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# Frequently Asked Questions (FAQs) Pertaining to *Penaeus vannamei* Shrimp Farming



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### **Foreword**

Pacific white shrimp *Penaeus vannamei* has emerged as the choice of the Indian shrimp farming since its introduction in 2009. The culture of P. vannamei has been in a progressive pace and its production has reached an all-time high of 3,51,413 metric tonnes in the year 2014-15 (MPEDA, 2016). Presently P. vannamei is being farmed from very low to oceanic salinities with different levels of technology adoption ranging from extensive, zero water exchange to biofloc based intensive systems. Equally *P. vannamei* farming is also beset with production risks of disease and pond health issues at every stage of farming and our farmers need to be sensitized about these risk factors and adoption of appropriate management practices. The ICAR-Central Institute of Brackishwater Aquaculture (ICAR-CIBA), Chennai played the pivotal and timely role in the introduction and farming of the exotic white shrimp *P.vannamei* in conducting risk assessment & developing guidelines for biosecurity, seed production and farming. Similarly, ICAR-CIBA has been monitoring the farming of *P. vannamei* across the country through its flagship programmes on National Surveillance and All India Network project on Fish Health with an active support of National Fisheries Development Board (NFDB) and the Indian Council of Agriculture (ICAR) from time to time. Likewise, ICAR-CIBA since the introduction of *P. vannamei* shrimp has been conducting sensitization workshops and capacity enhancement programmes for the fisheries extension officers of the coastal states and farmers to enhance their capacities on the emerging issues in production systems, providing technological support and guidelines to deal with them. As part of our capacity development intervention this publication on "Frequently Asked Questions (FAOs) Pertaining to Penaeus vannamei Shrimp Farming" along with answers is brought out based on the queries collected from the shrimp farmers and interactions with farmers across the coastal states. Authors deserve appreciation for their painstaking efforts and bringing out this farmer friendly publication. I am sure this book will be of immense use to the field level extension workers as well as the shrimp farmers in the country.

> K.K.VIJAYAN DIRECTOR

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### I. POND PREPARATION

1. How many days of pond drying between two cultures is required in *P. vannamei* farming?

After the final harvest of a crop, sun drying of pond bottom for 3-4 weeks is essential till it cracks or loses its moisture content. Drying and cracking of pond bottom helps in microbial decomposition of soil organic matter and mineralization of organic nutrients (nitrogen and phosphorus). If the previous culture was affected by white spot syndrome virus, it has been



Adequately dried pond

observed that the virus remains in the soil for 19-21days in drainable ponds whereas in case of non-drainable ponds, it remained infective for 35 days. In such cases, 5-6 weeks of drying is recommended to eradicate the pathogens. Moreover, screening of pond sediment is also essential to detect the presence of viral pathogens in such ponds. Further, investigations on duration of drying period revealed that farms practicing drying duration of 3, 5 and 10 days were affected with RMS and white gut diseases and harvested within 60 to 70 days of stocking whereas the farms that adopted a drying period of 30 and 45 days had successful harvest.

#### 2. What measures need to be taken to disinfect the pond bottom in case of non-drainable ponds?

In case of completely non-drainable ponds, the accumulated black soil in the pond bottom is flushed in the form of thin slurry using a sludge pump. Organic biodegradable piscicides like Mahua Oil Cake (100-150 ppm) or Tea Seed Cake (15-20 ppm) or chlorine @ 20 ppm can be used for elimination of unwanted organisms. Formalin or potassium permanganate or povodine iodine can also be used to kill the bacteria and external parasites as these compounds are degradable within the system and do



Non-drainable pond

not cause water pollution. Calcium Oxide (CaO) is recommended for enhancing the oxidation of organic matter and also is a disinfectant.

# 3. Is scraping of pond bottom essential? What would happen if a farmer does not do so?

Organic wastes in the form of a black layer at the pond bottom is formed due to the accumulation of decayed uneaten feed, dead and decaying plankton/algae and fecal matter of shrimp during culture. This type of waste material releases toxic organic metabolites like ammonia and hydrogen sulphide into the pond water and causes stress or death to the shrimp. Therefore,



Scrapping of pond bottom

this black soil layer to a level of 5-10 cm depth should be scraped and dumped away from the farm. If a farmer does not remove the black soil before starting the next crop, the possibility of having higher toxic metabolites at the pond bottom remains very high which would affect the pond environment thereby causing stress to shrimp.

### Whether ploughing before and after liming is necessary? What type of plough should be used?

The main purpose of ploughing is to expose the black soil underneath the bottom to sunlight and atmospheric oxygen. This results in the organic wastes in the pond bottom getting oxidized and making them as available nutrients. Presence of moisture in soil (i.e., under wet soil conditions) during ploughing, allows bacteria to work better in breaking down the black organic matter, thus making the ploughing process more effective. Ploughing before liming is more useful and Ploughing of pond bottom using disc plough



tilling after liming could enhance the soil pH. Ploughing the bottom soil with cultivator first and thereafter with rotavator proved better for oxidation of the soil and enhanced the availability of nutrients, released obnoxious gases and decomposition was greater. Rotavator makes smaller lumps of soil which provides better oxidation and minimizes seepage.

### How should one decide the disinfection dose for initial pond water? How should bleaching be done?

Disinfection of initial water taken to the pond is essential to kill unwanted organisms, pathogens and their hosts. Otherwise there is every chance that vectors of pathogens could enter through the water. Generally 20-30 ppm chlorine (600 kg of bleaching powder/ha) is used as no microbes or its hosts could survive such a chlorination dose. However, ascertaining the organic load in the source water (chlorine demand) and chlorine percentage in the bleaching powder would be better indicators to decide the chlorination dose.



Disinfection of initial pond water

### Does bleaching affect the efficiency of minerals present in the water during disinfection?

No. Disinfection of pond water with bleaching powder will not affect the efficiency of minerals present in the water.

#### Does the ground water also require disinfection? 7.

Disinfection is carried out to kill unwanted organisms, pathogens and their hosts. Since bore well water may not have any of these, disinfection is not required for ground water.

#### 8. Do we need to apply lime after disinfecting the initial pond water to adjust the pH?

Liming of shrimp ponds is done to neutralize soil acidity, increase the total alkalinity, total hardness concentration and enhance the primary food productivity in the pond water. When the soil pH is below 7 and the total alkalinity is less than 60 ppm liming is required. Agricultural limestone (CaCO<sub>3</sub>) is a safe product. Extra care is required if lime is applied in the form of burnt lime (CaO) and hydrated lime (calcium hydroxide<sub>3</sub>. If they are excessively used, the pH would tend to be very high which is dangerous. To increase the pH by 0.1, we need to apply approximately 50 kg of CaO or Ca(OH)<sub>2</sub> or 100 kg of CaCO<sub>3</sub> If the soil pH is more than 7 and the total alkalinity

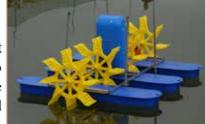


Liming of pond bottom

more than 60 ppm application of lime is not necessary. In case of slight adjustment of pH during pre-stocking preparations, dolomite (calcium and magnesium carbonate) can be used this would help in plankton development. In low saline waters, liming can improve phosphorus availability and enhance the pond productivity.

### 9. Is aeration before stocking is necessary?

Farmers apply fermented organic juice for the development of phytoplankton and minerals in case of low saline cultures to keep the optimum mineral requirements. In order to mix these uniformly throughout the pond, minimum aeration is required with at least 2 paddle wheel aerators at the corners of the pond.



Paddle wheel aerator

# 10. How does application of fermented juice help in improving the water quality?

Application of fermented juice enhances the population of beneficial bacteria. The bacteria require protein for its metabolic activity and the protein is synthesized by using the ammoniacal nitrogen present in the pond water. Thereby it reduces the ammonia level in the pond and improves water quality. Further, the fermented juice helps in lowering the high



Application of fermented juice

pH to the desired level and enhances the development of plankton food.

# 11. Farmers apply molasses instead of fermented juice. Which one is more effective for water culture?

Molasses contain more carbohydrates thereby enabling the growth of beneficial bacteria in higher density. Since molasses is not freely available to the farmers coupled with a higher requirement of aeration after its application, the use of fermented juice is more acceptable to the farmers.

### 12. What is C: N ratio and its significance in *P. vannamei* shrimp farming?

C: N ratio is the ratio of carbon and nitrogen in the pond water and a ratio of 10:1 normally exists in shrimp ponds. However, in case of *P. vannamei* due to the high stocking densities, more nitrogenous waste is generated in the form of ammonia and nitrite which deteriorates the water quality. To minimize this problem, carbon sources in the form of fermented juice or molasses is

applied which act as a medium for the growth of beneficial bacteria enhancing the C: N ratio to the level of 20:1. These beneficial bacteria require nitrogen to produce proteins for their metabolic activity and development and absorb nitrogen from the ammonia or from the fecal waste which is the source of nitrogen. Due to this, the harmful ammonia and nitrite are converted to proteins by the bacteria. Further, this bacteria forms "bio-floc" by adhering to the dead cells, inert materials thereby becomes the feed for



Molasses

*P. vannamei* shrimp. Hence, the feed quantity requirement can also be reduced considerably. The *C*: N ratio needs to be checked fortnightly and has to be maintained by periodical addition of fermented juice or molasses. It is generally useful for Biofloc system.

