SOME FACTORS AFFECTING THE GROWTH AND SURVIVAL OF <u>PSEUDOMONAS CEPACIA</u> IN CHLORHEXIDINE SOLUTIONS

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<u>Pseudomonas cepacia</u> has been shown to be a frequent contaminant of user strength chlorhexidine solution (0.05% w/v) and has been implicated in infections arising from the use of such solutions (Burdon and Whitby 1967). In the laboratory it has often proved difficult to induce resistant strains of <u>Pseudomonas cepacia</u> to grow in these solutions (Bassett 1971).

We have found that pH, adsorption onto glass surfaces and temperature are all factors involved in the ability of Pseudomonas cepacia to grow and survive in the chlorhexidine solution (0.05% w/v). The addition of gluconolactone to the solutions enabled the Pseudomonas cepacia to grow and survive for long periods. The possibility that the organisms were using the glyconolactone in their survival was investigated and it was found that the role of gluconolactone was to reduce the pH of the solutions. This turned out to be one of the significant factors involved in their survival in chlorhexidine solutions. The pH aspects were therefore further investigated by adding gluconolactone to the solutions. In the presence of water gluconolactone is hydrolysed to gluconic acid, which then dissociates to release protons thereby lowering the pH of the solution. Chlorhexidine normally binds to the negatively charged groups on the cell surface and exerts its action causing lysis of the cell (Hugo and Longworth 1964). The presence of protons will therefore reduce the numbers of the negatively charged groups available on the cell surface and thereby prevent the chlorhexidine from gaining access to the cell. Growth was obtained at pH values below pH 6.2, but the greatest numbers were found at a pH value of pH 4.3.

We also observed rapid colonisation of the internal glass surfaces of the containers when a culture was introduced to either fresh distilled water or to fresh chlorhexidine solution (0.05% w/v). When an inoculum is added to either of the above, a fall in viable count in the liquid media is observed over the initial time period. It is probable that the cells are adsorbed onto the glass surfaces fairly quickly and this is the cause of the apparent loss of viability. Using glass microscope slides we were able to inoculate fresh chlorhexidine solution (0.05% w/v) with Pseudomonas cepacia growing in a similar condition. The size and shape of the glass containers were also a factor involved in the demonstration of resistance.

A greater incidence of resistance was seen when the cultures were incubated at room temperature than at 32°C. Further investigation revealed that the chlorhexidine solutions were less active at the same concentrations when the temperature was lowered. Growth was not always observed at these lower temperatures but survival was always longer.

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