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# Determination of Fertilization Rate for Optimum Pond Productivity and Fish Growth in Inland Saline Groundwater Ponds: Monoculture of Grey Mullet and Milkfish

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## Abstract

The effects of four rates of application of organic fertilizer (cow-dung) along with supplementary feeding were studied on water quality and pond productivity in terms of plankton production and fish biomass in inland saline groundwater ponds. Two experiments were conducted. In experiment 1, ponds were fertilized (at 5000, 7500, 10000 and 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) and stocked (at 6000 ha<sup>-1</sup>) with *Mugil cephalus*, while in experiment II, *Chanos chanos* was stocked (at 6000 ha<sup>-1</sup>) and ponds were fertilized at 2500, 5000, 7500 and 10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>. The growout period for both the experiments was 100 days. Irrespective of the species stocked, significantly (P<0.05) high growth performance of grey mullet and milkfish in terms of biomass and mean fish weight gain were observed in ponds fertilized at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup> of cow-dung. Significantly (P<0.05) high values of parameters indicative of productivity (viz. turbidity, alkalinity, net primary productivity-NPP, Chlorophyll *a* concentration and plankton population) also coincided with the high fish growth. Nutrient release (o-PO<sub>4</sub>, NO<sub>3</sub>-N and total kjeldahl nitrogen) also remained significantly (P<0.05) high in ponds fertilized at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>. At higher or lower rates of fertilization, the values of most of these parameters remained low. Application of stepwise multiple regression models showed a significant and positive correlation of nutrients, NPP and plankton population with fish growth. Statistically also fish weight gain showed a significant positive correlation with nutrients [viz. NO<sub>3</sub>-N (r=0.66, P<0.001; r=0.36, P<0.05), o-PO<sub>4</sub> (r=0.70, P<0.001; r=0.27, P<0.05), alkalinity (r=0.45, P<0.001; r=0.50; P<0.001) and Kjeldahl-N (r=0.37, P<0.05; r=0.45, P<0.001)], net primary productivity (r=0.56, P<0.001, r=0.49, P<0.001) and plankton density (r=0.51, P<0.001; r=0.73, P<0.001) in both the experiments, respectively. A decline in fish biomass and pond productivity at higher rates of fertilization (10000 or 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) has been attributed to high NH<sub>4</sub>-N and BOD<sub>5</sub> levels. Further, based on estimated production function relating pond productivity (fish production, dissolved oxygen concentration and net primary productivity) to fertilizer loading rate (dosages of cow-dung), optimum fertilizer loading rates were computed and found to be 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>. Therefore, a dosage of 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup> of cow-dung along with supplementary feeding appears to be optimum for obtaining high productivity in inland saline water ponds with salinity levels ranging between 13.0-21.0 ppt.

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## Introduction

The use of different animal manures as the principal nutrient input to the fish ponds for enhancing pond productivity has been reviewed by different workers (Wohlfarth and Schroeder 1979; Edwards 1980; Wohlfarth and Hulata 1987). The type of fertilizer and the quantities to be used differ considerably from one region to another and even between different aquaculture management practices. Further, much of technical literature on manure utilization in fish ponds has been concerned mainly with temperate regions using carps as a main species (Hopkins et al. 1980). For sustaining pond productivity and for high fish production in subtropical regions, it is imperative to evaluate the fertilization rates for maximizing pond productivity, while maintaining optimal hydrobiological conditions. Based on soil and water quality, dosages have been worked out for both organic and inorganic fertilizers, however, the response to fertilization has been varying. Few studies that too under fresh water conditions have been carried out to determine the fertilizer dosage required in fish ponds for obtaining optimum pond productivity without adversely affecting pond ecology (Hopkins and Cruz 1982; Wohlfarth and Hulata 1987; Garg and Bhatnagar 1996) while, not much information on these lines is available from brackishwater ponds (Garg and Bhatnagar 1999). Johnson and Guenzi (1963) had stated that mineralization of organic matter is affected by salinity and SAR values of water. Further, decomposition of organic manures in brackishwater ponds is likely to be influenced not only by the prevailing anaerobic conditions in the pond soils, but also by the water salinities. Recent studies of Bhatnagar et al. (1994) have shown that the rate of degradation in terms of viable bacterial counts and nutrient release decreases with an increase in salinity. Since a slight increase in manure loading rate can cause water quality to deteriorate especially in shallow ponds, which may add up to the dissolved oxygen demand in the early hours of the morning and also when ambient temperature is high resulting in retardation of fish growth. Since dissolved oxygen (DO) shows an inverse relationship with salinity, therefore, the demand of oxygen in such manure loaded ponds may become more acute.

Therefore, to optimize the use of organic fertilizer in inland saline groundwater fish ponds, present studies were conducted. The objectives of the study were to examine the impact of four different dosages of organic fertilizer (cow-dung) on (i) Physico-chemical characteristics of pond water (ii) their nutrient status (iii) impact on pond productivity and plankton production in stagnant fish ponds with high salinity (13.0-21.0 ppt).

## Materials and Methods

Experiments were conducted in earthen ponds each measuring 15m×25m (area 375m<sup>2</sup>, depth 1.2 m) at the brackishwater fish pond facility attached to the Department of Zoology and Aquaculture, CCS Haryana Agricultural University, Hisar, India. Ponds were cleaned, and quick lime (CaO) at 200 kg•ha<sup>-1</sup>•y<sup>-1</sup> was applied. After the addition of first dose of fertilizer (cow-dung), ponds were filled with inland saline groundwater by pumping from borewells and allowed to stabilize for about two weeks. To maintain the desired level (90 cms) water was replenished as often as required. Semi-dry cow-dung (humidity: 30-40%) was thoroughly mixed in water (in the ratio 1:3 w/v) before spreading the same on water surface. Two experiments were conducted.

**Experiment I: Effect of four different dosages (5000, 7500, 10000 and 12500 kg·ha<sup>-1</sup>·y<sup>-1</sup>) of organic fertilizer (cow-dung) on pond productivity and growth performance of grey mullet, *Mugil cephalus*.**

In this experiment grey mullet fry (mean BW 1.13 g, length 4.15 cm) were stocked in April 2003. Water salinity fluctuated between 13.5-15.0 ppt during the experimental period of 100 days.

