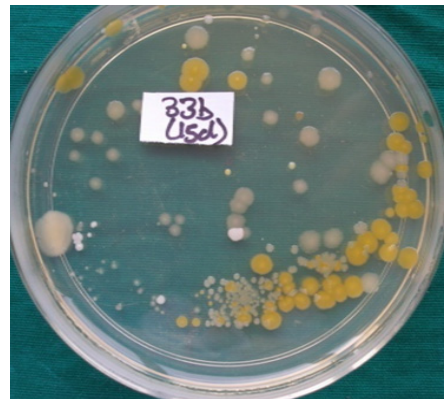


Fig 3: Output distilled water sample: Medium, cream and yellow, circular colonies seen

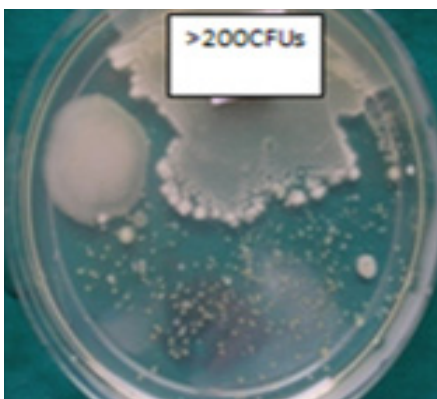


Fig 4: Biofilm samples from tubings of DUWLs using distilled water: Pinpoint cream and yellow colonies seen

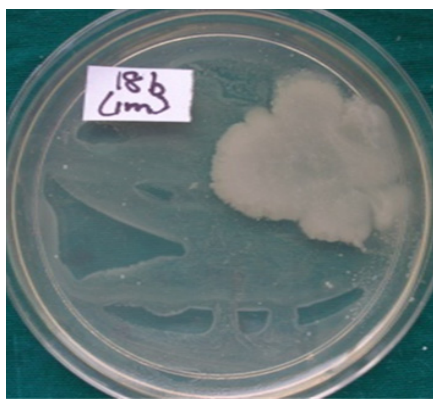


Fig 5: Input distilled water sample: No bacterial contamination seen



Fig 6: Input borewell water sample: Bacterial contamination seen



Fig.7: Input bore well water sample tested for presence of E.coli:No contamination seen

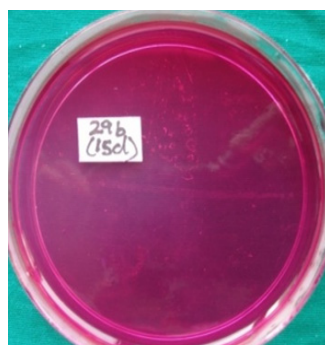


Fig 8:Input distilled water sample tested for presence of E.coli:No contamination seen

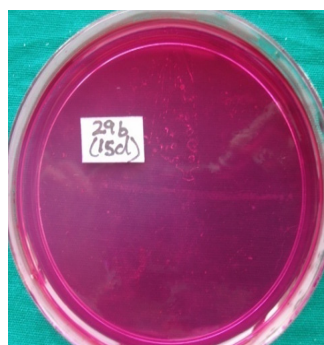


Fig9:Water and biofilm samples fromDUWLs using bore well water tested for E.coli:Small, pink, round, smooth colonies seen

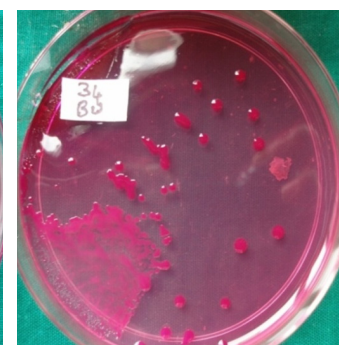


Fig 10:Water and biofilm samples fromDUWLs using distilled water tested for E.coli: No contamination seen