EFFECT OF PHYTOPLANKTON INOCULATION WITH SOME CHEMICAL FERTILIZERS ON WATER QUALITY AND GROWTH OF TILAPIA IN AQUACULTURE

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Abstract

Fingerlings of hybrid tilapia (Oreochromis niloticus x Oreochromis aureus) were cultured in 18 glass aquaria (75 x 40 x 60 cm). Two species of phytoplankton (Chlorella vulgaris & Scenedesmus spp) were used. The experiment included six treatments; each in three replicate aquaria. The 1st treatment (Urea) was fertilization by urea at the rate of 435 mg commercial urea / aquarium. The 2nd treatment (Urea+phy.) was the same rate of urea plus the addition of phytoplankton at a rate of 5X10⁴ cells/ml of water. The 3rd treatment (MSP) was fertilization by mono superphosphate (MSP) at a rate of 830 mg commercial MSP / aquarium. The 4th treatment (MSP+phy.) was the same rate of MSP plus the addition of phytoplankton at a rate of 5X10⁴ cells/ml. The 5th treatment (Urea+MSP) was fertilization with urea and MSP mixture at the rates mentioned above for each fertilizer. The 6th treatment was the Control; without any fertilization or phytoplankton application. Phytoplankton played an important role in removing ammonia and nitrite from the water milieu. The average of individual fish body weight (ABW) at the end of the experiment was (29.37 ± 0.60 g) in the Urea+phy. treatment which was significantly (P<0.05) greater than that of the Control (25.22 ± 0.84 g), and the other treatments. The fish in Urea treatment had the lowest value of ABW (19.22 ± 1.30 g), this is ascribed to the elevated values of un-ionized ammonia (NH₃) and nitrite concentrations in the water of this treatment. On the contrary Urea+phy. treatment had the lowest values of un-ionized ammonia and nitrite due to uptake of the nitrogenous compounds (urea in this study) from the water milieu by phytoplankton, consequently, the medium become more appropriate for fish growth.

Introduction

All algae including phytoplankton can take up nitrate and ammonia from the surrounding water, but ammonia is the preferred form for their growth because the incorporation of nitrate requires additional metabolic energy and enzymatic activity. The flourishing of several algae species reduces the concentration of these chemical elements, thus improving water quality for fish life and production. Fish aquaculture makes use of this phenomenon in fish natural feeding. Nile tilapia has emerged as the single most important species
The global production of farmed tilapia has increased more than three-folds since 1984, from 186,544 m.t. to 659,000 m.t. in 1995.

The ultimate goal of the present study was to find out the effect of phytoplankton inoculation on the thriveness of fish. This effect was sought through the amelioration of water quality by reducing the disastrous elements.

**Materials and Methods**

This experiment was conducted in the Central Laboratory for Aquaculture Research (CLAR), Abbassa, Sharkia Governorate, Egypt. The experimental work was carried out for 16 weeks, from 5 Sep. 1999 to 26 Dec. 1999. Eighteen glass aquaria (75 x 40 x 60 cm) were used for the experimental work. Each aquarium contained 100 liters tap water aerated continuously by air pumps.

