Chronic ammonia toxicity to duckweed-fed tilapia
(Oreochromis niloticus)

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Abstract

The effects of prolonged exposure to sub-lethal un-ionised ammonia nitrogen (UIA-N) concentrations on the growth performance of juvenile Nile tilapia (Oreochromis niloticus) fed on fresh duckweed (Lemna gibba) grown on pretreated domestic sewage have been investigated. The experiment was conducted over 75 days using juveniles with a mean body weight of 20 g. Five nominal, total ammonia nitrogen concentrations (control, 2.5, 5, 7.5 and 10 mg N l⁻¹) were established as treatment groups. Statistical analysis of the specific growth rate (SGR) showed no significant (p>0.05) differences between the SGR (0.71) of tilapia in the control (0.004 mg UIA-N l⁻¹) and the SGR (0.67) of those exposed to 0.068 mg UIA-N l⁻¹. The SGR of tilapia exposed to un-ionised ammonia nitrogen over 0.068 mg UIA-N l⁻¹ (0.144, 0.262 and 0.434 mg UIA-N l⁻¹) was significantly reduced (p<0.01). The no-observable effect concentration was 0.068 mg UIA-N l⁻¹, while the lowest observable effect concentration was 0.144 mg UIA-N l⁻¹. Increasing the un-ionised ammonia concentration increased the feed conversion ratio (FCR). At 0.144 mg UIA-N l⁻¹, the FCR increased by a factor 1.6 of the value observed in the control, while at 0.262 mg UIA-N l⁻¹ the FCR increased by a factor of 2.7. At 0.434 mg UIA-N l⁻¹, the FCR increased by a factor of 4.3. Protein efficiency ratio (PER) was also negatively correlated with the un-ionised ammonia concentration above 0.068 mg UIA-N l⁻¹. This study concluded that, for raising Nile tilapia in fishponds fed on fresh duckweed or other feed, the toxic level of UIA-N and its negative effect on the
growth performance lies between 0.07 and 0.14 mg UIA-N l\(^{-1}\). It is recommended that the UIA-N concentration be maintained below 0.1 mg l\(^{-1}\).

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Keywords: Chronic ammonia toxicity; Un-ionised ammonia; Oreochromis niloticus; Duckweed; Lemna gibba; Domestic sewage

1. Introduction

Ammonia is toxic, not only to fish but also to all aquatic animals (Baird et al., 1979; Zhao et al., 1997; Harris et al., 1998), especially in pond aquaculture at low concentrations of dissolved oxygen (Alabaster et al., 1983). The toxic levels of un-ionised ammonia for short-term exposure usually are reported to lie between 0.6 and 2 mg/l, while some consider the maximum tolerable concentration to be 0.1 mg l\(^{-1}\) (Pillay, 1992). Feed efficiency and body composition of fish are negatively affected by ambient ammonia concentration. The main change in the body composition includes an increase in the water content (Person-Le Ruyet et al., 1997b).

Two primary processes affect ammonia concentration in ponds, ammonia excretion rate by fish (indigenous ammonia) and sediment diffusion (Hargreaves, 1997) that represents one part of the exogenous ammonia in the pond. The production of indigenous ammonia is related to the quantity of nitrogen supplied by dietary protein (Buttle et al., 1995; Brunty et al., 1997; Chakraborty and Chakraborty, 1998; Thomas and Piedrahita, 1997). In tilapia, a significant increase in total ammonia nitrogen production was observed during the 24-h period following feeding as the dietary protein increased (Brunty et al., 1997). In Clarias gariepinus, small differences in relative protein may evoke distinct patterns of ammonia efflux over a range of dietary protein levels (Buttle et al., 1995). Different species of fish and fish at different life stages within the same species are differentially able to utilise the dietary protein and the relationship between dietary protein levels and total ammonia production for fish species and life stages within the same species may vary.