### 13. What is biofloc and explain the procedure to develop and maintain the same in simple terms?

Biofloc is a consortium of macro-aggregates consisting of diatoms, macroalgae, fecal pellets, exoskeleton, remnants of dead organisms, bacteria, protest and invertebrates. A 10-15ml/litre level of (imhoff cone) biofloc should be maintained in the shrimp pond. Addition of organic carbon in the form of fermented solution or molasses acts as a medium for the enhanced production of beneficial bacteria which absorb inorganic nitrogen from the pond water immobilizing the potentially toxic ammonia



Biofloc based Shrimp pond

and nitrite. This system reduces the total ammonia nitrogen (TAN) thereby decreasing the organic load at the pond bottom which helps to keep hydrogen sulphide under control. Typically a biofloc pond starts as an autotrophic/algal dominated system and after a few days the water turns brown and foamy as floc develops and the system shifts to a heterotrophic system without much algae. The advantages of this system are: zero/minimal water exchange system, *insitu* bioremediation to maintain water quality, augmentation of natural food and improvement of FCR, non-use of chemicals and other medicines and negligible environmental impact. Protein requirements in feed could be brought down in these systems and the cost of production in terms of feed would be less. Biofloc system may have 5-10% excess carrying capacity, gain by 2 g in ABW, low FCR and lower production cost by 10-15%. However, it requires, high aeration up to 28HP/ha, continuous (24x7) electricity, full/semi HDPE lined ponds and trained technicians.

# 14. Whether biofloc system is feasible for a small scale *P. vannamei* culture?

In case of small scale farming with a moderate stocking density of 20-30 PL/m² it may not be required. Optimum stocking, proper feed management and efficient water quality (pH, DO, minerals, bottom metabolites) management are sufficient for problem-free shrimp farming.



Shrimp pond with central drainage

#### 15. Whether *P. vannamei* farming requires deep ponds with a central drainage system?

White shrimp *P. vannamei* is a column dweller and not a bottom dweller like tiger shrimp. Further, in case of *P. vannamei*, high stocking density up to 60 PL/m<sup>2</sup> is practiced, hence it requires deep ponds having 1.5 m water column. Central drainage system would help in removing accumulated sludge.

# 16. What is the carrying capacity of a shrimp pond and how do you arrive at an optimum stocking density for a pond culturing *P. vannamei*?

It is the optimal biomass (tonnes of shrimp/ha) holding capacity of the pond. Carrying capacity of a shrimp pond is dependent upon many factors like biosecurity, electricity, aeration, water quality, quality of inputs, practices followed, technical and management capacity of the individual farmer and other extraneous factors. Based on the experience, a farmer should decide the optimum stocking density for the pond within the permitted level.

### 17. Whether *P. vannamei* farming needs a modification/alteration in pond design? If yes, how?

Yes. *P. vannamei* culture pond should have a water column of 1.5 m depth. Since *P. vannamei* is stocked at higher densities, it requires higher level of aeration for a longer time for water circulation and maintaining the optimum required level. Continuous aeration might cause erosion of bunds and hence adequate compaction of the dykes is essential. HDPE lining of dykes would minimize erosion. Besides, full time (24x7) electricity, generator back up and biosecurity measures are essential.



HDPE lined shrimp pond

### 18. How does ageing of pond water prior to stocking help in culture and what are its advantages?

The white spot virus can survive as free living form in water up to 12 days. Ageing of pond water after chlorination and de-chlorination will help in removing them. After 12 days, addition of fermented juice is beneficial due to enhanced bacterial growth in adequate quantity and has abundant primary productivity.

### II. SEED SELECTION AND STOCKING

### 19. What is SPF *P. vannamei* shrimp?

The seeds produced from the Specific pathogen free (SPF) *P. vannamei* shrimp brood stock are known as SPF *P. vannamei*. These SPF broodstocks are developed under rigorous quarantine and screening of pathogens over several generations from the SPF mother/founder broodstocks. Countries or regions which still do not have this species can be reasonably sure that the importation of SPF animals will not result in the introduction of the specified pathogens for which the animal is declared



P. vannamei shrimp seed

"free". This does not, however, guarantee against the animal being infected with known /unknown pathogens which are not screened against during the SPF programme.

#### 20. How far SPF *P. vannamei* status last long?

SPF means that the animals have been assured of being free from specific pathogens at the particular point of time. It is a wrong belief that SPF animals are resistant to and cannot become infected by any viral pathogens that shrimp encounter during culture. Once the animals are removed from the SPF production facilities, they can no-longer be referred to as SPF, even though they may remain pathogen free. Once outside the SPF facility, the shrimp may be designated as High Health



P. vannamei shrimp broodstock

(HH) as they are now subject to a greater risk of infection, but only if they are placed into a well-established facility with a history of disease surveillance and biosecurity protocols. If the shrimp are put anywhere else, for example into a non-biosecure maturation unit, hatchery or farm, they can no longer be called SPF or HH as they are now exposed to a high risk of infection. It was reported that SPF *P. vannamei* broodstocks are easier to spawn than pond-raised brood stock and their offspring perform better in terms of higher survival rates.

# 21. What are all the parameters a farmer should look for while selecting, *P. vannamei* shrimp seeds for stocking?

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S.No	Parameter	Standard
1	Colour	light to dark brown
2	Activity	Very active
3	Feeding behaviour	Readily accept and eat feed
4	Gut	Full gut and with a tail muscle to hind gut ratio (MGR) of 4:1or more
5	Hepatopancreas	Developed in at least 90% of the sample given and full with oil globules.
6	Rostral spines	More than 5 should be observed
7	Body Length	12 mm and above
8	Size variation	Less than 10%
9	Appendages	Intact without any deformity
10	PCR screening for viral and bacterial diseases	Negative

S.No	Parameter	Standard
11	Pigmentation	Chromotophores well defined and located along the mid-ventral line
12	Stress test with survival	100% for salinity reduction and above 90% for formalin test.
13	PL stage	PL 12 and above. Adequate gill development will not be there in small sized PL.
14	WSSV, EHP	Absent based on Real time PCR – two step nested PCR.
15	Necrosis	Absent
16	Fouling	Clean without any fouling organism

It is to be noted that after the introduction of SPF *P. vannamei* in to the country and due to its increasing demand, both the hatchery operators and farmers are not testing the quality of the shrimp seed unlike they did for tiger shrimp. Hence, the shrimp farmers should adopt the following measures to ensure the selection of quality seed.

- The hatchery should have registration for *P. vannamei* seed production and import of brood stock.
- Physical stress test: Collect a larval sample (approximately 100 PLs) from the bottom of the identified larval tank put them in a tub and rotates the water. If many seeds are concentrating at the center of the tub, reject the animals from this larval tank.
- Salinity stress test: Collect about 100 PL in a beaker with animals from a tank having the optimum salinity. Pour equal quantity of freshwater and wait for half an hour. If you find any mortality reject the animals from that tank.



Shrimp seed quality checking

- Formalin stress test: Collect 100 PL of P. vannamei shrimp seed from the selected tank and put them in 100 ppm formalin water and wait for an hour. If more than 90% of the seed survive, select that batch.
- The hatchery should have quarantine, adequate bio-security, required infrastructure and follow the standard operating procedures fully.
- The seed selected should be from a single spawning and should be from the first or second spawning of the specified mother brooder. Seeds from mixed spawning lead to low survival and size variation.

- Seed should be PCR screened for known pathogens (WSSV, EMS etc.) before selection.
- After selecting the seed lot, it should be gradually acclimatized (2 to 3 salinity in a day) to the pond water salinity. Rapid acclimatization leads to stress and low survival in the farm.
- Obtain 100 PL from the selected stock and put in a hapa in the pond and if the lot has a survival of ≥ 90% only then should the farmer buy the seed. Otherwise choose another lot and repeat the process until the above condition is met.
- Farmers from a shrimp cluster should procure seeds collectively through their association from a reputed and trusted hatchery. They should stay in the hatchery to closely monitor the seed production process. PCR screen



Hapa rearing for survival assessment

- to closely monitor the seed production process. PCR screening of mother brooder before and after spawning would be extremely helpful in preventing diseases.
- 22. Is it possible to differentiate SPF seed vis-à-vis seed from a pond-reared brood stock? It is not possible to differentiate SPF and non-SPF seed by observation.
- 23. Whether nursery rearing enhances the survival and growth in *P. vannamei* farming? If so, give the details of nursery rearing?

