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Temporal variation of phytoplankton populations in response to granular and liquid fertilizers

Variación temporal de poblaciones de fitoplancton en respuesta a la fertilización granular y líquida

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Abstract. The aim of this study was to determine the effect of a granular or liquid fertilizer on the temporal variation of phytoplankton. Enrichment of the pond water with nitrogen and phosphorus resulted in an increased gross photosynthesis and biomass of the dominant algal species. Nutrient concentrations and dilution rates can determine the temporal variation in phytoplankton abundance and primary production. The variation may be largely maintained by nutrient regeneration or turnover from different forms.

Keywords: Phytoplankton; Fertilizer; Granular; Liquid; Phosphorus; Nitrogen.

Resumen. La finalidad del presente estudio fue determinar los efectos de un fertilizante granular y otro líquido sobre la variación temporal del fitoplancton. El enriquecimiento del agua del estanque con nitrógeno y fósforo resultó en un incremento en la fotosíntesis bruta y la biomasa de las especies de alga dominantes. La concentración de nutrientes y las tasas de dilución pueden determinar la variación temporal de la abundancia del fitoplancton y producción primaria. La variación puede ser mantenida principalmente por la regeneración de nutrientes o la tasa de renovación de diferentes formas.

Palabras clave: Fitoplancton; Fertilizante; Granular; Líquido; Fósforo, Nitrógeno.

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INTRODUCTION

Growth of phytoplankton in water bodies and ponds primarily depends on the availability of nutrients. Phosphorus and nitrogen are the main nutrients limiting phytoplankton in freshwater bodies (Schindler, 1977; Elser et al., 1990). In general, studies on the effects of mineral nutrients on phytoplankton have been conducted on water bodies of oligotrophic, mesotrophic and eutrophic types (Petrova et al., 1977; Pavlova, 2002) in surface waters and shallow ponds (Petrova et al., 1977). In these waters, the limitation of phytoplankton by nutrients is considered unlikely because light and temperature conditions are more important (Golterman, 1983). However, few studies have addressed the effects of nitrogen and phosphorus application on the temporal changes in the phytoplankton composition of eutrophic waters.

Fertilizers have been used in various combinations to increase the number of phytoplankton and zooplankton available to fish as their primary food (Klimczyk, 1964; Geiger, 1983; Farquhar, 1987; Ponce-Palafox et al., 2010). The use of both organic and inorganic fertilizers increases the amount of phytoplankton developing within ponds, which causes a significant increase in the growth of fish and crustaceans that are reared. Farquhar (1987) found that granular fertilizers were as effective as liquid fertilizers when they were allowed to dissolve from a floating screen. Knowledge concerning the natural food preferences of fish and crustaceans can be used for both wildlife management in eutrophic bodies and for the management of the natural productivity of ponds containing fish and crustaceans. The aim of this study was to determine the effect of granular *versus* liquid fertilizers on the temporal variation of phytoplankton populations.

MATERIALS AND METHODS

The experiment was conducted at the Islamic Azad University over twenty-one days. Nine 300-L fiberglass ponds were distributed in a randomized design, and three types of fertilizer were added to the water in these ponds: granular fertilizer (GF), liquid fertilizer at low concentration (LFL) and liquid fertilizer at high concentration (LFH). The ponds were located outdoors, and each contained 150 L of water derived from an irrigation canal. For the initial elimination of zooplankton that would prevent the development of phytoplankton, 1 mg/L of trichlorofom was added shortly after the water was introduced into the ponds. After one hour of detoxification, phosphate and nitrogen fertilizers at a 1:1 ratio were added to the ponds. The amount of fertilizer used was estimated based on the tank volume and the phosphate and nitrogen contents of the fertilizers.

The chemical fertilizer composition was analyzed in the laboratories of the Islamic Azad University. Composition was as follows: LFL= NO_3 = 8%, P_2O_5 = 4%, K_2O = 5%, Fe = 3000 ppm, Zn = 1500 ppm, Mn = 1500 ppm, Cu = 1200 ppm and B = 1000 ppm; LFH= NO_3 = 16%, P_2O_5 = 8%, K_2O = 8%, Fe = 3000 ppm, Zn = 1500 ppm, Mn = 1500 ppm, Cu = 1200 ppm and B = 1000 ppm; GF= urea plus diammonium phosphate, $(NH_2)2CO$ + $(NH_4)2HPO_4$. The total mineral levels were determined through acid extraction (using nitric acid with perchloric acid). Nitrogen was determined using the Kjeldahl method, and carbon monoxide was measured using the Walkley-Black method. Phosphorus content was determined by a colorimetric method using vitamin C as the reductant. Calcium, magnesium, copper, iron, manganese and zinc levels were determined using an atomic absorption spectrophotometer, and sulfur and boron levels were determined by colorimetry.

To characterize the physical and chemical properties of the water, temperature and dissolved oxygen were measured with a digital oximeter; pH was measured with a digital pH meter, and transparency was measured using a Secchi disc. Ammonia, nitrogen, and orthophosphate levels were quantified in the laboratory (three replicates). Net and gross photosynthetic rates were determined using previously outlined procedures (Arredondo-Figueroa & Ponce-Palafox, 1998; Dodds & Whiles, 2010).