**Experiment II: Effect of four different dosages (2500, 5000, 7500 and 10000 kg·ha<sup>-1</sup>·y<sup>-1</sup>) of organic fertilizer (cow-dung) on pond productivity and growth performance of milkfish, *Chanos chanos*.**

In this experiment milkfish fry (mean BW 0.61 g, length 4.57 cm) were stocked in July 2003. Water salinity fluctuated between 13.0-21.0 ppt during the experimental period of 100 days.

Stocking density in both experiments was kept at 6000 ha<sup>-1</sup>. Two replicates of each treatment were maintained. Ponds were fertilized using cow-dung at biweekly interval using four different fertilizer dosages (Table 1). Supplementary feed at 4% BW·d<sup>-1</sup> was given daily once only between 08h00 and 09h00 to the fish and the feeding rate was adjusted every 15 day after weighing a representative sample of about 50-60 fish from each replicate pond per treatment.

Table 1. Experimental protocol and dosages of cow-dung\*

Treatment no.	Expt. I ( <i>Mugil cephalus</i> )	Expt. II ( <i>Chanos chanos</i> )
	Dosage of fertilizer kg·ha <sup>-1</sup> ·y <sup>-1</sup> (kg 375 m <sup>-2</sup> w <sup>-2</sup> )	Dosage of fertilizer kg·ha <sup>-1</sup> ·y <sup>-1</sup> (kg 375 m <sup>-2</sup> w <sup>-2</sup> )
1	5000 (7.8)	2500 (3.9)
2	7500 (11.7)	5000 (7.8)
3	10000 (15.6)	7500 (11.7)
4	12500 (19.5)	10000 (15.6)

\* Approximate composition (%) of cow-dung was: dry matter 89.00; crude protein 9.56; nitrogen extract, 45.00; free soluble ash 1.08; nitrogen 0.59; phosphorus 0.33 and potassium 0.83 (Garg and Bhatnagar 2000).

### Water quality monitoring

For the determination of water quality parameters, water samples in replicates of four were obtained from each pond (i.e. 8 samples per treatment) before sunrise. Water temperature (°C), salinity (ppt), conductivity (dSm<sup>-1</sup>) and pH were recorded daily. All other water quality parameters were monitored only twice (50 and 100 days).

For estimating the total assimilation rate by the producer organisms in ponds, net primary productivity (NPP) was determined following the light and dark bottle method and also by studying diurnal changes in dissolved oxygen (DO) concentrations (APHA 1998). For qualitative and quantitative estimations of phyto- and zooplankton, water samples were also collected in replicates of four from each pond (i.e. 8 samples per treatment) at the end of 50 and 100 days interval. Plankton samples were obtained by passing 20 L of water taken from five different locations through a plankton net (mesh size 125 µm). Plankton densities were estimated using a Sedgwick-Rafter cell (Wetzel and Likens 1979) under a binocular microscope. Plankton species diversity ( $\bar{d}$ ) was determined using Shanon and Weaver's diversity index formula (Washington 1984). For the determination of chlorophyll *a* concentrations, a known amount (10L from five different locations from each treatment) of

water was filtered through Whatman filter paper (No. 40) and was determined following Boyd (1990).

### ***Fish growth***

Fish growth (length and weight) was monitored at the end of 50 and 100 days in both the experiments. At one hundred days post-stocking, ponds were completely drained and the fish were harvested and counted. Thereafter, individual length (cm) and weight (g) were recorded and length-weight relationship (LWR) was calculated according to the following equation.

$$W = cL^n \text{ (Logarithmic form of equation is : } \log W = \log c + n \log L \text{)}$$

Where,

W= weight in kg, c=constant, n=exponential value of length, L=length of fish in cm.

Plankton species diversity was determined using the diversity index formula.

$$\bar{d} = -\sum(n_i/N) \log_2(n_i/N)$$

Where,

$\bar{d}$  = species diversity,  $n_i$ =no. of individual of  $i^{\text{th}}$  species

N=total No. of individuals

### ***Statistical analysis***

The data were subjected to ANOVA to test the effect of treatment using the following model:

$$Y_{ij} = \mu + T_i + e_{ij}$$

$Y_{ij}$  =  $j^{\text{th}}$  observation of  $i^{\text{th}}$  treatment

$\mu$  = overall mean

$T_i$  = effect due to  $i^{\text{th}}$  treatment

$e_{ij}$  = random error NID ( $\sigma$ ,  $\sigma^2$ )

Data were further subjected to orthogonal polynomials for trend analysis. Means were compared using Tukey's test as described by Snedecor and Cochran (1982). Coefficient of correlation between different parameters and multiple regression between independent (hydrochemical) and dependent (biological and productivity) parameters were determined.

## **Results**

### ***Experiment I: Effect of four different dosages (5000, 7500, 10000 and 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of organic fertilizer (cow-dung) on pond productivity and growth performance of Mugil cephalus***

#### ***Fish growth***

Survival appeared to be independent of the treatments and varied between 75.0-86.0%. ANOVA revealed a significant ( $P < 0.05$ ) high growth [measured in terms of mean weight (g) and length (cm) gain, biomass and growth day<sup>-1</sup>] in ponds fertilized at the dosage level of 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>. A decrease in survival and growth performance was observed in ponds receiving high dosages (10000 and 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of organic fertilizer (Table 2). The exponential value ('n') of length-weight relationship (LWR) was 2.94 in ponds fertilized at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>, whereas, in other treatments the values of 'n' varied between 2.84-2.88. A detailed presentation of fish production in relation to fertilizer inputs is shown in Fig. 1.