It was observed that nursery rearing enhances the seed survival up to 95-99% by making the seed healthy and strong. Prepare the pond similar to that for grow out culture pond preparation with scraping, ploughing, liming, water in-letting, chlorination, de-chlorination, plankton production by applying dolomite, fermented juice, required minerals and probiotics. Stock quality *P. vannamei* PL 10-12 size @ 1000/m² and feed @ 1kg of starter feed per one lakh PL from the day one. Increase



Nursery rearing of P. vannamei

daily feeding rate @200 g per lakh seed. Give feed no.1 for initial 10 days, a combination of feed no.1 and 2 during 10-20 days and after 20 days provide feed no.2 only. Provide aeration for 4 hours in the forenoon and 8 hours from 10.00P.M. It was observed that a growth rate of 2.5 to 3 g is achieved in 25-30 days of nursery rearing. Water quality monitoring for DO, pH, bottom metabolites-ammonia, nitrite and  $H_2S$  is very crucial. After 30 days, shift the seed from the nursery to grow-out pond using drag or scoop net.

24. What are the reasons for low seed survival in *P. vannamei* ponds? How can you ensure better/higher survival rate?

Poor seed quality, poor acclimatization and poor water quality are the reasons for low survival. Select and procure quality seed by strictly following the CAA guidelines and the standard protocol. Gradual acclimatization of PL to pond salinity is very important and should be achieved to the level of 1-2 ppt per day at the hatchery itself before packing. Maintain the pond water quality parameters within the optimum range.

#### III. FEEDING AND FEED MANAGEMENT

25. Is it correct to use high protein feed initially and switch over to low protein feed? If yes, what is the advantage?

It is good to use a high protein feed during the initial 30 DOC (pre-starter & starter) since the PL/early juveniles require more protein (38%) than the juveniles and it gives better growth.

26. Is it correct to stop feeding by 7.00 PM? Would not the *P. vannamei* shrimp starve for feed during night hours?

Yes. *P. Vannamei* are diurnal feeders hence need more feeding in the day time and it is always advisable to complete the feeding by 7.00 P.M. Due to high stocking density, the chance of reduction in DO during the night time might result in stress to the animal and affect the feeding which results in less feed consumption and more wastage. This unutilized feed



Boat feeding

would deteriorate the water quality. Wide feed distribution with multiple feedings also helps in the breakdown of organic wastes by beneficial bacteria and reduces ammonia and prevents water quality problems.

27. What is the required quantity of feed to be kept in check tray for feed monitoring at different stages of culture?

Generally to start with (30 DOC) 4-6 g per kg of feed is kept in check tray for monitoring. As the animals grow, the quantity has to be increased slightly and it will reach a maximum level of 8-10 g per kg of feed in the final stages of culture (25-30 g ABW at 100-120 DOC).

28. Why does size variation occur in *P. vannamei* shrimp? How would you minimize the problem through feed management?

Seed quality is the major factor for size variation. If the quality of the seed is good and

it is well fed with a balanced diet, there is less chance for size variation. Underfeeding may lead to size variation. In this condition the dominant and active shrimp will consume enough quantity and would finish the feed quickly while the sluggish and less active shrimp will not get the required quantity resulting in decreased growth. This issue can be managed to a certain extent by means of gap feeding i.e giving the previous grade feed after one hour of routine feeding and mixing the



Size variation in P. vannamei shrimp

two different sizes of feed (currently using size and a lesser size). During the course of culture, the increase in body weight should correlate with the feed given and the FCR should be in the expected range. If there is any deviation, then the actual reason has to be explored and rectified. If the growth is disproportional then the pond requires adequate attention.

29. Is it correct to adopt feeding after one week of stocking only and how can we determine the feeding rate initially?

Initial feeding depends on the natural (plankton) productivity of the pond. As *P. vannamei* larvae are voracious feeders and are fed @ 6-7 times/ day at the hatchery, it is advisable to start feeding from the date of stocking itself.

30. What are all the aspects to be kept in mind while adopting the feed dispenser for *P. vannamei* pond?

Even though *P. vannamei* can come to any place for feeding, if it is habituated, it is advisable to keep the feed dispenser at the centre as it facilitates the animals from all sides to come and take the feed. However, the total quantity feed to be fed for the day should be uniformly distributed. The timing between the successive feedings and gap between the rotations are very important in feed dispensers.



Automatic feed dispenser

31. What time of feeding would be appropriate to mix probiotics/supplements for application?

The first meal of the day should be mixed with probiotics/supplements as the hungry animal would consume it fully.

32. How much of nutrients applied through feed are actually converted into biomass?

Generally aquatic organisms convert 25% of nitrogen and phosphorous and less than 10% of the carbohydrate matter applied in the feed to biomass. The remaining N, P and organic matter enters the pond as faecal matter, uneaten feed, ammonia, phosphate and carbon dioxide. For every kg of feed fed, about 30 g of ammonia will be excreted by shrimp into the pond. Ammonia and phosphate stimulate phytoplankton production and increase the organic matter as phytoplankton continually die and settle at the bottom as organic matter.

33. Whether shrimp feed can be evaluated physically? If yes how?

A good quality shrimp feed should have a fishy odour and the feed pellet has to be of uniform size. There should not be any clump formation and musty odour. The bottom of the bag should be free from fine powder.



P. vannamei shrimp feed

34. How can we ascertain the nutrient content of the feed used by a farmer?

About 200 g of the feed sample has to be collected and sent to the feed laboratory to evaluate the nutrients viz., protein, fat, fiber, total ash and moisture. If the values are lower than the values given in the feed bag,the feed is of lower quality.

35. What is the standard water stability time for *P. vannamei* shrimp feed?

Even though *P. vannamei* is a fast feeder and it will consume the feed quickly, feed should be water stable for a period of 1-2 hours.

#### 36. Can we use natural feedstuff like clams to improve the shrimp growth rate?

No. Since the *P. vannamei* shrimp is SPF and reared under strict biosecurity protocol, use of live/fresh natural feedstuffs are to be avoided. This will ensure safety to the animal as there is a possibility of introducing disease causing agents through these sources. Moreover, use of natural feedstuffs will lead to deterioration of water quality in the pond.

### 37. How long can the feed be stored?

A properly preserved feed can be stored safely for a period of 60-75 days. However it is always advisable to use freshly processed feed to the extent possible.

### 38. Can we use different brands of feed in a single culture?

There is no harm in using different brands as long as the quality is good and the shrimp consumes the feed resulting in good FCR. However if you want to change the feed, it has to be done gradually and not abruptly as the animal has to adapt to a particular feed.

### 39. Is there any farm-made feed available for shrimp?

*P. vannamei* shrimp can be fed with feed prepared at the farm. However the feed has to be prepared scientifically. The details of farm made feed can be obtained from the Central Institute of Brackishwater Aquaculture, Chennai.

### **SOIL AND WATER QUALITY MANAGEMENT**

# 40. What are ionized and unionized forms of ammonia and what is their significance in *P. vannamei* shrimp culture?

Ionized forms are the readily absorbable forms of minerals and nutrients by the shrimp. Ammonia is present in two forms, ionized (NH<sub>4</sub>) which is nontoxic, and the toxic form -unionized ammonia (NH<sub>3</sub>). The shrimp utilize only ionic forms for their metabolic activity. Unionized ammonia (NH<sub>3</sub>) has been reported to be more toxic than ionized ammonia (NH<sub>4</sub>) due to its ability to diffuse across cell membranes. The total of both the ionized and un-ionized forms is the total ammonia nitrogen or TAN. The concentration of these depends on water temperature and pH. Higher the water temperature and pH, the greater the concentration of the toxic form of ammonia (NH<sub>3</sub>).



Ammonia checking chart

# 41. Whether application of probiotics and minerals are necessary for phytoplankton development? If so what is their actual role?

Probiotics could help in regulating the micro flora of the pond water, control pathogenic microorganisms, enhance the decomposition of undesirable organic substances and improve the pond environment. Application of plankton promoters enhances the availability of nitrates and phosphorus which



Application of probiotics

are important for the planktonic growth. The beneficial bacterial strains inhibit the pathogenic bacteria and help in better survival. In case of low saline water shrimp culture, the pond water has low level of essential minerals like calcium, magnesium, sodium and potassium which might cause stress to the larvae by disturbing the osmo-ionic balance of the shrimp larvae. Beneficial bacteria are helpful in nutrient recycling and organic matter degradation and thus keep the pond environment clean. Therefore, application of probiotics and minerals are essential.

# 42. Whether the lime applied helps in improving the plankton development also? Which lime material the best?

Yes. Finely crushed agricultural limestone is usually the best material to use at pH levels below 8.3. It is cost-effective and readily available. Both pond alkalinity and hardness can be increased by adding either CaCO<sub>3</sub> (agri-lime) or CaMg(CO<sub>3</sub>)<sub>2</sub> (dolomitic limestone). Both Agri-lime and dolomite are good. Dolomite which is calcium magnesium carbonate promotes plankton growth and helps in photosynthesis of planktons.



