Part III:

Discussion Group Summaries
Discussion Group Summaries

Introduction

Conference participants were given the opportunity to discuss a variety of shrimp disease-related topics in six 90-minute discussion groups. These sessions were designed to generate ideas, raise issues, solve problems and provide information to fellow experts. Relatively narrow topics were assigned to most sessions, and moderators were charged with facilitating the discussions.

In the context of the following pages, the term disease is defined as “any departure from normal structure or function” (after Sindermann, 1990). Furthermore, a pathogen is considered to be any biotic agent that may cause disease (as defined above). No attempt has been made to distinguish between obligate and facultative pathogens.

Discussion Group A was titled: “Country-by-Country Concerns.” Each participant listed his or her most important shrimp disease-related concerns. Common themes included the effects of shrimp culture on the natural environment, the indiscriminate use of drugs, and the need for more diagnostic techniques, better shrimp stocks, and improved husbandry techniques.

Subsequently, Discussion Group B focused on the “Prevention and Treatment of Diseases in Growout.” Biotic diseases were highlighted, and much of the time was devoted to preventing viral diseases. In addition, one of the concerns aired during this session was the quality of both wild-caught and hatchery-reared postlarvae. There was much discussion over how postlarval quality has changed over the past 10-15 years and what factors might be responsible for lowered performance.

‘Prevention and Treatment of Diseases in Hatcheries’ was the topic for Discussion Group C. Attention was given to the methods used to prevent and/or treat diseases of viral, bacterial, protozoan and fungal etiology. Most of the discussion centered around procedures used in P. monodon and P. vannamei hatcheries.

Issues relevant to the topic, “Disease Diagnosis” were dealt with in Discussion Group D. In particular, the problem of standardizing diagnostic methods and certifying diagnosticians, shrimp stocks, and culture facilities was discussed at length. The group issued a set of recommendations to address this issue.

Discussion Group E focused on an important issue raised in the previous discussion group, the certification and
possible quarantining of transported stocks and the certification of stocks and hatcheries as SPF.

In Discussion Group F, participants prioritized both the overall concerns and the research concerns that were raised during Discussion Group A. The need for SPF stocks and more diagnostic techniques were identified as two of the most important issues for researchers and farmers alike.

Discussion Group A: Country-by-Country Concerns

To begin the discussion group sessions, participants were asked, simply, what their primary concerns were with regard to diseases of cultured shrimp. As one might imagine, responses were varied, reflecting each individual’s home country and experience, in addition to their role as scientists, farmers or extension agents.

Central and South America

Because the majority of his experience has been with the shrimp culture industry in Central and South America, Rolland Laramore focused on the situation there. Topping his list was the problem of poor growth during dry seasons. Noting that several factors probably contribute to the phenomenon, Dr. Laramore was optimistic that genetic selection of robust and disease-resistant strains of Penaeus vannamei could alleviate some of the problems.

Other key avenues of research mentioned were developing bacterial vaccines for nauplii, postlarvae and broodstock, and “probiotics”, beneficial bacteria that displace or offset pathogens.

Japan

In Japan, P. japonicus is the dominant species of cultured shrimp. Kazuo Momoyama noted that the most economically important diseases of P. japonicus in Japan are caused by Vibrio spp. and Fusarium sp. Vibrio diseases have become increasingly important in recent years; they are estimated to cause 20% mortality during growout. In particular, there is a new, highly pathogenic Vibrio strain called “Vibrio sp. PJ” that is the subject of much concern in Japan. While there are two antibiotics sold in Japan to combat Vibrio diseases, their beneficial effects are only temporary.

Among penaeids, P. japonicus is known to be particularly susceptible to Fusarium disease, especially under intensive culture conditions. Reducing shrimp densities is the only effective means of controlling this fungus. Baculoviral midgut gland necrosis virus (BMNV) used to be a major problem, but, as a result of effective preventive measures, it is now relatively rare.

In more general terms, Tokuo Sano discussed the need for a clean culture environment to discourage facultative pathogens and the problems associated with developing bacterial vaccines, while Kazuo Momoyama aired con-
cerns about the international transport of shrimp stocks and feeds.

Malaysia

Speaking on behalf of both Malaysia and Asia in general, Mohd. Shariff called for more ecological studies of shrimp culture situations — studies that would enable researchers to monitor changes in potential pathogens. He further noted that chemotherapeutants are not necessarily the best means of combating shrimp diseases — not only can they harm the environment, but they are not always effective. Therefore, immunological studies should receive a high priority. Finally, Dr. Shariff called for the standardization of shrimp research methodologies. Perhaps a manual similar to the American Fisheries Society "Blue Book" could be issued for shrimp disease workers. Such a project would benefit from cooperation between the Asian Fisheries Society and the American Fisheries Society.

People's Republic of China

In what was to become a common theme, Dou Chen joined Mohd. Shariff in cautioning against the use of chemotherapeutants in shrimp culture. Stating that the most serious diseases of cultured shrimp in China are those caused by Vibrio spp., Prof. Chen listed several disadvantages to using antibiotics, including the possibility of promoting fungal diseases and fostering resistant strains of bacteria. For example, as a result of overuse/abuse, oxytetracycline is now completely use-

less. Since not all bacteria are harmful, Prof. Chen believes that the best way to control the potentially pathogenic species is by ecological or biological control.

