

# VARIED MICROBES IMPORTANT TO RECIRCULATING AQUACULTURE SYSTEMS

## Summary:

Many microbes facilitate high-intensity recirculating aquaculture systems. Oceanic Institute research on recirculation indicates that the microbial community changes throughout growout from stocking to harvest, both in terms of biomass and community function. The functional redundancy of the in situ microbial community provides rapid and efficient carbon cycling.

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A significant development in the optimization of high-intensity shrimp production systems in the United States is the movement toward closed or recirculating aquaculture systems (RAS).

The Oceanic Institute in Waimanalo, Hawaii, USA, currently cultures Pacific white shrimp, *Litopenaeus vannamei*, under intensive conditions in recirculating systems that rely solely on the microbial community for the removal and sequestration of dissolved nitrogen species that are toxic to shrimp.

## Shrimp RAS

The use of recirculation for shrimp production represents a major paradigm shift from methods that commonly rely on open coastal ponds and flow-through water exchange as a management strategy to improve water quality. Recirculation alleviates problems associated with effluent discharge and reduces the risk of pathogen introduction from water-borne vectors.

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Recirculating systems also have the added advantage of moving shrimp production inland from coastal areas and within closer proximity to markets. Inland recirculating systems ultimately depend on a highly functional in situ microbial community to maintain acceptable



The varied make-up of microbial communities provides supplemental nutrition and other functions for shrimp.

water quality. In addition, bacteria, microalgae, and microbial-detrital aggregates provide supplemental nutrition for the shrimp.

## RAS Research

Oceanic Institute research on recirculation indicates that the microbial community changes throughout growout from stocking to harvest, both in terms of biomass and community function. Typically, the bacteria exhibit a characteristic increase in cell density from 10 million cells/ml at the start to roughly 300 million cells/ml within the first five weeks. Correspondingly, the biological

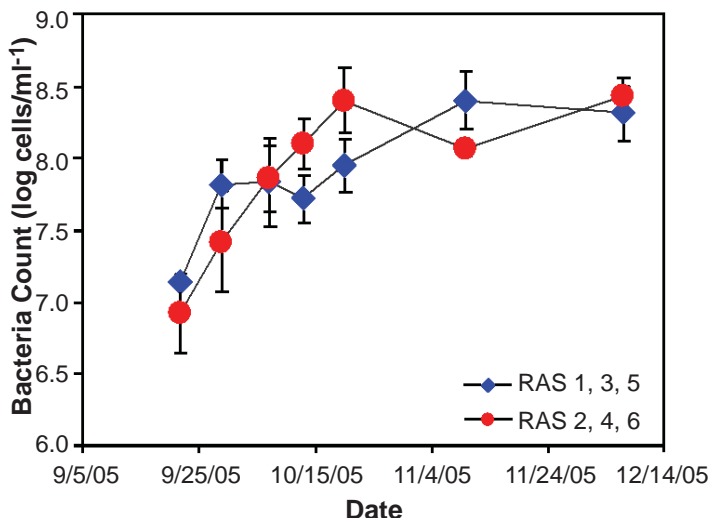


Figure 1. Total bacterial cell count over time in RAS. Error bars represent the mean standard deviation for three replicate water samples.

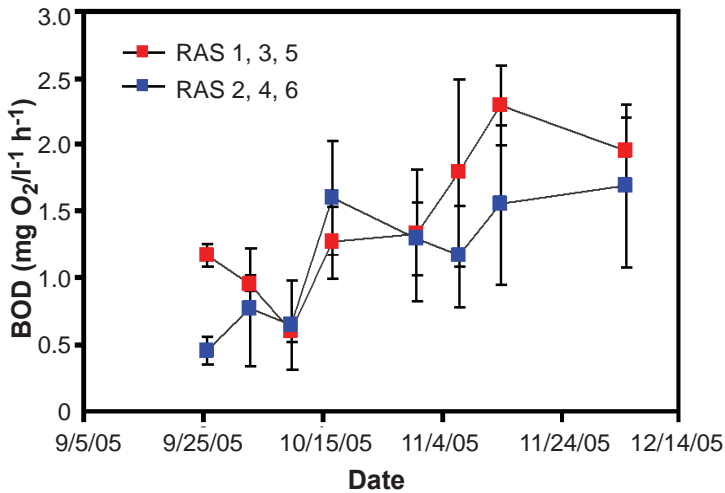


Figure 2. Biological oxygen demand of water column samples over time in RAS. Error bars represent the mean standard deviation for three replicate water samples.

oxygen demand in the water column increases with time (Figures 1 and 2). Results from a light/dark trial suggested that microalgae in raceways with light are important for the removal of nitrate and ammonia.

