Evaluation of a Bio- organic Catalyst in Channel Catfish Ponds

ABSSTRACT. A Bio-organic catalyst ***(Bio-Tech A , BTA)*** was tested in pond at Auburn, Alabama, for its effect on water quality, soil organic carbon, and channel catfish production. Ponds treated with ***BTA*** had higher concentrations of dissolved oxygen (DO) than control ponds during summer months even though all ponds were aerated mechanically. Data on water quality and soil organic carbon suggested that the primary influence of ***BTA*** was to inhibit phytoplankton productivity which in turn lessened the nighttime oxygen demand. Although fish production did not increase as the result of greater DO availability, fish harvest and survival were higher in the treated ponds (P=0.1). At the maximum daily feeding rate of 75 kg/ha, water quality was not severely impaired in any of the ponds. The bio- organic catalyst product might have greater benefits in ponds with higher stocking and feeding rates. Information on the mechanism of action of bio-catalyst additions in pond ecosystems would be useful in determining their potential benefits to pond aquaculture.

INTRODUCTION

Amendments high in enzymatic activity such as bio-organic catalysts have been developed to accelerate natural microbial process in soils, water, or other media. These bio-organic catalysts products have been used in agriculture, wastewater treatment, clean up of oil spills, and remediation of sites contaminated with hazardous wastes. Enzymes are catalysts that accelerate biochemical reactions, and microorganisms excrete extracellular enzymes that are important in the initial stages of decomposition. Extracellular enzymes break large molecules into smaller fragments that can be absorbed by microbial cell (Anderson 1987), and they can cause transformations of various compounds to accelerate microbial processes (Dick and Tabatabai 1992). Enough success has been obtained with some of these amendments to encourage companies to produce and market them for a variety of applications (Glass 1992).

Some of these companies recommend the use of their products in pond aquaculture. According to information from one manufacturer (Biological Technology International Co., Inc.), their product can improve water quality though increasing the solubility of hydrophobic material, improving gas diffusion by increasing dissolved oxygen levels, and accelerating breakdown of organic matter. Therefore, the present study was initiated to evaluate the use of this bio-organic catalyst (BOC), which contains derived catalysts, surface modifying, synthetic compounds, and other proprietary ingredients, in channel catfish (Ictalurus punctuatus) pounds.

MATERIALS AND METHODS

Ponds

Nine, 400-m^2 levee ponds on the Auburn University Fisheries Research unit (FRU) were used in this study. Ponds were square with earthen bottoms that gradually sloped from depths of about 20 - 40 cm at the vertical edges supported by wooden or concrete Walls to 130 -150 cm at the drains. Pond volumes ranges from about 450 to 480 m^3. These ponds have been used annually for experiments on water quality and pond fertilization for 25 years.

Soils used for construction of these ponds were typic, Kandiudults (clayey, kaolinitic, and thermic) . They are acidic, reddish brown soil of low cation exchange capacity with base saturation less than 35%, and in their native state, these soil have low concentrations of phosphorus and organic matter (McNutt 1981). Auburn, Alabama, is located at 32.5 N latitude. The normal annual temperature is 17.2℃(normal minimum =11.1℃;normal maximum =23.2℃);normal annual rainfall is 1,434 mm.

Ponds are supplied by water from a reservoir filled by runoff from woodland (Boyd 1990). This water has total alkalinity and total hardness values below 20 mg/L as $CaCO_3$ and soluble reactive phosphorus concentration less than 0.005 mg/L. These pond and treated most year with 500 to 1,000 kg/ha of agricultural limestone to maintain adequate pH , alkalinity ,and hardness.