Dolomite lime

### 43. Why does the pH change in *P. vannamei* ponds?

Fluctuations in pH are the result of the interplay of photosynthesis and respiration. At night time, respiration increases CO<sub>2</sub> concentrations, creating carbonic acid and causing pH to fall. During the day, phytoplanktons absorb CO<sub>2</sub> for photosynthesis, causing pH to rise. Large daily changes in pH can stress aquatic animals. Most aquaculture species can live in a broad range of alkalinity concentrations, but the desired alkalinity for many animals is 50 mg/L or higher. Liming to raise total alkalinity to the required or preferred range buffers the water and reduces fluctuations in pH.

# 44. What are the optimal water quality parameters? How can we correct (low/high) them during fluctuation?

The optimal range of water quality parameters to be maintained in *P. vannamei* pond, reasons for fluctuations in the water quality parameters and their management measures are given in the Table.

Low DO and higher DO fluctuations, Low and high pH, high diurnal pH fluctuations, bottom metabolites – ammonia, nitrite and hydrogen sulphide, higher water temperature, alkalinity, shortage of minerals and their dis-proportions, low alkalinity and higher hardness are the water quality related problems in *P. vannamei* farming.

Parameter	Optimal Range	Reasons for increase/decrease	Management measures
Dissolved oxygen	> 4 mg/L	<ul> <li>Overstocking and excessive feeding rates.</li> <li>Excessive algae bloom and high respiration.</li> <li>Plankton die-off, BOD increases due to bacterial decomposition of algae</li> <li>Heavy rainfall/cloudy weather</li> <li>Long periods of hot weather</li> </ul>	<ul> <li>Proper feed management (ration &amp; timing)</li> <li>Proper aeration.</li> </ul>
Temperature	28-32°C	Prolonged sunny weather –     affects shrimp metabolism,     stress, low feed intake.	<ul><li>Water-topping</li><li>Better aeration</li></ul>
рН	7.5-8.5	<ul> <li>Increase due to warm weather</li> <li>Decreases during night due to respiration of planktons and increased CO<sub>2</sub> levels.</li> <li>Diurnal fluctuation should not be more than 0.1-0.2</li> </ul>	<ul> <li>Add lime stone.</li> <li>To reduce pH, carry out water exchange/apply jaggery solution or fermented juices.</li> </ul>
Total Ammonia Nitrogen (TAN)	< 1mg/L ( NH <sub>3</sub> :< 0.1 mg/L)	<ul> <li>High temp and high pH lead to toxic ammonia increase-causes gill damage.</li> <li>Excessive feeding, high water temp, high pH and low algal bloom.</li> </ul>	<ul> <li>Maintain proper algal bloom</li> <li>Decrease the feeding</li> </ul>
H <sub>2</sub> S	< 0.03mg/L	<ul> <li>By-product of breaking down of organic matter under anaerobic conditions</li> <li>( rotten egg smell)</li> <li>Deceased water pH</li> <li>Increase in temp</li> </ul>	<ul><li>Sludge removal</li><li>Proper drying and tilling</li></ul>
Salinity	28-32 ppt	High / Low temperature     Heavy rainfall	Water exchange/top up with ground water of suitable salinity.
Total Alkalinity	> 50 mg/L as CaCO <sub>3</sub>	Affects primary productivity and water pH	Prefer dolomite which increases alkalinity, water pH and enhances the availability of nutrients.

# 45. On what basis should a farmer decide the dosage and frequency of minerals application during culture?

Shrimp pond water should have mineral nutrients viz. calcium, magnesium and potassium in the ratio of 1:3:1 with a minimum of 200 ppm calcium. In full strength sea water, Na: K ratio is 28:1. It is observed that lowering the Na: K ratio by increasing the K concentration enhances the survival and growth of shrimp. The farmer should check the mineral concentration at weekly intervals and carry out the necessary measures if the levels are low.



Mineral - Magnesium chloride

# 46. What is the basis to decide the duration of aeration required for *P. vannamei* shrimp at different DOC?

Adequate DO is required for the higher rate of organic matter decomposition by aerobic microorganisms, support ammonia oxidation by denitrifying bacteria, maintain aerobic conditions at the sediment-water interface and better feed utilization. Aerators are required for optimizing the dissolved oxygen and circulation of the pond water. A general guideline for aeration duration is given in the Table.

Day of Culture	Duration
Up to 30 DOC	4-6 hours
30-60 DOC	6-8 hours
60-90 DOC	8-12 hours
Above 90 DOC	12-16 hours

After 30 DOC aerators should be added @ 1 HP per 300-400 kg of biomass. Aerators are operated 6-8 hours mostly during night hours initially and increased gradually to 10-12 hours during mid phase and 14-16 hours per day during the final phase.

### 47. Which type of aerators should be used for better results?

Several studies reported that paddle-wheel aerators or long arm paddle wheel aerators are efficient in providing aeration and circulation of water. It is also reported that long arm and spiral aerators can circulate oxygen to the pond bottom and are more efficient. The aerators should have a rotation per minute (RPM) of 90-120. It is expressed by the farmers that combining long arm paddle wheel and spiral aerators provided better efficiency in terms of circulation and dissolved oxygen



levels. Air diffusers (blowers) are also used for providing aeration Combined long arm and spiral aerator to ponds by placing circular aeration stones. It was reported that 1 HP air diffuser is equal to 3 HP aerators and is felt that air blowers along with minimum aerators could provide better performance as it minimises the number of aerators and cost of electricity.

#### 48. How can we position the aerators for better performance?

Aerators are to be placed in such a way that entire pond water is circulated in a circular motion sweeping all the waste material to the central part of the pond. Some farms adopted sludge pumps or central drainage system to suck the sludge deposited in the centre of the pond. Positioning of paddle wheel aerators diagonally at four corners and long arm paddle wheel /paddle wheel cum spiral aerators in the middle level position respectively helps in water



Positioning of aerators

circulation and increasing the aeration. The aerators should be placed 4-5 meter distance from the dykes to prevent erosion.

### 49. What is total alkalinity and how does it differ from hardness?

Alkalinity (the concentration of carbonates, bicarbonates and hydroxyl ions in water) is a measure of the buffering capacity of water to resist change in pH. Hardness is often defined as the sum of polyvalent cat-ion concentrations dissolved in the water. The most common polyvalent cat-ions in fresh water are calcium (Ca<sup>++</sup>) and magnesium (Mg<sup>++</sup>). Alkalinity is important for pH optimization and it is essential for plankton development, metabolic activity of the shrimp and affects bottom metabolites. Optimum alkalinity has direct relationship with animal growth and survival. Hardness is the overall concentration of divalent salts (calcium, magnesium, iron, etc.)

### 50. Whether *P. vannamei* shrimp needs micro nutrients too? If yes, what are they?

The enriched trace minerals/micronutrients have a significant impact on immune functions, disease resistance, and stress release of shrimp/ fish. The micro-elements are important components of hormones and enzymes, and serve as cofactors and/or activators of a variety of enzymes. The quantum of their requirement is relatively low and the requirement is taken care through the feeds. Hence, farmers need not apply trace elements separately.

# 51. What is the weekly growth rate for *P. vannamei* and what should be done in case of stunted growth?

A weekly growth rate of 1.3-1.6 g is optimum for *P. vannamei* and it depends on the seed quality, water quality and feeding efficiency. If the water quality is optimum and growth is not proportional to feed provided, then feeding behavior needs to be checked. Giving one or two doses of feed probiotic could be beneficial and if it is not improving the growth rate then harvesting would be the right choice.

Assessing shrimp growth

# 52. What are water soluble and insoluble forms of minerals in *P. vannamei* culture? Explain in simple terms.

The water soluble fraction of minerals is the bio-available form which is readily available to the shrimp. This bio-available form of minerals from most of the commercial mineral mixtures is very low and it is advisable to test the compounds before application.

### 53. Why are minerals so important for *P. vannamei* shrimp alone vis-à-vis tiger shrimp?

Minerals are important for both tiger and *P. vannamei* shrimps. Since *P. vannamei* is cultured in low saline waters which have a relatively low amount of minerals vis-à-vis brackish/saline waters, external supplementation mineral is required only for low saline waters.

- 54. What are the important roles of minerals in *P. vannamei* farming? What is the optimum requirement level of essential minerals?
  - Minerals are essential constituents of skeletal structures such as bones and teeth.
  - Minerals play a key role in the maintenance of osmotic pressure, and thus regulate the exchange of water and solutes within the animal body.
  - Minerals serve as structural constituents of soft tissues.
  - Minerals are essential for the transmission of nerve impulses and muscle contraction.
  - Minerals play a vital role in the acid-base equilibrium of the body thereby regulating the pH of the blood and other body fluids.
  - Minerals serve as essential components of many enzymes, vitamins, hormones, and respiratory pigments, or as cofactors in metabolism, catalysts and enzyme activators.

