

- Moav, R., G. Wohlfarth, G.L. Schroeder, G. Hulata and H. Barash. 1977. Intensive polyculture of fish in freshwater ponds, 1. Substitution of expensive feeds by liquid cow manure. *Aquaculture* 10:25-43.
- Moriarty, D.J.W. 1987. Methodology for determining biomass and productivity of microorganisms in detrital food webs, p. 4-31. In D.J.W. Moriarty and R.S.V. Pullin (eds.) *Detritus and microbial ecology in aquaculture*. ICLARM Conf. Proc. 14, 420 p.
- Pearl, H.W. 1978. Microbial organic carbon recovery in aquatic ecosystems. *Limnol. Oceanogr.* 23:927-935.
- Ramesh, M.R. 1994. Influence of different substrates on heterotrophic food production and fish growth. University of Agricultural Sciences, Bangalore, India. 109 p. M.F.Sc. thesis.
- Schroeder, G.L. 1978. Autotrophic and heterotrophic production of microorganisms in intensely manured fish ponds and related fish yields. *Aquaculture* 14:303-325.
- Schroeder, G.L. 1983. Natural food web contributions to fish growth in manured ponds as indicated by stable carbon isotopes. *J. World Maricult. Soc.* 14:505-509.
- Schroeder, G.L. 1987. Carbon and nitrogen budgets in manured fish ponds on Israel's coastal plain. *Aquaculture* 62:259-279.
- Shankar, K.M., N.R. Umesh and C.V. Mohan. 1993. Development of bacterial biofilm in ponds for possible application in oral vaccination of carps. In *Second Symposium on Diseases in Asian Aquaculture*, Fish Health Section, Asian Fisheries Society, Phuket, Thailand (abstract).
- Shrestha, M.V. and C.F. Knud-Hansen. 1994. Increasing attached microorganism biomass as a management strategy for Nile tilapia (*Oreochromis niloticus*) production. *Aquacult. Eng.* 13:101-108.
- Umesh, N.R. 1993. Promotion and quantification of microbial biofilm for increasing fish production. University of Agricultural Sciences, Bangalore, India. 119 p. M.F.Sc. thesis.
- Welcomme, R.L. 1972. An evaluation of the acadja methods of fishing as practiced in the coastal lagoons of Dahomey (West Africa). *J. Fish Biol.* 4:39-45.
- Zhu, Y., Y. Yang, J. Wan, D. Hua and J.A. Mathias. 1990. The effect of manure application rate and frequency on fish yield in integrated fish farm ponds. *Aquaculture* 91:233-251.

K.M. SHANKAR, C.V. MOHAN and M.C. NANDEESHA are from the Department of Aquaculture, College of Fisheries, Mangalore-575002, India.

The Potential for Crop Rotation in Controlling Diseases in Shrimp Culture

Jose O. Paclibare, Marc C.J. Verdegem,
Willem B. van Muiswinkel and Bram E.A. Huisman

Abstract

The use of antibiotics and other chemicals in controlling shrimp pathogens becomes ineffective as the strains grow more resistant to these chemicals. Moreover, the bacterial pathogen (*Vibrio harveyi*) produces biofilm coating that protects it from drying and disinfection procedures that are followed during pond preparation. Biological control is being considered as an alternative means of preventing shrimp disease outbreak. The main principle behind biological control is to enhance the growth of beneficial microorganisms which serve as antagonists of target pathogens. The paper discusses shrimp and tilapia crop rotation as a form of effective biological control, a technique which is already being practiced in Indonesia and the Philippines.

The farming of tiger shrimp (*Penaeus monodon*) contributes significantly to the economies of the Philippines and other countries in the Asia-Pacific region. However, in recent years its production in many of these countries has declined due

to persistent disease problems. For example, in the Philippines, shrimp exports dropped from 30 462 t in 1991 to 18 275 t in 1995. In Negros Occidental, the major shrimp-farming area of the Philippines, only 10 out of 200 intensive

shrimp farms are currently operating. The rest of the farms ceased operation because of disease problems. The luminous bacterium *Vibrio harveyi* has been associated with many of these disease outbreaks.

Although *V. harveyi* has been considered a part of the normal microflora of shrimp and its environment, certain strain(s) may be more pathogenic than the others. When these pathogenic strains are plentiful, they can overwhelm the immune system of shrimp, allowing diseases to develop. In naturally diseased *P. monodon*, *V. harveyi* invades the hepatopancreatic tubules and causes extensive lesions even in the absence of other pathogens such as baculovirus and parasites (Jiravanichpaisal et al. 1994). *V. harveyi* produces proteases, phospholipases or hemolysins which may play important roles in the pathogenicity of *P. monodon* (Liu et al. 1996).

