# **Student Summer Scholar Program Application Examples**

## A1. Project Goals and Scope

## Example 1

#### a. Background:

Each year global estimates suggest that  $\geq 50$  million cases of disease affect swimmers and beach going activity(1). These occurrences are correlated to polluted waters caused by fecal contamination, either from point (e.g. wastewater facilities) or non-point (e.g. agriculture) sources(2-6). Abdelzaher et al. (2010) suggest that human health concerns are often associated with fecal coliforms, e.g. *Escherichia coli*, and/or parasites and viruses. Over the years microbial source tracking (MST) tools have been developed to establish regulatory guidelines for beach closures.

In Michigan, there are  $\sim 1,390$  recognized public beaches, of which  $\sim 450$  are monitored for *E. coli*(8,9). Traditional monitoring consists of culture-based monitoring requiring 18-24 hrs. to complete. Problems with this technology include the lack of rapid tests delaying safety concerns allowing beach goers to be exposed to pathogens and the lack of day-to-day comparisons as microbial numbers change every few hrs(10,8). More recent advances in monitoring consists of genetics associated with quantitative polymerase chain reaction (qPCR) using DNA markers. Advantages of this method include rapid analysis (3-4 hrs.) and accuracy. Some disadvantages include false positives/negatives, the inability to quantify more than one species per sample, and the lack of discrimination between live/dead cells. Hence, a beach closure could occur, not because of a health hazard but rather, the detection of dead cells (which pose no risk).

In this study we propose to use imaging flow cytometry (IFCM) associated with DNA markers. Flow cytometry (FC) technology dates back to ~ mid-sixties(II) and was mostly associated with the medical industry (e.g. cancer, viruses, etc.). Traditional FC used protein staining associated with cells of interest. Flow cytometry today uses DNA markers identical to qPCR with applications ranging from human health, ocean sciences, agricultural and freshwater applications. Newer developments, namely IFCM, use microscope objective lenses to both view and accumulate pictures of any particle detected. Its advantages include simultaneous multiple monitoring of species, DNA labelling of cells, rapid results (seconds/minutes to less than a few hrs.), and the detection of live vs. dead cells. Here, we propose to use IFCM to monitor beaches associated with inland lakes; although monitoring of Michigan Lake beaches have occurred, fewer studies have examined inland lakes.

#### b. Big picture:

Inland lakes are categorized according to surface area. In Michigan there are ~5 categories of inland lakes: (1) 62,798 lakes 0.1 acres or larger, (2) 11,037 lakes 5 acres or larger, (3) 1,148 lakes exceeding 100 acres, (4) 98 lakes larger than 1,000 acres, and (5) 10 lakes greater than 10,000 acres in size(12). Collectively, Michigan inland lakes generate ~\$15 billion in direct and indirect economic activity(13). Our study is part of a much larger project examining human health associated with beaches and MST. Our collaborators include at Grand Valley State University who has developed methods to monitor *E. coli* on beaches, and (EAGL) who runs the Michigan network of scientists and health providers monitoring human health affected by *E. coli* in the Great Lakes. Our specific goal in this study is to assess the

feasibility of using IFCM as a comparative method, coupled with qPCR, to characterize concentrations of  $E.\ coli$  in inland lakes. This project is significant as development of IFCM may eventually be viewed as a comparative MST method across the US.