Although there is concern about viruses in China, viral diseases are not as economically significant as those caused by bacteria. Anxiety is increasing, however, over a number of diseases of unknown etiology, including black-white spot disease (see D. Chen, this volume). Finally, epichommensal diseases also significantly impact shrimp culture in China.

Philippines

José Natividad was another who spoke out against the indiscriminate use of antibiotics in shrimp culture. He also shared his concerns about the current widespread use of probiotics and other drugs in the Philippines. Because the impact of probiotics is largely unknown, and because great quantities of probiotics, as well as other drugs, are present on the farms, Dr. Natividad called for some sort of government control or clearance process for probiotics and other compounds that are now unregulated. Secondly, Dr. Natividad said that he needed field diagnostic techniques to help him quickly assess production problems. Finally, some diseases that affect the marketability of shrimp, most notably "black meat disease" and "tail rot disease", have resulted in rejection of shipments, alarming shrimp farmers.
South Korea

Most shrimp culture in South Korea is extensive; as a result, relatively few disease problems have been encountered. Myoung Ae Park did, however, list some disease agents that had been encountered in farms and hatcheries in Korea: BMNV (in larval *P. japonicus*), hepatopancreatic parvo-like virus (HPV; in *P. chinensis*) and *Vibrio* spp. Ms. Park pointed out that better water quality management can alleviate some disease-related problems.

Thailand

When it comes to shrimp diseases, Taiwanese farmers and researchers have, unfortunately, gained a great deal of knowledge through experience. According to S.N. Chen, the most important problem is environmental impact. How does shrimp culture impact the surrounding environment, and, in turn, how do environmental changes affect cultured shrimp? Vibriosis is also a key concern, and while progress is being made in the area of vaccines, delivery technology has been problematic. Shrimp viruses also need to receive more attention, as does the problem of drug residues in harvested shrimp in some Asian countries.

As noted by Cheng-Fang Chang, intensification is responsible for many of the problems encountered on shrimp farms. In intensive systems, epicomensal diseases are important; water quality management is key to controlling fouling organisms. Interestingly, Mr. Chang also listed gregarines as a major concern. While their affect on shrimp performance is unknown, a recent survey in Taiwan found that 80% of cultured *P. monodon* were infected with these intestinal parasites (see Liao et al., this volume). Finally, Mr. Chang reminded the group that the relationship between the culture environment and diseases should not be ignored.

United States

Recognizing that many diseases can be avoided by improved husbandry techniques, a number of the experts from the United States called for studies to address issues such as sustainability, the ramifications of intensification, site selection/environmental planning, feeds, preventive health maintenance, and the impact of certain culture practices on the environment. Carl Sindermann challenged researchers to help
farmers "manage around pathogens," while James Wyban said the shrimp culture industry had a great deal to learn from other meat production industries such as the cattle, poultry and swine industries.

The two commercial representatives, Nick Carpenter and Fritz Jaenike, were among several participants who shared concerns about the spread of viral diseases. The movement of stocks threatens efforts to establish disease-free facilities and may also impact populations of wild shrimp. In a related topic, SPF technology was mentioned by several members of the U.S. contingent as a means of improving the shrimp industry's image, increasing production, and beginning the process of domestication.

Bacterial diseases, however, were also listed as concerns by three U.S. participants. For example, Donald Lightner pointed out that farmers need to have effective, government-approved chemotherapeutics, and Nick Carpenter wondered whether vaccines could be a realistic solution to the problem of bacterial diseases. Finally, Carl Sindermann strongly encouraged researchers studying shrimp diseases to quantify the economic impact of disease using standardized measures. This is the only way researchers are going to receive needed support, said Dr. Sindermann; scientists need to convince the shrimp industry, governments and funding agencies that their work is economically significant. He encouraged consultation with economists and experts in population management and epidemiology, and the uniform statistical treatment of data.

Discussion Group B: Prevention and Treatment of Diseases in Growout

Although the varied means by which viral, bacterial and protozoan diseases are prevented and treated was the topic of this session, most of the discussion centered around two issues: preventing viral diseases, and poor performance of animals in growout in many areas, possibly as a result of 10 - 15 years of culture activities.

Virus Prevention

As evidenced in Discussion Group A, viruses are not the only important pathogens of penaeid shrimp. The relative importance of viruses to cultured shrimp depends on a number of factors, including the type of shrimp cultured, the type of viral and nonviral pathogens present, the stressors present in the culture environment, and how heavily infected a given shrimp population is. There are environmental and regulatory issues as well. If a certain viral pathogen is absent from a given area, it is important to exclude that virus from nearby culture facilities.

In many cases, the best way to prevent viral diseases is to use certified SPF broodstock. An SPF program for *Penaeus vannamei* has already been implemented in the United States (see
Wyban, this volume). Maintaining SPF P. monodon in Southeast Asia may prove much more difficult, however. Steve Psinakis, a shrimp farmer from the Philippines that participated in the workshop, asked about the possibility of breeding disease-resistant strains, thereby eliminating the need to keep animals isolated from some pathogens.

