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Examination of factors that influence residual chlorine concentration in chlorine-based sanitising solutions: implications for ward disinfection

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Chlorine-based sanitising agents are commonly used in healthcare sanitising protocols and rely on the availability of residual free chlorine (RFC) to kill vegetative microorganisms as well as bacterial endospores. Such formulations are commercially available from several manufacturers and are generally delivered in dissolvable preweighed tablet format. Simple and unambiguous reconstitution instructions accompany such tablets, detailing the number of tablets and volume of water required to obtain levels of free chlorine for various sanitising scenarios. In addition, instructions are given to dissolve such tablets in warm water.

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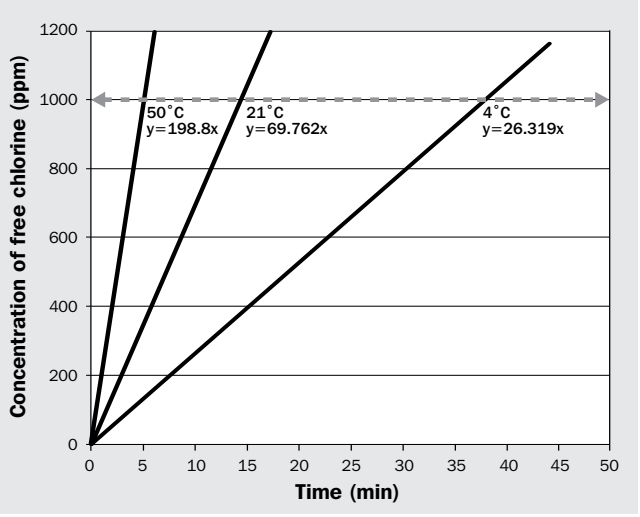

Fig. 1. Effect of water temperature on release of residual free chlorine (RFC) concentration on dissolving chlorine-based sanitising tablets.

The combination of optimal concentration and contact time in ward sanitising solutions forms an important critical control in the killing of nosocomial bacterial pathogens. Any deviation from these optimal values, as specified by the manufacturers of such sanitisers, will compromise their bactericidal efficacy. Minor deviations may not be materially important, but any major deviation may result in application of sanitising solutions with little or no killing ability. Deviation in reaching optimal concentration may result from i) underdosing due to the addition of an incorrect (and lesser) number of tablets by domestic staff, due to error or lack of understanding/education, ii) excessive organic interaction, or iii) transient underdosing due to the dissolving of such tablets at cold or ambient temperature.

Several factors routinely encountered during normal cleaning procedures by domestic staff may compromise the optimal concentration of residual chlorine during such operations. To date, no report has quantitatively examined the correlation between the temperature of water used to dissolve chorine-based sanitisers and concentration of free chlorine. In addition, there are limited data available on the quantitative correlation between type of clinical soil (e.g., blood, sputum, faeces, urine, saliva) and deactivation of residual chlorine, as well as a comparison between deactivation capacity of such individual clinical soils. Therefore, it is the aim of this short study to i) examine the relationship between various temperatures, emulating potential scenarios at ward level by domestic staff and the resulting availability of free chlorine in resulting sanitising solutions, and ii) examine the correlation between different clinical soils and deactivation of residual chlorine concentration in chorine-based sanitising solutions.

Chlorine-based sanitising tablets, based on sodium dichloroisocyanurate (NaDCC; 1,3-dichloro-1,3,5-triazinane-2,4,6-trione) were purchased from a commercial source. The appropriate number of tablets were employed, in accordance with the manufacturer's instructions, to make up solutions of 1000 parts per million (ppm) free Cl_2, which

is recommended for sanitising the general clinical environment and equipment. Tablets were dissolved in deionised water at 4°C (emulating cold tap water from water mains), 21°C (emulating tap water at ambient temperature) and 50°C (emulating warm tap water), and 10-mL volumes were removed at 3-min intervals between 0 and 30 min, or until all the tablets had dissolved completely, as determined by a lack of effervescence. The concentration of free chlorine was determined by stoichiometric titration with excess 0.5 mol/L iodide solution (KI) and sodium thiosulphate solution (0.01 mol/L), employing the following equations (A–F):

A $4H^+ (aq) + 2ClO^- (aq) + 2e^- \rightarrow Cl_2 (aq) + 2H_2O (l)$

B $2H^+ (aq) + ClO^- (aq) + 2e^- \rightarrow Cl^- (aq) + H_2O (l)$

C $2I^- (aq) \rightarrow I_2 (aq) + 2e^-$

D $2H^+ (aq) + ClO^- (aq) + 2I^- (aq) \rightarrow Cl^- (aq) + I_2 (aq) + H_2O (l)$

E $2S_2O_3^{2-} (aq) \rightarrow 2e^- + S_4O_6^{2-} (aq)$

F $I_2 (aq) + 2S_2O_3^{2-} (aq) \rightarrow 2I^- (aq) + S_4O_6^{2-} (aq)$

Five common clinical soils were compared, including blood (lysed horse blood, Oxoid, Basingstoke, UK), sputum (obtained from patients with cystic fibrosis), faeces, urine and saliva. For comparison, clinical soils were examined individually by inclusion in a 1000 ppm RFC solution at a concentration of 10% (v/v) and allowed to interact for 5 min. After this period, RFC concentration was determined, as outline above. In addition, calibration curves were obtained individually for each soil, by inclusion of clinical material in incremental concentrations in 1000 ppm RFC, allowing 5-min interaction time, prior to determination of RFC by titration, as detailed above.

