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Relevance of Heterotrophic Biofilms on the Agglomeration of *Helicobacter pylori* in Water Environments: Implications for Transmission

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The role of water and water-associated biofilms on the transmission of *Helicobacter pylori* has been under debate for the last 10 years. We have previously shown that the ability of this bacterium to form monospecies biofilms when exposed to water is quite limited. However, molecular detection methods have been used to demonstrate that *H. pylori* is present in water microenvironments. Here, we have tested the cultivability and viability of several strains of *H. pylori* in water under different conditions to check whether a large increase in cultivability could be observed. Interestingly, the absence of light during water exposure appeared to be highly beneficial for a longer cultivability of the bacterium, with the ability of some strains to form colonies increasing from 24 to 96 hours. Nevertheless, recovering the bacterium from heterotrophic environments continues to pose a challenge, even when using selective culture media. Hence, we have applied specific PNA fluorescent in situ hybridization probes to detect the presence of the bacterium on heterotrophic biofilms. Results show that *H. pylori* is able to form aggregates within biofilm structures under several conditions (high and low shear stress, 15 °C and 20 °C, addition of carbon source). Addition of chlorine, however, inhibited the formation of these *H. pylori* agglomerates, and the bacterium was found to be more evenly dispersed along the support. This work suggests that the implementation of chlorine as a disinfection method has hindered the transmission of the pathogen and hence is partly responsible for the decreased prevalence observed in the more developed countries.