Water samples were collected once every seven days to specify the phytoplankton composition and abundance within the experimental ponds during the interval between the two fertilizing instances. After fixation of the samples with a 10% formalin solution, cell counts and determination of the dominant phytoplankton species were conducted in the laboratory. Abundance of the taxa was expressed as the number of cells per liter (Utermöhl, 1956; Aktan et al., 2005). The total number of cells was determined from the cell counts according to Stirling (1985) using the formula:

$$N = \frac{(A \times 1000 \times C)}{(v \times F \times V)}$$

where

N = the number of phytoplankton cells/L of original water,

A = the number of phytoplankton cells counted,

C = the volume of the final concentrate of the samples (mL),

v = the volume of a field (mm^3),

F = the number of fields counted, and

V = the volume of original water (L).

Statistical analyses were performed using STATISTICA software version 10 for PC (StatSoft Inc., USA). An analysis of variance was used to detect significant differences in the physicochemical and biological factors among the treatments. After F tests were significant, treatment means were compared using Tukey´s test ($p<0.05$). The data were standardized (to a mean of zero and a standard deviation of 1) before performing a principal components analysis (PCA, Digby &

Kempton, 1987). All algal data were $\log_{10}$ transformed to normalize the data before conducting regression and correlation analyses. Varimax-rotated principal component analysis was applied to maximize the variance between resulting components to make them as distinctive as possible.

RESULTS

The pH, concentration of dissolved oxygen and gross photosynthesis were significantly higher in the LFH than in the other treatments (Table 1). The pH and net photosynthesis values were significantly lower in the GF treatment. The diurnal variation in the air and in the average tank water temperatures showed a conventional pattern throughout the study. The pH in all water ponds tended toward alkalinity during the study. A significantly higher water transparency was recorded in the GF than in the LFH treatment (Table 1).

A total of 24 genera of phytoplankton, which belonged to the Bacillariophyta, Chlorophyta and Cyanophyta, were identified. The main genera were *Oscillatoria* (42%), *Schroederia* (42%) and *Melosira* (9%) in the LFL treatment; *Scenedesmus* (48%), *Micractinium* (14%), *Ankistrodesmus* (11%), *Chlorogonium* (10%) and *Schroederia* (95%) in the LFH treatment; and *Oscillatoria* (49%) and *Spirulina* (45%) in the GF treatment.

Overall, the highest abundance of Bacillariophyta was recorded at 14 days from study initiation (Fig. 1) in the LFL and LFH treatments. The greatest abundance of Chlorophyta occurred after 21 days in the LFL treatment, and after 14 days in the LFH treatment. The Cyanophyta were most abundant after 21 days in the GF treatment. The greatest abundance of *Nitzschia* was recorded after 14 days in the LFL and LFH treatments, and after 21 days in the GF treatment (Fig. 2). *Ankistrodesmus* was most abundant after 14 days in all three treatments (Fig. 3). The greatest abundance of *Scenedesmus* and *Micractinium* occurred at 7 to 21 days from study initiation depending on the fertilizer treatment. *Schroederia* was the most abundant microalga within 14 days of the initial fertilizer application for all but the LFH treatment. *Anabaena*, *Gomphosphaeria* and *Merismopedia* were most abundant at 7 days after the fertilizer applications (Fig. 4). The response of *Oscillatoria* depended upon the fertilizer treatment. This genus was most abundant after 14, 7 and 21 days from study initiation in the LFL, LFH and GF treatments, respectively. The greatest abundance of *Spirulina* was after 7 days from initiation of the study in the liquid fertilizer treatments, and after 14 and 21 days in the GF treatment.

The combination of the PC1 (34.65%) and PC2 (28.25%) site scores accounted for 62.9% of the variation in the data set. PC1 accounted for a large amount of the total variation in both of the algal-abundance data sets, but only PC1 was related to the Chlorophyta abundance (LFL), whereas PC2 was related to the Bacillariophyta abundance (LFH). The results of the principal component analysis (Fig. 5) indicated that the algal response to the GF was directly proportional to components I and II, and was associated with the Cyanophyta. The response to the LFH was inversely proportional to component I and associated with the Bacillariophyta and Chlorophyta. The algal response to the LFL was indirectly proportional to component II and associated with the Chlorophyta.

DISCUSSION

In this research, we found that the water pH level increased more with the LFL and LFH treatments than with the GF treatment. This effect is attributable to the higher photosynthetic capacities of the treatments where the liquid fertilizers were used, which may be a result of the lower oxygen solubility and the abundance of organic materials, including the phytoplankton (Ghosh & Chattopadhyay, 2005). Jana and Patel (1984) showed that increased algal blooms are associated with an increased water pH. Although the phosphorous and nitrogen contents of the LFL and GF treatments were

Table 1. Physico-chemical Parameters of the experimental system under investigation.
Tabla 1. Parámetros fisico-químicos del sistema experimental bajo investigación.