Ammonia toxicity is a problem relevant to the practice of fish farming, since the growth yield may be depressed by usual ambient ammonia concentrations, under intensive farming conditions (Person-Le Ruyet et al., 1997a) and in sewage-fed aquaculture. Wrigley et al. (1988) reported that high ammonia and low dissolved oxygen concentration during the summer and spring were the major factors responsible for mortality in sewage-fed fishponds. Others reported that ammonia toxicity was the main reason of fish mortality in sewage oxidation ponds (El-Gohary et al., 1995; Nasr et al., 1998). In sewage-fed fish farms, sewage ammonia is the main, if not only, source of pond fertilisers. Algae utilise ammonia to grow and produce oxygen, whereas fish require both. The presence of more nitrogen loads to the pond increases both primary and secondary productivity of ponds and so more feed is available for fish for maximum growth (Diana et al., 1997). On the contrary, algae increase biodegradable organic matter and consume more oxygen during the night and create water hypoxia (Pillay, 1992; Diana et al., 1997). The algae bloom also elevates the water pH value and
shifts the ammonia balance towards more un-ionised ammonia, which results in greater toxicity for fish (Pillay, 1992; Diana et al., 1997). In sewage-fed fish aquaculture systems, the ammonia concentration will increase, decrease, or remain stable over time if ammonia excretion by fish and ammonia in the inlet water, are greater than, less than, or equal to the nutrient assimilation by phytoplankton and nutrient losses, respectively (Seawright et al., 1998). In the sewage-fed fish farm, a dynamic balance between ammonia and algae concentrations should be optimised for optimal growth conditions. A stocking density between 10,000 and 30,000 fingerlings per hectare in a sewage-fed pond is recommended with an optimal nitrogen loading rate of 4 kg N/ha/day (Mara et al., 1993).

Worldwide use of duckweed in wastewater treatment means that it is available at a reasonable price for use in fish aquaculture since it has a high protein content. However, ammonia causes moderate and severe physiological disturbance, depending on the level of ammonia and exposure duration. The chronic toxicity level of ammonia in duckweed-fed tilapia has to be defined since deficiency of duckweed in certain nutritional elements might affect the immune system and resistance of tilapia to un-ionised ammonia. The main objective of the current research was to study the effect of chronic ammonia toxicity on the growth performance of tilapia (Oreochromis niloticus) fed on fresh duckweed. The exposure-time was 2 1/2 months, which was more than the minimum of 1 month recommended for chronic toxicity tests on fish growth (APHA, 1998).

2. Material and methods

2.1. Fish species and test conditions

The experiment was conducted using juvenile tilapia (O. niloticus) with 20 g mean body weight, all from the same parental stock, obtained from a commercial fish farm, Cairo, Egypt. For 2 weeks prior to the experiment, the fish were adapted to the pre-selected experimental conditions in one tank with an effective water volume of 480 l and 1 m² surface area. The tank was supplied with running de-chlorinated and continuously aerated city tap water. The de-chlorination was performed using sodium thiosulfate solution (0.025 M). The water flow rate was 50 l/day. Tilapia were fed with fresh duckweed harvested from a duckweed pond system treating effluent from an Up-flow Anaerobic Sludge Blanket (UASB) reactor fed with domestic sewage (El-Shafai et al., submitted for publication). The daily feeding was 250 g fresh duckweed (Lemna gibba) per kg of fish. Daily maintenance and cleaning of the fish tank from faeces was done.

Two days before the experiment, the fish were randomly distributed between the experimental tanks. Each tank had 100-l effective water volume and 0.26 m² surface area. The tanks were arranged in two sets of five tanks each. The maximum recommended fish densities for toxicity testing at a temperature >20 °C is 2.5 g l⁻¹ (APHA, 1998). In this experiment, initial stocking-density was four fish per each tank (20 g average weight each) (Table 1). The fish were allowed to adapt for 2 days, during
which feeding was performed using a 25% feeding rate based on fresh duckweed (L. gibba) and live body weight of fish. The tanks were fed with de-chlorinated city tap water at 25 l daily.

2.2. Experimental design and analytical procedures

In addition to the control, four treatments with nominal total ammonia nitrogen (TAN) concentrations of 2.5, 5, 7.5 and 10 mg N l$^{-1}$ were tested with duplicates for each treatment. Ammonium chloride stock solution (25 g l$^{-1}$) was used as a source of ammonia. Each tank was fed with de-chlorinated city tap water containing the desired ammonia concentrations. When necessary, the ammonia concentration in the water flow was adjusted to the desired concentration. The experiment lasted 75 days during which total feed intake was recorded for each treatment. The fish in each treatment were weighed monthly and at the end of the trial. Daily observation of the fish behaviour was performed.