**Phytoplankton culture and utilization**

A set of collected Nile water samples were poured in Petri dishes containing solid Bold’s basal medium (Bischoff and Bold, 1963) as a nutrient medium for flourishing algal organisms. These Petri dishes were incubated in the algae culture incubator in the cultivation Lab at 25 ± 1 °C under photoperiods 14/10 light/dark cycles with light intensity 5000 lux. After 6-7 days smears of the grown organisms were examined microscopically to identify the different algal spp. *Chlorella vulgaris* and *Scenedesmus* spp. were the most dominant species in the Nile water samples. These two species were preferred for the intended experimental work since they are slightly digestible by fish as reported by Soeder (1979). These organisms were isolated using standard sterile microbiological techniques according to Guillard (1973). By the help of microscopic examination unialgal organisms of these selected spp. were isolated and passed into a sterile culture solution. Continuous dilution and examination for unialgal organism was done until a pure stock of each organism was obtained. The two algal species were individually subcultured in a solid Bold’s basal medium. The medium was previously autoclaved after the addition of about 20 g/l of agar and then poured in Petri dishes. Algal inoculum of pure slants of *Chlorella vulgaris* and *Scenedesmus* spp. were separately introduced to the surface of the solid medium of two groups of Petri dishes. The cultures were incubated in the algae culture incubator for 7 days. Incubated agar cultures were used to carry out mass cultivation of algae in tap water. Pumped air flow was pushed in these algae cultures. Mass algal cultivation took place in three successive stages; from the agar culture into 2-liter flasks, into 20-liter carboys and lastly into 100-liter aquaria. The Bold’s basal medium was used in all stages after being dissolved in the relevant water volume. When *Chlorella vulgaris* and *Scenedesmus* spp. cultures reached the harvest density (5X10^6 cells/ml), air pumping lines were closed and algal cells were left
for about 24 hours to precipitate then supernatant was siphoned and excluded. Distilled water was added to the phytoplankton sediment to maintain the same density of live algal cells (5X10^6 cells/ml), and to get rid of the chemicals used in the Bold’s basal media. One liter of that phytoplankton sediment (500 ml of *Chlorella vulgaris* + 500 ml of *Scenedesmus* spp.) was added to the 100 ml water of the algae treatment aquaria to give a density of live algae of 5X10^4 ± 933 cells/ml (initial count). All transfers were carried out in aseptic circumstances and all densities were adjusted using microscopic assessment.

**Fingerlings**

Fingerlings of tilapia hybrid (*Oreochromis niloticus* x *Oreochromis aureus*) were purchased from Abbassa Fish Hatchery. The fish were acclimated to laboratory conditions by holding 30 fingerlings in each aquarium for two weeks prior to the experimental execution. Fish were individually observed to select the more viable and healthy ones to stay in the same aquarium. Along the second week of acclimation 15 viable and healthy fingerlings were retained in each aquarium. The initial experimental body weight and body length ranges were from 8-12 g and 7-10 cm respectively.

**Experimental design**

The experiment included six treatments, each in three replicate aquaria as shown in the following table.

Concentrations of mono superphosphate (MSP) and urea per aquarium (mg/100 L tap water) and phytoplankton (phy.) cells (5 x 10^4/ml of water in each aquarium). (3 aquaria for each treatment).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Amount of fertilizers mg/aquarium</th>
<th>And phytoplankton cells/ml of aquarium water</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Urea</td>
<td>MSP</td>
</tr>
<tr>
<td>Control*</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Urea</td>
<td>435</td>
<td>-----</td>
</tr>
<tr>
<td>Urea+phy.</td>
<td>435</td>
<td>-----</td>
</tr>
<tr>
<td>MSP</td>
<td>-----</td>
<td>830</td>
</tr>
<tr>
<td>MSP+phy.</td>
<td>-----</td>
<td>830</td>
</tr>
<tr>
<td>Urea + MSP</td>
<td>435</td>
<td>830</td>
</tr>
</tbody>
</table>

* Nothing added
- All fish in all treatments (including Control) received daily 5% of their body weight artificial feed daily.

**Water quality tests**

Water from each aquarium was tested once a week to determine temperature (°C), pH, concentrations of dissolved oxygen (DO), total ammonia (NH₃ + NH₄), total phosphorus (TP) and nitrite-nitrogen (NO₂-N). All

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determinations were carried out according to the standard methods of the American Public Health Association (APHA) (1985) and Boyd (1990). Temperature and dissolved oxygen were measured at 9.00-10.00 h using yellow spring Instrument (YSI model 57) dissolved oxygen meter. The pH values were assessed by Hach Comparison Apparatus. Total phosphorus concentration was determined using spectrophotometer (model Milton Roy 21 D) at wavelength 690 nm. Total ammonia (NH$_4^+$ + NH$_3$) concentration was measured by Hach Comparison Method, then de-ionized ammonia (NH$_3$) was calculated from total ammonia according to Boyd (1990). Nitrite concentration was measured by the diazodizing method using spectrophotometer (model Milton Roy 21 D) at wavelength 543 nm.

**Phytoplankton dynamics**

Phytoplankton cells in each aquarium were counted twice weekly according to Boyd (1979) before each replacement of water, taking into consideration the adjustment of phytoplankton count to 5x10$^4$ cells/ml.