Parameter	Treatment (Mean ± SD)		
	GF	LFF	LFH
Temperature (°C)	24.28 ± 1.88a	24.44 ± 1.89a	24.60 ± 1.99a
pH	9.49 ± 0.42 b	9.81 ± 0.39 b	10.11 ± 0.59 a
Transparency (cm)	33.63 ± 1.64 a	31.70 ± 4.94 a	25.11 ± 7.58 b
Dissolved oxygen (mg/L)	11.49 ± 1.65 b	10.97 ± 2.35 b	13.23 ± 2.61 a
Ammonium nitrogen (mg/L)	0.34 ± 0.09 a	0.37 ± 0.16 a	0.40 ± 0.16 a
Orthophosphate (mg/L)	2.38 ± 2.41 a	1.57 ± 1.32 a	2.58 ± 2.52 a
Gross Photosynthesis (mg/cm^3.h)	89.00 ± 32.46 b	66.23 ± 30.80 b	114.76 ± 56.26 a
Net Photosynthesis (mg/cm^3.h)	28.25 ± 26.44 b	44.52 ± 35.53 a	32.73 ± 26.80 a

Different letters within the same row represent significant differences ($p<0.05$) among treatments.
Diferentes letras en una misma fila representan diferencias significativas (p<0,05) entre tratamientos.

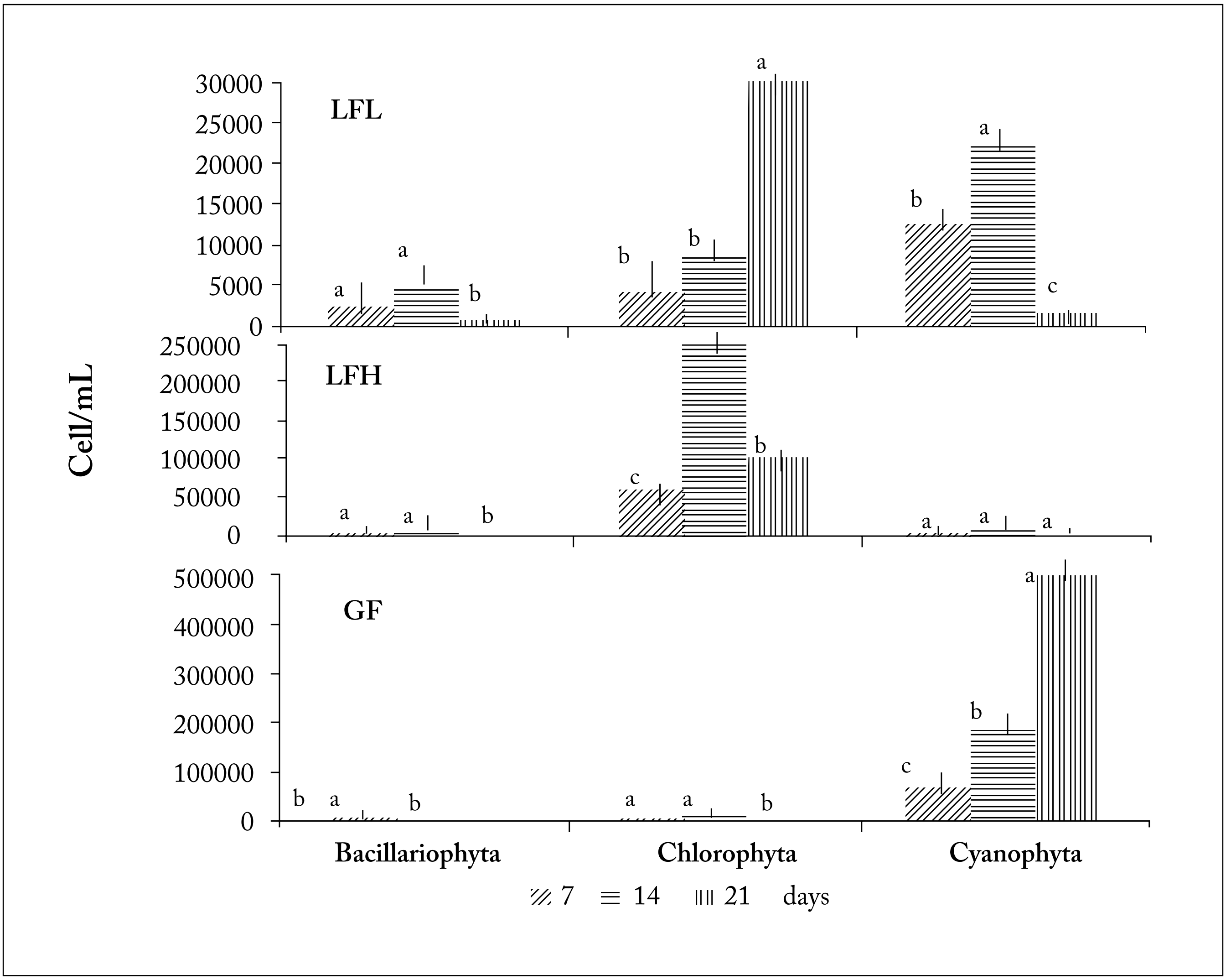

Fig. 1. Abundance of Bacillariophyta, Chlorophyta and Cyanophyta in the liquid and granular fertilizer treatments during 21 days.
Fig. 1. Abundancia de Bacillariofitas, Clorofitas y Cianofitas en los tratamientos de fertilizante líquido y granular durante 21 días.

considered to be equivalent, the dissolved ammonium nitrogen and orthophosphate resulting from the LFH fertilization were higher than those observed in the LFL and GF treatments. This result shows that the solubility of the nitrogen- and phosphorous-based biogenic materials was greater in the LFH treatment than in the two other fertilizer treatments. A downward trend in the amount of ammonium nitrogen and orthophosphate occurred in all of the treatment ponds after fertilization. However, this decline in biogenic substances was higher in the GF treatment than in the other two treatments. The intensity of the decline in the ammonium nitrogen in the water column is closely related to the algal density and the sedimentation rate of these substances on the bottom of the fish ponds (Jana & Roy, 1986).