What about the problem of introducing the progeny of SPF broodstock (hereafter referred to as high-health animals) into ponds that once held virus-infected shrimp? How do these animals perform? Do they become infected with viruses during the growout cycle? Preliminary results from several shrimp farms in the United States indicate that, in these situations, the incidence of viral disease is greatly reduced, significantly improving production. Factors to consider are the treatment of the pond bottom prior to stocking, and the nature of the virus(es) of interest. For example, it is likely that occluded viruses such as MBV will be more difficult to eliminate from ponds than nonoccluded viruses.

In Hawaii, growout ponds were dried for 10 to 14 days and treated with 800 lbs CaCO₃/acre prior to being stocked with high-health postlarval P. vannamei. Baculovirus pernæi (BP) and infectious hypodermal and hematopoietic necrosis virus (IHNV) were the viruses of concern. Shrimp yields were very good, and the incidence of runt-deformity syndrome was greatly reduced. Some of the shrimp, however, did test positive for BP (see Carpenter and Brock, this volume).

In Taiwan, growout ponds are routinely dried between harvests and then treated with chlorine (10 - 20 ppm for 48 h). These levels of chlorine are effective against some viruses and other pathogens; however, they probably also harm the natural environment. The results of a number of studies were discussed with regard to eliminating pathogenic viruses. For example, a treatment of 0.1 ppm iodine for 6 - 7 s eliminated 99.9% of the trout virus, IHN, from water (Batts et al., 1991). Studies on BP were performed at the Gulf Coast Research Laboratory (Le Blanc and Overstreet, 1991a, b). BP-infected hepatopancreases were subjected to a variety of treatments: desiccation, calcium hypochlorite, heating, pH extremes, etc. Though the results are not directly transferable to treating pond bottoms, the researchers found that BP could be inactivated rather easily using several methods. Finally, it was mentioned that the microbial activity in the pond bottom could be a significant factor in destroying infective viruses in the sediment. In a related question, participants discussed the best means to test sediments for the presence of viruses. Bioassay studies are presently being used in Mississippi for BP. Gene probe and
PCR (polymerase chain reaction) technology may also be applied to this problem in the future.

The Impact of Shrimp Farming on Postlarval Quality

During the course of the discussion, Steve Psinakis, a shrimp farmer from the Philippines, submitted the following hypothesis for discussion: “In many places around the world over the past 10 - 15 years, shrimp farming activities have done something to reduce the quality of both wild-caught and hatchery-reared postlarvae. Farms are experiencing higher feed conversion ratios, lower growth rates and lower survivals, and the problem is getting worse. The animals seem much more sensitive to perturbations than they used to be.” As evidence to support his hypothesis, Mr. Psinakis noted that despite improved culture techniques and better trained staff, production declines year by year. Furthermore, the phenomenon has been observed in ponds of varying ages using both wild seed and hatchery-raised seed from wild-caught spawners. Other participants related experiences that seemed to support the above hypothesis. For example, P. vannamei was once considered asymptomatic for the IHHN virus; now it is clearly not asymptomatic. Is this a result of deteriorating stocks? Also, while postlarval quality problems have not been observed in Panama over the past 15 years, farms in Guayaquil, Ecuador, have experienced lower production in recent years, and increased incidence of disease. Problems are worse in areas with a high density of farms.

What specific activities might be responsible for harming wild stocks of penaeids? Two possibilities were discussed, the use of chemotherapeutants, and the release of hatchery-reared postlarvae into the natural environment. Chemotherapy in the hatchery allows animals that are naturally weak and susceptible to disease to survive, thereby selecting for shrimp that are less fit. These animals typically perform poorly in growout. Most hatchery managers, moreover, have no incentive to produce animals that will perform well in growout, they profit by producing large numbers of healthy-appearing postlarvae. This makes it difficult to obtain animals that will remain healthy during growout.

In a related problem, it is a common perception in many parts of Asia that countless numbers of diseased, hatchery-reared postlarvae are regularly released into coastal areas. If some of these shrimp survive and reproduce, these artificially selected animals may alter the composition of the gene pool of the wild population. Secondly, because of the widespread practice of transporting stocks without regard to their disease status, viruses have been introduced to previously “clean” culture facilities. Furthermore, we are learning that a number of wild shrimp populations now carry pathogenic viruses (see Lotz, this volume), possibly
as a consequence of nearby shrimp culture activities. The impact of these viruses on natural stocks is unknown.

The above hypothesis has not been proven — it has not even been tested. Because there are so many interrelated factors, finding answers will be difficult. Clearly, though, more emphasis should be placed on determining the capacity of a given culture area to support shrimp. Also, precautions should be taken to avoid new viral introductions, and chemotherapeutants should be used more judiciously. Finally, the practice of releasing cultured animals into the natural environment should involve consideration of animal health and genetic diversity.

- Monitoring the culture environment; or
- Managing the animals.

Prevention

Viruses. No one in the group reported using specific water management techniques to prevent viral diseases. Many culturists, however, pretreat culture water, either with chemicals such as chlorine or iodine, or by other means such as ozonation or ultraviolet radiation (Table 1). Often culture water is filtered beforehand to increase the effectiveness of these other sterilization methods.

