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Microbial community management in aquaculture

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Abstract

Microbial community management in aquaculture creates benefits at the nutritional as well as at health level for cultured species. In addition, in case of biofloc application, it allows to link species at different trophic levels, making bioflocs the potential link in integrated multispecies aquaculture.

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1. Main text

The expansion of the aquaculture production is restricted due to the pressure it causes on the environment by the discharge of waste products in the water bodies and by its dependence on fish oil and fishmeal. Aquaculture using bio-floc technology (BFT) offers a solution to both problems. It combines the removal of nutrients from the water with the production of microbial biomass, which can in situ be used by the culture species as additional food source. Physicochemical measurements such as the level of protein, poly-β-hydroxybutyrate and fatty acids can be used to characterize microbial flocs. In this respect, the strategy to have a predominance of bacteria which can easily be digested by the aquaculture animals or which contain energy rich storage products such as the poly-β-hydroxybutyrate, appears to be of particular interest. The quality of biofloc as food depends on the nutritional composition, but may also be related to the size distribution. The latter may influence the efficiency by which

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cultured animals with different feeding behaviour (filter feeders, scavengers) utilize the flocs as feed. The effect of biofloc size on its utilization by white shrimp (*Litopenaeus vannamei*), red tilapia (*Oreochromis niloticus*) and mussels (*Perna viridis*) was verified. Bioflocs collected from a shrimp culture unit were labeled with N15. The flocs were sieved grouping them into 4 different size classes (un-sieved, <48 µm, 48 – 100 µm, and >100 µm) and subsequently offered to shrimp, red tilapia and mussel. Each biofloc size class showed different protein, lipid, and amino acids composition. All biofloc size classes were consumed and utilized by the shrimp, tilapia and mussel. The highest retention of nitrogen in the animal body, however, was consistently originating from the bioflocs larger than 100µm, i.e. 4.14 g N/kg shrimp, 7.22 g N/kg tilapia, and 1.2 g N/kg mussel, respectively. From this study it can be concluded that biofloc consumption by shrimp, tilapia and mussel occurs irrespective of floc size but that floc size can play an important role in the quality of biofloc in relation to the capacity of nitrogen retention by the animals [1,2]. Another study on a use of bacterial biomass was done focusing on *Artemia*. Bacteria (grown by adding sucrose in situ) enhanced the growth of *Artemia* and hence total biomass production especially when microalgae were offered in limited amount. The level of total fatty acid methyl ester (FAME) in *Artemia* was affected by feeding bacterial biomass. Particularly, the level of monounsaturated fatty acid (MUFA), typically present in bacterial cell walls, increased in *Artemia* from 30,0 to 38,4 mg/g DW in treatments supplemented with sucrose. Also 15N originating from the heterotrophic bacteria was found in high excess level in *Artemia* when fed on low density of algae in the diet [3,4].

Immunological effects of growing shrimp in biofloc systems were also observed. After 49 days, phenoloxidase (PO) activity of the shrimp grown in all biofloc systems was higher than that of the control. Following a challenge test, by injection with infectious myonecrosis virus (IMNV), the levels of PO and respiratory burst (RB) activity in shrimp of all biofloc treatments were higher than that of challenged control. Survival in the challenge tests with shrimp from the biofloc groups, was also significantly higher compared to the positive control [2].

Rather than trying to control microbial community composition, microbial activity can be steered. The disruption of quorum sensing, bacterial cell-to-cell communication, has been suggested as an alternative strategy to control infections in aquaculture [5]. Quorum sensing has been shown to regulate virulence expression in many bacteria in vitro (i.e. in bacteria grown in synthetic growth media). However, microbiologists are becoming more and more aware of the fact that bacteria behave differently in different environments. Hence, the question that arises is whether and how quorum sensing regulates virulence of pathogens where it really matters: in vivo during infection of a host.

We found that quorum sensing regulates the virulence of *Vibrio harveyi* towards gnotobiotic brine shrimp larvae [6] and rotifers [7]. We developed a method to monitor bacterial gene expression in vivo, during infection of gnotobiotic brine shrimp. Using this method, we found that there is a significant difference in the expression of quorum sensing-regulated virulence genes between virulent and non-virulent isolates [8]. Finally, we found that quorum sensing also affects survival of burbot challenged to *Aeromonas hydrophila* and *Aeromonas salmonicida* [9]. The most important quorum sensing-disrupting agents reported thus far include compounds that interfere with quorum sensing signal detection and signal transduction, and signal molecule-degrading bacteria. We found that signal molecule-degrading bacteria isolated from aquaculture settings have a positive effect on survival of turbot and giant river prawn larvae cultured in non-gnotobiotic conditions [10],[11]. But also synthetic and naturally occurring chemicals can interfer with quorum sensing [12], resulting in improved larval survival. Results of positive outcomes using cinnamaldehyde, furanones and thiophenones will be shown.