In this research, the effects of the nitrogenous and phosphorous liquid (LFL and LFH) *versus* granular (GF) fertilizers were examined with regard to the density and composition of the algae growing in ponds consisting of the water-filled fiberglass containers. The results indicated that the population densities of the different algal genera varied with the phytoplankton developmental stages observed within the study reservoirs or fish farming ponds (Boyd, 1995; Schneck et al., 2011).

Our examination of the phytoplankton in the experimental ponds revealed variation in the density and composition of the algal species among the three fertilizer treatments. The diversity and development of the algal populations within fish ponds are greatly affected by various factors, including temperature, type of water body, and biogenic constituents of the water supply (Stanley et al., 2000; Monika & Deborah, 2008). Among the algal species examined, *Scenedesmus*, *Micractinium* and *Ankistrodesmus* occurred with higher abundance in the LFH-treated pond than in the other treatments, whereas *Spirulina* and *Oscillatoria* were more prevalent in the GF-treated pond.

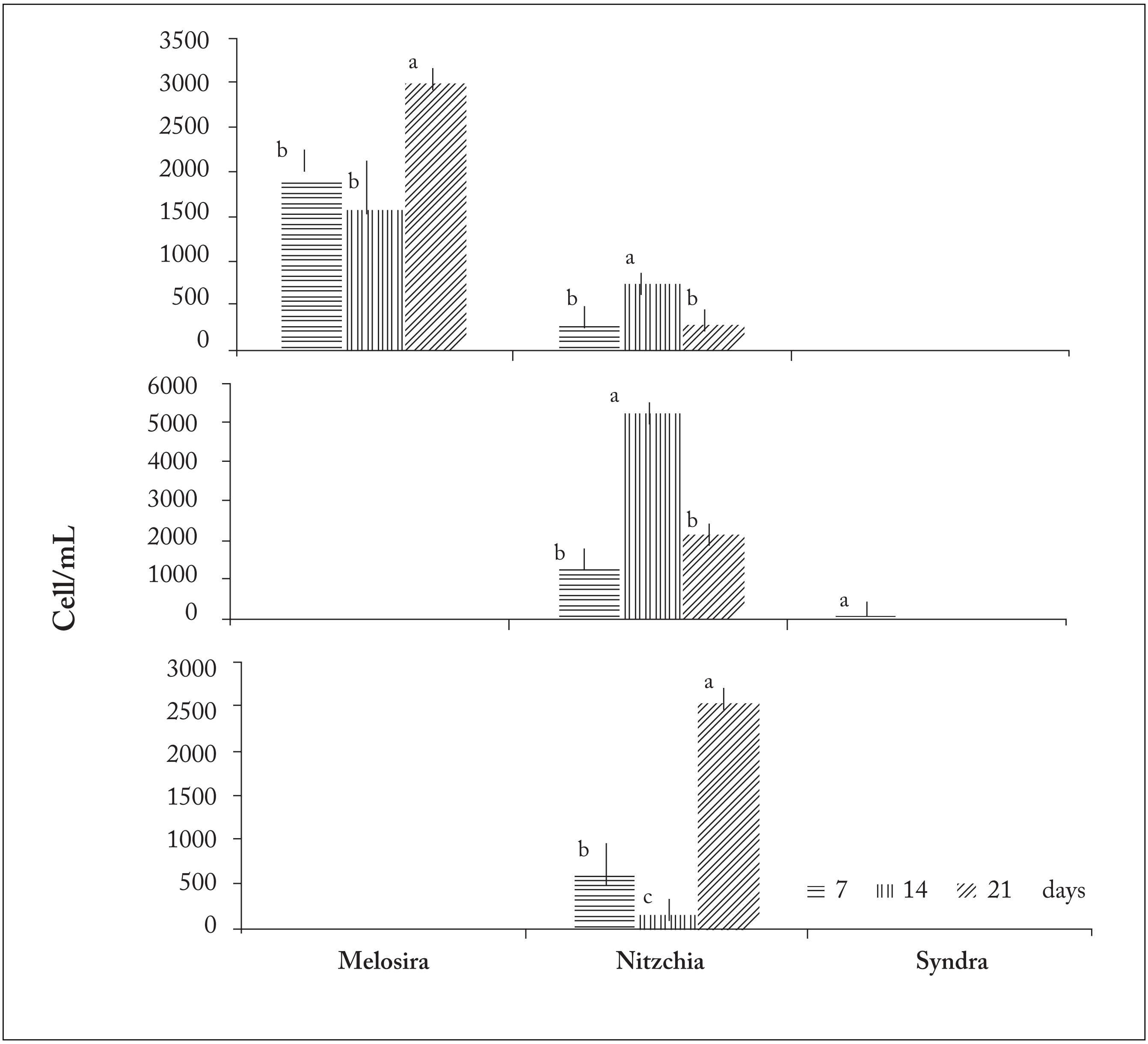

Fig. 2. Abundance of Bacillariophyta species in the three fertilizer treatments during 21 days.
Fig. 2. Abundancia de Bacilariofitas en los tres tratamientos de fertilizantes durante 21 días.