In some places, cultured shrimp are routinely monitored for the presence of viruses. Such measures, depending on the sensitivity of the assay used, could help prevent the spread of a virus from infected tanks to uninfected tanks. Animal management, however, is the best way to prevent viral diseases. Simply put, stocking with high-health postlarvae is the most effective means of keeping viruses out of the hatchery. Secondarily, eggs and/or nauplii can be rinsed or chemically treated to remove external virus particles. This has effectively prevented BMN outbreaks in Japan, and is also used to lower the incidence of MBV in P. monodon hatcheries and BP outbreaks in commercial shrimp hatcheries in South and Central America. Finally, many hatcheries these days are using batch methods; drying out their system in between cycles. While this is quite effective for
Table 1. Preventing diseases in hatcheries in the Americas and Asia.

<table>
<thead>
<tr>
<th>Potential pathogens</th>
<th>Water management</th>
<th>Chemicals</th>
<th>Monitoring</th>
<th>Animal management</th>
<th>Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Viruses None.</td>
<td>Pretreatment of water with ozone, chlorine, iodine, UV (filtration will enhance effectiveness of above).</td>
<td>Periodic diagnostic screening, broodstock history</td>
<td>Begin with high health animals, rinse or treat shrimp eggs and/or nauplii.</td>
<td>Use batch techniques with dry-outs in between.</td>
<td></td>
</tr>
<tr>
<td>Bacteria Increase exchange rate, use microalgae and other means to condition water, optimize temperature.</td>
<td>Disinfection with chemicals listed above, antibiotics, malachite green, vaccines, EDTA (5 ppm in Taiwan, 10 ppm in the Philippines).</td>
<td>Daily monitoring with agar plates, e.g., TCBS counts with water and larval homogenates.</td>
<td>Rinse shrimp nauplii and eggs, optimize stocking density.</td>
<td>Rinse Artemia nauplii, use batch techniques with dry-outs in between, optimize nutrition, add microalgae that have inhibitory effects.</td>
<td></td>
</tr>
<tr>
<td>Protozoa Increase exchange rate, optimize temperature.</td>
<td>Formalin, malachite green, copper sulfate, EDTA.</td>
<td>Observe water with microscope.</td>
<td>Rinse shrimp nauplii and eggs.</td>
<td>Rinse Artemia nauplii, optimize nutrition.</td>
<td></td>
</tr>
<tr>
<td>Fungi Teflaxan, malachite green, EDTA</td>
<td>Observation with microscope.</td>
<td></td>
<td></td>
<td>Optimize nutrition.</td>
<td></td>
</tr>
</tbody>
</table>

1 Preventive methods appearing in this table are used either singly or in combination in hatcheries producing *P. vannamei*, *P. monodon*, *P. chinensis* or *P. japonicus*. Some may be effective only for specific agents or shrimp species, and some may not be effective at all.

2 Sano and Momoyama, this volume; Liao et al., this volume.

3 The prophylactic use of antibiotics was strongly discouraged by most of the workshop participants.

Preventing bacterial problems, it may also destroy viruses.

**Bacteria.** Increasing the rate of water exchange can prevent the build-up of high levels of bacteria in a hatchery system. It is also important to maintain optimum temperatures (Table 1). Furthermore, many believe that healthy microalgae blooms can inhibit the growth of potentially pathogenic bacteria.

The chemical pretreatments used to prevent bacterial problems include those listed above for preventing viruses. Additionally, a number of different chemicals may be added to the culture water to inhibit bacterial growth, including malachite green, EDTA and antibiotics.

Most of the participants were strongly against the prophylactic use of antibiotics because of the dangers of selecting
for and disseminating antibiotic-resistant strains of bacteria, and because it may be detrimental to the shrimp in the long run. However, antibiotics are used regularly in *P. monodon* hatcheries in the Philippines, Taiwan and elsewhere to prevent bacterial diseases. S.N. Chen noted that much effort is being expended in Taiwan looking for alternatives to antibiotics. In general, hatcheries that do not use antibiotics produce animals that perform very well during growout, but production is reduced in these hatcheries. Timothy Flegel added that oxytetracycline (OTC) is used successfully to improve production in *P. monodon* hatcheries in Thailand, even though most of the bacterial strains isolated from diseased shrimp are resistant to OTC. He also noted that the addition of very small amounts of OTC improves the hatching rate of *P. monodon* eggs. He hypothesized that OTC was having some other effect; one that was unrelated to its antibiotic activity.

*Penaeus vannamei*, in contrast to *P. monodon*, appears to be much more tolerant of high levels of bacteria. Whereas antibiotics are sometimes used prophylactically in Central and South America, their use is not ubiquitous. Antibiotics cannot legally be used either to prevent or to treat bacterial diseases in the United States, and the commercial representatives present indicated that, under most circumstances, they would not consider using antibiotics prophylactically even if such use were legal.

In the People’s Republic of China, where the use of antibiotics is not regulated, *P. chinensis* hatcheries do not routinely add antibiotics to the culture water. Finally, in Japan, the use of antibiotics is strictly regulated. Prefectural extension agents instruct farmers on the proper usage of antibiotics (only a few of which have government approval) and antibiotics are not used prophylactically.

Many hatchery managers have pinned their hopes on vaccines to prevent bacterial diseases. While bacterial vaccines for penaeid shrimp are not now widely available, several researchers at the workshop had developed and tested vaccines that could eventually be useful in hatcheries (e.g., see Laramore, this volume).

