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# Phytoplanktonic composition of three cultivation systems used in *Litopenaeus vannamei* (BOONE, 1931) marine shrimp farms

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**ABSTRACT.** The aim of this work is to assess the different compositions of phytoplankton in three cultivation systems of marine shrimps *Litopenaeus vannamei* (BOONE, 1931), denominated as organic, intensive and semi intensive. The samples were done fortnightly, when phytoplankton was collected by a net for phytoplankton, 65 μm mesh, being then filtrated in a total volume of water of 100 L, and preserved in formaldehyde solution at 4% and identified according to the methodology of Cordeiro et al. (1997). The results show that the densities of Diatoms were of 16.65, 10.47 and 7.57 cel. 10<sup>3</sup> mL<sup>-1</sup> for the organic, intensive and semi intensive cultivations, respectively. As for cyanobacteria, the average figures were 42.06 cel. 10<sup>3</sup> mL<sup>-1</sup> for semi intensive 17.27 cel. 10<sup>3</sup> mL<sup>-1</sup>, in the intensive cultivation and 6.11 cel. 10<sup>3</sup> mL<sup>-1</sup> for the organic cultivation system. The dinoflagellates had the highest cellular density in the phytoplankton community analyzed with 61.9 cel. 10<sup>3</sup> mL<sup>-1</sup> in the intensive cultivation, 0.33 and 0.03 cel. 10<sup>3</sup> mL<sup>-1</sup> for both semi intensive and organic cultivation systems respectively. Euglenas presented the results of 4.98 and 14.86 cel. 10<sup>3</sup> mL<sup>-1</sup> only for semi intensive and intensive cultivations. It was then concluded that all cultivations presented average rates below recommended for such studied systems.

Key words: Floristic composition, different methods of producing, shrimp farms

RESUMO. Composição fitoplanctônica em três sistemas de cultivo do camarão marinho Litopenaeus vannamei (BOONE, 1931). Conduziu-se esse trabalho com o objetivo de avaliar as diferentes composições fitoplanctônicas em três sistemas de cultivo para o camarão marinho Litopenaeus vannamei (BOONE, 1931), denominados de orgânico, intensivo e semiintensivo. As amostragens foram realizadas quinzenalmente, onde o fitoplâncton foi coletado através de uma rede de plâncton, com malha de 65  $\mu$ m, sendo filtrado um volume de água total de 100 litros, que foram preservadas em solução de formol a 4% e identificadas segundo a metodologia de Cordeiro et al. (1997). Os resultados mostram que as densidades de diatomáceas foram de 16,65; 10,47 e 7,57 cel. 10<sup>3</sup> mL<sup>-1</sup>, respectivamente, para os cultivos orgânico, intensivo e semi-intensivo. Para as cianofíceas as médias foram 42,06 cel. 10<sup>3</sup> mL<sup>-1</sup> no semi-intensivo, 17,27 cel. 10<sup>3</sup> mL<sup>-1</sup> intensivo e 6,11 cel. 10<sup>3</sup> mL<sup>-1</sup>no sistema orgânico de criação. Os dinoflagelados obtiveram a maior densidade celular dentre a comunidade fitoplanctônica analisada de 61,9 cel. 10<sup>3</sup> mL<sup>-1</sup> no cultivo intensivo, 0,33 e 0,03 cel. 10<sup>3</sup> mL<sup>-1</sup> para o semi-intensivo e orgânico, respectivamente. As euglenas apresentaram densidades apenas para os cultivos semi-intensivo e intensivo de 4,98 e 14,86 cel. 10<sup>3</sup> mL<sup>-1</sup>. Conclui-se que as densidades fitoplantônicas em todos os cultivos estiveram abaixo daquelas recomendadas para os sistemas estudados.

Palavras-chave: Composição floristíca, diferente métodos de produção, fazenda de camarão

# Introdution

Among all species of crustaceans, marine shrimps, mainly of the species *Litopenaeus vannamei* (BOONE, 1931), show the fastest growth rates (BORGHETTI et al., 2003) and are now both the most popular and world commercial value species. For this reason, the principal worry of the shrimp-farming industry is to improve its cultivation methods in order to achieve the highest possible levels of productivity per hectare (SANCHEZ, 2003).