The three weekly observations indicated that the LFH treatment produced more Chlorophyta than it was observed in the other two fertilization treatments. This result could have occurred because of the high nutrient concentrations and the immediate availability of the inorganic nitrates and phosphates in the tank, encouraging the growth of the Chlorophyta species. This is consistent with previous studies (Becker, 1986; Ansa & Akinwale, 2007). Moreover, the Chlorophyta species belong to the "r" group of algae, which blooms in response to pulses of high-quality fertilizer, such as the LFH, in contrast to the "K" group of algae, such as the cyanophyte species, which bloom and develop rapidly in response to low-quality fertilizers. Therefore, under identical climatic conditions and biogenic consumption rates, the LFH treatment could produce more Chlorophyta biomass. The Chlorophyta are among the most important algae consumed by fish feeding in phytoplankton (Seymour, 1980; Ludwig et al., 1998).

Shrimp and fish are better able to digest Chlorophyta and Bacillariophyta than Cyanophyta (Villalon, 1991; Stockner & Cronberg, 2000), which has been clearly demonstrated for silver carp (Smith, 1989; Radke & Kahl, 2002). Because the total algal population density in the LFH-treated tanks was greater than that observed in the ponds treated with the GF, the Chlorophyta must account for most of the algal biomass in the LFH

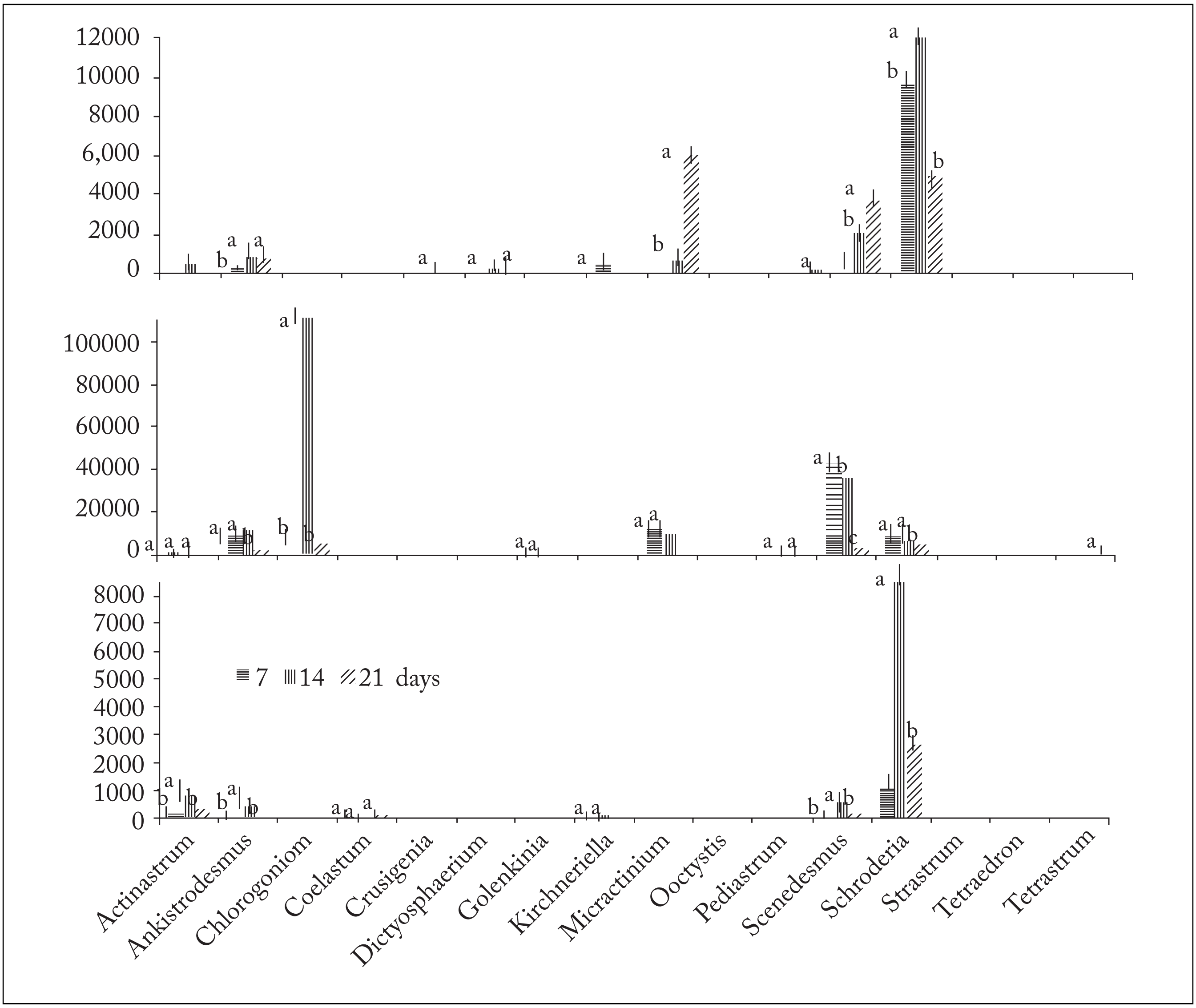

Fig. 3. Abundance of Chlorophyta species in the three fertilizer treatments during 21 days.
Fig. 3. Abundancia de Clorofitas en los tres tratamientos de fertilizantes durante 21 días.

treatment. However, despite the greater phytoplankton biomass observed in the GF treatment, Cyanophyta constituted the bulk of the algal population in the GF-treated pond, and these algae have a lower digestibility and are absorbed at a lower rate (Starling & Rocha, 1990). The results showed that the LFH treatment strongly induced the Chlorophyte population within the algal biomass (92.49%). Based on these findings, we recommend that nursery ponds be fertilized with either granular or liquid fertilizer depending on the requirements of cyanophyte or chlorophyte species, respectively, for raising finfish and crustacean larvae or for the extensive culture of algae as live food organisms. The main genera that should be used to feed fish and crustacean are (1) *Scenedesmus*, which is best fertilized with LFH, (2) *Nitzschia*, which is best fertilized with LFL, and (3) *Spirulina*, which is best fertilized with GF. In general, it was found that the temporal variability in the phytoplankton species composition fit expectations. Studies performed in lakes, reservoirs, shallow waters and farm ponds identified temporal variation in phytoplankton populations caused by physical (temperature and light), chemical (nutrients, mainly nitrogen, phosphorus and silica), biological (nutritional selectivity) and population factors (zooplankton).

CONCLUSIONS

The present study examined the effect of the nutrient supply, and its liquid or solid composition, on the phytoplankton populations. It was found that the nutrient concentration and dilution rate can determine a temporal variation in the phytoplankton abundance and primary production, and

