Collecting and counting aerobic anoxygenic phototrophic (AAP) Bacteria

Sample collection

1- Label a 125 ml bottle

2- Add 45 mL seawater

3- Add 5 mL 20% paraformaldehyde

4- Put in refrigerator labeled with sample number

5- Fix for 1 hour.

6- Load filter manifold with a Millipore HA filter and black 0.2 µm filters (polycarbonate).

7- Filter the sample

   10 mL in estuary
   50 mL offshore surface
   100 mL deeper than 100 m

8- Put the black filter in a 2-mL tube (orange cap) labeled with the sample number.

9- Store at -80°C

10- Record in log

Sample analysis

1- Cut 1/12 piece of the filter.

2- Stain with DAPI (1 µg/mL) in 1X PBS.

3- Mount on a slide with oil.

3- Count on microscope with IR sensitive camera immediately or store slide at -80°C.