Table 2. Effect of four different dosages of organic fertilizer (cow-dung) on growth performance and length-weight relationship (LWR) in *Mugil cephalus* and *Chanos chanos* (Stocking density 6000 kg•ha<sup>-1</sup>•y<sup>-1</sup>) under monoculture (Duration of experiment = 100 days)

Treatment cow-dung (kg•ha <sup>-1</sup> •y <sup>-1</sup> )	Initial fish stock		Final fish stock (after 100 days)		Increase in mean fish wt (g) (mean fish length cm)	Growth / day	Length-weight relationship (LWR)
	Stocking density/ 375 m <sup>2</sup>	Mean fish weight (g) (Mean fish length)	Survival (%)	Total Biomass (kg)			
<i>Mugil cephalus</i> (Experiment I)							
5000	225	1.11±0.11 (4.17±0.19)	80.0	43.05 <sub>B</sub> ±0.90	239.15 <sub>B</sub> ±4.60 (28.11 <sub>A</sub> ±0.22)	238.05 (23.94)	2.38 <sub>B</sub> W=0.000017×L2.88 log W= -4.76+2.88 log L
7500	225	1.10±0.09 (4.07±0.13)	86.0	51.05 <sub>A</sub> ±1.00	263.84 <sub>A</sub> ±4.78 (28.40 <sub>A</sub> ±0.22)	262.74 (24.33)	2.63 <sub>A</sub> W=0.000016×L2.94 log W= -4.76+2.94 log L
10000	225	1.15±0.09 (4.25±0.15)	81.0	25.59 <sub>C</sub> ±1.0	140.67 <sub>C</sub> ±5.01 (22.15 <sub>B</sub> ±0.33)	139.52 (19.16)	1.40 <sub>C</sub> W=0.000018×L2.85 log W= -4.75+2.85 log L
12500	225	1.14±0.10 (4.09±0.15)	75.0	20.32 <sub>D</sub> ±0.80	120.58 <sub>D</sub> ±4.37 (22.61 <sub>B</sub> ±0.46)	119.44 (18.52)	1.19 <sub>D</sub> W=0.000019×L2.84 log W= -4.73+2.84 log L
<i>Chanos chanos</i> (Experiment II)							
2500	225	0.66±0.05 (4.69±0.13)	93.2	9.54 <sub>D</sub> ±0.34	45.48 <sub>D</sub> ±1.79 (18.46 <sub>D</sub> ±0.15)	44.83 (13.77)	0.45 <sub>D</sub> W=0.0000045×L3.13 log W= -5.35+3.13 log L
5000	225	0.62±0.05 (4.51±0.13)	93.2	17.90 <sub>B</sub> ±0.58	85.38 <sub>B</sub> ±3.01 (23.15 <sub>B</sub> ±0.40)	84.76 (18.64)	0.85 <sub>B</sub> W=0.0000033×L3.29 log W= -5.48+3.29 log L
7500	225	0.58±0.04 (4.49±0.12)	94.2	23.11 <sub>A</sub> ±0.46	109.03 <sub>A</sub> ±2.39 (25.65 <sub>A</sub> ±0.31)	108.45 (21.16)	1.10 <sub>A</sub> W=0.0000035×L3.30 log W= -5.45+3.30 log L
10000	225	0.59±0.05 (4.57±0.12)	89.8	11.28 <sub>C</sub> ±0.38	55.81 <sub>C</sub> ±2.03 (19.86 <sub>C</sub> ±0.26)	55.22 (15.29)	0.55 <sub>C</sub> W=0.0000043×L3.17 log W= -5.37+3.17 log L

All values are mean±SE of mean. Means bearing different letters in the same column differ significantly (P<0.05)

### Physico-chemical characteristics of pond waters

Water conductivity fluctuated between 21.22±0.51 to 23.89±0.95 dSm<sup>-1</sup>, pH remained alkaline in all the treatments, slight but significantly (P<0.05) higher values were observed in ponds fertilized with the highest dosage of fertilizer. Dissolved oxygen (DO) concentration remained at optimal levels in all the treatments, slightly high values, however, were observed in ponds fertilized at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>. A detailed presentation of dissolved oxygen concentration in relation to fertilizer inputs is shown in Fig. 2. Total alkalinity, turbidity, orthophosphate (o-PO<sub>4</sub>), NO<sub>3</sub>-N and Kjeldahl-nitrogen increased with each increase in the fertilizer dosage up to the second treatment (7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>), and with further increase in the fertilizer dosage a decline in these parameters was observed. On the other hand, BOD<sub>5</sub> values increased significantly (P<0.05) with each increase in the dosage of fertilizer. NO<sub>2</sub>-N, Chlorides, hardness and total dissolved solids (TDS) showed no definite trend and remained high in all the treatments. Ammonical nitrogen (NH<sub>4</sub>-N) and sulphates showed a significant (P<0.05) increase from 7500 to 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup> (Table 3).

### Primary productivity, pigment concentration and biotic community

Parameters indicative of productivity viz. NPP, GPP, chlorophyll *a*, phytoplankton and zooplankton population and their species diversity ( $\bar{d}$ ) increased with each increase in the fertilizer dosages up to the second treatment (7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) and with further increase in the dosage of fertilizer a decrease in their values was observed. A detailed presentation of net primary productivity in relation to fertilizer inputs is shown in Fig. 3. Abundance and distribution of different taxa of phyto- and zooplankton showed that Chlorophyceae (5 taxa) and Bacillariophyceae (3 taxa) were the dominant groups, followed by Cladocera (4 taxa), Copepoda (3 taxa) and Rotifera (2 taxa). Further, none of these groups showed a stable community (Data not shown).

Table 3. Mean values (50 and 100 days sampling) of physico-chemical and biological characteristics of ponds stocked with mullet, *Mugil cephalus* (Experiment I) : Effect of four different dosages (5000, 7500, 10000 and 12500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of organic fertilizer (cow-dung)

