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Aquaculture

Science & Solutions

Following Norway's Example

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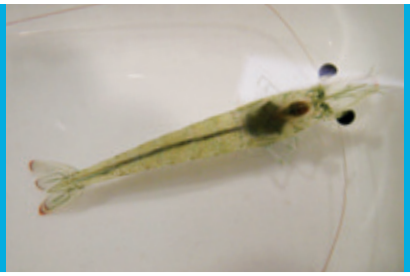
Sustainability Using Feed Additives



Benefits of Shrimp Hatchery Probiotics

Editorial

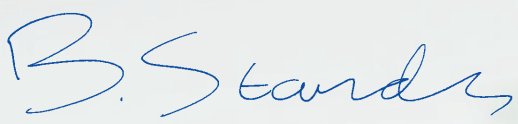
Following Norway's Example

The aquaculture industry has received a lot of bad press, often due to a lack of information or common misconceptions. One popular myth, for example, is that salmon is full of antibiotics. The facts, however, speak for themselves. A recent report by the Norwegian Veterinary Institute demonstrated that in Norway over the course of 2015, the domestic salmon industry used 273 kg of antibiotics, compared with 5850 kg for terrestrial livestock species. It should be noted that salmon production in Norway is four times higher than terrestrial animal production, which puts antibiotic use at 80 times lower in salmon culture. The Norwegian salmon industry has drastically reduced its antibiotic use over the years, largely through a combination of improved nutrition, rigorous biosecurity measures, stronger fry and the application of feed additives.

Feed (and water) additives have not only reduced the need for medicines; they have also enabled producers to improve pond management, feed efficiency and limit the negative effects of mycotoxins. In this issue of **Science & Solutions**, our first article focuses on the contribution of BIOMIN feed additives to a more sustainable aquaculture industry.

In order to maximize production, it is essential that our animals are given the best start in life. This is particularly true in aquatic species, where larvae are often under developed in terms of immunity. Our second article highlights how probiotics provide extra protection to shrimp during this critical period.

Happy reading!



Benedict STANDEN
Product Manager Aquaculture

Contents

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Driving the Blue Revolution with Green Feed Additives

By **Benedict Standen**, Product Manager, **Rui Gonçalves, Scientist**, and **Gonçalo Santos**, R&D Manager

Feed additives can help producers achieve better aquaculture practices that enhance growth, survival and feed conversion, increase tolerance to diseases and improve environmental conditions.

This article initially appeared in Aquafeed magazine.

With an estimated two billion more mouths to feed by 2050, aquaculture will account for a greater share of a growing animal production industry: this is the blue revolution! Industry growth will not come without challenges: disease and environmental discharge are ever-present concerns. The increased price of key feed components has led to the adoption of alternative materials that can introduce contaminant and nutrition challenges. At the same time, consumers, retailers and regulators increasingly demand sustainable production practices, driving the growth of widely recognized certification schemes.

One thing can be guaranteed: sustainability will accompany the goal of long-term profitability in aquaculture. With an extensive portfolio of natural, or 'green', feed additives, BIOMIN is committed to driving sustainable aquaculture.

Probiotics for improved pond management

Intensive farming produces large amounts of organic waste which accumulate in the pond environment. Oxidation of these organic waste compounds depletes the dissolved oxygen and leads to the formation of toxic metabolites. Considering the intimate relationship between aquatic animals and their environment, poor water quality is directly related to poor growth and poor health in fish and shrimp.

Probiotics are a useful, sustainable tool for managing the pond environment. The BIOMIN AquaStar® line contains unique probiotic strains and enzymes which control pathogens, eliminate undesirable waste compounds and reduce pond sludge.

To support this, four earthen ponds in China were stocked with juvenile shrimp at a density of 50 shrimp/m^2. Ponds were split into two treatments: control ponds with normal production operations and treatment ponds that received AquaStar® Pond and AquaStar® PondZyme. After 57 days the treatment pond soil was a yellow color which is regarded as the best bottom type, while control ponds exhibited dark grey and black soils—indicative of dead and decaying organic matter (*Figure 1*).

