Microbial Community Screening in a Host-Pathogen Relationship

Methods

1. 4 Experimental scenarios: 1)Oyster + V. tubiashii, 2) oyster, 3) Vt, 4)water only
   1. 5 experimental replicates per scenario
   2. Each sampling date represents 3 replicate samples from each experimental replicate: 3 samples x 5 replicates x 4 scenarios = 60
   3. Larval and water samples taken coincidentally and seawater replenished commensurate to volume taken (vol. = mL). Seawater strained from scenarios 1 & 2 through a 60 micron mesh strainer into a clean beaker. Larvae then transferred from strainer into RNAlater for further analysis.
   4. Upon collection, water samples are immediately filtered through Isopore membrane filters, 0.2 uM (Millipore) and filters are folded and stored in screw-cap tubes at -20C.
2. Recently spawned larvae acquired from Taylor Shellfish Hatchery in Quilcene, WA
3. Day 0: Larvae transferred to experimental conditions, salinity = , T = (no tubiashii) and Vt added to Vt only (3). **Sample 0**
4. After 24 hours take **Sample 1** and add tubiashii to set-up for oyster + Vt (1)
5. Sample daily (replacing water each time with fresh) for 7 days total, **Samples 2-7.** For oysters, also note growth & percent mortality for each sampling date.
6. Particularly interesting ERISAs could be sequenced for further identification at a later date.