TAN was determined using the boric acid–sulphuric acid titration method (APHA, 1998). Un-ionised ammonia nitrogen (UIA-N) concentrations were calculated using the general equation of bases (Albert, 1973):

$$\text{NH}_3 = \frac{[\text{NH}_3 + \text{NH}_4^+]}{[1 + 10^{pK_a - \text{pH}}]} \quad (1)$$

In fresh water, the calculation of $pK_a$ is based on the equation developed by Emerson et al. (1975):

$$pK_a = 0.09018 + 2729.92/T \quad (2)$$

($T$=Kelvin=$273 + T$ °C) Daily (at 10:30 AM) pH was determined electrochemically using a “Radio pH meter model 62”. BOD, nitrate nitrogen, nitrite nitrogen, total hardness and total alkalinity were measured according to the Standard Methods for the Examination of Water and Wastewater (APHA, 1998). BOD, nitrate nitrogen, total hardness and total alkalinity were measured weekly while nitrite was measured twice a week.

<table>
<thead>
<tr>
<th>Item</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Surface area of tank (m$^2$)</td>
<td>0.26</td>
<td>0.26</td>
<td>0.26</td>
<td>0.26</td>
<td>0.26</td>
</tr>
<tr>
<td>Working volume (l)</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Initial mean body weight (g fish$^{-1}$) (± S.D.)</td>
<td>21.2 ± 5.5</td>
<td>21.5 ± 6.5</td>
<td>24.3 ± 7.4</td>
<td>26.2 ± 5.7</td>
<td>26.1 ± 9.3</td>
</tr>
<tr>
<td>Stocking density (fish tank$^{-1}$)</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
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<tr>
<td>Feeding rate (%)</td>
<td>25%</td>
<td>25%</td>
<td>25%</td>
<td>25%</td>
<td>25%</td>
</tr>
<tr>
<td>Water flow (l day$^{-1}$)</td>
<td>25</td>
<td>25</td>
<td>25</td>
<td>25</td>
<td>25</td>
</tr>
</tbody>
</table>

Table 1
Operational conditions in fish tanks
2.3. Studied parameters

Mortalities were counted daily for different exposure doses. Every month and at the end of the trial, the fish in each tank were weighed individually and specific growth rates (SGR) were calculated using the following expression:

$$\text{SGR} = \left[ \frac{\ln \frac{W_f}{W_i}}{\text{days}} \right] \times 100$$

(3)

Where, $W_i$ and $W_f$ are initial and final mean body weight, respectively.

The total feed intake was recorded in each treatment, and the feed conversion ratio (FCR) and protein efficiency ratio (PER) were calculated based on the following expression:

$$\text{FCR} = \frac{\text{total feed ingested (dry weight)}}{\text{fish weight gain (wet weight)}}$$

(4)

$$\text{Protein efficiency ratio (PER)} = \frac{\text{fish weight gain}}{\text{total protein ingested}}$$

(5)

Composition of duckweed was analysed for dry matter and protein content. Dry matter content was determined by drying at 70 °C overnight. The protein content was calculated from the organic nitrogen (org-N × 6.25) using the macro-Kjeldahl method (APHA, 1998).

2.4. Data analysis

All the results were presented as mean values and standard deviation. The influence of ammonia concentration was analysed by one-way analysis of variance (one-way