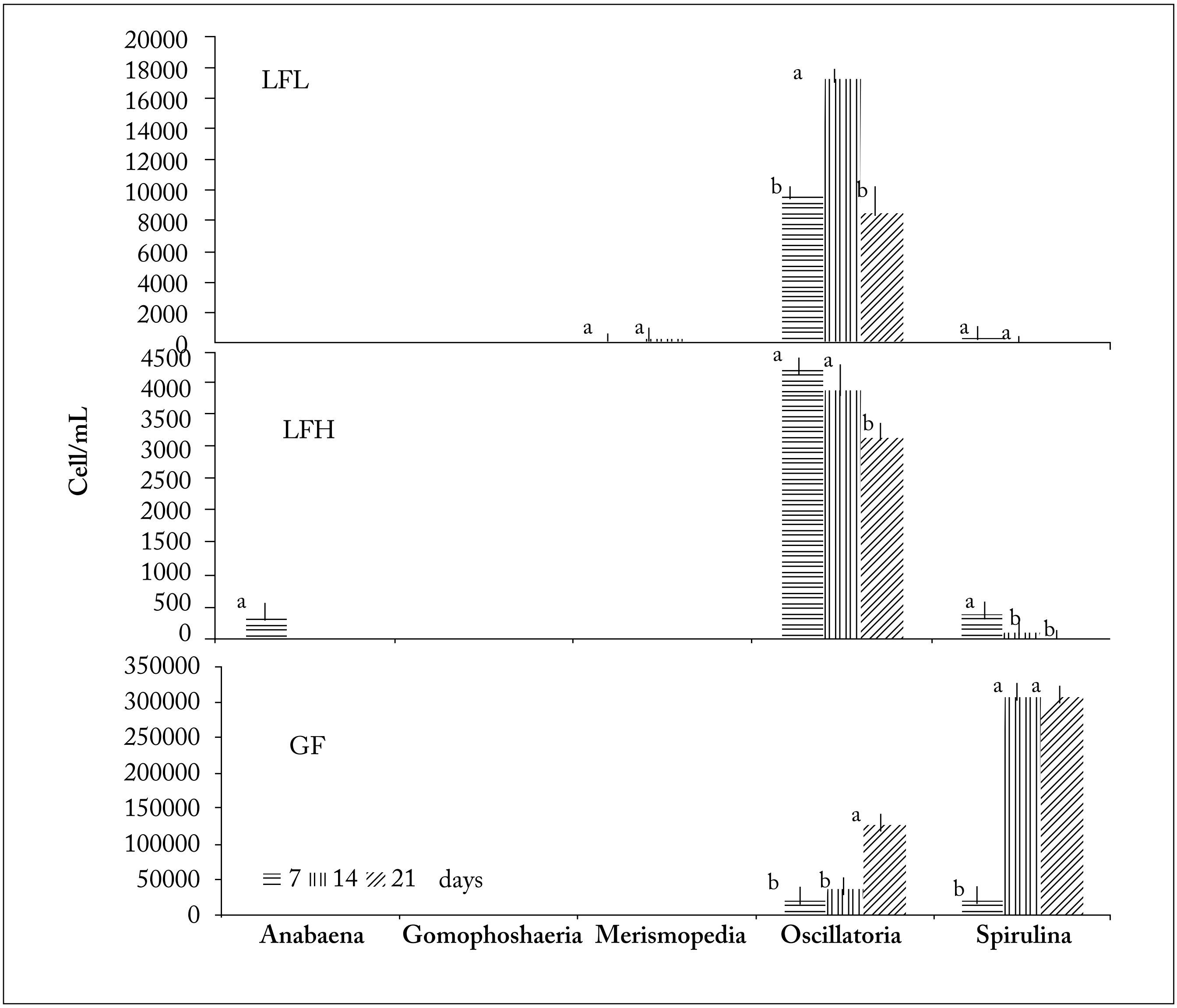

Fig. 4. Abundance of Cyanophyta species in the three fertilizer treatments during 21 days.
Fig. 4. Abundancia de Cianofitas en los tres tratamientos de fertilizantes durante 21 días.

it can be maintained largely by nutrient renewal or turnover from different nutrient forms. The results indicated that the concentration and availability of phosphorus are major factors regulating algal succession. As phosphorus concentrations increased, the biomass of algal groups indicative of eutrophic conditions also increased. Applying fertilizer to the water allowed to know that eutrophication tendency increased from treatment LFL to GF. Again, this finding supports the view that an increase in nutrient concentrations increases the biomass of those algal species that are less efficient in utilizing nutrients. Some species (such as the Cyanophyta) appear to be able to rapidly exploit a nutrient-rich environment, while other species, such as the Chlorophyta and Bacillariophyta, were more efficient in utilizing nutrients present at low concentrations. The characterization "efficient in utilizing nutrients" possibly also applies to some of the "problem species" in the eutrophication process, i.e., those species constituting the largest biomasses in lakes, reservoirs, shallow waters and farm ponds.

REFERENCES

Aktan, Y., V. Tufekci, H. Tufekci & G. Aykulu (2005). Distribution patterns, biomass estimates and diversity of phytoplankton in Izmit Bay (Turkey). *Estuarine, Coastal and Shelf Science* 64: 72-384.