Management

The ponds were stocked at a rate equal to 15,000 channel catfish fingerlings per hectare in April 1996. The average fish at stocking was 10.7 kg per thousand fingerlings. A 32% crude protein, pelleted feed was offered 7 day per week at 3% of body weight per day. Feeding rates were increased weekly according to an assumed feed conversion (weight of feed applied/weight gained by the fish) of 1.6. Pond were inspected daily, and any dead fish were removed. Daily feeding rate did not exceed 75 kg/ha. When this rate was reached, it was continued unit fish harvest. All ponds had a 0.25-kW vertical pump aerator (Air-O-Lator Corporation, Kansas City, Missouri) connected to a timer. Aerators were operated from dusk to dawn from the end of June unit harvest in October. Water levels in ponds were maintained 10-12 cm below the tops of standing drain pipes to prevent to prevent overflow after rains. Water was added from a pipeline when necessary to replace evaporation and seepage losses.

Ponds were drained and drained and fish were harvested and weighed on 15 October.

Bio-organic Catalyst treatment

A bio-organic catalyst product marketed under the trade name ***BTA*** was obtained from Biological Technology International, Co., Inc.. The experiment was designed so that the treatment applied to any given pond was not revealed until after the ponds were harvested. The researchers assigned each pond a latter (A-I) by ballot. The manufacturer bottled 960-mL aliquots of BTA to provide 2 mg/L in ponds (based on 480m^3 volume) and equal volumes of a placebo consisting of herbal tea looked like ***BTA***. Bottles were identified by letters A to I to correspond to letters assigned to ponds and the application date and shipped to Auburn University. This provided three treatments of three replications each as follows: high ***BTA*** treatment – 2 mg/L ***BTA*** weekly (8 mg/L per month); low ***BTA*** treatment – 2mg/L ***BTA*** one week followed by application of placebo for three weeks (2mg/L per month); control – placebo at weekly intervals. The manufacturer sent the code identifying bottle contents to Dr. Craig S. Tucker, Delta Research and Extension Center, Stoneville, Mississippi, and he revealed it to the researchers at the end of study. These treatments were applied between 13 May 1996 and 11 October 1996. The placebo and ***BTA*** were splashed over surfaces, and the aerator was then operated for 1 hour to mix the materials with the pond water.

Measurements

Dissolved oxygen and temperature were measured one or more time per day with a polarographic oxygen meter (Yellow Spring Instrument Company, Yellow Springs, Ohio) to verify that fish were not subjected to low oxygen stress. Water samples were collected between 0630 and 0700hr at 2-week intervals with a 90-cm water column sampler (Boyd and Tucker 1992). Samples were taken from three places in each pond and combined to give a composite fir analysis. Analyses were conducted for pH (glass electrode), total alkalinity (acidimetry), total hardness (EDTA titration), specific conductance (conductivity meter), chemical oxygen demand (potassium dichromate-sulfuric acid oxidation), biochemical oxygen demand (standard 5-day test), soluble reactive phosphorus (ascorbic acid method), total ammonia nitrogen (phenate method),chlorophyll a (membrane filtration, acetone extraction, and spectroscopy) and bacteria abundance (standard plate count). Standard protocol was followed for these analyses (Eaton et al.1989). Nitrate was followed for these analyses (Eaton et al. 1989). Nitrate was determined by the NAS reagent method (van Rijn 1993). Before stocking and 1 week before harvest, five soil samples (upper 5 cm layers) were collected from each pond with a 5-cm diameter core sampler. On each samples, the sampling from a given pond were combined to make a composite sample. Soil sample were dried at 60℃ and pulverized in a hammer mill. They were analyzed for carbon by an induction furnace carbon analyzer (Leco Corporation, St. Joseph, Michigan). At the harvest time, the following fish production data were corporation, St. Joseph, Michigan). At the harvest time, the following fish production data were collected: number and total weight of fish, percentage survival, average weight of individual fish, and food conversion ratio.

Data Analysis

Means were tested for statistical differences with Duncan's multiple range test. A large degree of variation is typically encountered in water quality and fish production variables in experiments conducted in earthen, aquaculture ponds (Boyd et al. 1994). Because of this large variation, the amount of replication necessary to declare significant differences at the 0.05 probability level in pond aquaculture experiments is often prohibitive. Therefor, in this study, we declared means to be different if the probability was 0.1 or less.