This is particularly dangerous in shrimp culture which, due to their biology, spend most of their time at the bottom of the pond. An improvement in pond sediments can manifest in improved shrimp growth. In the same trial, FCR was reduced by 9% and growth rate was increased by 36% in BIOMIN ponds.

Phytogenics for improved feed efficiency

Given that feed generally constitutes over 70% of total production costs, many producers increasingly rely upon less costly protein sources and less nutrient dense diets. This typically entails greater use of raw materials with lower protein digestibility, higher amino acid imbalance, and higher carbohydrate and fiber content. These dietary changes can lead to inefficient use of nutrients in the feed, increased feed usage and a decline in animal performance.

Figure 1. Pond sediments after 57 days of semi intensive shrimp culture with (a, b) and without (c,d) AquaStar® probiotics.

Photo: ideation90

Scientifically-developed and field tested feed additives can be effective tools for feed aquaculture industry while meeting the demanding targets imposed by consumers and

Table 1. Growth performance parameters of juvenile *L. vannamei* after eight weeks of feeding experimental diets with and without Digestarom®.

Treatment	Fish meal (%)	Digestarom® P.E.P. MGE	Final Weight (g)	PER	Survival (%)	FCR	SGR (%/day)
FM25	25	0	15.36	2.52	98.66	1.02	6.78
FM22 FM22 + P.E.P.	22 22	22 200g/t	12.31 13.70	2.04 2.17	98.67 98.67	1.26 1.17	6.37 6.55
FM19 FM19 + P.E.P.	19 19	0 200g/t	12.24 13.45	1.96 2.10	98.00 96.67	1.29 1.17	6.36 6.54

Source: BIOMIN

Table 2. Growth performance and intestinal bacterial counts of pangasius after eight weeks of feeding control, Biotronic® or antibiotic diets.

	Control	Biotronic® (0.5g/kg)	Flavomycin (20mg/kg)
FCR	1.39	1.37	1.43
SGR	1.16	1.18	1.17
Total intestinal bacteria (CFU/g)	17×10^{12}	4×10^{12}	7×10^{12}

Source: BIOMIN

Phytogenics are known to stimulate digestive secretions and increase the abundance and length of mucosal folds. As a result, phytogenics can be used to improve feed digestibility and efficiency.

A recent trial in China demonstrated how to reduce fish meal levels in shrimp feeds without compromising growth performance or feed efficiency. Five diets containing varying levels of fishmeal were formulated and fed to Pacific white shrimp for eight weeks. Three levels of fish meal were used; 25% (FM25), 22% (FM22) and 19% (FM19).

Digestarom® P.E.P. MGE was supplemented to the reduced FM diets. The addition of Digestarom® P.E.P. MGE to shrimp diets improved growth performance even when fishmeal components were reduced, as seen by comparing FM19 + P.E.P. with FM22 (without P.E.P.): all performance parameters were improved (final weight = +8.5%; PER = +6%; FCR = -7%; SGR = +3%; *Table 1*).

Acidifiers to reduce medicines

The use of antibiotic growth promoters (AGPs) is a major concern, as it allows bacteria to build transferable immune resistance, creating 'superbugs' that can endanger human health. Biotronic® Top3 is an enhanced acidifier combining three organic acids with phytochemical extracts which have quorum quenching properties. It also contains a unique permeabilizing complex which specifically targets Gram-negative pathogens, weakening the cell wall and enabling a greater bactericidal effect.

A trial was conducted in Thailand using pangasius, split into three treatments: control, flavomycin (20mg/kg) and Biotronic® PX Top3 (0.5g/kg). After eight weeks, growth performance and feed efficiency was slightly improved in the acidifier group. In addition, fish fed Biotronic® supplemented diets had a 76% reduction in total intestinal bacteria versus the control group and a 43% reduction versus the antibiotic group (*Table 2*).