**Fish tests**

Weight gain of fish was determined by the difference between the initial and final weights of fish at the end of the experimental time (16 week). Relative growth rate (RGR) was determined as a percentage of fish weight gain to the initial weight and Specific Growth Rate (SGR) (growth rate/day) was determined as a percentage of body weight gain/day according to the equation by Allen and Wootton (1982). The general condition of the fish was described by the condition factor (K) calculated for each individual fish from the formula recommended by Bishai (1976)

Statistical analysis was performed using ANOVA and Duncan’s multiple Range Test to determine differences between treatments, means at P< 0.05. All statistics were carried out using Statistical Analysis Systems (SAS) program (SAS, 2000)(R).

**Results and Discussion**

Although the same concentration of phytoplankton inoculation was applied in both treatments with urea or MSP (5 x 10$^5$ ± 933 cells/ml water) the Urea+phy. treatment had phytoplankton concentration of about 75% of that in MSP+phy. treatment throughout the experimental time (16 week). The final overall average of phytoplankton abundance in MSP+phy. treatment was 38,735 cells/ml, while that of Urea+phy. treatment was 29,555 cells/ml (Table 1). This could not be due to the consumption by the fish. Soeder (1979) Stated that these algae species are indigestible due to the polysaccharides complex contained in its cells’ walls, which hinder the penetration of proteolytic enzymes into the interior of these cells within the gastrointestinal tract of the fish. This greater abundance
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of phytoplankton in MSP+phy. treatment compared with Urea+phy. treatment, suggests that fertilization by MSP, creates a more appropriate medium for phytoplankton, which means that, phosphorus is a more effective nutrient for phytoplankton growth. A similar result was obtained by Boyd and Musig (1981) who advocated that phosphorus is a key nutrient in ponds fertilization, and its dynamic has a practical value to pond managers. Saha et al. (1978) found that phosphate is an important nutrient in fertilizers for Indian fish ponds.

In the present study, in both treatments phytoplankton abundance reached the lowest value in week 12 where it reached 27,130 cells/ml in Urea+phy. treatment and 32,644 cell/ml in MSP+phy. Treatment. This phenomenon could be explained by the decrease in the natural water temperature to the lowest degree in that week (14.92 ± 0.07 °C) (Fig. 1). This result may confirm the vital effect of the temperature on phytoplankton growth. This is in agreement with Van Nguyen and Wood (1979) who found that collapse in algae growth is proximally triggered by meteorological changes such as cloudy weather or a decline in water temperature. Knud-Hansen and Batterson (1994) confirmed the important role of temperature in flourishing of phytoplankton; they stated that algal productivity is primarily a function of nutrients, light availability and temperature. It is clear that during the period of the present study there was no significant difference in water temperature among the six treatments (P>0.05). Figure (1) shows that there was a gradual decrease in water temperature from the start of the experiment (1st of Sep.) (25.43 ± 0.10 °C) to reach a plateau in Nov. and Dec. (15.06 ± 0.05 °C). Water temperature in aquaria was related to the room air temperature throughout the experimental time.

There was no significant difference in the dissolved oxygen content between all treatments, along the experimental time (Fig. 1). However an increase of oxygen content in water of Urea treatment was observed compared with that of MSP treatment, slightly after 4 w (2%) to reach 13% and 11% after 12 and 16 w, respectively. This could be attributed to the less fish biomass (171.33 ± 31.63 g) in the former treatment than in the other (335.47 ± 58.46 g), consequently less respiration, and slower consumption of oxygen were attained. The addition of phytoplankton with urea caused a clear drop in DO, while its addition to MSP caused an increase in DO, which is more obvious in the last 8 weeks.

