

PCR and Dead Cell Detection

One criticism of PCR is that it amplifies DNA from both live and dead cells. This could be problematic if a PCR method reports positive results based on harmless dead cells, and food is then unnecessarily destroyed.

This potential problem is generally not an issue when using the BAX[®] System because food or environmental samples are enriched prior to testing. Enrichment provides the nutrients for live bacterial growth; dead cells are not affected. Therefore, dead cells would need to be present at a sufficient level prior to enrichment to yield the concentration required for BAX[®] System detection (approximately 10,000 cells/mL).

Further, the BAX[®] System protocol calls for 1:10 dilution of original sample into the enrichment broth. Thus, if 100 mL of enrichment broth were used, the true minimum concentration requirement for detection by the BAX[®] System would be 100,000 dead cells per gram of food or 1,000,000 dead cells per sponge.

Additionally, cells must provide good quality DNA for detection. It is highly unlikely that the DNA of dead cells will remain intact through the industrial processes that lead up to enrichment. Thus, the actual level of dead cells required to yield detectable DNA would need to be even higher.

This calculation applies only to samples tested after a single-stage enrichment. Samples that require an additional re-growth step or secondary enrichment would need between 10 and 100 times the concentration of dead cells calculated above due to the additional enrichment steps.