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The truth is in the eye of the beholder; opportunistic pathogens in the dental unit

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Infection control measures in the dental office are meant to reduce the risk of cross-contamination. A known source of cross-contamination is the dental unit. This medical device is prone to biofilm formation as the potable water used to cool the rotating instruments and ultrasonic devices contains microorganisms. Once adhered to the tubing surface, these waterborne micro-organisms form a matrix-encapsulated biofilm. This biofilm subsequently acts as a scaffold for the incorporation of bloodborne pathogens, introduced into the unit due to failing anti-retraction valves [1]. When released from the biofilm, these water- and bloodborne micro-organisms may be aerosolized and subsequently inhaled or ingested by patients and dental staff.

Immunocompromised patients run a risk from exposure to these micro-organisms. With the current COVID-19 pandemic, a new population of patients has arisen with long-term pulmonary complications, making prevention of bacterial and fungal aerosol exposure of prime importance [2,3]. To prevent

exposure to these micro-organisms, infection control protocols are in place.

Within the Netherlands, infection control guidelines are issued to ensure microbiological water quality in the dental unit: dentists perform a bi-annual risk assessment, based on an effluent water sample [4]. This sample, collected 30 s post flushing, is analysed using heterotrophic plate counts (HPC) to determine the number of viable bacteria. Units are deemed safe to use if the HPC are <100 cfu/mL. Additional testing for legionella has to be performed when >10⁴ cfu/mL are detected and practices must halt patient treatment when >100 cfu/L *Legionella* spp. are found.

In a recent study on 213 dental units, we showed that 84 units contained HPC <100 cfu/mL, while 54 units contained >10⁴ cfu/mL [5]. Additional to the regular effluent water sample, a 'proxy' biofilm sample was taken. This sample, taken after an overnight stagnancy period and prior to any flushing or hygiene measures, was analysed using molecular techniques. We found that, besides bacteria, 98% and 43% of the units contained fungal and amoeba DNA, respectively. Of the units containing HPC <10⁴ cfu/mL, 65% contained >2000 genomic units per litre *Legionella* spp. Even units without detectable HPC contained considerable concentrations of *Legionella* spp. Microbiome analysis revealed the presence of bacterial genera frequently associated with community-acquired pneumonia [5].

The risk assessment for dental units is primarily based on the HPC and less on the presence of (opportunistic) pathogens. Hydrodynamic principles, however, drive a favourable outcome of the water quality. During use, the fluid column in the lumen of the tubing is replaced with relatively clean input water, while creating a hydrodynamic boundary layer on the tubing wall with little or no exchange of micro-organisms between the biofilm and the effluent. Hence, a microbiological evaluation of the effluent, immediately after flushing, would more likely reflect the microbiological quality of the input water. Biofilm sloughing, the random process in which high numbers of potentially

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pathogenic micro-organisms detach from the biofilm, is also unlikely to be detected due to the low sampling frequency. These factors, combined with the use of suboptimal growth media, make a risk assessment using HPC questionable [6,7].

In our view, a risk assessment should be based on the 'proxy' biofilm sample using, for instance, molecular techniques. Biofilms contain a factor of $10^2 - 10^3$ more cells than the effluent, and opportunistic pathogens, such as legionella, reside preferably in the biofilm [8,9]. This alternative sample, taken from the stagnant water column, before flushing and hygiene measures, contains micro-organisms which diffused or migrated from the biofilm, in its relaxed state, into the lumen. Additionally, the sudden increase in shear stress at sampling gives the highest chance of biofilm clumps sloughing off [10]. Whereas more research would be needed to establish new risk thresholds, the 'proxy' biofilm sample does give the best prediction of the worst-case scenario to which a patient can be subjected. In our view, this risk assessment, together with strict adherence to infection control guidelines, is needed to guarantee patient safety.

If no changes are made, continued use of a post-flush effluent in combination with HPC may result in the dentist believing that the dental unit is safe to use, while in reality exposing vulnerable patients to the risk of contracting pulmonary infections.

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