Parameters	'n'	Cow-dung (kg•ha <sup>-1</sup> •y <sup>-1</sup> )			
		5000	7500	10000	12500
Conductivity dSm <sup>-1</sup>	100	23.89 <sub>A</sub> ±0.95	21.22 <sub>B</sub> ±0.51	22.51 <sub>AB</sub> ±0.92	21.96 <sub>AB</sub> ±0.36
pH	100	8.38 <sub>B</sub> ±0.05	8.36 <sub>B</sub> ±0.05	8.27 <sub>B</sub> ±0.04	8.68 <sub>A</sub> ±0.02
Dissolved oxygen mg•L <sup>-1</sup>	16	7.15 <sub>A</sub> ±0.17	7.90 <sub>A</sub> ±0.12	7.48 <sub>A</sub> ±0.31	7.55 <sub>A</sub> ±0.32
BOD mg•L <sup>-1</sup>	16	5.18 <sub>C</sub> ±0.32	5.78 <sub>BC</sub> ±0.35	6.63 <sub>AB</sub> ±0.35	7.40 <sub>A</sub> ±0.34
Carbonates mg•L <sup>-1</sup>	16	13.75 <sub>A</sub> ±2.01	11.13 <sub>A</sub> ±0.47	13.13 <sub>A</sub> ±2.13	13.50 <sub>A</sub> ±2.22
Bicarbonates mg•L <sup>-1</sup>	16	191.13 <sub>B</sub> ±4.16	209.88 <sub>A</sub> ±5.30	178.50 <sub>B</sub> ±6.32	176.50 <sub>B</sub> ±6.73
Total alkalinity mg•L <sup>-1</sup>	16	204.88 <sub>B</sub> ±2.26	221.00 <sub>A</sub> ±4.39	191.63 <sub>C</sub> ±4.24	190.00 <sub>C</sub> ±4.56
Chlorides mg•L <sup>-1</sup>	16	5566.36 <sub>C</sub> ±62.21	5712.39 <sub>BC</sub> ±74.34	5821.13 <sub>AB</sub> ±57.37	5973.28 <sub>A</sub> ±65.31
Total hardness mg•L <sup>-1</sup>	16	4362.50 <sub>B</sub> ±30.10	4306.25 <sub>B</sub> ±52.81	4625.00 <sub>A</sub> ±34.76	4681.25 <sub>A</sub> ±72.58
Calcium mg•L <sup>-1</sup>	16	565.63 <sub>C</sub> ±10.13	575.43 <sub>C</sub> ±11.32	662.31 <sub>B</sub> ±9.00	701.73 <sub>A</sub> ±7.41
Magnesium mg•L <sup>-1</sup>	16	720.16 <sub>A</sub> ±9.15	700.00 <sub>A</sub> ±13.04	725.00 <sub>A</sub> ±11.31	723.86 <sub>A</sub> ±21.07
Total Kjeldahl-N mg•L <sup>-1</sup>	16	9.57 <sub>B</sub> ±0.60	12.09 <sub>A</sub> ±0.26	8.56 <sub>B</sub> ±0.50	8.70 <sub>B</sub> ±0.51
NH <sub>4</sub> -N mg•L <sup>-1</sup>	16	0.43 <sub>A</sub> ±0.01	0.33 <sub>B</sub> ±0.03	0.42 <sub>A</sub> ±0.03	0.51 <sub>A</sub> ±0.04
NO <sub>2</sub> -N mg•L <sup>-1</sup>	16	0.38 <sub>A</sub> ±0.01	0.43 <sub>A</sub> ±0.05	0.45 <sub>A</sub> ±0.02	0.45 <sub>A</sub> ±0.01
NO <sub>3</sub> -N mg•L <sup>-1</sup>	16	1.37 <sub>B</sub> ±0.04	1.50 <sub>A</sub> ±0.03	1.32 <sub>B</sub> ±0.02	1.36 <sub>B</sub> ±0.03
o-PO <sub>4</sub> mg•L <sup>-1</sup>	16	0.22 <sub>A</sub> ±0.01	0.25 <sub>A</sub> ±0.03	0.20 <sub>AB</sub> ±0.02	0.15 <sub>B</sub> ±0.02
Sulphate mg•L <sup>-1</sup>	16	80.83 <sub>B</sub> ±1.64	78.15 <sub>B</sub> ±0.08	93.26 <sub>A</sub> ±0.95	93.87 <sub>A</sub> ±0.87
Turbidity (NTU)	16	13.63 <sub>B</sub> ±0.12	16.88 <sub>A</sub> ±0.24	12.88 <sub>B</sub> ±0.74	10.38 <sub>C</sub> ±0.26
TDS mg•L <sup>-1</sup>	16	8991.25 <sub>B</sub> ±139.13	9250.00 <sub>AB</sub> ±131.02	9512.50 <sub>A</sub> ±42.20	9251.25 <sub>AB</sub> ±144.83
Phytoplankton numbers•L <sup>-1</sup>	16	6210 <sub>B</sub> ±300	10220 <sub>A</sub> ±630	6500 <sub>B</sub> ±510	5380 <sub>B</sub> ±200
Zooplankton numbers•L <sup>-1</sup>	16	6560 <sub>B</sub> ±45	9250 <sub>A</sub> ±300	5660 <sub>C</sub> ±270	4880 <sub>C</sub> ±190
Phytoplankton $\bar{d}$	16	1.45 <sub>AB</sub> ±0.10	1.94 <sub>B</sub> ±0.49	1.31 <sub>A</sub> ±0.23	1.51 <sub>AB</sub> ±0.03
Zooplankton $\bar{d}$	16	1.77 <sub>AB</sub> ±0.07	2.25 <sub>B</sub> ±0.00	1.52 <sub>A</sub> ±0.01	1.49 <sub>AB</sub> ±0.36
Chlorophyll 'a' µg•L <sup>-1</sup>	16	6.18 <sub>B</sub> ±0.12	7.11 <sub>A</sub> ±0.11	5.43 <sub>C</sub> ±0.17	5.44 <sub>C</sub> ±0.15
NPP mg C•L <sup>-1</sup> •d <sup>-1</sup> *	16	0.56 <sub>B</sub> ±0.07	0.75 <sub>A</sub> ±0.08	0.32 <sub>C</sub> ±0.04	0.34 <sub>C</sub> ±0.04
GPP mg C•L <sup>-1</sup> •d <sup>-1</sup>	16	1.60 <sub>B</sub> ±0.13	2.51 <sub>A</sub> ±0.06	1.45 <sub>B</sub> ±0.13	1.52 <sub>B</sub> ±0.11

All values are mean ± SE of mean of eight replicate and two sampling dates (n=16). Means bearing different letters in the same row differ significantly (P<0.05). Water temperature during the experimental period ranged from 27.6-33.8°C. \*Determined through light and dark bottle method (APHA 1998).

