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Influence of Physico-Chemical Parameters on the Survival of *Helicobacter pylori* in Drinking Water Biofilms

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The route of transmission for *Helicobacter pylori* is not well-known, but one of the suggested possibilities is via drinking water and associated biofilms. As such, the aim of this work is to study the influence of several physico-chemical parameters, including temperature, shear-stress and carbon concentration, on the prevalence and survival of *H. pylori* in drinking water biofilms. The biofilm studies were carried out using a two-stage chemostat system. The outflow culture of the first vessel fed three secondary chemostats in parallel and under different conditions of shear stress and carbon concentration. After 10 days the chemostats reached steady conditions, and the second stage chemostats were spiked with an inoculum of *H. pylori* NCTC 11637 (of approximately 10^6 cells ml^{-1}) and PVC coupons were then immersed to allow biofilm formation. The coupons were removed at different times (up to 32 days) and biofilms detached with sterile glass beads. Planktonic and sessile cells were quantified by standard cultivation techniques (R2A and HPSPA) and SYTO9 staining. Remarkably, *H. pylori* lost cultivability under all conditions in less than 1 h which compares with 24-75 h that the pathogen usually takes to lose cultivability in pure culture at these temperatures. This suggests that *H. pylori* is negatively affected by the presence of heterotrophic microbial consortium; alternatively, overgrowth of other species might hinder colony development of *H. pylori*. Current studies are tracking the uncultivable *H. pylori* in the biofilms using peptide nucleic acid probes in a high performance fluorescence in situ hybridisation assay. The data indicate that the cells continue to label with high fluorescence