The pH value was significantly higher in Urea treatment than in Control and Urea+MSP treatments (P<0.05). Application of MSP in both MSP and MSP+phy. treatments, as well as the application of phytoplankton in Urea+phy. treatment caused significantly lower pH values than that obtained in the control and Urea+MSP treatments (P<0.05) (Fig.1). Contrary to the Urea treatment, the pH value was gradually decreased in MSP, MSP+phy. and Urea+phy. treatments. The decrease in the pH value in the MSP and Urea+MSP treatments refers to MSP fertilizer, where the MSP is ionized in water to give the negative acidic ion H₂PO₄⁻ (Boyd, 1990). The addition of phytoplankton to urea caused a drop (10.4%) in the pH value than treatment with urea alone (7.47 vs. 8.34). Addition

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of phytoplankton to MSP did not affect the pH value. The fact that the phytoplankton decreased the effect of urea on the pH value is due to uptake of urea by phytoplankton from the ambient water. Healey (1977) stated that urea can be directly utilized by many species of algae.

Table (1) Average phytoplankton (Chlorella vulgaris and Scenedesmus spp.) count (cells/ml) in water treated with urea+phy. and MSP+phy. during the experimental time (16 weeks).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Week within Experimental Periods</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Period</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Urea+Phy.</td>
<td>1st</td>
<td>29902</td>
<td>36125</td>
<td>32653</td>
<td>27828</td>
</tr>
<tr>
<td></td>
<td>2nd</td>
<td>26751</td>
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<td></td>
<td>3rd</td>
<td>27808</td>
<td>34047</td>
<td>24523</td>
<td>30332</td>
</tr>
<tr>
<td></td>
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<td>27808</td>
<td>34047</td>
<td>24523</td>
<td>30332</td>
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<tr>
<td></td>
<td>4th</td>
<td>34575</td>
<td>26370</td>
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<td>29915</td>
<td>32668</td>
<td>27130</td>
<td>28507</td>
</tr>
<tr>
<td>MSP+Phy.</td>
<td>1st</td>
<td>37930</td>
<td>46079</td>
<td>34018</td>
<td>31729</td>
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<tr>
<td></td>
<td>2nd</td>
<td>45145</td>
<td>40667</td>
<td>28317</td>
<td>40084</td>
</tr>
<tr>
<td></td>
<td>3rd</td>
<td>48106</td>
<td>43450</td>
<td>33159</td>
<td>37488</td>
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<tr>
<td></td>
<td>4th</td>
<td>40943</td>
<td>37918</td>
<td>36041</td>
<td>41765</td>
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<tr>
<td>Average</td>
<td></td>
<td>43613</td>
<td>40325</td>
<td>32644</td>
<td>38359</td>
</tr>
</tbody>
</table>

- Initial count of Phytoplankton for both treatments was 50,000 ± 933 cell/ml.
* The assessment of phytoplankton number was carried out twice per week after 3 - 4 days after replenishing the water with the same treatment in each aquarium.
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Fig (1) Water temperature (a), Dissolved oxygen (b) and pH (c) of the six experimental treatments during the experimental periods (16 weeks).

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The total phosphorus concentrations were significantly higher in the MSP and Urea+MSP treatments than in the other treatments (P<0.05) (Fig. 2). At the end of the experimental time (week 16) the total phosphorus concentration in MSP+phy. treatment (0.321 ± 0.108 mg/l) was about 50% lower than that of treatment by MSP alone (0.678 ± 0.01 mg/l), although the same dose of MSP fertilizer was applied in both treatments. It is worth noticing that the phosphorus concentration in this MSP+phy. treatment showed close concentration to that of control (0.296 ± 0.083 mg/l) which contained phosphorus from the artificial feeding. These results are due to the uptake of phosphorus from the water by phytoplankton. Rodina (1966) and Fry (1987) stated that phytoplankton species are certainly capable of taking up soluble inorganic phosphorus from the water, as well as, many forms of soluble organic phosphorus produced by the enzymatic breakdown of organic molecules.