RESULTS

Total alkalinity and total hardness concentrations in pond were between 25 and 30 mg/L in May, and they increased to 35 to 40 mg/L in October. Specific conductance of pond waters was within the range 75 to 105 μ mhos/cm, and morning pH values were between 7.7 and 8.2. Morning water temperature ponds increased from 23℃ on 16 May to 29℃ on 8 August and declined to 18℃ by 14 October. No influence of ***BTA*** treatment could be detected on pH, specific conductance, water temperature, alkalinity, and hardness.

Because ponds were aerated nightly, dissolved oxygen (DO) concentrations were seldom less than 3 mg/L. From July to September, when water temperature and feeding rates were high, DO concentration often were 1 or 2 mg/L greater in ponds treated with ***BTA*** than in control Ponds (figure 1), but the high ***BTA*** and treatment were similar in DO concentration. Concentrations of soluble reactive phosphorus (SRP) were quit similar in all ponds during most of the study, but there were large increases in SRP concentration in the high ***BTA*** treatment on two dates and in the low ***BTA*** treatment on one data (figure 1). On a few dates, nitrate-nitrogen ($NO_3 - N$) concentration also tended to be higher in ponds treated with ***BTA*** than in control ponds. During the summer, there was a trend of greater of greater concentrations of total ammonia nitrogen (TAN) in ***BTA*** ponds than in ponds than in ponds (Figure 1). Also, biochemical oxygen demand (BOD) and chemical oxygen demand (COD) concentrations often were slightly lower during the summer in ponds treated with ***BTA*** as compared to controls (Figure 1). Chlorophyll a concentration (an index of phytoplankton abundance) tented to be higher during August and September in control ponds of the ***BTA*** treatments. From June through August, there was a trend of greater bacteria abundance in control ponds than in ***BTA*** ponds (Figure 1). Even though trends of difference in water quality between control and BTA ponds were detectable at certain times, grand means for treatments were similar (Table 1). The only significant difference was for SRP concentration, and the difference resulted from high concentrations of SRP in ***BTA*** ponds on a few dates.

Pond soils on the FRU do not naturally contain free calcium carbonate (McNutt 1981), and only small amounts of residual calcium carbonate could have resulted from agricultural limestone applications. Therefor, the total carbon concentrations in pond soil were considered to result primarily from organic carbon. In May, averages and standard deviations for percentages of soil carbon were as follows: control, 0.81±0.15; low ***BTA***, 1.00±0.30;high ***BTA***, 0.70±0.18. Soil carbon increased in all ponds during the study, and percentages found in October were: control, 1.26±0.47; low ***BTA***, 1.40±0.91;high BTA, 1.21±0.59. These data suggest that ***BTA*** treatment did influence the rate of soil organic carbon in pond bottoms.

Fish production data are summarized in Table 2. A portion of the fish in all ponds were infected by proliferative gill disease and enteric septicemia of catfish (Edwardsiela ictaluri), and survival ranged from 56.1% in controls to 75.7% in the high ***BTA*** pond. Nevertheless, similar amounts of feed were applied in all ponds, and fish tented to be larger at harvest in ponds that had the lowest survival. Feed conversion ratio and net fish production did not net fish production did not differ among treatments ($P<0.1$).

DISCUSSION

Even though ponds were aerated during the night, there was a trend of higher DO concentrations in the ponds treated with ***BTA*** as compared to control ponds. Differences in DO concentration were most obvious in August and September. This difference resulted because higher concentrations of COD and BOD and more phytoplankton in control ponds than in ***BTA*** ponds caused a greater nighttime demand for oxygen in control ponds.

One explanation for the lower oxygen demand in ***BTA*** ponds is that additions of bio-organic catalysts accelerated the decomposition of organic matter to low BOD and COD. This explanation is consistent with the observation that SRP, TAN, and NO_3-N concentrations were greater in ***BTA*** ponds, because greater decomposition wound have caused more nutrient recycling. However, greater nutrient recycling should have stimulated phytoplankton productivity in the ***BTA*** ponds, but this effect was not observed. Also, similar rates of organic carbon accumulation in pond soils of the three treatments is not consistent with enhanced decomposition of organic matter in ***BTA*** ponds.