Worldwide, there are four methods of producing shrimps: the extensive, semi intensive, intensive and organic cultivation systems. The extensive, semi intensive, intensive and organic cultivation systems are characterized by a progressive reduction in the size of growout ponds, progressive rise in the cost of production and construction, higher population density, intensification in the cultivation methods used (including the preparation of the ponds, water drainage, fertilization, the use of balanced food, among

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other methods) (JORY, 2001). Organic cultivation is a system characterized by reproduction in growout ponds, an environment similar to the natural habitat in which the animals grow free from chemical products, having reduced stressed and natural growth in a healthy way, and as a result, increasing survival capacity (CHELLAPA et al., 2007).

Therefore, so that shrimp cultivation becomes an economically viable and eco-friendly sustainable activity, some obstacles still need to be overcome, among which the importance of understanding the role of phytoplankton when it comes to food chain.

Phytoplankton is the first link in most of the food chain in water environments; its composition is widely dependent on several variants in those systems, which include both biological and non-biological factors in the environment (BOYD, 1995).

However, it is not every type of algae that is suitable for cultivation in ponds. Diatoms and dinoflagellates are the most desirable; some of them have been even used in cultivation as food for larvae of mollusks, early life stage crustaceans and fish (MOURA et al., 2006). On the other hand, some species of cyanobacteria are considered not suitable as they release toxins that contaminate the cultivated organism, cause bad odor, as well as environmental imbalances when dumped into adjacent environments when drainage occurs (MARTINEZ-CÓRDOVA et al., 2003).

The knowledge of the diversity of the phytoplankton community will benefit both the environment in which it occurs and also the environments in which cultivation is introduced, thus resulting in improvements through phytoplankton knowledge and, by means of this, the knowledge of zooplankton, bacteria related to detritus, hydrophytes, benthos and shrimps and, as a consequence, of all the flux of energy in ponds (BEYRUTH et al., 1998).

The objective of this work was to verify the gradient occurrence in the concentration of phytoplankton in the three marine shrimp cultivation systems *Litopenaeus vannamei*, and the quantification, identification of the main groups of phytoplankton.

# Material and methods

The study took place in three shrimp farming compounds for marine shrimps *Litopenaeus vannamei*, located in the municipalities of João Pessoa and Lucena, in the state of Paraíba, and in Tibaú do Sul, in the state of Rio Grande do Norte, Brazil.

"Aquamaris" farm, which was represented as F1 farm, is located in the municipality of João Pessoa, presented the semi intensive cultivation system, with a density of 15 shrimps m<sup>-2</sup>; 'Costa Azul' farm,

represented as F2 farm, is located in the municipality of Lucena and represented the intensive cultivation system, with a density of 300 shrimps m<sup>-3</sup>; and 'Primar' farm, in the municipality of Tibaú do Sul, denominated F3 farm, represented the organic system, with a density of 3 shrimps m<sup>-2</sup>.

The samples were collected close to sluice gates and in the middle of the ponds, where phytoplankton was collected with a net for phytoplankton, 65  $\mu$ m mesh, through filtration 100 L of water from cultivation. The filtrated phytoplankton was kept in plastic bottles of 200 mL and preserved in a solution of formaldehyde at 4%. Later, the samples were sent to the laboratory of coastal ecosystem analysis at the centre of Natural and Exact Sciences (CCEN), at the Federal University of Paraíba (UFPB).

Two collection points were established for each farm, the samples were collected fortnightly and distributed as to follow all the stages of fattening of shrimps, altogether 15 samples.

The qualitative analyses were done with the help of an inverted microscope (Zeiss Axiovert), equipped with camera so that the algae could be recorded. The counting method followed the procedures described by Cordeiro et al. (1997), which is an adaptation of the traditional method proposed by Utermohl (1958).

The identification of the species was done with a video camera and with the help of specialized literature by Bold and Wynne (1985), Balech (1988) and Chretiènnot-Dinet et al. (1990). All results found were measured and photographed and recorded for further taxonomic studies. The calculations to determine the amount of organisms per liter were done using arithmetic patterns. The phytoplankton densities were treated statistically by using Anova, followed by Tukey's test at 5% probability

## Results and discussion

Table 1 qualitative composition of phytoplankton found in the semi-intensive, intensive and organic cultivations during the study.