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Table 2

Water quality in fish tanks during chronic ammonia toxicity experiment

<table>
<thead>
<tr>
<th>Parameter</th>
<th>1 (Control)</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature ($^\circ$C)</td>
<td>26–34</td>
<td>26–34</td>
<td>26–34</td>
<td>26–34</td>
<td>26–34</td>
</tr>
<tr>
<td>pH</td>
<td>7.2–7.5</td>
<td>7.3–7.6</td>
<td>7.3–7.7</td>
<td>7.4–7.8</td>
<td>7.4–7.9</td>
</tr>
<tr>
<td>BOD (mg O$_2$ l$^{-1}$)</td>
<td>5.2 ± 0.6</td>
<td>4.9 ± 0.5</td>
<td>5.3 ± 1.3</td>
<td>6.0 ± 0.4</td>
<td>6.5 ± 1.5</td>
</tr>
<tr>
<td>DO (mg O$_2$ l$^{-1}$)</td>
<td>7.2 ± 1</td>
<td>7.5 ± 0.8</td>
<td>7.7 ± 0.8</td>
<td>8.0 ± 0.7</td>
<td>8.3 ± 0.8</td>
</tr>
<tr>
<td>TAN$^a$ (mg N l$^{-1}$)</td>
<td>0.2 ± 0.04</td>
<td>2.9 ± 0.7</td>
<td>5.3 ± 0.6</td>
<td>7.5 ± 0.5</td>
<td>10 ± 0.6</td>
</tr>
<tr>
<td>UIA-N$^b$ (mg N l$^{-1}$)</td>
<td>0.004 ± 0.008</td>
<td>0.068 ± 0.03</td>
<td>0.144 ± 0.06</td>
<td>0.262 ± 0.1</td>
<td>0.434 ± 0.18</td>
</tr>
<tr>
<td>Nitrite (mg N l$^{-1}$)</td>
<td>0.04 ± 0.03</td>
<td>0.036 ± 0.028</td>
<td>0.043 ± 0.03</td>
<td>0.037 ± 0.026</td>
<td>0.033 ± 0.025</td>
</tr>
<tr>
<td>Nitrate (mg N l$^{-1}$)</td>
<td>0.067 ± 0.03</td>
<td>0.072 ± 0.04</td>
<td>0.085 ± 0.05</td>
<td>0.088 ± 0.05</td>
<td>0.095 ± 0.08</td>
</tr>
<tr>
<td>T hardness (mg CaCO$_3$ l$^{-1}$)</td>
<td>120 ± 1</td>
<td>121 ± 1</td>
<td>120 ± 2</td>
<td>121 ± 2</td>
<td>121 ± 1</td>
</tr>
<tr>
<td>Total alkalinity</td>
<td>127 ± 3</td>
<td>124 ± 6</td>
<td>128 ± 4</td>
<td>129 ± 5</td>
<td>129 ± 5</td>
</tr>
</tbody>
</table>

$^a$ Total ammonia nitrogen.

$^b$ Un-ionised ammonia nitrogen.
ANOVA). The lowest-observable-effect concentration (LOEC) was estimated (Person-Le Ruyet et al., 1997a).

3. Results

3.1. Water quality

The experiment extended for 75 days during which the temperature was the same in all tanks, but it varied during this period in the range of 26–34 °C. The pH values in the treatment tanks are presented in Table 2. The DO was above 5 mg O₂ l⁻¹ in all tanks. The calculated un-ionised ammonia based on the temperature, pH and total ammonia concentration was 0.004, 0.068, 0.144, 0.262 and 0.434 mg UIA-N l⁻¹ in the control (treatment 1), treatments 2, 3, 4 and 5, respectively (Table 2). The total hardness and total alkalinity were

<table>
<thead>
<tr>
<th>Item</th>
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<th>2</th>
<th>3</th>
<th>4</th>
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</tr>
</thead>
<tbody>
<tr>
<td>Initial mean body weight (g fish⁻¹)</td>
<td>21.2 ± 5.5</td>
<td>21.5 ± 6.5</td>
<td>24.3 ± 7.4</td>
<td>26.2 ± 5.7</td>
<td>26.1 ± 9.3</td>
</tr>
<tr>
<td>Final mean body weight (g fish⁻¹)</td>
<td>36.2 ± 15</td>
<td>35.5 ± 13.3</td>
<td>30.1 ± 9.5</td>
<td>30.4 ± 7.7</td>
<td>29 ± 10.8</td>
</tr>
<tr>
<td>Fish weight gain (g fish⁻¹)</td>
<td>15</td>
<td>14</td>
<td>5.8</td>
<td>4.2</td>
<td>2.9</td>
</tr>
<tr>
<td>Total feed intake (g fish⁻¹)²</td>
<td>22.3</td>
<td>22.1</td>
<td>22.6</td>
<td>23.6</td>
<td>23.2</td>
</tr>
<tr>
<td>SGR (± S.D.)</td>
<td>0.71 ± 0.13⁶</td>
<td>0.67 ± 0.1³</td>
<td>0.29 ± 0.04⁶</td>
<td>0.2 ± 0.05⁶</td>
<td>0.14 ± 0.03⁶</td>
</tr>
<tr>
<td>FCR (g feed g fish⁻¹)</td>
<td>1.5</td>
<td>1.6</td>
<td>3.9</td>
<td>5.6</td>
<td>8.0</td>
</tr>
<tr>
<td>Total protein intake (g fish⁻¹)</td>
<td>8.03</td>
<td>7.96</td>
<td>8.14</td>
<td>8.5</td>
<td>8.35</td>
</tr>
<tr>
<td>PER (g fish g protein⁻¹)</td>
<td>1.78</td>
<td>1.76</td>
<td>0.71</td>
<td>0.49</td>
<td>0.35</td>
</tr>
</tbody>
</table>

* a g dry duckweed/fish.
  b, c, d means indicated with a different letter are significantly different (p < 0.01; * indicates p < 0.05).
  * p < 0.05.