Mineral Name	Recommended level in water and feed		
Calcium	Above 150-200 mg/L in water ; 1.25 to 2% (max) in feed		
Magnesium	300-400 mg/L in water and 0.2% in feed.		
Potassium	150-200 mg/L in water and 0.5 to 1% in feed		
Sodium	0.5% in feed		
Phosphorous	1 to 1.5% in feed		
Chlorine	Depends on the salinity of the water		

# 55. How do the open and ground waters from the coastal and inland areas differ in mineral composition?

Mineral composition in open source waters from coastal areas is mainly dependent on water salinity. For example, Ca, Mg and Na are more in saline waters compared to low saline waters. The composition of underground bore-well waters depends on the location of the bore well and the depth from which water is extracted. For example, bore wells in coastal areas are fresh/low saline to high saline in nature. Recently shrimp farming has spread to inland saline arid and semi-arid regions of the country viz., Haryana and Rajasthan with the use of ground water. These waters are deficient in K and Mg unlike coastal ground waters.

# 56. Whether water source has a role in the availability of minerals like bore water, creek water, sea water etc?

Yes. The sea/brackishwater has adequate quantity of minerals. In case of low saline waters, they are relatively low in amount, hence require external supplementation. The minerals in

the pond water have certain key functions and contribute for the survival and growth. Among the minerals, calcium, magnesium and potassium are important for *P. vannamei* shrimp. The *P. vannamei* require calcium, magnesium and potassium in a ratio of 1:3:1 which is the ratio in sea water. We have to maintain this ratio in the low saline waters by adding the minerals externally. The minimum requirements of these minerals at 1 ppt salinity are given below and we need to multiply with the pond salinity.



Water quality test report

Minerals	Required level in 1 ppt salinity	Required level in 5 ppt salinity	Required level in 10 ppt salinity	Required level in 15 ppt salinity
Calcium (mg/L)	11.6	58.0	116.0	174.0
Magnesium (mg/L)	39.1	195.5	391.0	586.5
Potassium (mg/L)	10.7	53.5	107.0	160.5

#### 57. Do brackishwater ponds require mineral supplementation?

Test the brackishwater pond water for minerals and if required only, one should add minerals. Normally brackishwater has enough amounts of minerals and may not require mineral supplementation.

#### 58. Do you think that the mineral composition changes as per the salinity?

Yes. The mineral composition changes as per the source water. Generally the sea water and brackishwater bodies have adequate amount of required minerals in required proportion (ratios). The inland waters do not have the required level of minerals in right proportions.

- 59. Do you feel that pond age has any influence on the availability of minerals? Not confirmed. The investigations are in progress to test this hypothesis.
- 60. What are the important soil parameters which indicate the deteriorating conditions of the pond bottom?

Farmers can test the shrimp pond bottom condition without any laboratory analysis. The bottom soil can be scooped and tested for any bad smell. If there is a smell of a rotten egg, it indicates the bad condition of the soil having hydrogen sulphide. Technically the redox-potential of the soil can be measured on site which gives an indication of oxidation/reduction condition of soil. Organic carbon content of the soil can be estimated in the laboratory for the quantification of organic load accumulation on the pond bottom.



Symptom of mineral deficiency

### 61. What kind of symptoms do shrimp show which indicate mineral deficiency?

The common symptoms of mineral deficiency are anorexia, reduced vigour, poor feed intake, shrimp body cramp/bend, at the place of cramp the muscle becomes white and juveniles

are more susceptible to ratio differences between the important minerals (Na, K, Ca & Mg). Mineral deficiency in the form of either presence of lesser amount or not in correct proportion might hamper osmo-ionic regulations and metabolic activities. The shrimp will be under stress, low feed intake, water quality deterioration and prone to manifestation of diseases.



Turbidity in shrimp pond

#### **62.** Is high turbidity in pond waters harmful to shrimp?

Turbidity in pond waters is generally caused by the planktonic organisms and silt particles. The planktonic turbidity is good to shrimps. The turbidity due to suspended silt particles is harmful and undesirable for a healthy pond. Proper drying of pond bottom after the harvest with recommended pond preparation practices including the strengthening of dykes and provision of optimum aeration minimizes the eroding of soil particles and reduced turbidity.

# 63. Whether pH and minerals are associated? At what pH are the essential minerals available at a maximum level?

The mineral composition and pH are related. At neutral to slightly alkaline pH (between 7 to 7.5) the availability of P, Ca, K and Mg is maximum and functions of the mineral nutrients are also adequate. The micro nutrients whose requirements are very small are also available at a higher pH though their availability is more at low pH.

### 64. Whether water temperature has any role in the availability and functions of essential minerals?

The optimum temperature for *P. vannamei* is 28°C to 32°C. Continued high or lower temperature vis-à-vis the optimum level negatively affects the metabolic activity of the shrimp and their growth is disturbed. Higher temperature increases the salinity of the water evaporation of pond water and thus indirectly increases the mineral composition.

# 65. On what basis one should apply mineral nutrients in *P. vannamei* pond?

Shrimp farmers need to test their shrimp pond water once in 10-15 days for their mineral profile and if required, they should apply minerals in an appropriate form based on the composition of the mineral salt. Mineral nutrients are to be supplemented only when there is a deficiency. The compositions of minerals in the inorganic fertilizers are given in the Table.



Water quality analysis report

### Properties of mineral salts for use in aquaculture

Mineral Salt	Formula	Common Name	Typical Composition
Calcium sulphate	CaSO <sub>4</sub> 2H <sub>2</sub> 0	Gypsum	22% Ca; 53% SO <sub>4</sub> ; 55% hardness
Potassium chloride	KCl	Muriate of Potash	50% K; 45% Cl
Potassium magnesium sulphate	K <sub>2</sub> SO <sub>4</sub> 2MgSO <sub>4</sub>	K-mag	17.8% K; 10.5% Mg; 63.6% SO <sub>4</sub>
Potassium sulphate	K <sub>2</sub> SO <sub>4</sub>		41.5% K; 50.9% SO <sub>4</sub>
Magnesium sulphate heptahydrate	MgSO <sub>4</sub> 7H <sub>2</sub> 0	Epsom salt	10% Mg; 39% SO <sub>4</sub>
Sodium chloride	NaCl	Rock salt,	39% Na; 61% Cl; 98%
Socium chioride	NaCi	mine run salt	Salinity

To calculate the dose rate of mineral salt for a desired concentration of any one of the variables listed in the last column, use the following equation:

Dose (g/m3) = Desired concentration of variable (mg/L) ÷ Percentage variable in salt/100

For example, if you want to use muriate of potash to increase potassium concentration by 25 mg/L:

Dose of muriate of potash = 25 mg K/L  $\div$  50% K/100 = 50 mg/L.

66. Whether the *P. vannamei* require same ratio of essential minerals from post larvae to adult? Or at what stages do it requires more/less?

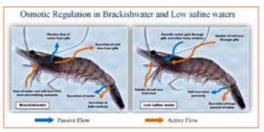
P. vannamei require same ratio of essential minerals from post larvae to adult.

67. Whether shrimp feed contains the required level of mineral nutrients which is essential for metabolic and physiological activity of the shrimp?

Normally, a balanced shrimp feed has the required level of minerals for the metabolic activity of shrimp. Brackishwater and high saline waters have the required minerals at optimum levels, hence additional supplementation of minerals is not required. However, in case of low saline waters, additional mineral supplementation is required to raise the levels at par with the mineral composition of shrimp. Otherwise, due to osmotic pressure, the minerals and ions from the shrimp move to the water and the animal will be under stress. The stressed shrimp cannot feed properly. The uneaten feed as a waste reaches the bottom and increases the bottom metabolites. Therefore, supplementation of minerals in low saline waters is necessary mainly to keep the rearing medium at par in mineral levels to that of shrimp.

# 68. What is the osmo-ionic regulation in *P. vannamei*? Whether external supplementation of mineral is required?

The shrimp lives in oceanic waters which has the optimum level of required minerals. The minerals calcium, magnesium, potassium, phosphorous, chlorine, sulphur and trace elements are essential for its exoskeleton and energy metabolism. Even though the mineral ions enter and exit through body exoskeleton and gills, the animal and the living medium are having



equal concentration of minerals, the ionic composition will not change. If the rearing medium is having lower proportion of required minerals then external application of minerals is required (especially in low saline soils).

69. Whether minerals in soil are as such available in water phase and available for shrimp?

All the portions of minerals present in the soil are not bio-available. When the water is added to soil, only the water soluble portion of the particular mineral is bio-available although the total quantity of mineral is quite high. This availability may vary based on the soil characteristics also.

70. Whether DO level is same in all the places at different depths of the *P. vannamei* pond? While measuring DO which is the correct way for measuring the DO of the pond?

Dissolved Oxygen levels may vary at the surface level in the dead corners where the water circulation is not effective. As the depth increases the level of DO might be gradually decreasing. Water bottles for DO measurement are available and the water should be drawn at least two feet below surface level.

71. When ground water is used for topping up of shrimp pond water what parameters need to be checked?

The mineral composition, alkalinity, hardness and iron content of the ground water are to be checked before its use. However, as per the CAA guidelines, ground water should not be used for shrimp culture.

72. When should a farmer apply gypsum salt? When should dolomite be preferred instead of agri-lime?

If the alkalinity level is more than 50 ppm, calcium carbonate, application of agri-lime to increase the water hardness to the optimum level will not work. Under this condition, application of Gypsum (CaSO<sub>4</sub>·2H<sub>2</sub>O) should be preferred. For one ha of shrimp pond, 19 kg of gypsum can enhance 1 ppm calcium carbonate.