Four taxonomic classes were identified with 29 taxon for farm 1, represented by the semi-intensive cultivation, being seven taxa for Cyanophyceae, 18 taxon for Diatomophyceae, two for Dynophyceae and one for Euglenophyceae. As to farm 2, which represented the intensive cultivation, four taxonomic classes were found, distributed in 21 taxon, and eight taxa represented the Cyanophyceae, ten taxa the Diatomophyceae, two taxa the Dynophyceae and one taxon the Euglenophyceae. As for farms 3, which had the organic cultivation, the number of taxa were 28

divided into three taxanomic classes, among these, six taxa of Cyanophyceae, 20 taxa of Diatomophyceae and two taxa of Dinophyceae (Table 2).

**Table 1.** Species of phytoplankton identified in the semiintensive, intensive and organic systems.

-	Study environment				
Taxonomic	Aquamaris	Costa Azul	Primar (Organic)		
groups	(Semi-intensive)	(Intensive)			
Diatomo-					
phyceae	Amphiprora sp.	Amphora alata	Amphora alata		
• /	Amphora alata	Coscinidiscus centrales	Astrorenella sp.		
	Anabaenopsis	Coscinidiscus sp	Biddulphia sp.		
	Chaetoceros gracilis	Navícula sp.1	Coscinodiscus centrales		
	Coscinodiscus centrales	Navícula sp.7	Coscinodiscus sp.2		
	Coscinodiscus sp.2	Navícula sp.9	Gramatophora marinha		
	Entomoneis alata	Niztcha sp	Gramatophora sp.2		
	Navícula directa	Rizolenia robusta	Navícula sp.10		
	Gimnodinium sp.	Thalassiosira sp.1	Navícula sp.12		
	Navícula sp.1	Thalassiosira sp.2	Navícula sp.14		
	Navícula sp.11		Navícula sp.15		
	Navícula sp.12		Navícula sp.7		
	Navícula sp.14		Navícula sp.9		
	Navícula sp.15		Nitzcha longuissima		
	Navícula sp.2		Nitzcha sp.1		
	Nitzcha longuissima		Pleuro-girosigma sp.1		
	Nitzcha sp.1		Pleuro-girosigma sp.2		
	Pleuro-girosigma sp.1		Rizolenia sp.2		
	Pleuro-girosigma sp.2		Synedra sp.		
	Thalassiosira sp.1		Thalassiosira sp.1		
Cyanophyceae		Anabaena sp.	Anabaena sp.		
	Lyngbia sp.2	Lyngbia sp.1	Lyngbia sp.1		
	Mycrosistis sp.	Lyngbia sp.2	Oscilatória laetivirens		
	Oscilatória laetivirens	Oscilatória laetivirens	Oscilatória sp.3		
	Oscilatória sp.2	Oscilatória sp.2	Spirulina sp.1		
	Oscilatória sp.3	Oscilatória sp.3	Spirulina sp.2		
		Spirulina sp.1			
		Spirulina sp.2			
Dinophyceae	Peridinium sp.	Peridinium sp.	Peridinium sp.		
		Gimnodinium sp.	Gimnodinium sp.		
Eugleno-					
phyceae	Euglena sp.	Euglena sp.			

**Table 2.** distribution of taxonomic classes according to density and relative abundance scores followed by the same letter, per column, do not change significantly in Tukey's test (p < 0.05).

Site	Taxonomic classe	Mean density	Relative abundance	of taxa
		cel. 10 <sup>3</sup> mL <sup>-1</sup>	%	
Farm 1	Cyanophyceae <sup>a</sup>	42.06	76	7
Farm 2	Cyanophyceae b	17.27	17	8
Farm 3	Cyanophyceae c	6.11	24	6
Farm 1	Diatomophyceae c	7.57	14	18
Farm 2	Diatomophyceae b	10.47	10	10
Farm 3	Diatomophyceae a	19.65	76	20
Farm 1	Dynophyceae b	0.33	1	2
Farm 2	Dynophyceae a	61.9	59	2
Farm 3	Dynophyceaee c	0.03	0	2
Farm 1	Euglenophyceae a	14.86	14	1
Farm 2	Euglenophyceae b	4.98	9	1

Farm 1 = semi-intensive cultivation; Farm 2 = intensive cultivation; Farm 3 = organic cultivation

As for cyanobacteria, the highest final average density was of 42.06 cel. 10<sup>3</sup> mL<sup>-1</sup> in the semi-intensive cultivation system, followed by 17.27 cel. 10<sup>3</sup> mL<sup>-1</sup> in the intensive cultivation system and of 6.11 cel. 10<sup>3</sup> mL<sup>-1</sup> in the organic one.