There are a number of ways to monitor hatcheries for bacteria, including doing total plate counts or TCBS counts of water and/or larval homogenates (Table 1). This is usually only practiced routinely in areas where there is a high density of farms and/or the water available is not of optimal quality. The widespread practice of rinsing or treating shrimp eggs and/or nauplii to remove bacteria before stocking falls under the category “animal management”. Similarly, *Artemia* nauplii, a common feed in shrimp hatcheries, are routinely rinsed before use to minimize the spread of bacteria.

Employing batch techniques can also help minimize bacterial problems (Table 1). And, since bacteria are ordinarily considered facultative pathogens of shrimp, another good way to prevent
infection is to minimize stress. This can be accomplished, in part, by providing adequate quantities of high-quality feed and by optimizing stocking density. Finally, there is some evidence (see D. Chen, this volume) that certain species of microalgae inhibit the growth of Vibrio spp. and other potential pathogens — adding these microalgae to the culture water should, therefore, lower the density of these bacterial species in the tanks.

Protozoa. Many of the methods used to prevent bacterial diseases also apply to diseases caused by protozoa (Table 1). In addition, chemicals such as Formalin and copper sulfate are also used in hatcheries, and it is important to monitor the culture water regularly with the aid of a microscope to monitor protozoan populations.

Fungi. Larval mycosis is the most well-known fungal disease afflicting larval penaeids. This disease is usually prevented by adding Treflan® to the rearing water, although chemicals such as EDTA and malachite green have also been used (Table 1).

Treatment

Viruses. Once a shrimp population has been infected with a virus, there is no way to get rid of the virus. There are, however, techniques that can maximize the performance of virus-infected animals in culture. The most commonly cited method is simply to minimize the stress placed on the animals. For example, in the past, Nick Carpenter has effectively “managed around” BP at his P. vannamei hatchery by lowering shrimp densities (Table 2). In addition, monitoring the level of infection can help in making management decisions.

Bacteria. Increasing water exchange and optimizing temperature are water management techniques that can be used to treat diseases of bacterial etiology (Table 2). Antibiotics, of course, may also be effective, but, as Tokuo Sano noted, treatment should be preceded by sensitivity analyses and determination of minimum inhibitory concentrations. In Ecuador and elsewhere, hatchery managers are adding 1 - 10 ppm sucrose to culture tanks to encourage the growth of bacteria that might outcompete potentially pathogenic species and strains. Although the technique is still being refined, shifts in the types of bacteria revealed in TCBS counts have been documented. Sucrose is also being used in growout, but its effect in ponds is more difficult to document. Similarly, some culturists in Asia and in the Western Hemisphere are adding so-called “beneficial” bacteria, or “probiotics” to hatchery tanks, and even to growout ponds, in the hope that they will prosper and outcompete Vibrio spp. and other potential pathogens.

Protozoa. Flushing to lower the densities of microbes on which epicommensal protozoa feed is one management method for protozoan fouling disease (Table 2). Alternatively, chemicals such as copper sulfate, malachite green and EDTA can be added to tanks. In fact,
### Table 2. Treating hatchery diseases in the Americas and Asia.

<table>
<thead>
<tr>
<th>Potential pathogens</th>
<th>Water management</th>
<th>Chemicals</th>
<th>Monitoring</th>
<th>Animal management</th>
<th>Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Viruses</td>
<td>None.</td>
<td>None.</td>
<td>Periodic diagnostic screening to make treatment decisions.</td>
<td>Avoid stress, lower or optimize densities.</td>
<td></td>
</tr>
<tr>
<td>Bacteria</td>
<td>Increase water exchange, optimize temperature.</td>
<td>Antibiotics, 1 - 10 ppm sucrose (to encourage growth of &quot;beneficial&quot; bacteria.</td>
<td>Antibiotic sensitivity profiles to make treatment decisions.</td>
<td>Do not overfeed if animals are sick.</td>
<td></td>
</tr>
<tr>
<td>Protozoa</td>
<td>Increase rate of exchange.</td>
<td>Teflan®, copper sulfate, malachite green, EDTA.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fungi</td>
<td>Increase rate of exchange.</td>
<td>Copper sulfate, malachite green, EDTA.</td>
<td></td>
<td>Lower density.</td>
<td></td>
</tr>
</tbody>
</table>

Cutrine®-Plus, a copper sulfate compound, is the only substance that has received formal approval in the United States to treat cultured shrimp (see Bell, this volume).

**Fungi.** Larval mycosis is variously treated by increasing the rate of water exchange, by using chemicals such as malachite green and EDTA, and by lowering larval densities (Table 2).

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**Discussion Group D: Disease Diagnosis**

One of the key disease-related issues facing the global shrimp industry is certifying stock and hatcheries as SPF. For example, stock that is certified as "virus-free" is worthless without knowledge of the diagnostic procedures used to detect viruses. Furthermore, all economically important infectious agents should be targeted, not only viruses. Another topic discussed was the development of new diagnostic techniques and guidelines for investigating idiopathic observations and cases in which there are multiple pathogens.