Availability of free chlorine increased linearly with NaDCC tablet dissolving time. Concentration of free chlorine (ppm) was determined by titration at each time point and at each temperature, and these are shown in Figure 1, along with the calculated equation of the line for each temperature. At 50°C, 21°C and 4°C, the times required for complete dissolution of the tablets were 3 min, 21 min and 45 min, respectively, while the times required to reach optimal RFC concentration were 3 min, 14 min and 38 min, respectively.

Under standardised conditions, as detailed above, blood had the greatest deactivating ability on RFC (89.6%), followed in order by faeces (53.4%), sputum (44.6%), urine (36.6%) and saliva (24.0%) (Fig. 2).

Chlorine-based disinfection of healthcare environments is a major critical control of infection preventative measure.[1] Any effect or activity which has the potential to interfere with or reduce free residual chlorine concentration is important, and the dynamics of such interference needs to be mapped carefully by microbiologists and infection prevention practitioners, in order to perform risk assessments and adopt risk management strategies, to allow for any such events and maintain free chlorine concentrations at optimal levels.

The active ingredient in the majority of chlorine-based sanitising tablets employed in healthcare cleaning protocols is sodium dichloroisocyanurate, with a solubility of 22.7 g/100 mL water (25°C). A review on the comparison of sodium hypochlorite and sodium dichloroisocyanurate has been published.[1] One factor that may alter the dynamics of free chlorine in sanitising solutions is the solubility of NaDCC at different temperatures. While clear reconstitution

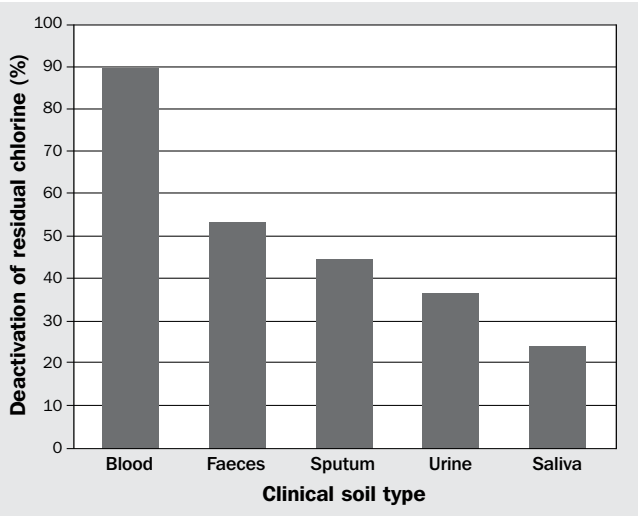

Fig. 2. Comparison of deactivating ability of five common clinical soils on RFC concentration.

instructions are provided by the manufacturer, and accompany batches of tablets, which indicate the need to dissolve the tablets in warm water, various human behavioural factors may confound this, including i) misreading such instructions, ii) ignoring the instructions, iii) a lack of education and awareness of the importance of such instructions among domestic staff, and iv) lack of available warm water to reconstitute the tablets.

This short study arbitrarily selected three temperatures to emulate potential healthcare scenarios and showed that dissolving NaDCC tablets is most effective at 50°C, compared to dissolving at 21°C or 4°C, which increased the time to reach optimal concentration (RFC=1000 ppm) times by approximately 3.5-fold and 14.5-fold, respectively (Fig. 1). Equations of the line for each temperature are also given in Figure 1 to allow hypothetical free chlorine calculations to be made. Furthermore, the increasing burden of clinical soil during sanitising procedures will also have an adverse effect on RFC concentration. This study demonstrated that an increasing burden of blood in particular resulted in a significant reduction in RFC concentration (Figs. 2 and 3).