A more feasible explanation for the high concentrations of chlorophyll a, BOD, COD at times in the control ponds is that ***BTA*** have reduced the phytoplankton growth rate. Boyd (1973) and Boyd et al. (1978) demonstrated that BOD and COD concentrations in pond waters had a strong positive correlation with chlorophyll a concentrations (phytoplankton abundance). An increase in chlorophyll a concentration should cause an increase in BOD and COD concentration. Nutrient inputs to all treatments in feed were similar, and the tendency of greater nutrient concentration in ***BTA*** ponds relative to controls could have resulted from less uptake of nutrients by smaller phytoplankton biomass in ***BTA*** ponds instead of greater rates of nutrient recycling in ***BTA*** ponds. Boyd and Musing (1981) showed that the rate of decline in spikes of SRP made to pond waters increased with increasing phytoplankton abundance. Tucker et al. (1984) demonstrated that TAN concentrations in fish pond waters decreased as phytoplankton abundance increased. Thus, lower nutrient concentrations and higher BOD and COD values in control ponds are consistent with explanation that ***BTA*** have reduced the phytoplankton abundance. Further support of the effects of ***BTA*** on phytoplankton growth rates comes from the claim by the manufacturer that ***BTA*** can be used to reduce the abundance of nuisance algal growth in water. Because of the proprietary name of ***BTA***, its composition was not divulged by Biological Technology

International Co., Inc. Without this information, one cannot speculate on the mechanism by which ***BTA*** might have influenced phytoplankton growth.

Differences on fish harvest and survival were observed (Table 2). These results could be related to the actions of the ***BTA***, however the high variation between the pond due to the few replicates used and the disease that has occurred requires further investigations.

The improvement in DO concentration in ***BTA*** ponds did not cause greater fish production. This is not surprising because the stocking rate was conservative and the feeding rate did not exceed 75 kg/ha per day. When mechanical aeration is used, dangerously low DO concentrations, excessive TAN concentrations, and other water quality impairment seldom occurs unless feeding rates exceed 100 to 120 kg/ha per day (Boyd 1990; Tucker and Boyd 1995). The likelihood of water quality improvement and enhanced fish production through the use of bio-organic catalysts supplementation would be greater in ponds where stocking and feeding rates are high enough to cause severe water quality deterioration than observed in the present study. Although there were not market differences in water quality or fish production data between the low and high ***BTA*** treatments, it seems better to use weekly treatments. The bio-catalyst molecules probably slowly degrade to non-functional from, and more frequent application would provide amore constant concentration of bio-catalysts.

This study shows that bio-organic catalysts supplementation can effect improvement in water quality even in aquaculture ponds stocked and fed at conservative rates. Further studies are needed to show if larger benefit can be achieved in ponds where stocking and feeding rates are higher and water quality conditions are more critical. Information on the mechanisms of action of extracellular enzyme additions in ponds would be useful in assessing the potential benefits of these products and defining the conditions under which they can be effective.

REFERENCE

Anderson, J.M.1987. Production and decomposition in aquatic ecosystem and implications for aquaculture. pages 123-147 in D.J.W. Moriarty and R. S. V. Pullin, eds. Detritus and Microbial Ecology in Aquaculture. ICLARM Conference Proceedings 14,International Center for Living Aquatic Resources Management, Manila, Philippines.

Boyd, C. E. 1973. The chemical oxygen demand of waters and biological materials from ponds. Transactions of the American Fisheries of American Fisheries Society 102:606-611.

Boyd, C. E. 1990. Water Quality in Ponds for Aquaculture. Alabama Agricultural Experiment Station, Auburn University, Alabama.

Boyd,C.E.,and Y. Musig. 1980. Orthophosphate uptake by phytoplankton and sediment. Aquaculture 22:165-173.