Total ammonia concentration was significantly higher in the Urea treatment (P<0.05), followed by Urea+MSP, MSP, control, Urea+phy. and MSP+phy. treatments, respectively. Although the same dose of urea was applied in urea and Urea+MSP treatments, yet the ammonia was reduced by the presence of MSP which demonstrate the role of MSP in reducing the dissociation of urea towards formation of ammonia in water. Phytoplankton application with urea (Urea+phy.) induced a dramatic reduction in total ammonia concentration. The overall average of total ammonia was significantly lower (P<0.05) in Urea+phy. treatment (1.40 ± 0.30 mg/l) than in Urea treatment (4.29 ± 0.39). This result confirms the fact that urea and/or ammonia were uptaken by phytoplankton from the water. Healey (1977) stated that urea can be directly utilized by many species of algae. Abdalla (1989) concluded that uptake of total ammonia by phytoplankton accounted for 37% to 100% during daylight hours. The concentration of total ammonia in urea and Urea+MSP treatments showed great increase from the start of the experiment (1.13 ± 0.09 and 1.10 ± 0.00 mg/l respectively) to the end of the experiment (5.47 ± 0.58 and 3.73 ± 0.42 mg/l, respectively) (Fig. 2). This could be ascribed to the release of more ammonia from the grown fish as a result to urea application in both treatments. Colt and Armstrong (1981) recorded that ammonia is the principal nitrogenous compound excreted by aquatic animals. The major source of ammonia in pond’s water is the direct excretion of ammonia by fish (Tucker and Boyd, 1985). Wang and Walsh (2000) reported that fish are both ureotelic (excreting urea) and ammoniotelic (excreting ammonia). Engin and Carter (2001) revealed that the higher rates of urea-nitrogen excreted by teleost fishes indicates that urea is an important excretory end-product in these species. In the control, MSP and MSP+phy. treatments, the average concentration of total ammonia was low at the end of the experiment (2.26 ± 0.76, 2.43 ± 0.39 and 1.80 ± 0.16 mg/l respectively). During the experimental time, the overall average of ammonia (NH₃) concentration was significantly higher (P<0.05) in Urea treatment (0.446 ± 0.07 mg/l) than in all other treatments (Fig., 2). At the end of the experiment the NH₃ reached the
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Fig. (2) Total phosphorus (a), ammonia NH$_3$ (b) and nitrite (c) concentrations of the six experimental treatments during the experimental periods (16 weeks).

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greatest contribution in total ammonia in the Urea treatment and the least in Urea+phy. treatment. The percentages of NH\textsubscript{3} were 13.51\%, 3.38\%, 3.36\%, 0.91\%, 0.89 and 0.88 from the total ammonia concentration in urea, Urea+MSP, Control, MSP, MSP+phy and Urea+phy., respectively. This caused a higher pH value (8.63) in the Urea treatment than that of control, Urea+MSP, MSP, MSP+phy. and Urea+phy. (7.96, 7.90, 7.40, 7.37 and 7.37, respectively). This is in agreement with Boyd (1990) who demonstrated that pH value decreases as the ratio of NH\textsubscript{3} : NH\textsubscript{4} decreases and vice versa. There was drastic increase in the concentration of ammonia in the Urea treatment throughout the experimental time. The concentration started with 0.042 ± 0.002 mg/l and reached 0.739 ± 0.036 mg/l at the end of the experiment. This was mainly due to several factors; notably, partial dissociation of the urea fertilizer and the more excretion of urea and ammonia fraction by the grown fish biomass. The increase of total ammonia and NH\textsubscript{3} concentration in the 3\textsuperscript{rd} and 4\textsuperscript{th} periods in the Urea+phy. and MSP+phy. treatments, was partly associated with the decrease in water temperature, which decreases the number of phytoplankton cells, as well as, retarding the activity of ammonia uptake by phytoplankton. Mitamura (1986) reported that the assimilation rate of ammonia nitrogen was highest in summer and lowest in winter, as expected from the seasonal variation in water temperature. Inoculation of these algae spp., by that experimentally applied density, in the aquaria water caused the useful effect of reducing the NH\textsubscript{3} concentration. This effect was immense in case of applying with Urea treatment, causing around 95\% reduction to that of treatment by urea alone. The effect was less obvious in case of adding the phytoplankton with MSP. It is interesting that the phytoplankton reduced NH\textsubscript{3} concentration in both cases to 0.009 mg/l during the 1\textsuperscript{st} and 2\textsuperscript{nd} periods and to 0.018 – 0.016 mg/l in the 3\textsuperscript{rd} and 4\textsuperscript{th} periods. This effect of these species, is of vital value since the NH\textsubscript{3} is toxic and high concentration of it causes stunted growth and/or death of fish in accordance with tolerance of fish spp. and age. Such a biological inference emphasizes its practical value in aquaculture projects, moreover in amelioration of water quality in cases of pollution. The toxicity of ammonia depends on the pH value (Tomasso \textit{et al}., 1980), total ammonia (Boyd, 1990), carbon dioxide (Lloyd & Herbert, 1960) and dissolved oxygen (Merkens & Downing, 1957).