Recent studies also indicate that opportunistic aquatic pathogens, such as *Vibrio* are also able to sense host clues such as stress hormones. We investigated the impact of catecholamine stress hormones on growth and virulence factor production of pathogenic vibrios. Both norepinephrine and dopamine (at 100 µM) significantly induced swimming motility of the tested strains. Pretreatment of pathogenic *V. campbellii* with catecholamines significantly increased mortality towards giant freshwater prawn larvae [13].

In conclusion, the further expansion of aquaculture needs to take integration in account. This means that nutrients should be recycled at the maximum. It appears that biofloc technology might make a substantial contribution to this. This however implies that much more attention will have to be given to the management of the microbial community composition and activity in addition to the management of the target aquaculture species.

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References

1. Ekasari, J., Azhar, M., Surawidjaja, E., Nuryati, S., De Schryver, P., Bossier, P., Immune response and disease resistance of shrimp fed biofloc grown on different carbon sources. *Fish & Shellfish Immunology*. 2014a. 41: 332-339.
2. Ekasari, J., Angela, D., Waluyo, S., Bachtiar, T., Surawidjaja, E., Bossier, P., De Schryver, P., The size of biofloc determines the nutritional composition and the nitrogen recovery by aquaculture animals. *Aquaculture*. 2014b. 426: 105-111
3. Toi, H., Boeckx, P., Sorgeloos, P., Bossier, P., Van Stappen, G., Bacteria contribute to *Artemia* nutrition in algae-limited conditions: A laboratory study. *Aquaculture*. 2013. 388: 1-7.
4. Toi, H., Boeckx, P., Sorgeloos, P., Bossier, P., Van Stappen, G., Co-feeding of microalgae and bacteria may result in increased N assimilation in *Artemia* as compared to mono-diets, as demonstrated by a N-15 isotope uptake laboratory study. *Aquaculture*. 2014. 422: 109-114.
5. Defoirdt, T., Boon, N., Bossier, P., Verstraete, W. Disruption of bacterial quorum sensing: an unexplored strategy to fight infections in aquaculture. *Aquaculture.* 2004. 240: 69-88.
6. Defoirdt, T., Bossier, P., Sorgeloos, P. and Verstraete, W. The impact of mutations in the quorum sensing systems of Aeromonas hydrophila, Vibrio anguillarum and Vibrio harveyi on their virulence towards axenically cultured *Artemia* franciscana. *Environmental Microbiology*. 2005.7 (8): 1239-1247
7. Tinh, N., Linh, N., Wood, T., Dierckens, K., Sorgeloos, P., Bossier, P., Interference with the quorum sensing systems in a Vibrio harveyi strain alters the growth rate of gnotobiotically cultured rotifer Brachionus plicatilis. *Journal of Applied Microbiology*. 2007. 103: 194-203
8. Ruwandeepika H, Defoirdt T, Bhowmick P, Karunasagar I, Bossier P. In vitro and in vivo expression of virulence genes in Vibrio isolates belonging to the Harveyi clade in relation to their virulence towards gnotobiotic brine shrimp (*Artemia* franciscana). *Environmental Microbiology*. 2014. 13: 506-517.
9. Natrah F, Ruwandeepika H, Pawar S, Karunasagar I, Sorgeloos P, Bossier P, Defoirdt T. Regulation of virulence factors by quorum sensing in Vibrio harveyi. *Veterinary Microbiology* .2011. 154: 124-129
10. Tinh, N., Yen, V., Dierckens, K., Sorgeloos, P., Bossier, P., An acyl homoserine lactone-degrading microbial community improves the survival of first-feeding turbot larvae (Scophthalmus maximus L.). *Aquaculture*. 2008. 285: 56-62
11. Nhan D, Cam D, Wille M, Defoirdt T, Bossier P, Sorgeloos P. Quorum quenching bacteria protect Macrobrachium rosenbergii larvae from Vibrio harveyi infection. *Journal of Applied Microbiology*. 2010. 109: 1007-1016.
12. Defoirdt, T. Antivirulence therapy for animal production: filling an arsenal with novel weapons for sustainable disease control. *PLoS Pathogens*. 2013. 9: e1003603
13. Pande, G., Suong, N., Bossier, P., Defoirdt, T. The catecholamine stress hormones norepinephrine and dopamine increase the virulence of pathogenic *Vibrio anguillarum* and *Vibrio campbellii*. *Fems Microbiology Ecology*. 2014.90: 761-769.