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millers, integrators and farmers to build a more sustainable certification organizations.

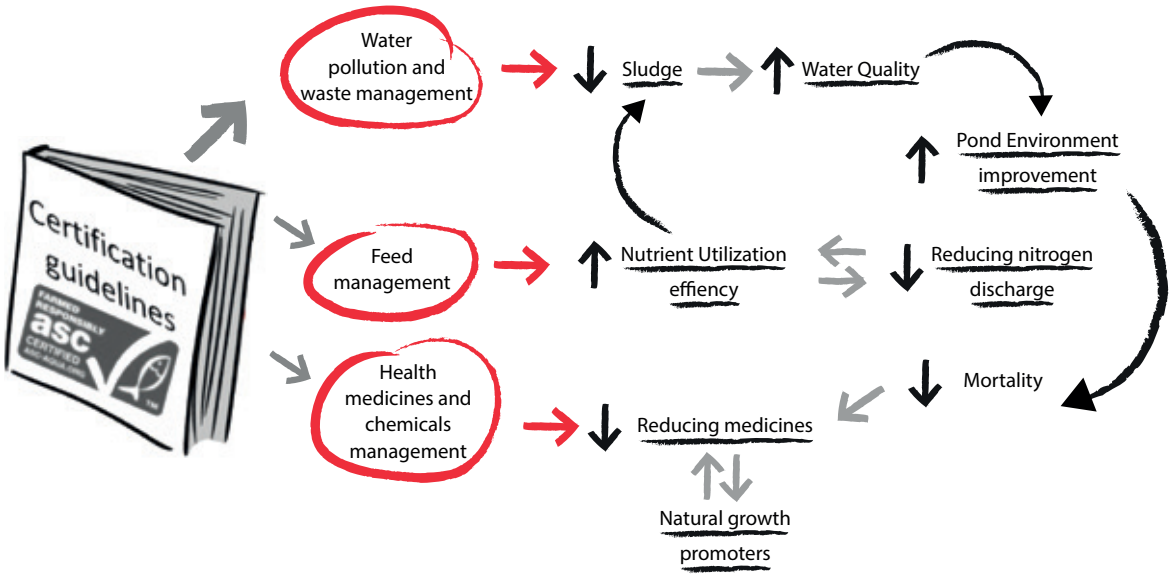


Figure 2. Simplified diagram demonstrating showing how sustainability issues are interlinked and the role feed additives have in improving fish and shrimp production.

Mycotoxin risk management

With increasing levels of plant-derived materials in aquafeed formulations, the risk of animals' exposure to mycotoxins increases. It is difficult to directly observe the negative effects of mycotoxins in aquaculture species, as most of the symptoms are subclinical and hard to detect. Several studies have highlighted the negative effects of mycotoxin-contaminated feeds in aquaculture. These effects are varied and include reduced growth, increased production costs, poor feed conversion and immune suppression.

The incorporation of Mycofix® product line in aquafeeds can help mitigate the negative effects of mycotoxins that can lead to a poor growth performance and disease susceptibility.

Recently, Dr Xinxia and co-authors (2016) studied the sensitivity of yellow catfish to dietary AFB_1 contamination, and the efficiency of Mycofix® Secure in offsetting the negative effects of AFB_1. The major impact was observed in FCR, where the presence of AFB_1 in the diet at levels of 500 µg/kg or higher led to a significant increase in FCR. At maximum level of AFB_1 in diet (1000 ppb), FCR increased 60% compared to control. When Mycofix® Secure was added to this diet, the FCR improved by 36%.

BIOMIN and sustainable aquaculture

While specific additives bring their own individual benefits, many issues surrounding aquaculture sustainability are intertwined (*Figure 2*). For example, better nutrient digestibility leads to improved feed efficiency, lowers feed costs and reduces environmental discharges thus improving water quality. Scientifically-developed and field tested feed additives can be effective tools for feed millers, integrators and farmers to build a more sustainable aquaculture industry while meeting the demanding targets imposed by consumers and certification organizations.