Nitrite concentration showed the greatest values, throughout the experimental time, in the two treatments urea and Urea+MSP, however, Urea treatment had higher values (P<0.05) (Fig., 2). The overall mean of nitrite in the Urea treatment was 0.848 ± 0.101 mg/l ranging from 0.601 ± 0.168 to 1.147 ± 0.145 mg/l, the overall average in the Urea+MSP treatment was 0.513 ± 0.078 mg/l ranging from 0.394 ± 0.200 to 0.627 ± 0.238 mg/l. The nitrite concentration in both urea and Urea+MSP treatments had an increasing trend during the course of the experiment, while that of Urea+phy. treatment had relatively stable value at that low level. Nitrite concentration in the Urea treatment was doubled from week 4 (0.601 ± 0.168 mg/l) to the end of the experiment (week 16) (1.147 ± 0.145 mg/l). This continuous increase combined with the elevated ammonia
concentration increased fish mortality than that in the other treatments, fish mortality in Urea treatment was the highest and reached 40%. Inoculation of phytoplankton in the Urea+phy. treatment induced the lowest concentration of nitri...e 0.086 ± 0.005 mg/l) (P<0.05) ranging from 0.073 ± 0.014 to 0.099 ± 0.024 mg/l. This result clarifies the beneficial role which the phytoplankton performs in such circumstances, where it uptakes the nitrogenous compounds, thus maintaining healthy conditions to the fish and other organisms. This phytoplankton function is helpful in resisting pollutants in water, as evidenced in Figure (3). Colt and Armstrong (1981) observed that tilapia growth was retarded when nitrite levels exceeded 1.6 mg/l. they reported that, trout is very sensitive being stressed by 0.15 mg/l and killed by 0.55 mg/l nitrite.

**Phytoplankton Amelioration of Water Quality:**

This study emphasized the importance of using these phytoplankton species in aquaculture since they were efficient in depriving the water from injurious elements. Figure (3) shows the efficiency of phytoplankton in reducing the concentration of NH₃, total ammonia and NO₂; the compounds which causes disastrous effect on the vitality and growth of fish, ultimately decreasing fish harvest in a culture.

Mitamura (1986) stated that the rate of nitrogen assimilation is regulated primarily by the phytoplankton standing crop. It is clear in the Figure (3) that phytoplankton clearance efficiency was very effective with urea fertilization than with MSP fertilization. This difference is of biological benefit since urea alone causes greater concentrations of these elements - up to detrimental or even fatal levels- than the lower levels with MSP alone (Figures, 4 & 5). Nitrogen uptake rates by algae depend on the concentration of nitrogen source (Haines & Wheeler, 1978 and Lobban & Harrison, 1994). McCarthy *et al.* (1977) stated that the phytoplankton preferentially utilizes ammonia as a nitrogen source and urea nitrogen is used second to ammonia.

**Fish performance:**

**Survivability:**

Figure (4) shows clearly that phytoplankton treatments maintained the highest survival rate. On the contrary, treatment with urea alone caused adverse effect on the vitality of fish causing disastrous mortality leaving only 58% live fish. It is of practical value that adding MSP to urea deleted, to a great extent, the negative effect of urea. The phytoplankton application induced excellent maintenance of the fish, keeping 96% survivals. The results of Urea treatment alone was confirmed by Abdalla *et al.* (1996) who reported that fish mortality in ponds with high weekly urea input (14.3 g N/m³) was 71%, while in ponds with low weekly urea input (1.2 g N/m³) fish mortality declined between 22 and 26%.
Total Weight Gain (TWG):

The TWG of fish, after 16 weeks (112 d), treated with different fertilization and phytoplankton treatments (Table 2) showed clear different responses. The greatest weight increment, throughout the experimental time (16 w) was obtained in Urea+phy. treatment (18.83 ± 0.89 g/fish) compared to the increments; 14.82 ± 2.30 g/fish in control, 14.30 ± 1.35 g/fish in MSP treatment, 11.43 ± 0.62 g/fish in Urea+MSP treatment and the least one 8.66 ± 0.61 g/fish in Urea treatment. There was a negative relationship between that gain and the concomitant ammonia (NH₃) concentration (Fig. 5).