**Standardization of Diagnostic Procedures and Certification of Stocks, Pathologists and Facilities**

With currently available technology, how do we standardize diagnostic procedures? Then, once we have standard procedures, how do we certify diagnosticians, and how do we certify shrimp stocks and culture facilities as SPF? Several of the researchers present were in favor of a committee approach; form-
Table 3. Recommendations.

1. Develop priority pathogens list containing pathogens that are economically or ecologically significant, that can be diagnosed with existing technology.
   - Should we apply certification? Where?
   - What will be the criteria for sensitivity (what level is acceptable or should it be zero tolerance)?

2. Select and organize committees comprised from members of the following: the World Aquaculture Society, the Asian Fisheries Society, the Office of Internationale Epizooties and the European Association of Fish Pathologists. Committees will review and standardize diagnostic procedures.
   - Committee must be representative.
   - Take advantage of computer networking to alleviate need for many formal meetings.
   - Handbooks need to be developed by committees and so do SPP procedures.

3. Reference labs. Specimen exchange will be very helpful in the diagnosis of certain diseases.

ing one or more representative committees of experts to:

- Develop or endorse handbooks that contain detailed diagnostic procedures; and

- Develop or endorse guidelines for certifying diagnosticians, shrimp stocks and culture facilities (Table 3).

Perhaps committees could be formed within each of the following organizations: the World Aquaculture Society, the Office Internationale des Epizooties, the European Association of Fish Pathologists and the Asian Fisheries Society.

Regional committees, it was decided, would probably be needed to develop lists of pathogens to be excluded from stocks. Different species and various regions are expected to have different lists. For example, if a certain pathogen is endemic to an area, that is, present in the wild shrimp population, it would be impractical to exclude that pathogen from that region.

Research and Development on New Diagnostic Techniques

Tissue Culture. There was general agreement that much more needs to be done in the area of shrimp tissue culture. The lack of progress in this area has hindered the development of new diagnostic techniques.

Idiopathic Syndromes. Some participants believed that the phrases "idiopathic lesions" and "idiopathic syndromes" are overused in the scientific literature. Furthermore, diagnosticians would benefit from a standard set of steps to be used to investigate idiopathic observations in cultured shrimp.

Multiple Pathogens. Similarly, diagnosticians often encounter shrimp that contain multiple pathogens. In this case, it is very difficult to determine if one pathogen is more important than
the other(s), and if so, which one(s). Perhaps a set of guidelines could be
developed to assist researchers who
encounter these cases.

Morphological Diagnosis vs. Etiological Diagnosis. Citing Texas as one
example, Ken Johnson observed that
hepatopancreatic granulomas are being
reported with increasing frequency in
many culture areas. Because shrimp
have a nonspecific immune response,
these granulomas are generally re-
garded to be caused by a number of
unrelated factors. In many cases, how-
ever, it is impossible to determine the
etiological agent. It is extremely impor-
tant for pathologists to carefully report
the structural changes they observe,
that is, to make a morphological diag-
nosis, if the disease agent is unknown.

Discussion Group E: Quarantine/SPF

Quarantine

Country Descriptions. To begin, the
group heard descriptions of the inspec-
tion/quarantine procedures used in Ha-
waii, South Korea, and the People’s
Republic of China.

In Hawaii, permit requirements for im-
ported shrimp have drastically reduced
introductions in the past seven years.
Importers are required to quarantine
imported stock, and, because the like-
lihood that the shrimp will carry obli-
gate, exotic pathogens is so great, shrimp introductions to Hawaii are
now rare. Most producers are not will-
ing to risk the financial losses that
would result from an infected ship-
ment.

In South Korea, fish health inspections
are conducted under the auspices of the
National Fisheries Research and Devel-
opment Agency. A serious hindrance,
however, is the lack of a specialized
quarantine facility. Right now, patholo-
gists check imported fish for bacteria
and parasites; they presently lack the
means to test for viruses.

There are also standardized procedures
in the People’s Republic of China for
importing shrimp stocks. Implementation,
however, is sometimes difficult
because the regulations are too broad.

Purpose. There are at least two rea-
sons to implement quarantine proce-
dures. First of all, many countries are
interested in protecting native shrimp
populations from diseases that may be
carried by imported shrimp. Quarant-
ine procedures may not be effective in
attaining this goal, however, if there are
nearby countries that import and cul-
ture shrimp without regard to their
disease status. Secondly, quarantining
can protect the shrimp culture industry
from the potentially devastating effects
of new pathogens.

Implementation. The heart of the prob-
lem lies in implementing quarantine
procedures without crippling the
shrimp culture industry within a given
country or region. Many of those pre-
sent at this session were in favor of
establishing reliable domestic supplies of SPF stocks prior to the adoption of regulations. Another issue raised was the need for international cooperation in establishing workable, reasonable quarantine procedures.

Furthermore, as was pointed out in the previous discussion, before one can begin to certify stocks and hatcheries on a large scale, standard diagnostic procedures must be in place, and there must also be qualified diagnosticians to use those procedures.

In a related problem, how should we develop priority pathogen lists? This issue was touched on in the previous discussion. Some researchers wondered whether enough is known about the geographic ranges of pathogens to develop pathogen lists. Furthermore, any pathogens on an exclusion list must be able to be diagnosed with certainty. Other participants were worried that priority pathogen lists would be misused by governments in some countries and become regulatory lists.