In addition, microalgae incorporate carbon into the particulate fraction and therefore retain nitrogenous nutrients in algal biomass. Interestingly, measurements of the natural abundance of nitrogen and carbon-stable isotope show that shrimp did not incorporate these small (less than 3  $\mu$ ) algal cells as a supplementary food source during this study. However, shrimp survival was higher in the presence of light, suggesting the importance of phytoplankton to system stability.

### Trophic States

Recirculation systems are often categorized as net phototrophic, autotrophic, or heterotrophic. However, the authors' stable isotope data suggests this distinction is not clear and that all three trophic states exist simultaneously when light is available. Figure 3 shows the carbon-stable isotope signature of three discrete size fractions of

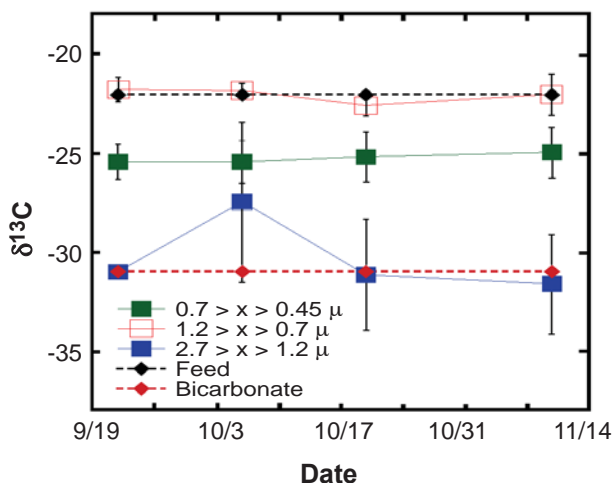


Figure 3. Carbon stable isotopic signature of size-fractionated water column particles from RAS over time. Isotopic composition of the feed and bicarbonate are added for reference. Error bars represent the mean standard deviation for three replicate water samples.

water column particles from the Oceanic Institute system.

The isotopic signature of the largest fraction clearly reflects photosynthetic incorporation of bicarbonate. Heterotrophic particles, as evidenced by a carbon signature closely resembling that of the feed, is seen in the 1.2 > x > 0.7- $\mu$  size fraction, whereas the smallest particles exhibit an autotrophic signature distinct from both heterotrophy and phototrophy.

The presence of all three trophic states in an RAS with ambient light suggests a functionally redundant microbial community that is prepared for carbon uptake and remineralization under fluctuating temperature, light, and pH conditions.

### Nitrification

Farmers know that nitrification, or the oxidation of nitrate and ammonia, is an integral microbial process in an RAS. An active nitrifying community ensures that toxic ammonia and nitrite are rapidly oxidized to nitrate, which is relatively harmless to shrimp.

Rate measurements suggest that both nitrification and denitrification potentially take place simultaneously in the Oceanic Institute systems during the later stages of the growout period. In unamended RAS water, a significant increase in nitrate concentration occurs after 24 hours of incubation under oxic conditions (Figure 4). When RAS water was amended with a nitrification inhibitor, there was no significant change in nitrate concentration under oxic conditions.

These results demonstrated nitrification in Ocean Institute systems. In addition, nitrate concentration decreased significantly under anoxic conditions, reflecting the potential impact of microbial denitrification on the removal of nitrogen from the RAS.

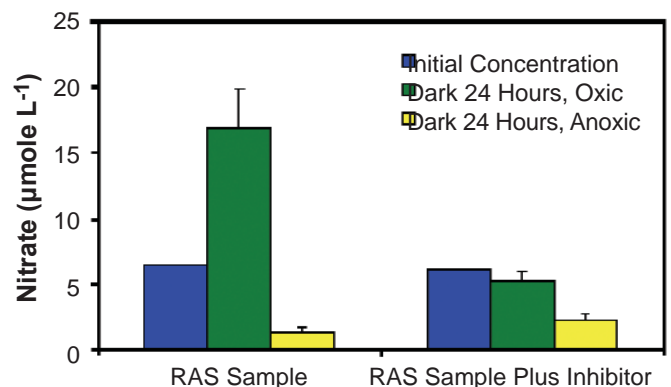


Figure 4. Nitrate concentration in water samples incubated with or without a nitrification inhibitor compared to the initial concentration. Error bars represent the mean standard deviation for three replicate incubations.

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