Shrimp Hatchery Probiotics

By **Benedict Standen**, Product Manager

Beneficial bacteria are key to improve PL survivability and profit in shrimp hatcheries.

Aquatic animal embryos are well-suited to bacterial colonization. Tank water will typically harbor rich microbial communities which can come into contact with the eggs. Thus, embryos are colonized by microorganisms rapidly; this community is termed the 'epibiota'. From a microbiology standpoint, most research has focused on pathogens in hatchery systems, as opposed to beneficial bacteria.

Disinfectants lack precision

Pathogens can be bacterial, viral or fungal and have different pathologies. For example, *Flexibacter* can dissolve the chorion via enzymatic means whereas other species can cause mortalities by overgrowth (and ultimately hypoxia), e.g. *Saprolegnia*. Initially, aquatic animals are extremely vulnerable as their immune system is not fully developed or functional and their gut is virtually sterile. Therefore, it is of paramount importance that


Photo: stanciuc

Boost Gut Performance and Profits

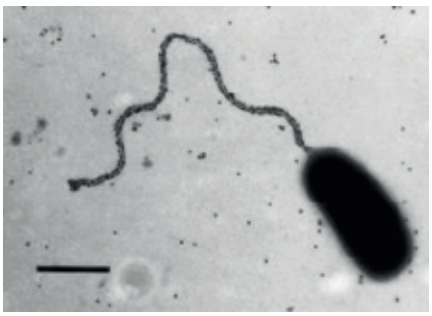
regardless of their type, pathogenic insults are minimized. In an attempt to reduce the microbial load during early life stages, embryos are often disinfected. However, since these techniques do not discriminate between good and bad bacteria, disinfection can be counterintuitive as the beneficial bacteria are also killed. It is these beneficial bacteria which would otherwise form the first line of defense. By removing beneficial bacteria, disinfectants increase an organism's susceptibility to disease and slows down the development and function of a proper gut structure, needed for nutrient absorption and growth. Therefore, when larvae hatch, it is important that the microbial balance is restored, and favors 'good' bacteria over 'bad'.

Probiotics for gut performance

Probiotics, or beneficial bacteria, can reduce the abundance of pathogens directly, or, enable larvae to fight pathogens themselves by stimulating the development

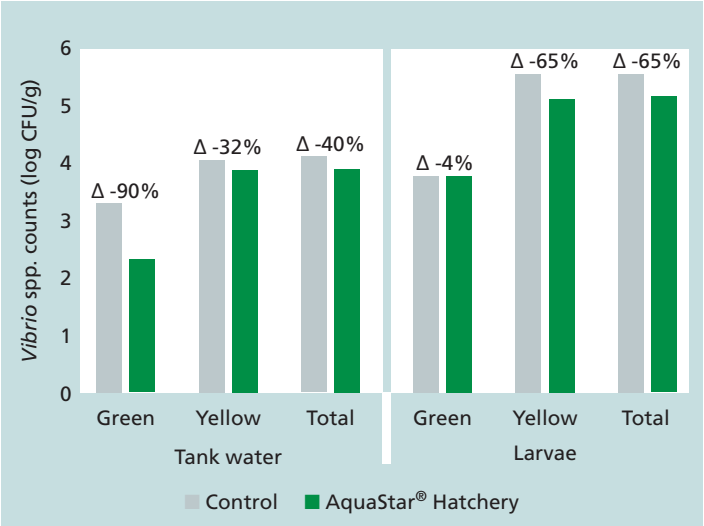
Probiotics, or beneficial bacteria, can reduce the abundance of pathogens directly, or, enable larvae to fight pathogens themselves by stimulating the development of the immune system.

Figure 1. Electron micrograph of a *Vibrio parahaemolyticus* cell. Scale bar = 1µm.