The relative abundance found was of 76% for the semi-intensive cultivation, 24% for the organic one and of 17% for the intensive one.

As for the diatoms, the highest final average density of 19.65 cel. 10<sup>3</sup> mL<sup>-1</sup> for the organic cultivation, followed by 10.47 cel. 10<sup>3</sup> mL<sup>-1</sup> for the intensive cultivation and by 7.57 cel. 10<sup>3</sup> mL<sup>-1</sup> for the semi-intensive cultivation.

The relative abundance was 14% for the semiintensive cultivation, 10% for the intensive cultivation and 76% for the organic cultivation. The figures found in the organic cultivation system shows the benefits for the group, as it is considered to be a great display of the fertility of the water environment, for the presence of these organisms in the growout ponds determine the quality of the water, and as a consequence, the good growth of the shrimps (OLIVEIRA et al., 1998).

As for the dinoflagellates, the intensive cultivation obtained 61.90 cel. 10<sup>3</sup> mL<sup>-1</sup> of final average density, followed by 0.33 cel. 10<sup>3</sup> mL<sup>-1</sup> for the semi-intensive one and only 0.03 cel. 10<sup>3</sup> mL<sup>-1</sup> for the organic one. The abundance for the intensive cultivation was 59% and of only 1% for the semi-intensive cultivation, not showing any figures for the organic cultivation. Campos et al. (2007) points out that high concentration of dinoflagellates can produce toxic substances which may cause harm to the cultivated animals or even to humans. Thus, one can claim that the use of chemical fertilizers made from nitrogen and phosphate composts, as well as the misuse of probiotics which aim to increase the levels of algae in ponds, might have propelled the levels of dinoflagellates in the intensive cultivation as showed.

As for euglenas, the level was of 14.86 cel. 10<sup>3</sup> mL<sup>-1</sup> for the final average density in the intensive cultivation, followed by 4.98 cel. 10<sup>3</sup> mL<sup>-1</sup> for the semi-intensive cultivation, not showing any levels for the organic cultivation. The relative abundance found was of 9% for the semi-intensive cultivation, 14% for the intensive cultivation, not being found any figures for the organic one.

These taxonomic groups, when found, can show that in the system studied, there is a high level of organic matter (KOENING, 1983), which was proved in this study, making us think that as the intensive system is the one in which there is a higher level of density of shrimps and, as a result, a higher level of organic matter, there will be a higher consumption of food and higher metabolic levels produced by the animals cultivated. This line of thought becomes even more justifiable when contrasted with the non-detection of euglenas in the organic cultivation, as this method does not make use of phytoplankton promoter, its density was, thus, low (WANG, 1990).

The diatoms of the species *Navicula* sp. were identified through the academic references showed in the methodology researched Balech (1988), Bold and Wynne (1985) and Chretiènnot -Dinet et al. (1990).

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Although, according to the database of pre- existent images from the laboratory of Systematic and Coastal Ecology of the Federal University of Paraíba. 15 different gender types were identified, only the genders classified as *Navicula* sp.1, *Navicula* sp.2, *Navicula* sp.4, *Navicula* sp.5, *Navicula* sp.7, *Navicula* sp.9, *Navicula* sp.10, *Navicula* sp.11, *Navicula* sp.12, *Navicula* sp.14 e *Navicula* sp.15 were found in this study.

Academic data about the quantity and composition of phytoplankton in the cultivation of shrimps varies significantly. Clifford (1994) and Nunes (2001) suggest that, in semi-intensive cultivations, the density of phytoplankton should remain in 80,000 and 120,000 cel. mL<sup>-1</sup>. The total figures of density of phytoplankton in the semi-intensive cultivation studied were of 54,940 cel. mL<sup>-1</sup>, thus, inferior to those described by the authors here mentioned.

But Chien (1992) says that, in Taiwan, in intensive cultivations, the total densities of algae were between 100,000 to 10,000,000 cel. mL<sup>-1</sup> in the cultivation analyzed, the total number of phytoplankton were of 10,450 cel. mL<sup>-1</sup>, therefore, quite inferior to what was said, as verified in Table 2.