The continuous monoculture of shrimp over the past few years may have caused the increase in shrimp-pathogenic *V. harveyi* in the culture environments. Although many of the farms may have employed thorough pond preparation techniques, these bacteria may have carried over into succeeding cultures as they may have been protected in bacterial biofilms. Bacterial biofilms are notably resistant to drying and disinfection. The role of biofilms in the development of bacterial diseases in shrimp ponds has not been investigated yet. However, the finding of Karunasagar et al. (1994, 1996a) may have some relevance to shrimp ponds. They reported mass mortality of *P. monodon* larvae due to an antibiotic-resistant *V. harveyi* infection and suggested that antibiotic-resistant, virulent strains of *V. harveyi* were colonizing larval tanks. In the follow-up study (Karunasagar et al. 1996a), they observed that *V. harveyi* can form biofilms on all three substrates they tested: cement slab, high density polyethylene plastic and steel surface. Furthermore, the bacteria in biofilm were found to be more resistant to chlorine disinfection than their planktonic counterparts. In shrimp ponds, Karunasagar et al. (1996b) found that *V. harveyi* can

survive even in sediments that are treated with high doses of disinfectants. The addition of lime at 100 ppm to microcosms containing pond soil and water affected only *V. harveyi* counts in the soil, and marginally at that. The addition of chlorine at 10 ppm also led to a slight reduction of *V. harveyi* counts followed by an increase. When experiments were conducted on bacterial populations suspended in seawater, *V. harveyi* was found to be completely eliminated after 30 min. exposure to 10 ppm chlorine.

Because of the difficulty in reducing the concentration of pathogenic bacteria in shrimp ponds by conventional chemical disinfection, other effective means such as biological control should be explored. Biological control may be categorized into two approaches: a) the addition into the environment of beneficial microorganisms that serve as antagonists of the target pathogens and b) the manipulation of the environment in such a way that beneficial microorganisms are favored to proliferate. Examples of these approaches are the use of probiotics and crop rotation, respectively. Experiences on the use of probiotics in shrimp culture indicate conflicting results. For instance, many commercial probiotics that were claimed by their producers to be effective did not perform well in Negros Occidental. Assuming probiotics can reduce pathogenic bacteria in shrimp ponds, the method may still not be cost-effective because high amounts of the costly probiotic products must be added to the ponds frequently. The second approach, i.e., crop rotation for disease control in shrimp cultures is not yet widely recognized although it is already an established practice in agriculture.

The Incas, for instance, established mandatory seven-year rotations for potatoes before the arrival of the Spanish. It is now known that this practice was used to control potato cyst-nematodes (Sieczka

1989). The merit of crop rotation has also been proved in the potato industry in the Netherlands (Molendijk and Mulder 1996). As an example for other crops, rotation of lupin and wheat resulted in only an 18% brown spot leaf infection rate in lupin in contrast to a 63% rate for continuous lupin cultivation in Australia (Reeves et al. 1984). Kommendahl and Todd (1991) list approximately 64 fungal, 19 nematode, 1 viral and 16 bacterial diseases in plants in which crop rotation was found to be an effective control practice.

Crop rotation is a type of "sanitation" practice and is an integral part of plant health management. A sanitation practice should reduce the initial inoculum to a sufficiently low level so that the normal development of disease will not reach a high enough level to cause appreciable yield loss, provided unusual influx was avoided (Berger 1977).

Crop rotation in shrimp aquaculture is worth exploring and may prove feasible in view of the recent findings on the host specificity/preference of certain strains of *V. harveyi*. Liu et al. (1996) found differences in the pathogenicity of *V. harveyi* isolated from penaeid and non-penaeid sources. The results of the study of Owens et al. (1996) on the siderophore production in *V. harveyi* isolates suggest differences between isolates from finfish and penaeids. There is now increasing evidence that *V. harveyi* has two major biotypes. Pizzutto and Hirst (1995) found two major DNA and protein profiles for Australian strains of *V. harveyi*. Apparently, most of the pathogenic strains of *V. harveyi* of shrimp are sucrose-negative while the sucrose-positive strains are benign and even used as probiotics (Owens et al. 1996). Based on our observations in the Philippines, the sucrose-positive vibrios usually dominate ponds of healthy tilapia.