### **Experiment II: Effect of four different dosages (2500, 5000, 7500 and 10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of organic fertilizer (cow-dung) on pond productivity and growth performance of *Chanos chanos***

#### *Fish growth*

Milkfish survival (90.0-94.0%)/mortality was independent of the various fertilizer loading rates/treatments. ANOVA revealed a gradual and significant (P<0.05) increase in growth performance [measured in terms of mean weight (g) and length (cm) gain, biomass and growth day<sup>-1</sup>] from the lowest to the third highest dosage (7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of cow-dung, thereafter, at higher dosage (10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>) a significant (P<0.05) decline in fish growth parameters was observed. Length-weight relationship (LWR) calculated at the end of 100 days had revealed that the exponential value 'n' was 3.30 in ponds fertilized at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>, while slightly low (3.13-3.25) values of 'n' were recorded from other treatments (Table 2). Fertilizer loading at 7500 kg•ha<sup>-1</sup>•y<sup>-1</sup> produced a higher fish production of milkfish (Table 2). A detailed presentation of fish production in relation to fertilizer inputs is shown in Fig. 4.

#### *Physico-chemical characteristics of pond waters*

Water conductivity varied between 20.31±1.00 to 23.10±0.06 dSm<sup>-1</sup>. The DO concentrations were at optimal levels and no significant variations among different treatments

during the entire period of investigation was observed. A detailed presentation of dissolved oxygen concentration in relation to fertilizer inputs is shown in Fig. 5. The BOD<sub>5</sub> increased with each increase in the dosage of fertilizer and significantly ( $P<0.05$ ) higher values were observed in ponds fertilized with the highest dosage of fertilizer (10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>). The pH remained alkaline during the entire period of investigation and varied between 8.60±0.04 to 8.68±0.03. Total alkalinity, turbidity, orthophosphate (o-PO<sub>4</sub>) and nitrate (NO<sub>3</sub>-N) increased significantly ( $P<0.05$ ) with each increase in the fertilizer dosage up to the third treatment (7500 kg•ha<sup>-1</sup>•y<sup>-1</sup>) and a significant ( $P<0.05$ ) decline in these parameters took place at the highest fertilizer dosage. Chlorides, calcium, magnesium, total hardness and total dissolved solids (TDS) showed no definite trend and remained high in all the treatments (Table 4).

Concentrations of sulphates were low at the lowest fertilizer dosage and high at the highest fertilizer dosage, while, no variation at the intermediate dosages (5000-7500 kg) was observed. Total Kjeldahl nitrogen and NH<sub>4</sub>-N, in general, increased significantly ( $P<0.05$ ) with each increase in the dosage of fertilizer. No significant trend in NO<sub>2</sub>-N values was observed, however, the values increased with respect to time (Table 4).

Table 4. Mean values (50 and 100 days sampling) of physico-chemical and biological characteristics of pond waters stocked with milkfish, *Chanos chanos* (Experiment II): Effect of four different dosages (2500, 5000, 7500 and 10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>) of organic fertilizer (cow-dung)