Fig. (3) Clearance efficiencies of total ammonia, ammonia (NH₃) and Nitrite (NO₂) by phytoplankton from water treated with urea+phy. and MSP+phy. treatments

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Fig. (4) Survival percentage of Tilapia (*Oreochromis niloticus* X *Oreochromis aureus*) exposed to water fertilization and phytoplankton treatments, at the end of the experimental time (16 weeks).

Fig. (5) Relation between un-ionized ammonia (NH$_3$) concentrations and individual weight gain of tilapia *Oreochromis niloticus* X *O. aureus* exposed to water fertilization and phytoplankton treatments. For 16 weeks.
**Table (2) Fish growth traits: Initial weight (iw), final weight (fw), total weight again (TWG), specific growth rate (SGR), relative growth rate (RGR) and condition factor (k) of Tilapia (*Oreochromis niloticus X Oreochromis aureus*) exposed to water fertilization and phytoplankton treatments. for 16 weeks. (Mean ± S.E.)**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>iw (g/fish) ± S.E.</th>
<th>fw (g/fish) ± S.E.</th>
<th>TWG (g/fish) ± S.E.</th>
<th>SGR (%BW/day) ± S.E.</th>
<th>RGR (% IW) ± S.E.</th>
<th>Condition factor (k) ± S.E.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10.4 ± 0.78 a</td>
<td>25.22 ± 0.84 b</td>
<td>14.82 ± 2.30 ab</td>
<td>0.78 ± 0.12 ab</td>
<td>150.09 ± 27.12 ab</td>
<td>1.53 ± 0.02 ab</td>
</tr>
<tr>
<td>Urea</td>
<td>10.56 ± 0.84 c</td>
<td>19.22 ± 1.30 c</td>
<td>8.66 ± 0.61 c</td>
<td>0.53 ± 0.03 c</td>
<td>81.61 ± 5.52 c</td>
<td>1.44 ± 0.02 b</td>
</tr>
<tr>
<td>Urea+phy.</td>
<td>10.54 ± 1.26 a</td>
<td>29.37 ± 0.60 a</td>
<td>18.83 ± 0.89 a</td>
<td>0.93 ± 0.07 a</td>
<td>188.14 ± 11.22 a</td>
<td>1.59 ± 0.04 a</td>
</tr>
<tr>
<td>MSP</td>
<td>10.53 ± 0.10 a</td>
<td>24.83 ± 1.67 b</td>
<td>14.30 ± 1.35 b</td>
<td>0.76 ± 0.05 ab</td>
<td>135.44 ± 12.82 ab</td>
<td>1.64 ± 0.06 a</td>
</tr>
<tr>
<td>MSP+phy.</td>
<td>10.45 ± 0.78 a</td>
<td>23.32 ± 0.66 b</td>
<td>12.87 ± 0.80 bc</td>
<td>0.72 ± 0.06 ab</td>
<td>126.22 ± 12.04 bc</td>
<td>1.57 ± 0.04 a</td>
</tr>
<tr>
<td>Urea+MSP</td>
<td>10.49 ± 0.85 b</td>
<td>21.92 ± 0.73 bc</td>
<td>11.43 ± 0.62 bc</td>
<td>0.66 ± 0.05 bc</td>
<td>111.44 ± 9.37 bc</td>
<td>1.52 ± 0.04 ab</td>
</tr>
</tbody>
</table>

- a,b,c: Means in the same column followed by different letters denote significant treatment difference (P < 0.05).