Personal observations on crop rotation with shrimp and tilapia

indicate that the practice reduces disease incidence in shrimp culture. In Indonesia, some shrimp farmers had relatively good harvests of shrimp after their ponds were used to produce one crop of tilapia. In general, the greater the phylogenetic differences between the culture organisms used in crop rotation, the better the sanitary effect (Francis and Clegg 1990). Since shrimp and tilapia belong to different orders within the animal kingdom, they are considered good candidates to test the sanitary effect of crop rotation on shrimp culture. In addition, crop rotation may be effective in preventing not only bacterial diseases but viral diseases as well, as proven in agriculture (Fyedorov-Davydov et al. 1989; Rist and Lorbeer 1991).

The potential of crop rotation in controlling diseases in shrimp culture is now a subject of a collaborative research project in the Philippines between the Bureau of Fisheries and Aquatic Resources (BFAR) and the Wageningen Agricultural University (WAU) in the Netherlands.

References

Berger, R.D. 1977. Application of epidemiological principles to achieve plant disease control. *Ann. Rev. Phytopath.* 15:165-183.

Francis, C.A. and M.D. Clegg. 1990. Crop rotations in sustainable production systems. *In Sustainable Agricultural Systems. Soil and Water Conservation Society, Iowa.* 696 p.

Fyedorov-Davydov, D.G., E.N. Mishustin and G.A. Bonartseva. 1989. Dynamics over many years for nodule bacteria in food

legume grasses with and without the presence of the host plant. *Agrokemia es Talajtan* 39(3-1):316-320.

Jiravanichpaisal, P., T. Miyazaki and C. Limsuwan. 1994. Histopathology, biochemistry and pathogenicity of *Vibrio harveyi* infecting black tiger prawn *Penaeus monodon*. *J. Aquat. Anim. Health* 6:27-35.

Karunasagar, I., R. Pai, G.R. Malathi and I. Karunasagar. 1994. Mass mortality of *Penaeus monodon* larvae due to antibiotic-resistant *Vibrio harveyi* infection. *Aquaculture* 128:203-209.

Karunasagar, I., S.K. Otta and I. Karunasagar. 1996a. Biofilm formation by *Vibrio harveyi* on surfaces. *Aquaculture* 140:241-245.

Karunasagar, I., S.K. Otta and I. Karunasagar. 1996b. Effect of chlorination on shrimp pathogenic *Vibrio harveyi*. *Book of Abstracts, World Aquaculture '96, Thailand.*

Kommendahl, T. and L.A. Todd. 1991. The environmental control of plant pathogens using eradication. *In D. Pimentel (ed.) CRC handbook of pest management in agriculture. CRC Press, Boca Raton, Florida.*

Liu, P.C., K.K. Lee and S.N. Chen. 1996. Pathogenicity of different isolates of *Vibrio harveyi* in tiger prawn *Penaeus monodon*. *Lett. Appl. Microbiol.* 22(6):413-416.

Molendijk, L.P.G. and A. Mulder. 1996. The Netherlands, nematodes and potatoes: Old problems are here again. *Potato Res.* 39 (Supplement): 171-177.

Pizzutto, M. and R.G. Hirst. 1995. Classification of isolates of *Vibrio harveyi* virulent to *Penaeus monodon* larvae by protein analysis and M13 DNA fingerprinting. *Dis. Aquat. Org.* 21:61-68.

Owens, L., D.A. Austin and B. Austin. 1996. Effect of strain origin on siderophore production in *Vibrio harveyi* isolates. *Dis. Aquat. Org.* 27:157-160.

Reeves, T.G., A. Ellington and H.D. Brooke. 1984. Effects of lupin-wheat rotations on soil fertility, crop disease and crop yields. *Aust. J. Agric. Anim. Husb.* 24:595-600.

Rist, D.L. and J.W. Lorbeer. 1991. Relationships of weed reservoirs of cucumber mosaic virus (CMV) and broad bean wilt virus (BBWW) in commercial lettuce field in New York. *Phytopathology* 81(1):367-371.

Sieczka, J.B. 1989. Some negative aspects of crop rotation. *In J. Vos, C.D. van Loon and G.J. Bollen (eds.) Effects of crop rotation on potato production in the temperate zones. Kluwer Academic Publishers, The Netherlands.* 310 p.

J.O. PACLIBARE is from the Bureau of Fisheries and Aquatic Resources, Arcadia Building, 860 Quezon Avenue, Quezon City, Philippines. M.C.J. VERDEGEM and B.E.A. HUISMAN are from the Department of Fish Culture and Fisheries and W.B. VAN MUISWINKEL is from the Department of Cell Biology and Immunology, Wageningen Institute of Animal Sciences, Wageningen Agricultural University, PO Box 338, 6700 AH Wageningen, The Netherlands.