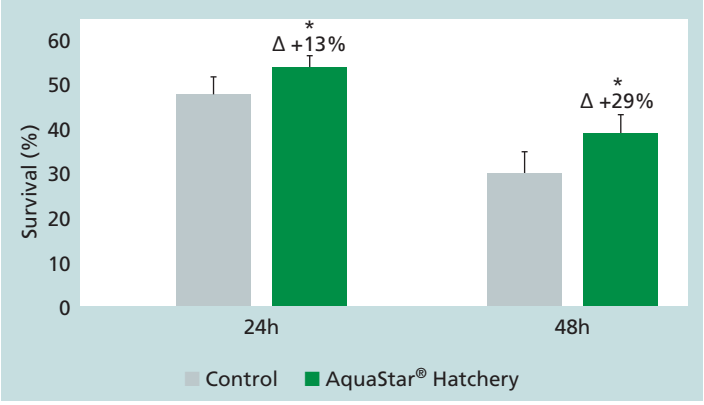
Source: BIOMIN

Figure 2. *Vibrio* counts in tank water and shrimp larvae from control and probiotic treatments.



Source: BIOMIN

Figure 3. Effect of probiotic supplementation on the survival of shrimp PL's after 24 and 48 hours exposure to ammonium chloride (20 ppm). Asterisk indicates significant difference within the same time point (P < 0.05).



Source: BIOMIN

of the immune system. Intestinal probiotics can be given a strategic advantage if they are applied at an early stage, in effect by arriving at, and colonizing the gut first. Consequently, they are a popular choice for hatchery managers. A number of trials demonstrate how probiotics can support pathogen control, environmental stress resistance, survivability and a reduction in production costs.

Probiotics for *Vibrio* control

Controlling *Vibrio* spp. is a major challenge for hatchery operations (*Figure 1*). Two recent trials demonstrate that probiotics can reduce the abundance of potentially pathogenic *Vibrio*, and consequently improve survival. In the first, Pacific white shrimp nauplii were split into two treatments: control nauplii were fed with live algae and *Artemia* until PL10, and a commercial feed until PL15. Shrimp in the probiotic treatment received a similar dietary regime but with AquaStar® Hatchery supplemented via the water (at 2.5 g/1000 l). *Vibrio* spp. were enumerated using widely recognized culture based techniques. Results showed that *Vibrio* counts were significantly reduced in the probiotic treated water. This could explain the survival results, where a 20% improvement was demonstrated at the nauplii phase, and a 30% improvement at PL15.

Similar results were obtained in the second trial. Although *Vibrio* spp. were detected in control and probiotic tank water, AquaStar® Hatchery appeared to reduce the total *Vibrio* abundance by 40% (*Figure 2*). *Vibrio* spp. were further differentiated according to their colony appearance. In the probiotic group, there were 90% fewer green colonies (*V. parahaemolyticus*, *V. harveyi* and *V. fischeri*) and 32% fewer yellow colonies (*V. alginolyticus* and *V. anguillarum*). A similar pattern emerged when analyzing *Vibrio* loading in shrimp larvae. *Vibrio* counts were generally higher in the larvae when compared with the water, indicating an accumulation effect. However, the probiotic reduced the total *Vibrio* count by 65%. This reduction was

Benedict Standen
Product Manager

mainly down to fewer yellow colonies, whereas green colonies were only reduced marginally (-4%, *Figure 2*).

Probiotics for ammonia stress resistance

Ammonia can build up quickly in hatchery systems, particularly those using live foods such as *Artemia*, as dead organisms add to the organic load. Furthermore, hatchery systems have very little water turnover, to prevent loss of the live food, allowing toxic metabolites to accumulate. Concentrations of ammonia >0.2mg/l may interfere with the animals ability to respire, and causes high mortalities. It is likely that physiologically immature larvae, are more sensitive to low levels, when compared to their adult counterparts. A short-term ammonia challenge (20 ppm ammonium chloride added to PL tanks) was carried out to investigate whether the probiotic could alleviate this stress. After 24 and 48 hours, the survival of shrimp was significantly higher in the probiotic treatment when compared to the control group (increased by 13% and 29%, respectively; *Figure 3*).