**TWG** = Final weight - Initial weight

\[
TWG = \frac{fw - iw}{iw} \times 100
\]

**SGR** = Specific Growth Rate

\[
SGR = \frac{Ln fw - Ln iw}{Time(days)} \times 100
\]

**RGR** = Relative Growth Rate

\[
RGR = \frac{fw - iw}{iw} \times 100
\]

**Condition factor** = Condition factor

\[
Condition\ factor = \frac{Weight(g)}{Length\ (cm)} \times 100
\]

**Specific Growth Rate (SGR) & Relative Growth Rate (RGR):**

Table (2) demonstrates that SGR and RGR followed the same trend of treatments effects as TWG caused the highest SGR and RGR (0.93 ± 0.07 % BW/day and 188.14 ± 11.22 %IW respectively) followed by; control (0.78 ± 0.12 and 150.09 ± 27.12), MSP (0.76 ± 0.05 and 135.44 ± 12.82), MSP+phy. (0.72 ± 0.06 and 126.22 ± 12.04), Urea+MSP (0.66 ± 0.05 and 111.44 ± 9.37). The lowest rates were that in Urea (0.53 ± 0.03 and 81.61 ± 5.52).

**Condition Factor (K):**

As shown in Table (2) MSP treatment caused the significantly (P<0.05) highest (K) value where it was 1.64 ± 0.06, while the lowest (K) was observed in Urea treatment (1.44 ± 0.02). There was no significant difference between MSP treatment and Urea+phy., MSP+phy., control and Urea+MSP treatments respectively.

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Conclusion

The results of this study confirm that it is not recommended to use urea as a unique fertilizer, with the same dose used in this study, where it has deleterious effects in the water environment, such as; the release of toxic forms of ammonia (NH₃) and nitrite (NO₂). Consequently great percentage of fish died in this treatment, and the remaining fish had a decreased daily weight gain, ultimately fish biomass was decreased.

The addition of MSP (mono superphosphate) to urea reduced the concentrations of ammonia and nitrite, compared to urea alone, although the same dose of urea was applied in both treatments, which demonstrate the role of MSP in reducing the dissociation of urea towards formation of ammonia and nitrite in water.

Phytoplankton application – by inoculation in the water- induced a marvelous effect either on the water quality and fish thriveness. This application ameliorated the quality of water; consequently the fish response was augmented towards better thriveness.

References


تأثير التلقيح بالهائمات النباتية مع إضافة بعض الأسمدة الكيميائية على جودة المياه ونمو أسماك البلطي

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تم استخدام إصبعيات البلطي الهجين (البلطي النيلي x البلطي الأزرق) و نوعين من الأسمدة الكيميائية (سماد السوبرفوسفات الأحادي و سماد البوريا) و نوعين من الطحالب الخضراء (الكلوريل فاجر المنسمس) في 18 حوض زجاجي بقياس (75 x 40 x 60 سم). شملت هذه التجربة ستة معاملات لكل منها ثلاثة تكرارات وكانت المعاملة الأولى هي التسخين بسماد البوريا بعدين 435 ملجم/حوض. المعاملة الثانية هي التسخين بالبوريا بنفس المعدل السابق بالإضافة إلى إضافة الطحالب إليها بتركيز 5 خلايا/مل. المعاملة الثالثة هي التسخين بالسوبرفوسفات الأحادي بعدين 830 ملجم/حوض. المعاملة الرابعة هي التسخين بسماد السوبرفوسفات الأحادي بنفس المعدل السابق بالإضافة إلى إضافة الطحالب إليها بتركيز 5 خلايا/مل. المعاملة الخامسة هي التسخين بمخلوط سوادياً البوريا والسورفوسفات بدون إضافة أي سماد أو طحالب. لعبت الطحالب دوراً هاماً في إزالة كل من الأمونيا والنيتروجين من المياه. كان متوسط الوزن الفردي للأسماك في نهاية التجربة في معاملة 29.37±0.60 جم (أعلى معيار). وبلغ متوسطة قل وزن urea+phy (19.22±1.30 جم) وهو لونز المعاملات (22±0.84 جم). وخلايا المعاملات كانت صاحبة أقل وزن NH3 كلاً من الأمونيا غير المتذبذبة (NH3) ونترتبت في هذه المعاملة. وعلى النقيض من هذا فإن معاملة control كانت صاحبة أقل تركيز من الأمونيا ونترتبت كنتيجة لامتصاص الطحالب لهذه المركبات النيتروجينية (البوريا في هذه التجربة) من الوسط المائي مما جعله أكثر ملاءمة لنمو الأسماك.