Fig. 1. PER and FCR in relation to UIA-N concentrations.
stable. Except ammonia, no significant differences in the water quality were detected between the treatment tanks. Statistical analysis of water quality showed no significant differences between the parallel treatments in the two sets of experiment.

3.2. Growth performance

Growth performance of tilapia in different treatment tanks is presented in Table 3. No mortality was detected in any treatment during the experimental period. The results of the specific growth rates (SGR) showed mean values of 0.71, 0.67, 0.29, 0.2 and 0.14 in treatments 1, 2, 3, 4 and 5, respectively. The statistical analysis showed no significant difference \( p > 0.05 \) between the control group \( (0.004 \text{ mg UIA-N l}^{-1}) \) and tilapia exposed to \( 0.068 \text{ mg UIA-N l}^{-1} \). The SGR of treatments 3 \( (0.144 \text{ mg UIA-N l}^{-1}) \), 4 \( (0.262 \text{ mg UIA-N l}^{-1}) \) and 5 \( (0.434 \text{ mg UIA-N l}^{-1}) \) were significantly lower \( p < 0.01 \) than the control and treatment 2. There was a significant difference \( p < 0.05 \) between the SGR of tilapia exposed to \( 0.144 \) and \( 0.262 \text{ mg UIA-N l}^{-1} \) but no difference \( p > 0.05 \) was detected between the SGR of tilapia in group 4 and those in group 5.

No effect on the feed intake was detected in any of the treatment groups. The feed conversion was affected by ammonia concentrations over \( 0.068 \text{ mg UIA-N l}^{-1} \). There was no difference between the FCR of the control and group 2. The FCR increased by 4.3 fold in treatment 5 \( (0.434 \text{ mg UIA-N l}^{-1}) \) as compared to the control (Fig. 1). Increasing the UIA-N over \( 0.068 \text{ mg l}^{-1} \) progressively decreased the protein efficiency ratio (Fig. 1). The negative effect of un-ionised ammonia on the growth performance of tilapia increased with the exposure time (Fig. 2).

4. Discussion

Considerable information on the sensitivity of fish and aquatic animals to ammonia has been reported but not for particular feed like sewage-grown duckweed. El-Gohary et al.
(1995) studied fish production in sewage oxidation ponds and reported 100% mortality for silver carp, which was attributed to an un-ionised ammonia concentration of 0.41 mg N/l. In the current experiment, no mortality was detected at exposure levels up to 0.434 mg UIA-N l⁻¹. The low level of lethal ammonia toxicity reported by El-Gohary et al. (1995) was attributed to the high sensitivity of silver carp, since tilapia were stocked in the same pond with a calculated 0.52 mg UIA-N l⁻¹ for 92 days without mortality. Chronic toxicity of constant exogenous ammonia concentrations was studied in two different batches of turbot juveniles (Person-Le Ruyet et al., 1997a). Additionally, they reported maximal survival up to 0.33 mg UIA-N l⁻¹ while at 0.73 mg UIA-N l⁻¹ 50% mortality was observed. They estimated a 28-day exposure concentration that results in 50% of the SGR of the control between 0.6 and 0.75 mg UIA-N l⁻¹. In three batches of trout exposed to chronic ammonia toxicity, Person-Le Ruyet et al. (1997b) reported no mass mortality up to 0.4 mg UIA-N l⁻¹. In treated sewage-fed ponds, tilapia suffered clear skin ulcers, necrosis and haemorrhage associated with mass mortality at 0.45 mg UIA-N l⁻¹ (Nasr et al., 1998). On the other hand, no effects were observed on the growth or survival of fathead minnows exposed to 0.44 mg UIA-N l⁻¹ but clear negative effects on growth and survival were detected at 0.91 mg UIA-N l⁻¹ (Thurston et al., 1986) and the same was observed for striped bass (Hargreaves and Kucuk, 2001).

In fresh water crustaceans, *Eriocheir sinensis*, results (Zhao et al., 1997) showed that the tolerance to ammonia increased as the larvae developed to juveniles and decreased by increasing the exposure time, which is similar to our results (Fig. 2). Similarly, Smith and Piper (1975) reported that the growth performance of trout exposed to 0.033 mg UIA-N l⁻¹, showed no change in growth rate within 4 months, however, there was a significant reduction after 6 and 12 months.