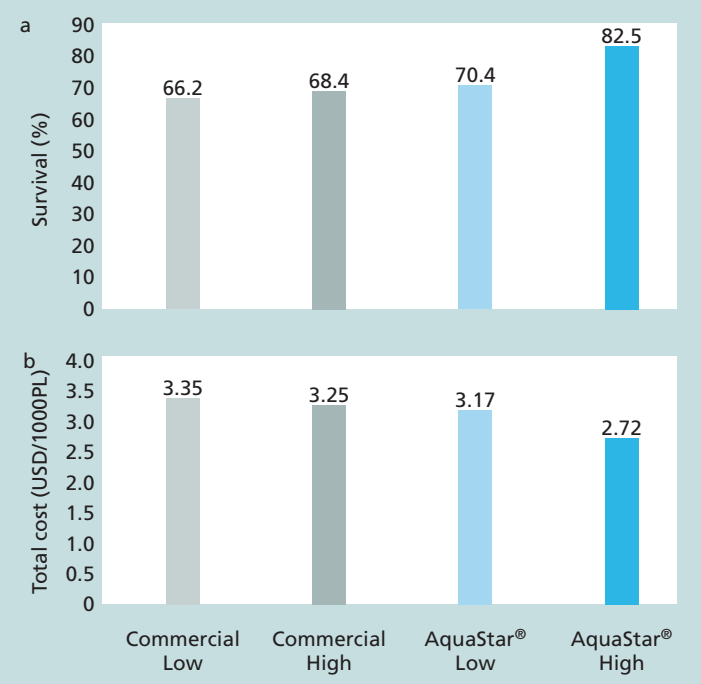
Probiotics in low salinities

Obtaining salt water for shrimp production can be a challenge, especially for inland hatcheries. Consequently, many producers use water with a lower salinity, which is not ideal for most shrimp species. However, probiotics can help alleviate this stress. For example, during a salinity stress test, shrimp produced at 0.3 ppt showed significantly higher survival rates (>96%) when produced with AquaStar® Hatchery, compared to those produced without the probiotic.

Probiotics make economic sense

Probiotic applications can be very successful, and consequently, there are a number of commercial probiotics available. It is important that each commercial formulation is supported by sound scientific principles, is safe to use in larvae as well as effective. A recent trial demonstrated the increased effectiveness of AquaStar® Hatchery in shrimp, when compared to another commercial probiotic product. After 12 days of culture (DOC), regardless of dosage, the survival was considerably higher in AquaStar® treatments, when compared to the other probiotic (*Figure 4a*). Even at a lower dosage (1 ppm), AquaStar® Hatchery supplementation resulted in higher survival when compared to the second product applied at 2 ppm. Higher survival, and consequently higher productivity has clear economic benefits; these can be visualized in *Figure 4b*. Ultimately, an effective probiotic can lower operating costs and increase survivability—improving a hatchery's profitability.

Figure 4. PL survival after 12 DOC (a) and the total cost to produce 1,000 PLs (b) when using different probiotic applications.



Source: BIOMIN

Ultimately, an effective probiotic can lower operating costs and increase survivability—improving a hatchery's profitability.

Conclusions

It is clear that probiotics can support aquatic animals during the vulnerable hatchery phase by controlling pathogens, improving disease resistance and alleviating environmental stressors (e.g. ammonia, low salinity). This results in improved survival, increased production and ultimately higher profitability of hatchery operations.

Biotronic® Top3

the breakthrough in pathogen control

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The **Biomin® Permeabilizing Complex** in Biotronic® Top3 damages the outer membrane of Gram-negative bacteria thus boosting the synergistic effect of its components, the organic acids and the phytochemical.

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- Improved feed conversion
- Maximized economical benefit

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