Boyd, C. E., and C. S. Tucker. 1992. Water Quality and Pond Soil Analyses for Aquaculture. Alabama Agricultural Experiment Station, Auburn University, Alabama.

Boyd, C. E., and C. S. Tucker 1995. Sustainability of channel catfish farming. World Aquaculture 26:45-53.

Boyd, C. E., R. P. Romaire, and E.Johnston.1978. Predicting early morning dissolved oxygen concentrations in channel catfish ponds. Transactions of the American Fisheries Society 107:484-492.

Boyd, C. E., E. Hernandez, J. C. Williams, and R. P. Romaire. 1994. Effects of sampling technique on precision estimates for water quality variables in fish culture ponds. Journal of Applied Aquaculture 4:1-18.

Dick, W. A., and M. A. Tabatabai. 1992. Significance and potential uses of soil enzymes. Pages 95-127 in F.V. Meting, ed.. Significance and potential uses of soil enzymes. Pages 95-127 in F.V. Meting, ed.. Soil Microbial Ecology. Marcel Dekker, Inc., New York, New York.

Eaton, A. D., L. S. Clesceri, and A. E. Greenburg. 1995. Standard Methods for the Examination of water and Wastewater, 19 ed. American Public Health Association, Washington, D.C.

Glass,D.J.1992. Commercialization of soil microbial technologies. Pages 595-618 in F. B. Meeting, ed. Soil Microbial Ecology. Marcel Dekker, Inc., New York, New York.

Mcnutt, R. B.1981. Soil Survey of Lee County, Alabama. National Cooperative Soil Survey, Soil Conservation Service, United States Department of Agriculture, Washington, D.C.

Tucker, C. S. W. Lloyd, and R. L. Busch.1984. Relationships between phytoplankton periodicity and the concentrations of total and un-ionized ammonia in channel catfish ponds. Hydrobiologia 111:75-79.

Van Rijn, J. 1993.Methods to Evaluate Water Quality in Aquaculture. Faculty of Agriculture, The Hebrew University of Jerusalem, Rehovot, Israel.

TABLE 1. Grand means of water quality data collected from channel catfish ponds with extracellular enzymes at 4-week intervals (low ***BTA***), weekly intervals (high ***BTA***),and controls. There were three replications of each treatment.

Variables	Treatment		
	Low ***BTA***	High ***BTA***	Control
Dissolved oxygen (mg/L)	5.25a	5.59a	5.26a
Soluble reactive phosphorus (mg/L)	0.039a	0.038a	0.020b
Total ammonia nitrogen (mg/L)	0.45a	0.35a	0.28a
Nitrate nitrogen (mg/L)	0.18a	0.13a	0.09a
Chemical oxygen demand (mg/L)	23.5a	19.0a	25.1a
Biochemical oxygen demand (mg/L)	7.8a	7.8a	8.12a
Chlorophyll a (μ g/L)	64.5a	54.5a	62.2a
Bacterial abundance (cells/mL x 10^2)	10.5a	11.7a	13.0a

[a,b]Means indicated by the same letter did not differ significantly at P=0.1. Horizontal comparisons only.

TABLE 2. Average channel catfish production data for ponds treated with extracellular enzymes at 4-week intervals (low ***BTA***), weekly intervals (high ***BTA***),and controls. There were three replications of each treatment.

Variables	Treatment		
	Low ***BTA***	High ***BTA***	Control
Stocking rate(number/ha)	15,000a	15,000a	15,000a
Feed input(kg/ha)	6,128a	6,298a	6,008a
Fish harvested (number/ha)	10,225a	11,350a	8,425b
Survival(%)	68.2ab	75.7a	56.1b
Average harvest weight weight per fish	342a	328a	400a
Net production(kg/ha)	3,502a	3,728a	3,301a
Feed conversion	1.75a	1.77a	1.82a

[a,b]Means indicated by the same letter did not differ significantly at P=0.1. Horizontal comparisons only.