Parameters	'n'	Cow-dung (kg•ha <sup>-1</sup> •y <sup>-1</sup> )			
		2500	5000	7500	10000
Conductivity dSm <sup>-1</sup>	100	20.73 <sub>B</sub> ±0.13	20.31 <sub>B</sub> ±1.00	22.89 <sub>A</sub> ±0.18	23.10 <sub>A</sub> ±0.06
pH	100	8.63 <sub>A</sub> ±0.05	8.68 <sub>A</sub> ±0.03	8.60 <sub>A</sub> ±0.04	8.64 <sub>A</sub> ±0.01
Dissolved oxygen mg•L <sup>-1</sup>	16	4.95 <sub>B</sub> ±0.11	5.05 <sub>B</sub> ±0.13	5.50 <sub>A</sub> ±0.04	4.95 <sub>B</sub> ±0.11
BOD mg•L <sup>-1</sup>	16	3.28 <sub>C</sub> ±0.12	3.52 <sub>BC</sub> ±0.10	3.75 <sub>B</sub> ±0.14	4.43 <sub>A</sub> ±0.12
Carbonates mg•L <sup>-1</sup>	16	11.63 <sub>A</sub> ±1.25	11.50 <sub>A</sub> ±1.28	9.50 <sub>A</sub> ±1.02	11.13 <sub>A</sub> ±0.55
Bicarbonates mg•L <sup>-1</sup>	16	180.00 <sub>C</sub> ±6.48	197.00 <sub>AB</sub> ±4.92	208.38 <sub>A</sub> ±5.33	185.25 <sub>BC</sub> ±2.97
Total alkalinity mg•L <sup>-1</sup>	16	191.63 <sub>B</sub> ±5.30	208.50 <sub>A</sub> ±3.70	217.88 <sub>A</sub> ±4.38	195.75 <sub>B</sub> ±3.14
Chlorides mg•L <sup>-1</sup>	16	7455.00 <sub>A</sub> ±36.86	7358.67 <sub>A</sub> ±84.82	7293.48 <sub>A</sub> ±61.96	7444.84 <sub>A</sub> ±47.13
Total hardness mg•L <sup>-1</sup>	16	3862.50 <sub>A</sub> ±297.47	3475.00 <sub>AB</sub> ±38.19	3393.75 <sub>B</sub> ±34.72	3637.50 <sub>AB</sub> ±42.70
Calcium mg•L <sup>-1</sup>	16	501.96 <sub>B</sub> ±13.01	509.87 <sub>B</sub> ±10.77	494.11 <sub>B</sub> ±11.19	546.67 <sub>A</sub> ±8.58
Magnesium mg•L <sup>-1</sup>	16	548.16 <sub>A</sub> ±10.59	542.01 <sub>A</sub> ±11.58	527.08 <sub>A</sub> ±13.10	548.08 <sub>A</sub> ±11.43
Total Kjeldahl-N mg•L <sup>-1</sup>	16	6.17 <sub>B</sub> ±0.21	7.50 <sub>B</sub> ±0.27	10.06 <sub>A</sub> ±0.30	11.52 <sub>A</sub> ±1.04
NH <sub>4</sub> -N mg•L <sup>-1</sup>	16	1.00 <sub>B</sub> ±0.04	0.87 <sub>C</sub> ±0.02	0.77 <sub>D</sub> ±0.03	1.27 <sub>A</sub> ±0.01
NO <sub>2</sub> -N mg•L <sup>-1</sup>	16	0.69 <sub>B</sub> ±0.04	0.67 <sub>B</sub> ±0.01	0.58 <sub>C</sub> ±0.01	0.76 <sub>A</sub> ±0.03
NO <sub>3</sub> -N mg•L <sup>-1</sup>	16	1.61 <sub>C</sub> ±0.02	1.78 <sub>B</sub> ±0.02	1.90 <sub>A</sub> ±0.03	1.50 <sub>D</sub> ±0.02
o-PO <sub>4</sub> mg•L <sup>-1</sup>	16	0.11 <sub>C</sub> ±0.01	0.16 <sub>B</sub> ±0.00	0.22 <sub>A</sub> ±0.01	0.10 <sub>C</sub> ±0.01
Sulphate mg•L <sup>-1</sup>	16	84.04 <sub>C</sub> ±2.80	105.07 <sub>B</sub> ±5.84	105.41 <sub>B</sub> ±5.21	130.56 <sub>A</sub> ±6.65
Turbidity (NTU)	16	17.88 <sub>C</sub> ±2.44	27.00 <sub>B</sub> ±2.78	35.38 <sub>A</sub> ±1.72	18.25 <sub>C</sub> ±0.52
TDS mg•L <sup>-1</sup>	16	11660.00 <sub>B</sub> ±398.26	12126.25 <sub>B</sub> ±518.71	13718.75 <sub>A</sub> ±100.25	13923.75 <sub>A</sub> ±67.48
Phytoplankton numbers•L <sup>-1</sup>	16	8690 <sub>C</sub> ±300	10400 <sub>B</sub> ±200	12820 <sub>A</sub> ±100	9750 <sub>BC</sub> ±220
Zooplankton numbers•L <sup>-1</sup>	16	1280 <sub>C</sub> ±30	1970 <sub>C</sub> ±70	3030 <sub>B</sub> ±80	4060 <sub>A</sub> ±70
Phytoplankton $\bar{d}$	16	2.28 <sub>A</sub> ±0.04	2.20 <sub>A</sub> ±0.06	2.37 <sub>A</sub> ±0.12	2.41 <sub>A</sub> ±0.11
Zooplankton $\bar{d}$	16	0.00 <sub>D</sub> ±0.00	0.25 <sub>C</sub> ±0.06	0.97 <sub>B</sub> ±0.16	1.20 <sub>A</sub> ±0.14
Chlorophyll 'a' $\mu\text{g}\cdot\text{L}^{-1}$	16	3.69 <sub>C</sub> ±0.13	4.97 <sub>B</sub> ±0.05	6.42 <sub>A</sub> ±0.20	3.97 <sub>C</sub> ±0.08
NPP mg C•L <sup>-1</sup> •d <sup>-1</sup> *	16	0.79 <sub>B</sub> ±0.06	1.10 <sub>A</sub> ±0.07	1.29 <sub>A</sub> ±0.10	0.51 <sub>C</sub> ±0.07
GPP mg C•L <sup>-1</sup> •d <sup>-1</sup>	16	1.88 <sub>C</sub> ±0.09	2.45 <sub>B</sub> ±0.12	2.40 <sub>A</sub> ±0.13	1.80 <sub>C</sub> ±0.09

All values are mean ± SE of mean of eight replicate and two sampling dates (n=16). Means bearing different letters in the same row differ significantly ( $P<0.05$ ). Water temperature during the experimental period ranged from 27.7-32.8°C. \* Determined through light and dark bottle method (APHA 1998).

### Primary productivity, pigment concentration and biotic community

Initially, NPP, GPP, chlorophyll *a* and phytoplankton population increased significantly ( $P < 0.05$ ) with each increase in the fertilizer dosage up to the third treatment ( $7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ); thereafter, with further increase in the dosage of fertilizer, a significant ( $P < 0.05$ ) decline in these parameters was observed. A detailed presentation of net primary productivity in relation to fertilizer inputs is shown in Fig. 6. Zooplankton population showed a gradual and significant ( $P < 0.05$ ) increase with each increase in the fertilizer dosage throughout the experiment. No significant variation in phytoplankton species diversity was observed, while zooplankton species diversity increased with each increase in the fertilizer dosage. Abundance and distribution of different taxa of phyto- and zooplankton showed that Chlorophyceae (5 taxa) and Bacillariophyceae (2 taxa) were the dominant phytoplankton groups. Among the zooplanktons, Copepoda (3 taxa) was the dominant group, followed by Rotifera (1 taxa). *Navicula* and *Closterium* among the phytoplanktons and Cyclops among zooplanktons showed a stable community (Data not shown).

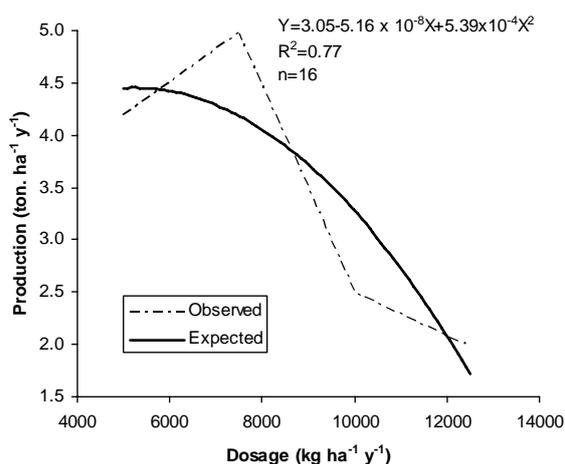


Fig. 1. Relationship between net fish production and fertilizer loading rates in brackish water ponds stocked with mullet. Where,  $Y$ =net production ( $\text{ton} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ),  $X$ =fertilizer loading rates ( $\text{kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and  $n$ =mean of total number of observations (16).