The SPF Issue

Motivations. There are a number of reasons countries or groups might want to develop SPF shrimp stocks. One is the shortage of broodstock in some areas. Domesticated stocks are needed because wild sources of high-quality broodstock are becoming scarce. Logically, if one is going to begin a domestic stock program, he or she should begin with SPF animals. One could also argue that domesticated stocks are needed so that genetically "superior" strains of animals can be developed. Alternatively, some companies simply want to have a reliable supply of high-health animals for growout. Finally, other companies may be motivated by the economic incentive of selling high-health seed to other farms.

Approaches to Certification. Finally, when it is time to certify animals and hatcheries with regard to their pathogen status, what approach should be taken? Some fish hatcheries are classified based on the number of pathogens present in their stocks. For example, a Class A hatchery may contain SPF animals, whereas animals in a Class B hatchery might carry one known pathogen, and so on.

It may also be desirable to categorize the pathogens themselves. For a given area, disease agents might be divided into groups based on 1) the presence or absence of the agent in the natural environment, and 2) the threat posed by the agent in question. Finding answers to the above mentioned questions for all of the known shrimp pathogens affecting all the various species and culture regions will certainly require a great deal of study. The Fish Disease Commission of the Office International des Epizooties has already begun to develop a list of excludable fish, shrimp and mollusc pathogens (see Sano and Momoyama, this volume).
Table 4. Overall disease-related concerns, in order of decreasing importance.

1. The need for SPF stocks.
2. The use/abuse of chemotherapeutants, including antibiotics.
3. The need for rapid diagnostic techniques.
4. The natural environment — the need for ecological studies.
5. Problems associated with movement of shrimp.
7. Bacterial diseases.
8. Vaccines/preventives

Discussion Group F: Open Session and Wrap-Up

This session was devoted to prioritizing the concerns raised in Discussion Group A. Two summary lists were made that reflected 1) the participants' overall disease-related concerns, and 2) their priorities for future disease research. There was some overlap; for example, the need for SPF stocks was both a research priority and an overall concern. Everyone was asked to individually prioritize the issues on the two lists. The results are in Tables 4 and 5.

The need for SPF stocks was ranked highest among the overall shrimp disease-related concerns and was also second on the research priority list. This reflects, in large part, the perceived threat of viral diseases to the global shrimp culture industry, and the need for a reliable supply of seed. The use or abuse of chemotherapeutants was the second most important overall concern. In particular, the prophylactic use of antibiotics and the presence of drug residues in harvested shrimp were at issue. Third on the list was the need for rapid diagnostic techniques. Those present emphasized the need for two different types of techniques, those that could be used on farms and others that could be applied at “minimal clinical labs.”

The need for rapid diagnostic techniques was also the highest research priority (Table 5). SPF stocks were the second priority, and the need for more studies on probiotics was third. Finally, the commercial representatives stated that they wanted more research efforts to be directed toward developing more and better chemical preventives, including vaccines (number eight and four on the overall concerns and research priority lists, respectively). Better disinfection methods are needed, in addition to antibiotics that are designed for aquaculture, and immunostimulants.

Table 5. Research priorities, in order of decreasing importance.

1. Rapid diagnostic techniques.
2. SPF stocks.
3. Probiotics.
4. Vaccines/preventives
5. Effects of disease on wild shrimp populations
Literature Cited


Appendices
Appendix I: Workshop Participants

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U.S.A.
Appendix II: Agenda

April 27, 1992
East-West Center, Jefferson Hall, Asia Room

8:30 am Dr. Paul Bienfang
The Oceanic Institute
Introduction and Welcome

8:45 am Dr. Mohammed Shariff
Universiti Pertanian, Malaysia
An Overview of the Shrimp Disease Situation in Asia

9:15 am Dr. Donald Lightner
University of Arizona
Arizona, USA
New Developments in Penaeid Virology: Application of Biotechnology in Research and Disease Diagnosis for Shrimp Viruses of Concern in the Americas

10:00 am Dr. Jeffrey Lotz
Gulf Coast Research Lab
Mississippi, USA
Developing Specific-Pathogen-Free (SPF) Animal Populations for Use in Aquaculture: A Case Study for IHHN Virus of Penaeid Shrimp

10:45 am Dr. James Brock
Anuenue Fisheries Research Center
Hawaii, USA
Current Diagnostic Procedures for Diseases of Marine Shrimp

11:30 am Dr. José Natividad
Bureau of Fisheries and Aquatic Res.
Philippines
Prevalence and Geographical Distribution of Penaeus monodon baculovirus (MBV) and Other Diseases in Hatchery-Reared and Pond-Cultured Giant Tiger Prawn (Penaeus monodon Fabricius) in the Philippines

12:30 pm LUNCH—Garden level, Jefferson Hall

2:00 pm Discussion Group A — Sarimanok Room (Third floor)
Country by Country Concerns

3:30 pm Discussion Group B — Sarimanok Room
Prevention and Treatment of Diseases in Growout

5:00 pm RETURN TO HOTEL

6:30 pm DINNER — The Oceanarium
2490 Kalakaua Ave.
Pacific Beach Hotel
April 28, 1992
East-West Center, Jefferson Hall, Arts Room