73. When the rapid fluctuation in temperature/diurnal variation is high what are the parameters need to be checked?

The term diurnal more often refers to the change of temperature from the daytime high to the night time low. The important parameters to be measured are total available nitrogen and pH which are correlated with temperature.

# 74. How does sudden heavy rain fall affect the pond condition and what needs to be done?

During heavy rainfall, the pond water becomes acidic and the pH goes down and the plankton crashes. Therefore, lime has to be applied to enhance the pH and plankton production. During heavy rain, the shrimp may be under stress due to low temperature and change in salinity. Therefore, feeding should be checked based on the check tray and behaviour of shrimp in the pond.



Water quality management during rain

#### 75. Whether aeration helps in ammonia management in the pond water?

Yes. Aeration keeps the water in motion and increases the dissolved oxygen in the pond water. Aeration may help in immobilization of ammonia and its release from the pond due to the circulation of pond water. The aerators stimulate bacterial growth and activity which consequently maintain the quality of water. Placement of aerators in the corners allows for solid accumulation in the centre of the pond, which can then be removed if a centre drain is used or will facilitate solids removal during pond drying time. Additional aerators can be placed in the middle of the pond length providing for additional oxygenation and circulation.

# 76. Some technicians opine that ammonia upto a certain level is required for shrimp growth. Is it true? Please give explanation.

No. The technicians might have opined that nitrogen from ammonia may be useful for the growth of beneficial bacteria and phytoplankton growth. It is actually the ammoniacal form of N (NH<sub>4</sub>) which is essential for this purpose. However, ammonia beyond permissible limit is toxic to the shrimp and at low level could affect the growth of shrimp.

### 77. How far are water quality testing kits in the market accurate?

Water quality test kits cannot be 100% accurate like laboratory results. However,  $\pm$  5% deviation is allowed for field measurement to take immediate management measures.

Water quality testing using kit

# 78. Farmers started using chemical fertilizers like Ammonium chloride/Ammonium sulphate, DAP, MOP etc. for water culture in *P. vannamei* farming? Is it necessary?

Application of fertilizers is mostly done in low saline *P. vannamei* culture. Nitrogenous fertilizers like Ammonium chloride/Ammonium sulphate are being applied to provide nitrogen for better phytoplankton production. The compound fertilizers like Di-ammonium phosphate (DAP) will provide both nitrogen and phosphorous required for the bloom development. The Murate of Potash (MOP) might be applied as a mineral nutrient to enhance the potassium content in the low saline waters.



Chemical fertilizers

79. What should be the optimal diurnal DO variation expected? What does it indicate if it is too high or too low and what measures need to be taken to correct the same?

Diurnal variation in DO should not be too high or too low. A diurnal variation of 3 to 4 ppm is fine. If the variation is too high, then it indicates over bloom and if it is too low it shows low bloom. Accordingly, if it is high bloom carry out 10-20%water exchange. Incase low bloom, apply a dose of fermented juice or dolomite @ 40 kg/ha.

80. Whether shrimp pond water pH varies diurnally? If so what needs to be done?

The pH should be monitored before dawn for the low level and in the afternoon for high level. The magnitude of diurnal fluctuation is dependent upon the density of organisms producing and consuming CO<sub>2</sub> and on the buffering capacity of pond water (greater buffer capacity at higher alkalinity). i.e., diurnal fluctuation of pH is not great in pond water of higher alkalinity. An alkalinity above 50 ppm CaCO<sub>3</sub> is preferred in prawn/shrimp ponds. Intervention, such as flushing of ponds to reduce the pH, is advisable when the magnitude of diurnal fluctuation in pH is great. Nevertheless, one should notice that the drastic fluctuation of pH would cause stress to culture organisms. Normally, it should maintain the daily fluctuation within a range of 0.5 difference. Control of pH is essential for minimizing ammonia and H<sub>2</sub>S toxicity.

81. Whether application of jaggery for pH reduction, garlic for gut digestibility etc. really works?

It has been observed that farmers based on their Indigenous Technical Knowledge (ITK) do use jaggery which helps in reducing pH as it creates fermented condition in the pond, turmeric powder for disinfection and garlic paste for enhancing gut digestibility. It seems they are facilitating the purpose for which they are applied.

82. What are all the management measures required to keep the pond bottom clean and prevent its deterioration?

Maintenance of bloom and optimum feed management are the key factors to keep the pond bottom clean. In order to prevent the deterioration of pond bottom, chain dragging is recommended from the beginning of the crop after 15 DOC It is advisable not to practice chain dragging after 60 DOC as it will create more harm.

Mixing of soil probiotic

### **HEALTH MANAGEMENT**

83. Whether PCR screening protocol is available for diseases other than WSSV? Whether screening for WSSV is necessary for *P. vannamei* also?

Yes, PCR test is available for IHHNV, MBV, HPV, TSV, YHV (Viruses), EHP and for many of the vibrios, particularly for the EMS caused by *V. parahaemolyticus*. Screening of *P. vannamei* for WSSV is necessary as non-SPF pond reared broodstocks could have been used for seed production.

# 84. What are all the morphological parameters/features a farmer should look to assess the health of *P. vannamei* shrimp?

Farmers should always monitor the color, activity, swimming pattern, appendages, gill, gut and hepatopancreas of the shrimp. If there is an antenna cut, bacterial infection is suspected. Color change or pinkish coloration will indicate WSSV/ bacterial infection. Gill choking is expected if too much plankton is there in the pond. Status of gut and hepatopancreas should be seen, if



Morphological features of P. vannamei

they are white in color and empty, then EMS or bacterial infection is suspected.

# 85. Running mortality occurs after 60 DOC in culture systems on a daily basis, what could be the cause for this and how can we control this?

As of now it appears that RMS is due to high stocking density and inadequate pond preparation. Select healthy post larvae by carefully adopting the suggested screening procedures. It is preferable to stock *P. vannamei* in low density and give sufficient gap (3-4 weeks) between two cultures.

86. What are the diseases reported in *P. vannamei* farming in India, their causes and management measures?

Name of the Disease	Cause(s)	Symptoms	Prevention/ Management	Photograph
1. White Spot Disease	WSSV	<ul> <li>Reddened body</li> <li>Broken antennae</li> <li>White spots on the carapace</li> <li>Accumulation of water in the head portion</li> <li>Animal becomes lethargic</li> <li>Animal coming to the sides of the pond</li> </ul>	<ul> <li>Stocking of SPF quality seed after PCR screening from the CAA approved hatcheries.</li> <li>Adoption of BMPs and biosecurity</li> </ul>	
2. Running Mortality Syndrome	Unknown	<ul> <li>Antennae and tail portion become red in colour</li> <li>Distinct pattern of chromophores on periopods.</li> <li>Hepatopancreas become yellow or red in colour</li> <li>Yellowish white feaces</li> <li>Regular mortality of few shrimps or few kgs per day.</li> </ul>	<ul> <li>Optimum stocking density as per the carrying capacity of the pond</li> <li>Adoption of BMPs</li> </ul>	

Name of the Disease	Cause(s)	Symptoms	Prevention/ Management	Photograph
3.Infectious Hypodermal Haematopoietic Necrosis (IHHN)	IHHNV	<ul> <li>Bent rostrum</li> <li>Uneven growth &amp; size differences</li> <li>Shape of the animal body becomes uneven</li> <li>Broken antennae</li> </ul>	<ul> <li>Proper drying of the pond bottom</li> <li>Stocking SPF Quality seed</li> <li>BMPs and biosecurity</li> </ul>	
4. Vibriosis (Bacterial Scepticemia)	Vibrio sp.	<ul> <li>Reddening of the body</li> <li>Flared up exoskeleton</li> <li>Broken antennae</li> <li>Hepatopanchreas size increases or decreases</li> <li>White fecal matter</li> <li>Gills become black</li> <li>Black spots or blisters on the body of the animal</li> <li>Florescence appearance during the night.</li> </ul>	<ul> <li>Application of water probiotics at periodic intervals.</li> <li>Adoption of BMPs</li> </ul>	
5.Body cramp/ White muscle	Mineral deficiency/ high diurnal temperature variation	<ul> <li>Animal body cramp/bends</li> <li>At the place of cramp the muscle becomes white</li> <li>Juveniles are more susceptible</li> <li>Ratio differences between the important minerals (Na, K, Ca &amp; Mg)</li> </ul>	<ul> <li>Periodic monitoring of mineral compositions in the pond water.</li> <li>Maintenance of proper ratio of essential mineral nutrients</li> </ul>	San Die
6. White fecal matter	Bacteria/ Protozoan/ algal toxicity due to BGA	<ul> <li>Shrimp gut becomes white</li> <li>White feaces floating on water</li> <li>Hepatopancreas becomes white and accumulation of fat in HP</li> </ul>	<ul><li>Gut probiotics</li><li>Adoption of BMPs</li></ul>	
7. Black gill	Poor pond management and H2S formation /Deficiency in Vitamin-C/ Fungal infection/ Bacterial infection	<ul> <li>Gills turn to black colour (gill necrosis).</li> <li>Shrimp comes to the surface even though DO is at optimum levels.</li> <li>Mortality in severe cases</li> </ul>	❖ Adoption of BMPs and water quality management	