While vegetative cells are more susceptible to chlorine-

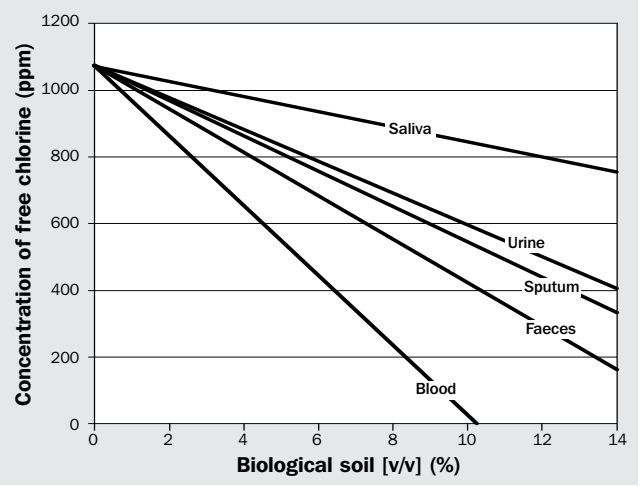

Fig. 3. Effect of increasing clinical soil concentration on RFC concentration with five common soils.

based disinfectants, spores are more resistant and therefore require either increased concentration of free chlorine or greater contact time, or both. Previous studies on the effect of chlorine-based disinfectants on *Clostridium difficile* spores have produced highly variable data. Wheeldon *et al*.[2] showed a 2.76–2.96 $\log_{10}$ spore reduction when treated with 1000 ppm available chlorine over a 15–30 min contact time in the presence of soil. Perez *et al*.[3] reported that in order to achieve >6 $\log_{10}$ reduction in viable spores on stainless steel surfaces in the presence of soil, a 1000 ppm free-chlorine solution needed to be applied for 15–20 min. More recently, Ungurs *et al*.[4] reported that precleaning with detergent followed by sufficient exposure time with at least a 1000 ppm free-chlorine solution resulted in a 4 $\log_{10}$ reduction. In a study by Speight *et al*.,[5] 32 sanitising agents were examined for their sporicidal activity under clean and dirty conditions and showed that three products failed to reduce the viability of spores by 10^3 under any test condition.

Overall, what is apparent from these studies is that 1000 ppm free-chlorine is the pivotal concentration. Hence, in the context of the current study, any application of sanitising solution prior to achieving this optimal (1000 ppm) concentration would result in a compromised ability to kill spores and a vulnerability relating to their survival and potential to infect new hosts.

What is currently lacking with the use of chlorine-based sanitising agents in healthcare cleaning regimes is a simple and effective means to aid domestic staff in their real-time assessment of RFC concentration in sanitising solutions. Simple real-time estimation methodology of approximate RFC concentration should be developed in the form of a rapid colorimetric determination, as is used for quality control purposes for RFC determination in swimming pools, or by using RFC probes as part of hand-held electronic devices. Such easy-to-perform and real-time adoption of these devices should be included as part of the cleaning regimes within healthcare and domestic staff trained and educated to ensure optimal maintenance of RFC concentrations.

In conclusion, this study emphasises the importance of water temperature in dissolving chlorine-based NaDCC tablets in order to reach the optimum free-chlorine concentration as quickly as possible, and highlights the deactivating ability of clinical soil. Therefore, it is important that these simple messages are conveyed to domestic staff in order to optimise chlorine-based disinfection protocols employed in healthcare environments and ensure the effectiveness of this critical control in infection prevention. □

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Detecting HIV antibodies in oral fluid: validation of a commercial antigen-antibody assay

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The use of saliva and oral fluids for detecting antibodies to human immunodeficiency virus (HIV) has long been suggested as an alternative to the use of blood.[1–3] Oral fluid is a safe, simple and convenient sample to collect for this purpose for a number of reasons. First, the occupational risks associated with needlestick accidents and injuries from phlebotomy are eliminated. Second, although oral fluid from HIV-1-infected individuals contains antibodies to HIV-1, the presence of infectious virus is rare. Third, oral fluid samples are easy to collect and the procedure is non-invasive, increasing patient comfort, acceptability of the method, and compliance with repeated testing.[4,5]

Oral fluid is a mixture consisting of the secretions of the salivary glands together with oral cavity microorganisms, cells and a gingival-crevicular transudate (GCT). The GCT is a fluid that contains immunoglobulin (IgA and IgG) and other blood components which have passed through the mucosa into the oral cavity.[6,7] It has been shown that the GCT of HIV-infected individuals contains high concentrations of HIV-specific IgG antibodies.[8] This antibody concentration, although lower than that found in serum, is quite sufficient to render GCT an adequate sample for anti-HIV antibody detection in epidemiological studies.[9,10] Studies have shown that a modified serum HIV assay can be used with acceptable sensitivity and specificity to test for HIV antibodies in GCT, regardless of the rate of prevalence of HIV-1 infection in the population under study.[11–14]

The introduction of specialised collection devices designed to improve the suitability of samples for HIV testing has seen an improvement in the sensitivity compared to tests performed on whole saliva. This is attributed to the presence of preservative fluid in the collection device, which contains antibacterial and antiproteolytic substances that protect the IgG from proteolytic degradation.[15,16]

The HIV assay used by Quest Diagnostics for three years to test saliva samples from non-hospital sites was due to be phased out by the manufacturer (Adaltis) and there was a

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