

Effectiveness of DNase and washing steps in removing dead cells' DNA for PCR detection of viable Escherichia coli

Abstract

This study investigates the use of DNase and washing steps in removing dead cells' DNA during sample preparation and their effect on the detection of viable cells using polymerase chain reaction (PCR). The results indicated that the DNA from heat-killed cells could be completely removed by DNase; thus, would not be detected by PCR. Inclusion of washing steps in centrifugation during sample preparation fails to remove DNA from heat-killed cells, but it reduces the amount of DNA from dead cells as well as viable cells. DNase could selectively remove DNA of heat-killed cells in the water sample without influencing the PCR amplification of viable cells' DNA. The inclusion of washing steps in the centrifugation procedure was ineffective, because viable cells might be lost during washing steps. This method allows the detection of viable bacteria and subsequently contributes to research concerning environmental samples.