8:30 am  Dr. S.N. Chen
National University of Taiwan
Republic of China

9:15 am  Dr. James Wyban
The Oceanic Institute
Hawaii, USA

10:00 am Dr. Kazuo Momoyama
Naikai Fisheries Exp. Station
Japan

10:45 am Mr. Nick Carpenter
Amorient Aquafarm
Hawaii, USA

11:30 am Ms. Myong Ae Park
Natl. Fisheries Res. & Devel. Agency
Republic of Korea

8:30 am  Studies on the Epizootiology and Pathogenicity of Bacterial Infection in the Cultured Giant Tiger Prawn, Penaeus monodon, in Taiwan

9:15 am  Selected Breeding of SPF Shrimp for High Health and Increased Growth

10:00 am  Viral Diseases of Cultured Penaeid Shrimp in Japan

10:45 am  A Comparison of Virus-Diseased vs. SPF Penaeus vannamei on a Commercial Scale in Hawaii

11:30 am  An Overview of the Shrimp Disease Situation and Diagnostic Techniques and Methods of Treatment Used on Shrimp Farms in Korea

12:30 pm  LUNCH—Garden level, Jefferson Hall

1:30 pm  GROUP PHOTO — Garden behind Jefferson Hall

2:00 pm  Discussion Group C — Sarimanok Room
Prevention and Treatment of Diseases in Hatcheries

3:30 pm  Discussion Group D — Sarimanok Room
Disease Diagnosis

5:00 pm  RETURN TO HOTEL

6:30 pm  DINNER — Camellia Restaurant
2460 Koa Ave.
Waikiki Resort Hotel
April 29, 1992
East-West Center, Jefferson Hall, Asia Room

8:30 am  Dr. Tokuo Sano  
Tokyo University of Fisheries  
Japan

9:15 am  Mr. Fritz Jaenike  
Harlingen Shrimp Farms, Ltd.  
Texas, USA

10:00 am  Dr. Brad LeaMaster  
The Oceanic Institute  
Hawaii, USA

10:45 am  Professor Dou Chen  
Academia Sinica Inst. of Oceanology  
People’s Republic of China

11:30 am  Dr. S. Ken Johnson  
Texas A & M University  
Texas, USA

12:30 pm  LUNCH—Garden level, Jefferson Hall

1:30 pm  RETURN TO HOTEL — Free time

Baculoviral Infections of Cultured Shrimp in Japan

Shrimp Production in Texas Using SPF Stocks

Shrimp Health Management Procedures

An Overview of the Shrimp Disease Situation, Diagnostic Techniques and Methods of Treatment Used on Shrimp Farms in China

A Review of the Regulatory Issues Related to Treatment of Penaeid Shrimp Diseases in Texas
<table>
<thead>
<tr>
<th>Time</th>
<th>Speaker</th>
<th>Topic</th>
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<tbody>
<tr>
<td>8:30 am</td>
<td>Dr. Rolland Laramore</td>
<td>Shrimp Culture Technologies Inc.: Implementing Research to Improve Shrimp Genetics and Health</td>
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<td>Shrimp Culture Technologies Inc.</td>
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<td>Florida, USA</td>
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<td>9:15 am</td>
<td>Dr. Timothy Flegel</td>
<td>Occurrence, Diagnosis and Treatment of Shrimp Diseases in Thailand</td>
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<td>Thailand</td>
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<td>10:00 am</td>
<td>Dr. Carl Sindermann</td>
<td>Precautions to be Taken in Importing and Culturing Non-Native Shrimp</td>
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<td>National Marine Fisheries Service</td>
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<td>Maryland, USA</td>
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<td>10:45 am</td>
<td>Mr. Thomas Bell</td>
<td>Drugs and Chemotherapeutants for Shrimp Diseases: Their Present Status in the USA, With an Overview of Research and Approval Processes</td>
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<td>University of Arizona</td>
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<td>Arizona, USA</td>
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<tr>
<td>11:30 am</td>
<td>Mr. Cheng-Feng Chang</td>
<td>Diseases of Grass Prawn (Penaeus monodon) in Taiwan: A Review from 1977 to 1991</td>
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<td>Tungkang Marine Laboratory</td>
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<td>Republic of China</td>
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<tr>
<td>12:30 pm</td>
<td>LUNCH—Garden level, Jefferson Hall</td>
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<td>2:00 pm</td>
<td>Discussion Group E — Sarimanok Room</td>
<td>Quarantine/SPF</td>
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<td>3:30 pm</td>
<td>Discussion Group F — Sarimanok Room</td>
<td>Open Session and Wrap-up</td>
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<tr>
<td>5:00 pm</td>
<td>RETURN TO HOTEL</td>
<td></td>
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<tr>
<td>6:30 pm</td>
<td>DINNER —— Cannon Club</td>
<td>Diamond Head Road</td>
</tr>
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May 1, 1992
Optional Tour of Aquaculture Facilities on Oahu

7:45 am    DEPART HOTEL
8:30 am    The Oceanic Institute
11:30 am   Mariculture Research and Training Center
1:00 pm    LUNCH — Pat's at Punaluu
2:30 pm    Amorient Aquafarm
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