Ansa, E.J. & M.M.A. Akinwale (2007). The potential suitability of nocturnally occurring plankton flora in earthen freshwater nursery ponds. *Research Journal of Applied Science* 2: 697-703.

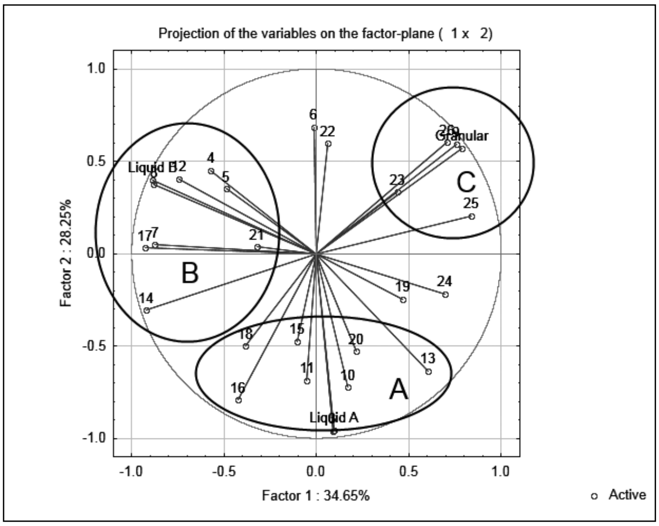

Fig. 5. PCA of the cell counts of phytoplankton species in the granular and liquid fertilizer treatments (A = LFL, B = LFH and C = GF).
Fig. 5. PCA de la abundancia de las especies del fitoplancton en los tratamientos de fertilizantes líquido o granular (A = LFL, B = LFH y C = GF).

Arredondo-Figueroa, J.L. & J.T. Ponce-Palafox (1998). La calidad del agua en acuicultura: Conceptos y Aplicaciones. AGT: Editors S.A. México. 250 p.

Becker, E.W. (1986). Nutritional properties of microalgae, potentials and constraints. In: Richmond, A. (Ed.). CRC Handbook of Microalgal Mass Culture. Boca Raton Florida. pp. 339-419.

Boyd, C.E. (1995). Bottom soils, sediment, and pond aquaculture. Chapman and Hall, New York. 301 p.

Digby, P.G.N. & R.A. Kempton (1987). Population and Community Biology Series: Multivariate Analysis of Ecological Communities. Chapman and Hall, London.

Dodds, W. & M. Whiles (2010). Freshwater Ecology: Concepts & environmental applications of Limnology. Academic Press. Burlington, MA 01803 USA.

Elser, J.J., E.R. Marzolf, & C.R. Goldman (1990). Phosphorus and Nitrogen Limitation of Phytoplankton Growth in the Freshwaters of North America: A Review and Critique of Experimental Enrichments. *Canadian Journal of Fisheries and Aquatic Sciences* 47: 1468–1477.

