THE RELATIVE IMPORTANCE OF PARTICLE-ASSOCIATED FECAL INDICATOR BACTERIA FOR MICROBIAL WATER QUALITY ASSESSMENT AND MICROBIAL INDICATOR QUALITY IN KANEOHE STREAM.

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ABSTRACT

The basis for recreational water quality assessment relies on the ability to measure a characteristic pre-determined water quality criterion, which can be related to the risk posed by the water body. In terms of microbial water quality, the criteria are the concentrations of microbial indicators (e.g. enterococcus, coliforms, etc.) and the risk is characterized by the rate of illness of a population exposed to the contaminated water. Two methods, the membrane filtration and the most probable number, are routinely used to monitor indicator density. This approach to water quality assessment, however, faces many challenges, amongst which are the questionable reliability of the indicator system and the inability of the available measurement methods to accurately quantify the density of the microbial indicators.

Hawaii's State Law provides recreational water quality standards for enterococci. Nonetheless, this indicator has been shown to be unreliable in tropical environments because levels of enterococcus in excess of the standards are consistently found in unpolluted locations. Identification of positive colonies isolated on enterococcus-specific agar from a control (conservation land) and a contaminated (urban) station shows that at both stations, the majority of the enterococcus species recovered are common inhabitants of the soil and therefore cannot be used for water quality assessment. A minority of enterococcus species with hygienic importance (e.g. *E. faecalis*) was recovered from the urban site, however. Since the membrane filtration assay for enterococcus is not exclusive to the enterococcus species with hygienic significance, enterococci cannot be used unambiguously as water quality indicators in Kaneohe Stream.

The association of cells with aggregates is a known fact in microbial ecology. The culture-dependant methods for microbial water quality assessment, however, are not able to distinguish between multiple cells associated with aggregates. Instead, the colonies originating from each cell merge into one, such that only one colony forming unit per aggregate is detected. Computer simulations reveal that the problem of colony merging does not only occur because of the presence of aggregates, but that colony merging occurs also by chance alone, from the filtration of a randomly distributed cell population, inducing measurement error of typically <20%, depending on the size and number of the colonies. Efforts to assay the measurement error arising from the presence of aggregates were made both theoretically and empirically. Both methods show that this type of measurement error is typically on the order of a few tens of percent. The error, however, varies with the type of microorganism, being largest for heterotrophic plate count (HPC) bacteria and enterococci, and smallest for *C. perfringens*. Proportionally, enterococci were found to associate more readily with large aggregates (>5 μm) than *C. perfringens*, whose spores are primarily free-living, or than HPC bacteria. Overall, however, small aggregates (<5 μm) are responsible for most of the measurement error due to the inability of the membrane filtration method to separate multiple colonies growing from clusters.