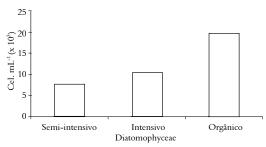
Campos et al. (2007) in studies on the taxonomic groups, describe that the ideal groups to be in ponds should be diatoms with minimum density of 20,000, cyanobacteria, with maximum levels of 50,000 cel. mL<sup>-1</sup>. As for dinoflagellates and chlorophyta, the maximum recommended levels of density should be, respectively, 500 and 50,000 cel. mL<sup>-1</sup>

All data obtained from the cultivation systems analyzed show that the all levels related to diatoms are under the recommended interval, only in the organic cultivation system the average figures found were close to what was described (Figure 1).

The dinoflagellates in the intensive cultivation obtained much higher levels than recommended by Nunes (2001), reaching average figures superior to 61,000 cel. mL<sup>-1</sup>. Yan et al. (2003) says that massive groups of dinoflagellates were found in various areas of cultivation of marine shrimps and that factors such as inappropriate fertilization, unfavorable temperature and salinity can be responsible for undesired groups of these organisms, affecting negatively the cultivation of shrimps Peneideos (Figure 2).

As to the levels of cyanobacteria, all the average figures were much below what was described, only the semi-intensive system showed levels close to the indexes mentioned (Figure 3).

As to chlorophyceae, the levels found in the intensive cultivation obtained the highest results, approximately 16,000 cel. mL<sup>-1</sup> (Figure 4).



**Figure 1.** Density of diatoms in of dinoflagellates in semi-intensive, intensive and intensive and organic cultivations.

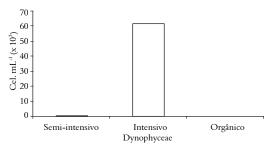


Figure 2. Density of dinoflagellates in semi-intensive, organic cultivations.

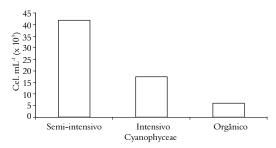
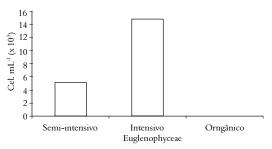


Figure 3. Density of cyanobacteria in semi-intensive, intensive and organic cultivations.



**Figure 4.** Density of euglenas in semi-intensive, intensive and organic cultivations.

Diatoms and green algae were considered beneficial as they are the main food source for most water invertebrates and fish larvae, whereas blooms made of cyanobacteria and dinoflagellates are associated with poor quality of waters and eutrophication (YUSOFF et al., 2002). Therefore, you can say that the low levels of final average density of taxonomic groups detected are indicative of non-eutrophication in the systems

studied. Yet Santana et al. (2008) claims that the high density of cyanobacteria and low densities of diatoms can cause deformities in cultivated shrimps.

Brito et al. (2006), when verifying the effects of the use of sodium nitrate in the fertilization to increase the population of diatoms in semi-intensive ponds with *L. vannamei*, found in a controlled test (without the use of nitrate), the figures of 5,833 cel. mL<sup>-1</sup>, inferior to the three cultivation systems studied, and figures of 12,554 cel. mL<sup>-1</sup> with the use of sodium nitrate, higher than the figures found in both semi-intensive and intensive cultivations, which used probiotics for algae increase. Therefore, the average figure cited by the author was relatively closer to the organic cultivation, showing that this natural method provided, the cultivation, with suitable supply of nitrogen, which is the source of strength for phytoplankton and, as a consequence, caused a good level of diatoms to the system.

Thus, there are a number of obstacles for a satisfactory management of the levels of phytoplankton in growout ponds, such as, for instance, to stimulate the growth of desirable diatoms by the use of chemical fertilizers and probiotics, for not all groups of phytoplankton bring benefits to the ecosystem in ponds. But obviously, the bigger the density of shrimps cultivated, the more serious damage there will be for the cultivation in case of problems related to the overgrowth of a certain phytoplankton group, such as cyanobacteria and dinoflagellates . These may generate blooms (overpopulation) causing depreciation of the levels of oxygen and even causing the death of shrimps by toxins liberated (SHUMWAY, 1990).

So it is necessary to investigate which groups found in growout ponds would be the best to handle in order to keep excellent levels of phytoplankton.

### Conclusion

The densities of phytoplankton found in the systems studied were below those recommended for semi-intensive and intensive cultivations. As to organic cultivation, recommended levels of phytoplankton were not found in the literature researched. Within the systems studied, organic cultivation was the most abundant in diatoms, being such organisms said to be of relevant importance for the cultivation marine shrimps according to specialized literature.

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