Name of the Disease	Cause(s)	Symptoms	Prevention/ Management	Photograph
8. Brown gill	Protozoan – Zoothamnium/ water with more Iron content/ Bacterial infection	<ul> <li>Presence of dianoflagellates in the pond water</li> <li>Gills turn to brown colour</li> <li>Shrimp comes to the surface even though DO is at optimum levels</li> <li>Mortality in the morning</li> <li>More mortality if DO drops</li> </ul>	❖ Adoption of BMPs and water quality management	
9. Microsporidiosis (EHP)	Enterocytozoon hepatopenaei (Microsporidian parasite)	❖ It severely Damage Hepatopancreas (HP) and shrimp gut, causing severe growth retardation.	<ul> <li>A systematic pond preparation is very essential.</li> <li>PCR screening of the seed for presence of EHP is absolutely essential.</li> <li>Maintain strict biosecurity to prevent unwanted animals into the farm.</li> </ul>	

87. What is the reason for white muscle formation in *P. vannamei*? How can it be prevented and managed after its outbreak?

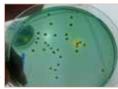
Any kind of stress, including DO problem can produce white muscle in *P. vannamei*. The shrimp should be examined further to know the cause and accordingly suggestions should be given.



White muscle formation in *P. vannamei* 

88. What are green and yellow colonies of *Vibrio* and what is the limit in *P. vannamei* farm? Green and yellow colonies are based on sucrose fermentation that is

present in media. Green colonies cannot ferment sucrose. In general, it is thought that green colonies are pathogens. However, yellow colonies can also be pathogens. If the number of colonies is less than 100 per ml, it would be good.



Green and yellow colonies of *Vibrio sp*.

### 89. What is Early Mortality Syndrome (EMS/AHPND), what are its symptoms? How to prevent/manage this?

In EMS, nearing 100% mortality occurs within 30-40 days of stocking. There will be white gut and HP size will also be very small and when pressed between the fingers, it will feel rubbery. EMS comes through larvae or brood stock. If EMS is already there in an area, it can also get transferred through water. Larvae should be tested (both with atrophied Hps indicative of EMS.



PCR and stress test) and pond should be well prepared. During infection, feed management should be done very carefully. Use of probiotics also helps to a great extent. So far, EMS has not been reported in India. Many other bacterial infections might also have majority of these symptoms.

### 90. What is EHP? Will it infect P. vannamei shrimp? If so, what are its symptoms? What measures can be undertaken to prevent/manage the entry and spread of EHP?

The full name of EHP is *Enterocytozoon hepatopenaei*. It is a spore forming unicellular parasite and belongs to a group called microsporidea. EHP is found to infect shrimps and recent survey by ICAR-CIBA confirmed the presence of EHP in the shrimp farms of India. EHP is not characterized by any specific clinical signs. Though EHP does not cause mortality, it severely damages Hepatopancreas (HP) and shrimp gut, causing severe growth retardation. EHP can also get associated with white faeces syndrome, EMS and WSSV etc. The disease is spread through the oral route when shrimps eat infected animals. So far EHP has been detected only from polycheats other than the shrimps. If the brooders are infected this can be transmitted through the larvae. EHP spores are thick and difficult to destroy even with higher chlorine concentration. The best way is to prevent the entry of EHP into the system. The following measures need to be adopted for the prevention of EHP pathogen in to the shrimp farm.

- A systematic pond preparation is very essential particularly when the previous crop is infected with EHP. Adequate drying of the pond for 4-5 weeks and apply CaO (lime) @ 6 tons/ha. Plough the pond for the lime to penetrate dip into pond bottom (1 foot or more). Moisten the lime for activation. Leave it for one week.
- > PCR test is the most suitable and sensitive method for detection of EHP. PCR screening of the seed for presence of EHP is absolutely essential.
- > Maintain strict biosecurity to prevent unwanted animals into the farm because the range of carriers for EHP is still unknown.
- 91. SPF *P. vannamei* is free from how many pathogens? What are they?

It depends from which companies you are getting the SPF and there is no limit to it. Presently a certificate for 7 pathogens is sought from the SPF broodstocks supplier (WSSV, IHHNV, TSV, IMNV, YHV, LSNV and EHP).

92. Is it possible to enhance the immunity of *P. vannamei* shrimp through use of immune stimulants? If yes, suggest what needs to checked in such products?

Yes. Many of the herbal products and yeast/bacteria derived products can be used for the purpose. CIBASTIM is a bacterial product developed by CIBA and tested widely in farms.

93. Is the Running Mortality (RMS) and water quality parameters related? If yes, how?

RMS starts after 45 days and till about 90 DOC there will be partial mortality in each day. RMS is directly related with ammonia, turbidity and nitrite. Temperature also has an influence on RMS to some extent.



Running mortality in P. vannamei

- 94. How to keep/ preserve an abnormal shrimp for sending the same for analysis/screening? Keep the animals in alcohol for PCR testing. Live moribund shrimps will be a good sample.
- 95. Is partial harvesting in *P. vannamei* farming essential? If so at what stage it has to be done and what gear should be used?

The biomass of the *P. vannamei* pond after 70 days of stocking is relatively high and maintenance of water quality parameters and feed management are very important. In order to minimize the risk, farmers can do partial harvesting upto 20-30% of the stock using drag net so that the load of the pond gets optimized. At 70 DOC the shrimp could have grown to 12-14 g size and they may fetch good price at the local market and by this, the farmer sometimes might get his break even. Similarly, one more partial Partial harvesting of P. vannamei shrimp



harvesting at 90 DOC with 15-18 gm size shrimps again might help in better farm management. This approach may help the farmer to avoid market price crash during the time of final harvest (June-July syndrome). Therefore, partial harvesting of shrimp helps in maintaining the optimum pond carrying capacity and minimizes the risks.

### REGULATION, FOOD SAFETY AND RECORD KEEPING

96. How P. vannamei farming in inland/low saline areas are regulated? Does this need to be registered?

The Govt. of India has notified that farmers who desired to culture P. vannamei outside the jurisdiction of CAA having the water salinity of above 0.5 ppt shall get registered with the Department of Fisheries (DoF) of the state government concerned. The farms should possess all the required infrastructure and biosecurity. The DoF may constitute a separate district level committee to inspect and give registration to the farms within a reasonable time frame of 60 days and other guidelines are same as that of brackishwater area. However, farms within the jurisdiction of CAA (up to 2 km from the high tide line) should get registered with the CAA only.

97. What is crop holiday? How it help? As such, does a shrimp pond require a holiday once in few/some years even otherwise also?

Leaving the shrimp pond without culture for a specified period is called crop holiday. It is generally declared whenever new emerging diseases appear on a large scale. It also helps in total eradication of pathogens or their hosts present in the pond sediments.

#### 98. What are periphyton and its significance in *P. vannamei* farming?

Use of submerged substrates like agriculture shade nets or dry wooden planks, which serve for the promotion of growth of mixed microalgae-bacteria mats (Periphyton). The Periphyton utilizes the dissolved waste products within the culture system by autotrophic bacteria and algae or through direct heterotrophic



conversion of organic and inorganic nitrogen species into microbial *P. vannamei* pond with periphyton net biomass which improves water quality and at the same time, the microbial biomass becomes an important direct or indirect source of natural feed for the farmed organisms.

#### 99. What are the responsibilities and do's and don'ts of a *P. vannamei* farmer?

Farmers should get registration from the CAA/DoF for the culture of *P. vannamei*. The farms should have adequate biosecurity measures and infrastructure to culture *P. vannamei* shrimp. Farmers should procure seed from the registered hatchery after screening for pathogens and restrict the stocking density as per the CAA guidelines. The farmer should adopt better management practices of farming and send the periodical reports to the CAA. Farmers should not carry out farming without registration and should not procure seed from a non-registered hatchery.

# 100. When or at what level of intensification, plastic lining of *P. vannamei* pond is necessary? What are its advantages?

Plastic lining of *P. vannamei* shrimp ponds is necessary in case of super intensive systems with very high stocking density of 250-300 PL/m<sup>2</sup> with biofloc technology and other high energy inputs. Also, pond lining can be done if a farmer wants to culture three or more crops in a year and the bottom lining can be cleaned using pressure pumps before starting the next culture. It helps in preventing soil erosion of bunds/dykes due to the use of more aerators continuously. Nevertheless, CAA has permitted a stocking

farmers may use lining for dykes to prevent erosion.



aerators continuously. Nevertheless, CAA has permitted a stocking Plastic lining of *P. vannamei* pond density to the maximum of 60 PL/m<sup>2</sup> and for this, density lining is not required. However,

#### 101. What are the food safety measures a farmer needs to adopt in *P. vannamei* farming?

Cleanliness and hygiene of farm workers and basic amenities for them are essential. Wild/pet animals should not be allowed entry into the farm. Chill killing of shrimp immediately after harvest and proper handling of harvested shrimp are required to maintain the quality. Farmed shrimp should be free from antibiotics, banned feed additives and should not have any discoloration or bad odor.