Water Quality Parameters in CatfishPonds (1996)

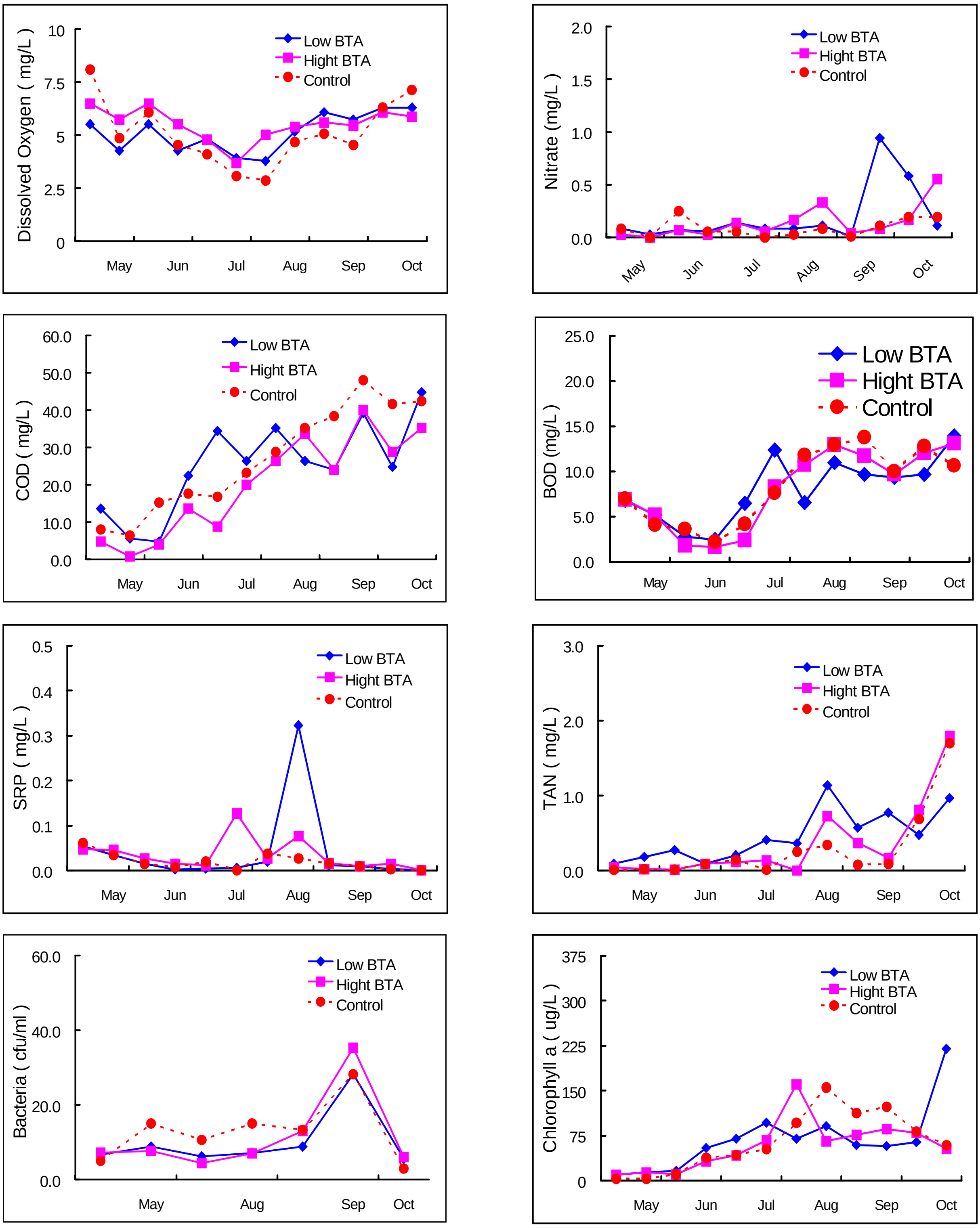

Fig1.Average water quality parameters that were measure biweekly in earthen catfish ponds in the fisheries research unit, Auburn Alabama between May to October 1996.