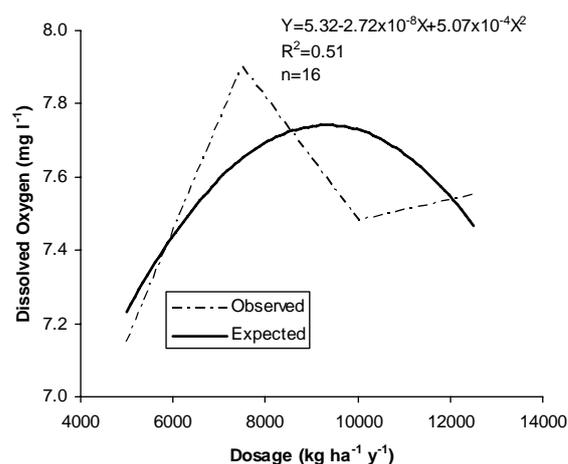


Fig. 2. Relationship between dissolved oxygen and fertilizer loading rate in brackishwater ponds stocked with mullet. Where,  $Y$ =dissolved oxygen production ( $\text{mg} \cdot \text{L}^{-1}$ ),  $X$ =fertilizer loading rates ( $\text{kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and  $n$ =mean of total number of observations (16).

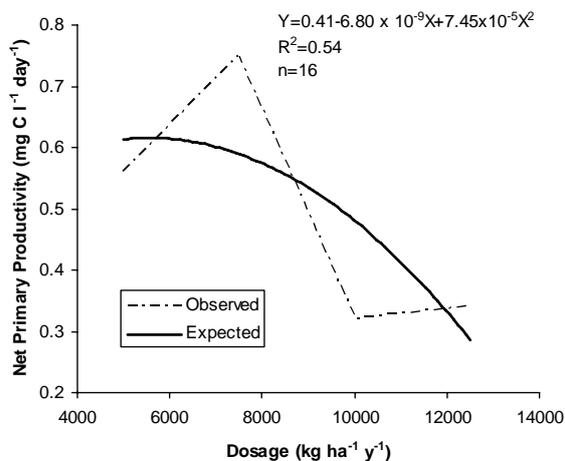


Fig. 3. Relationship between net primary productivity and fertilizer loading rates in brackish-water pond stocked with mullet. Where, Y=net primary productivity ( $\text{mg C}\cdot\text{L}^{-1}\cdot\text{day}^{-1}$ ), X=fertilizer loading rates ( $\text{kg}\cdot\text{ha}^{-1}\cdot\text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and n=mean of total number of observations (16).

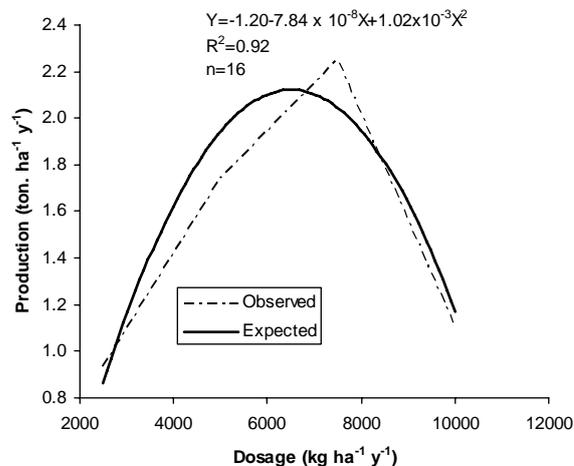


Fig. 4. Relationship between net fish production and fertilizer loading rates in brackish water ponds stocked with milkfish. Where, Y=net production ( $\text{ton}\cdot\text{ha}^{-1}\cdot\text{y}^{-1}$ ), X=fertilizer loading rates ( $\text{kg}\cdot\text{ha}^{-1}\cdot\text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and n=mean of total number of observations (16).

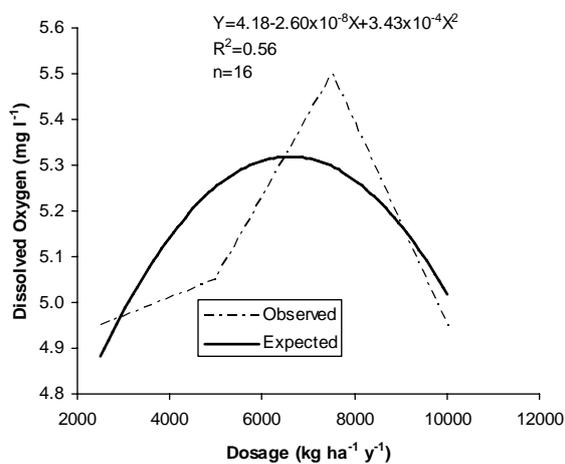


Fig. 5. Relationship between dissolved oxygen and fertilizer loading rate in brackishwater ponds stocked with milkfish. Where, y=dissolved oxygen production ( $\text{mg}\cdot\text{L}^{-1}$ ), X=fertilizer loading rates ( $\text{kg}\cdot\text{ha}^{-1}\cdot\text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and n=mean of total number of observations (16).

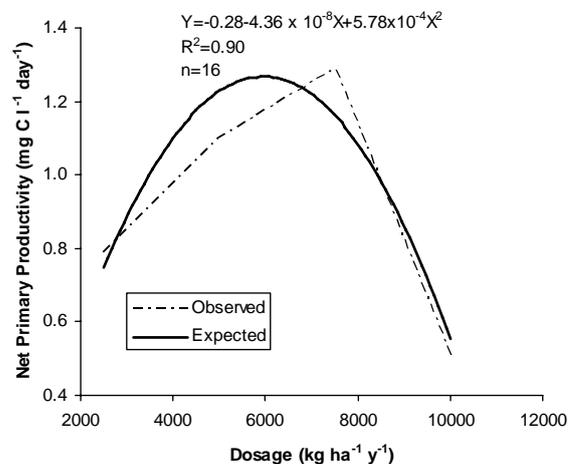


Fig. 6. Relationship between net primary productivity and fertilizer loading rates in brackish-water pond stocked with milkfish. Where, Y=net primary productivity ( $\text{mg C}\cdot\text{L}^{-1}\cdot\text{day}^{-1}$ ), X=fertilizer loading rates ( $\text{kg}\cdot\text{ha}^{-1}\cdot\text{y}^{-1}$ ),  $R^2$ =Coefficient of determination and n=mean of total number of observations (16).

