

## COMPLETION NOTICE

### SA ¶4(g) Study the effects of sampling holding time, sample storage, and preservation on sample integrity for future use (P16)

---

#### Summary of the Study

This study investigated how qPCR measurements were affected by holding fresh water samples for 24 and 48 hours. This study also investigated how qPCR measurements were affected by storing frozen samples for incremental time periods of 6, 12 and 24 months. Finally, this study evaluated the feasibility of using freezer-archived NEEAR epidemiologic study samples for determining health relationships using qPCR measurements from new and/or improved fecal indicators with the health data from the NEEAR epidemiologic studies. The integrity of frozen archived samples was investigated by measuring the fecal indicators (*Enterococci* and *Bacteroidales*) in freezer-archived water samples (using the same methods that were used in the NEEAR studies). To determine if the archived samples had degraded, the fecal indicator measurement results from the archived NEEAR samples were compared to the fecal indicator measurement results from the samples analyzed during the NEEAR studies using statistical analyses.

#### Summary of Findings

The summary of results from this study are:

- The 24-48 hour holding time results for fresh/refrigerated samples were limited, conflicting, and difficult to interpret.
- The 6, 12 and 24 month frozen samples significantly changed over the 24 months, but not consistently across indicators and not progressively over time.
- Reanalyzed archived frozen filters from prior NEEAR studies were degraded.

**This study has been completed as of December 15, 2010.**