Poor handling of harvested shrimp

# 102. What are all the aspects that need to be recorded for record keeping? Is it mandatory for marketing?

Record keeping is a good practice to record all the operations pond wise, quantity of inputs applied at each stage, seed stocking density, feeding - schedule, quantity, timing, sampling details, ABW, probiotics and minerals applied, water quality measurements and their reports which are very essential to take appropriate decisions as and when required. Record keeping is one of the BMPs and it is essential for certification of farmed shrimps.



Record keeping

#### 103. What is *P. vannamei* and what are the main advantages over *P. monodon*?

White legged shrimp *P. vannamei* is the native species of pacific coast of Mexico, Central and South America and presently it is the most widely cultured shrimp species worldwide. The supremacy of *P. vannamei* vis-à-vis other species is mainly due to the availability of Specific Pathogen Free (SPF) stocks, amenable for higher stocking density and culture in varying salinities. The comparative advantages of *P. vannamei* vis-à-vis *P.monodon* is given in the Table.

Sl.No	Characteristics	Tiger shrimp	White shrimp
		(P.monodon)	(P. vannamei)
1	Specific Pathogen Free (SPF) broodstocks	Not available	Available
2	Capacity to tolerate wide range of salinities	Yes	Yes
3	Capacity Low temperature	No	Yes. It can tolerate low temperature up to 15°C
4	Protein requirement in the feed	High (> 40%)	Low (30-35%)
5	Amenable stocking density per ha	1.0 – 1.5 Lakh/ha	Permitted up to 6.0 Lakh/ha
6	Larval survival at hatchery	Low (< 40%)	High 60-70%)
7	Growth rate	1.5 – 2.0 gm per week	1.5 – 2.0 gm per week
8	Average Productivity	Low (1.5-2.0 tonnes/ha)	High (5-6 tonnes/ha)
9	Culture pond depth	1 meter	1.5 to 2.0 meter
10	Infrastructure requirement	Relatively Low	High (Aerators, 24x7 electricity, generator back up etc.)

### 104. Is it necessary to take CAA registration for starting the *P. vannamei* farming?

Yes. The farmers interested to farm *P. vannamei* need to avail an exclusive registration even though they have already availed Coastal Aquaculture Authority (CAA) registration for the farming of tiger shrimp (*P. monodon*).

#### 105. What is the procedure for obtaining license for *P. vannamei* farming?

Farmers who have already have a CAA registration for tiger shrimp farming should send a request letter along with copy of the earlier registration to the CAA. In case of small farms up to 2 ha, the CAA would send this request to the concerned District Level Committee (DLC) of the Department of Fisheries (DoF) of the concerned State for inspection of the facility for the infrastructure and biosecurity requirements. Based on the recommendations of the DLC and SLC (State Level Committee) the registration for farming of *P. vannamei* will be issued to the concerned farmer.

# 106. Is it possible to do *P. vannamei* culture along with other indigenous shrimp species as mixed culture?

No. As per the CAA regulations/guidelines mixed culture of *P. vannamei* with other shrimp species is not permitted.

### 107. Is it possible to do *P. vannamei* culture in the tide fed traditional ponds, Pokkali fields, etc.,

As per the CAA farming of *P. vannamei* shrimp require strict biosecurity measures like disinfection of source water, filtrations, fencing (crab,men,bird) and disinfection protocol for labour and implements. Considering the limitations with the tide fed ponds and pokkali fields it is not possible to take up *P. vannamei* farming.

#### 108. Which is the ideal season for *P. vannamei* Culture?

Shrimp farming is being carried out in two seasons in India. The first season is between February/March to June/July. The second season is between August/September to November/December

#### 109. From where we can source *P. vannamei* seeds?

The CAA has permitted 180 shrimp hatcheries for import of SPF broodstock and seed production and supply of *P. vannamei*. This list is available in the website of CAA (www.caa. gov.in) and shrimp farmers should procure seeds only from the registered hatcheries following due quality screening procedures.

# 110. Is it possible to do *P. vannamei* culture in fresh water resources such as artificial ponds, tanks, etc? What is the package of practice for culture in fresh water systems?

*P. vannamei* shrimp is tolerant to low salinities but the rearing water should have a salinity of more than 0.5 ppt. The Govt. of India has notified that farmers who desired to culture *P. vannamei* outside the jurisdiction of CAA having the water salinity of above 0.5 ppt shall get registered with the Department of Fisheries (DoF) of the state government concerned. The farms should possess all the required infrastructure and biosecurity. The DoF may constitute a separate district level committee to inspect and give registration to the farms within a reasonable time frame of 60 days and other guidelines are same as that of brackishwater area.

*P. vannamei* farming in Low saline waters requires two critical practices to be adopted as below and remaining practices are more or less same as that of brackishwater.

- Acclimatization of shrimp seed to the pond conditions should be done gradually not suddenly. It is better to stock larger PL (above 15 PL) which has the capacity to acclimatize to low saline waters.
- The mineral composition of the rearing water should be maintained at an optimal level throughout the culture period. *P. vannamei* shrimp requires essential minerals viz., calcium, magnesium and potassium in the ratio of 1:3:1. The minimum requirement of calcium is 150-200 ppm. In 1 ppt salinity the required level of essential minerals should be @ calcium-11.6 mg/L; magnesium-39 mg/L and potassium-10.7 mg/L. According to the pond salinity this value can be multiplied and the required level of minerals in proportion should be maintained always for the better survival and growth of *P. vannamei* shrimps.

# 111. Is it possible to insure *P. vannamei* culture farms? What is the procedure for insuring the *P. vannamei* culture farms?

An insurance scheme for shrimp culture is available with the National Insurance Companies of India. However, crop insurance is not popular among shrimp farmers due to the following reasons.

- Insurance companies are reluctant to insure shrimp farms for disease outbreaks.
- They take in to account only the inputs costs not the actual biomass exist during the mishap.
- > The premium is relatively high.

### 112. Is there any government schemes or subsidies exist for doing *P. vannamei* culture?

The Marine Products Export Development Authority (MPEDA; www.mpeda.com), The National Fisheries Development Board (NFDB; www.nfdb.gov.in) and the DoFs of respective states do have shrimp farming development schemes for construction of new ponds, renovation of existing ponds, one time first-year inputs for fish/prawn culture and farm infrastructure development etc.

- 113. In a nut shell, what are all the infrastructure requirements and the salient management practices to be followed for *P. vannamei* farming?
  - Adequate farm Biosecurity, deep ponds, central drainage/sledge removal capacity, waste water pond, continuous electricity and exclusive permission from the Coastal Aquaculture Authority are essential requirements for *P. vannamei* farming.
  - Prepare the culture pond by adequate drying, scraping, ploughing, liming, strengthening of bunds and dykes. In case of white spot disease incidence in the previous crop, drying of soil for a minimum of 21-30 days is essential.

- Adopt filter screens to prevent the entry of vectors, disinfection with 20 ppm chlorine and allow ageing of water for 12-14 days to eliminate free living viral pathogens.
- Apply carbon source for development of adequate phytoplankton and beneficial bacteria.
- Select SPF *P. vannamei* seed from a registered shrimp hatchery after adequate screening for presence of pathogens. Above PL12 is the optimum size for stocking.
- Acclimatization of seed helps in better survival. Stock optimum density based on the pond carrying capacity, existing infrastructure and biosecurity. Moderate stocking minimizes the risks.
- Always Maintain dissolved oxygen level at above 4ppm. Artificial aeration is important to maintain the dissolved oxygen levels. Keep1HP aerator for every 300 kg biomass.
- Continue to maintain mineral nutrients at optimum levels (Ca & Mg at 1:3 ratio and Na & K at 28:1 ratio) based on periodic water quality tests.
- Maintain the pond water quality with total alkalinity between 50-150 ppm and hardness between 75 200 ppm. The safe levels for the metabolites are: Total Ammonia Nitrogen: <1ppm, Nitrite nitrogen: <0.5ppm and Hydrogen sulphide: <0.03 ppm.
- Optimize feeding regime by careful monitoring of check trays and shrimp biomass.
   Avoid feeding after 7.00 PM. Stop feeding when shrimp are in stress and the gut is half or empty. Adopt automatic feeders for better FCR.
- Monitor the health of shrimp by observing its behaviour, feed intake, gut, hepatopancreas, appendages, gills, body discoloration and patches. Contact scientists/consultants for technical advice in case of abnormality.
- Partial harvesting helps in maintaining the pond carrying capacity and minimize risks. Chill kill the shrimp immediately after the harvest to maintain the quality.



Filtration system



P. vannamei shrimp seed



Shrimp aeration with air diffusers



Chill killing of harvested shrimp