## Discussion

Highest mean fish weight and growth  $\cdot d^{-1}$  in both experiments were observed in ponds fertilized at  $7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ . High growth performance of fish at  $7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$  also coincided with high plankton population, NPP and chlorophyll *a*. Statistically fish weight gain also showed a significant ( $P < 0.05$ ) and positive correlation with phytoplankton ( $r = 0.41$ ,  $r = 0.69$ ), zooplankton ( $r = 0.60$ ,  $r = 0.77$ ), NPP ( $r = 0.56$ ,  $r = 0.49$ ), GPP ( $r = 0.44$ ,  $r = 0.59$ ) and chlorophyll *a* ( $r = 0.60$ ,  $r = 0.77$ ) in both the experiments, clearly revealing that fish growth is significantly ( $P < 0.05$ ) correlated with the trophic status of ponds. Knud-Hansen and Batterson (1994), Garg and Bhatnagar (1996, 1999, 2000) and Jana et al. (2004) have also reported that fish growth is positively correlated with the trophic status of the ponds.

The study of length-weight relationship is useful in evaluating experimental pond conditions, such as varying dosages of fertilization. According to Bishara (1978) increase in fish weight is related to increase in length as an indicator of growth. No significant differences for the slope of regression of fish weight or length were observed in both the experiments. However, the results confirmed that the best growth in terms of weight and length increase in *M. cephalus* ( $n = 2.94$ ) and *C. chanos* ( $n = 3.30$ ) were observed in ponds fertilized at  $7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ . Present results on length-weight relationship on *M. cephalus* appears to be in close proximity to those of Chow (1958) on *M. cephalus* ( $n = 3.025$ ). For *C. chanos* the value of 'n' was more than 3.00 (3.13-3.30), revealing that the fish growth followed the cube-law and inland saline groundwater appears to be conducive for the ideal growth of milkfish.

The DO concentrations remained within the range required for the optimal growth of *M. cephalus* and *C. chanos*. The pH was alkaline and alkalinity was high indicating that pond waters were well buffered. An increase in BOD<sub>5</sub> with each increase in the dosage of fertilizer in both the experiments indicated an accumulation of biodegradable material at higher dosages ( $> 7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ). An inverse relationship of BOD<sub>5</sub> with productivity indices viz. phytoplankton population ( $r = -0.58$ ,  $P < 0.001$ ), zooplankton diversity ( $r = -0.30$ ,  $P < 0.05$ ), chlorophyll *a* ( $r = -0.66$ ,  $P < 0.001$ ), NPP ( $r = -0.37$ ,  $P < 0.05$ ) and fish weight gain ( $r = -0.43$ ,  $P < 0.05$ ) further confirms the results. Nutrients (viz.  $\text{o-PO}_4$ ,  $\text{NO}_3\text{-N}$  and total Kjeldahl nitrogen) showed a significant ( $P < 0.05$ ) and positive correlation with phytoplankton ( $r = 0.65$ ,  $0.59$ ,  $0.32$ ,  $P < 0.001$ ) and zooplankton ( $r = 0.42$ ,  $0.44$ ,  $0.64$ ,  $P < 0.01$ ) populations in both the experiments. According to Boyd (1979), phytoplankton use nutrients, creating chemical gradients which in turn cause rapid flux from the soil, thus increasing nutrient concentration in the water.

Decrease in the concentration of  $\text{NO}_3\text{-N}$  and accumulation of  $\text{NH}_4\text{-N}$  at higher dosages ( $> 7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ) in both the experiments may probably be due to the process of denitrification or ammonification. Increase in  $\text{NH}_4\text{-N}$  during this period was found to be negatively correlated with the phytoplankton ( $r = -0.72$ ,  $-0.42$ ) population indicating its use by the autotrophs. Negative correlation of  $\text{NH}_4\text{-N}$  with weight gain ( $r = -0.39$ ,  $-0.53$ ) further supports the low growth of fishes at higher fertilizer dosages.

The downward slope of the curves at high fertilizer levels ( $> 7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ) in experiment indicated that excessive fertilizer depressed the production of mullet, whereas the downward slope of the curves at both high and low levels ( $< 7500$  or  $> 7500 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1}$ ) in

experiment II indicated that less or excessive fertilizer depressed the production of milkfish. In both the experiments, with increase in fertilizer loads average fish production increased while, at very high fertilizer loads average fish production begin to decrease, which is in broad agreement with the finding of Hopkins and Cruz (1982). These studies, thus, have established that a dose of 7500 ha<sup>-1</sup> y<sup>-1</sup> along with supplementary feeding appears to be optimum for obtaining high fish production in ponds with water salinity fluctuating between 13.0 to 21.0 ppt and water temperature between 25.1 to 32.8°C. Hopkins and Cruz (1982) have reported that optimum levels of manure loading are 101 - 110 kg•ha<sup>-1</sup>•d<sup>-1</sup> for pig, 82 kg•ha<sup>-1</sup>•d<sup>-1</sup> for duck and 100 kg•ha<sup>-1</sup>•d<sup>-1</sup> for chicken. Garg and Bhatnagar (1996) obtained high plankton population, species diversity and high fish production in brackish water ponds fertilized with cow-dung (10000 kg•ha<sup>-1</sup>•y<sup>-1</sup>) and SSP (1500 kg•ha<sup>-1</sup>•y<sup>-1</sup>). In freshwater fish ponds (Garg and Bhatnagar 1999) average plankton production and fish biomass were high when cow-dung was used at 15000 kg•ha<sup>-1</sup>•y<sup>-1</sup> than when used at higher rates (20000 or 24000 kg•ha<sup>-1</sup>•y<sup>-1</sup>).

As the decomposition processes of organic matter depends on microbial activity and prevailing environmental conditions (Seitzinger 1988), caution therefore, must be taken when recommending the dosage of organic fertilizer, especially in saline water ponds located in semi-arid areas as fertilizer optima is liable to change with quantity and frequency of application of fertilizer and also with change in season, temperature and salinity of the pond water.

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