Farquhar, B.W. (1987). Comparison of granular and liquid inorganic fertilizers used in striped bass and smallmouth bass rearing ponds. *The Progressive Fish Culturist* 49: 21-28.

Geiger, J.G. (1983). A review of pond zooplankton production and fertilization for the culture of larval and fingerling striped bass. *Aquaculture* 35: 353-369.

Ghosh, M. & N.R. Chattopadhyay (2005). Effects of carbon/nitrogen/phosphorus ratio on mineralizing bacterial population in aquaculture systems. *Journal of Applied Aquaculture* 17: 85-98.

Golterman, H.L. (1983) Algal Bioassays and Algal Growth Controlling Factors in Eutrophic Shallow Lakes. *Hydrobiologia* 100: 59–64.

Jana, B.B. & G.N. Patel (1984). Spatial and seasonal variations of phosphate solubilizing bacteria in fish ponds of varying fish farming managements. *Archives für Hydrobiologie* 10: 555-568.

Jana, B.B. & S.K. Roy (1986). Seasonal and spatial distribution pattern of nitrogen fixing bacteria in fish ponds under different management systems. *Hydrobiologi*a 137: 45-54.

Klimczyk, M. (1964). Zooplankton and its biomass in fertilized ponds. *Acta Hydrobiologia* 6: 187-205.

Ludwig, G.M., N.M. Stone & C. Collins (1998). Fertilization of fish fry ponds. Southern Regional Aquaculture Center publication #469. 25 p.

Monika, W. & A.H. Deborah (2008). Temporal organization of phytoplankton communities linked to physical forcing. *Oecologia* 156: 179-192.

Pavlova, O.A. (2002). An Experimental Study of the Effect of Addition of Biogenic Elements on the Phytoplankton of an Eutrophic Polluted Lake, in *Biologiya vnutrennikh vod: problemy ekologii i bioraznoobraziya: Mater. XII Mezhdunar. Konf. Molodykh Uchenykh* (Inland Water Biology: Problems of Ecology and Biodiversity. Proc. XII Int. Conf. Young Sci.), Borok, 2002, pp. 166–173.

Petrova, N.A., G.F. Raspletina & B.L. Gusakov (1977). The study of the demands of the phytoplankton of lakes of different types in biogenic elements by the method of planned additives. *Botanicheskij Zhurnal* 62: 984–989.

Ponce-Palafox, J.T., J.L. Arredondo-Figueroa, S.G. Castillo-Vargasmachuca, G. Rodríguez Chávez, A. Benítez-Valle, M.A. Regalado de Dios, F. Medina-Carrillo, R. Navarro-Villalobos, J.A. Gómez-Gurrola & P. López-Lugo. (2010). The effect of chemical and organic fertilization on phytoplankton and fish production in carp (cyprinidae) polyculture system. *Revista Bio Ciencias* 1: 44-50.

Radke, R. & U. Kahl (2002). Effects of a filter-feeding fish [silver carp, *Hypophthalmichthys molitrix* (Val.)] on phyto and zooplankton in a mesotrophic reservoir: results from an enclosure experiment. *Freshwater Biology* 47: 2337-2344.

Schindler, D.W. (1977). Evolution of phosphorus limitation in lakes. Natural mechanisms compensate for deficiencies of nitrogen and carbon in eutrophied lakes. *Science* 195: 260–262.

Schneck, F., A. Schwarzbold, S. Rodrigues & A.M. Melo (2011). Environmental variability drives phytoplankton assemblage persistence in a subtropical reservoir. *Austral Ecology* 36: 839-848.

Seymour, E.A. (1980). The effects and control of algal blooms in fish ponds. *Aquaculture* 19: 55-74.

Smith, D.W. (1989). The feeding selectivity of silver carp, *Hypophthalmichthys molitrix* Val. *Journal of Fish Biology* 34: 819-828.

Stanley, I.D., E.A. Shelley & L.C. Kathryn (2000). The relationship in lake communities between primary productivity and species richness. *Ecology* 81: 10, 2662-2679.

Starling, F.L.R.M. & A.J.A. Rocha (1990). Experimental study of the impacts of planktivorous fishes on plankton community and eutrophication of a tropical Brazilian Reservoir. *Hydrobiologia* 200/201: 581-591.

Stirling, H.P. (1985). Chemical and biological methods of water analysis for aquaculturists. Stirling: Institute of Aquaculture, University of Stirling. UK. 117 p.

Stockner, J.G. & G. Cronberg (2000). Picoplankton and other non-bloom forming Cyanobacteria in lakes. In: Whintton, B.A. & M. Potts (Eds.). The Ecology of Cyanobacteria. Kluwer Academic Publishers, the Netherlands, pp. 195-231.

Utermöhl, H. (1958). Zur vervollkommnung der quantitativen Phytoplankton-Methodik. *Mitteilungen Internationale Vereinigung für Theoretische und Angewandte Limnologie* 9: 1-38.

Villalon, J.R. (1991). Practical Manual for the Semi-intensive Commercial Production of Marine Shrimp. Texas A&M University Sea Grant College Program Publication. 104 p.