The non-effect concentration is the concentration of toxicant that can be present in water without affecting growth, reproduction or survival over the full life cycle of the test organism. The no-observable effect concentration in this study is 0.068 mg UIA-N l⁻¹ and the proposed safe level to be lower than 0.1 mg UIA-N l⁻¹. This level is lower than the recorded value (0.18–0.33) for turbot juveniles (Person-Le Ruyet et al., 1997b). Szumski et al. (1982), suggested that a non-effect ammonia criterion of 0.08 mg UIA-N l⁻¹ could be applied to warm water fish. Similarly, Alderson (1979) reported threshold levels of un-ionised ammonia below which no effect on the growth performance could be observed as 0.066 mg UIA-N l⁻¹ for Dover sole and 0.11 mg UIA-N l⁻¹ for turbot. Zhao et al. (1997) proposed a safe level of UIA-N for *E. sinensis* juveniles of 0.09 mg UIA-N l⁻¹. Our results showed that the lowest-observable effect concentration on the growth performance is 0.144 mg UIA-N l⁻¹. This value is comparable to what has been reported (0.1–0.4 mg UIA-N l⁻¹) for turbot juveniles with a body weight range of 14–104 g (Person-Le Ruyet et al., 1997a,b). On contrary, for juveniles of gilthead seabream (*Sparus aurata*), Wajsbrot et al. (1993) proposed the maximum acceptable toxic concentration for growth between 0.27 and 0.47 mg UIA-N l⁻¹. The growth rate of juvenile channel catfish was reduced when exposed to ammonia that ranged from 0.048 to 0.989 mg UIA-N l⁻¹ (Colt and Tchobalongoelous, 1978).

The presence of ammonia suppressed feed utilisation and increased water content of the body and urea excretion rate in turbot juveniles (Person-Le Ruyet et al., 1997a). Also, Colt and Tchobalongoelous (1978) showed an increase in the water content of channel catfish
exposed to a range of 0.048–0.989 mg UIA-N l\(^{-1}\). During this experiment, no reduction in feed intake has been recorded in ammonia exposed fish up to 0.434 mg UIA-N l\(^{-1}\). Person-Le Ruyet et al. (1997b), on the other hand reported that the first response to ammonia exposure was a change in turbot appetite, which recovered within one week, but full recovery of appetite was limited to 0.4 mg UIA-N l\(^{-1}\). Other mechanisms rather than reduction in feed intake could be the reason of the reduction in the growth rate. Histopathological and haematological effects, particularly those affecting gills and liver function may contribute to reduce fish growth through inducing tissue hypoxia (Wajsbrot et al., 1993). Bucher and Hofer (1993) observed a tissue alteration in the liver and kidney of brown trout reared in a mixture of ground water and treated sewage with an average un-ionised ammonia concentration of 0.13 mg UIA-N l\(^{-1}\). Thurston et al. (1984) find similar tissue alterations in gills, kidney and liver in trout exposed to 0.067 and 0.076 mg UIA-N l\(^{-1}\). Nasr et al. (1998) reported that exposure to 0.33 mg UIA-N l\(^{-1}\) induced gill damage and subsequent liver tissue hypoxia associated with blood anaemia. Also, significant increase in the plasma glucose was observed in Atlantic salmon exposed to a range of 0.014–0.08 mg UIA-N l\(^{-1}\) (Fivelstad et al., 1995). This increase was proposed to be due to enzymatic stimulation of glycolysis.

Hargreaves and Kucuk (2001) proposed that the digestibility of dietary protein and the energy source might have been affected by un-ionised ammonia. The biochemistry of energy derivation from fats, carbohydrate and especially from protein is compromised by the presence of ammonia. Additionally, ammonia detoxification by fish is energy dependent and can result in a 68% reduction in the normal rate of energy production (Zieve, 1966). The effects of ammonia on energy utilisation may explain growth suppression in fish exposed to ammonia. Other mechanisms may contribute to growth reduction caused by ammonia exposure. Liver glycogen depletion and consequent blood acidosis (Sousa and Meade, 1977; Chetty et al., 1980) may contribute to increased susceptibility to hypoxia.

5. Conclusions

The conclusion of this study is that un-ionised ammonia concentration should be kept lower than 0.1 mg UIA-N l\(^{-1}\) in tilapia ponds fed with fresh duckweed grown on sewage. There was no fish mortality at 0.144, 0.262 and 0.434 UIA-N l\(^{-1}\) while the growth rate was negatively affected at these levels.

Acknowledgements

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