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Effect of tank background colour on the hatchability of *O. niloticus* eggs and survival of fry

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Abstract
This study was conducted at University of Eldoret, Fisheries and Aquatic Science Department hatchery to investigate the effects of tank background colour on the hatching and survival performance of Nile tilapia (*Oreochromis niloticus*) fry in glass tanks. The experimental set up consisted of four treatments of blue, green, red and colourless tank backgrounds where 55 fertilized *Oreochromis niloticus* eggs were obtained and incubated. Tanks were closely monitored, cleaned with controlled feeding once the fish were at the swim up fry stage. The best hatching and survival performance was achieved in the blue background tank. Percentage hatching of the fish was not significantly affected ($p > 0.05$) by the tank background colour with fish reared in blue background showing the highest percent hatching (93.62±6.06). Survival was significantly higher ($p < 0.05$) by the tank that had background colour with highest survival in the blue tanks (86.36±8.49). The results from the present study clearly suggest that *O. niloticus* fry should be reared in blue backgrounds and the use of red tanks should be avoided.

Keywords: hatchability, survival, tank background colour.

1. Introduction
1.1 Background of study
Tilapia is the second species after carp that is most cultured in the world (Head & Malison, 2000) [14]. This is because its hardiness which makes it resistant to diseases and infection with an enhanced ability to adapt and live in most of the environments (Karakatsouli *et al.*, 2010) [21]. Apart from that the fish is preferred because of its ability to reproduce in captivity just as it does in the natural environment (Chinen *et al.*, 2005) [8]. Because of these factors the fish has significant economic importance as a source of food (FAO, 2006) [11] for the and also a source of capital as well for the farmers (Neumeyer, 1992) [30]. However a challenge to tilapia culture in artificial environments is the prolific breeding nature of the fish which makes it critical as the farmer is supposed to monitor the breeding of the fish (Brummet, 1995) [6]. If not monitored the end result is that the fish focuses more of the breeding and the end result is stunted growth which affects the quality of the fish in terms of flesh and the market value of the fish (Hinshaw, 1986) [17]. Fingerling production and availability of quality fish feeds have been bottlenecks towards aquaculture (Rad *et al.*, 2006) [35]. This has necessitated the adoption of private sector fingerling production which has hence significantly increased owing to the rising demand for the fingerlings by the farmers (Bromage *et al.*, 2001) [5]. The cultivation of economically important species such as tilapia and catfish has been helped greatly by the growing use of artificial fertilization and incubation (Endal *et al.*, 2000) [10]. Artificial spawning and fertilization of many species has been aided with the development of aquaculture techniques that have slowly been developing over the years (Bouef & Le Bai, 1999) [3].
Aquaculture development entails the development of hatchery techniques such as artificial propagation. This is achieved through the manipulation of the fish environment, water and the biological features to improve the productivity and profits (Volpato *et al.*, 2004) [47]. Artificial incubation of fish eggs has greatly helped in terms of allowing for the manipulation of the conditions such as the water conditions like temperature, salinity and light to accelerate the hatching, and survival of the eggs and larvae subsequently (Rad *et al.*, 2006) [35]. In considering this light has also been a critical component that has been considered as one of the components that can be manipulated to improved performance of Nile tilapia (FAO, 2007) [12].

1.2 Problem statement

In aquaculture, the price of fish is determined by the market demand and supply (Primavera *et al.*, 1992) [34] which depends largely on growth and survival of the fish. Therefore the factors that influence the production of fish need to be considered in any culture system. Although *O. niloticus* culture has effectively been done in tanks, the tank background colours for larval rearing of *O. niloticus* has not always been considered in terms of their effect on egg hatchability (Scott *et al.*, 1984) [40]. To achieve high hatchability rates and survival of tilapia fry especially in tank larviculture, optimization of the environmental conditions is necessary (Stanley *et al.*, 2003) [42]. This entails the determination of the best contrast between light and tank walls since light and background colour have an effect on the hatching of the *O. niloticus* eggs.

Several fish species prefer dark tank walls (Luchiari & Pirhonen, 2008) [24] as they promote a suitable breeding grounds while others prefer lighter or rather coloured backgrounds (Karakatsouli *et al*., 2007) [20]. While a number of studies have examined biological condition such as growth, survival and production of *O. niloticus* under different light colours in tanks (Britz & Pienaar, 1992) [4], little published information exists on the relationship between tank background colour on the hatchability of eggs and survival of *O. niloticus* fry. The purpose of this study is therefore to investigate the effect of tank background colour on the hatchability performance of *O. niloticus* eggs and survival of *O. niloticus* fry reared in static water conditions of tanks.

1.3. Objectives of the study

1.3.1. General objective

To investigate the effects tank background colour on the performance of *O. niloticus e*ggs.

1.3.2. Specific objectives

i. To determine the effects of tank background colour on the hatchability of *O. niloticus* eggs.
ii. To assess the effects of tank background colour on the survival of *O. niloticus* fry.

1.4 Research hypothesis

i. **HO:** Tank background colour has no effect on the hatchability of *O. niloticus* eggs.
ii. **HO:** Tank background colour has no effect on the survival *O. niloticus* eggs.

1.5 Justification of study

The production success of *O. niloticus* larvae on any culturing technique depends on the provision of suitable food, stocking density, suitable rearing environment and the acquisition of the food given (Rad *et al.*, 2006) [35]. In semi-intensive and intensive aquaculture, the fingerling availability is an important factor that determines the maximum productivity and viability of the farming systems. However, there is not much has been done on the effects of tank background colour on the hatchability of *O. niloticus* eggs and survival of *O. niloticus* fry (Volpato *et al.*, 2004) [47]. The results of this research will therefore be important to aquaculturists in both semi intensive and intensive systems in improving production of adequate quality fingerlings required for commercial farming of *O. niloticus* in Kenya and other developing countries. It will also help farmers improve the hatchability of *O. niloticus* eggs and survival rate of *O. niloticus* fry.

2. Literature Review

With the rise in population and continued depleted natural resources as a result of overexploitation the cost of living is rising exponentially and so are the rates of hunger and poverty which have surged to very high levels (Stanley *et al.*, 2003) [42]. As a result of this factors cost and demand for food especially the dietary fish protein has also risen considerably. As a result agriculture and science have been coming up with better ways of accelerating productivity to meet the population needs (Hogsetle *et al.*, 1999) [18]. In fisheries aquaculture has been the solution in the form of aquaculture. Despite use adoption of aquaculture as the remedy, it has not adequately done much because it is mainly done on a subsistence level and is facing quite a number of challenges which discourage other farmers from pursuing it on large scale (Ostrowski, 1989) [32]. However it is undisputed that the intensive aquaculture activities with major focus on the culture of local species using simple, readily available low cost technology has been a major step in an effort to overcome the challenges of production (Ruchin, 2004) [37].

Oreochromis niloticus fish is amongst the fish that are mostly cultured fish which are indigenous to Africa (FAO, 2008) [13]. It is most preferred by many farmer because of its special attributes especially its hardy nature which makes it unsusceptible to diseases due (Marchesan *et al.*, 2005) [26]. Other traits are its diverse feeding habits, they can feed on a very large variety of foods, both natural and supplemental feeds and their ability to stay in water with poor conditions because of its ability to tolerate low pH levels and low oxygen levels (Kusmic & Gualtieri, 2000) [22]. It has been a principal cultured species in Kenya for very many years mostly used to generate income and provide food for subsistence farmers (Papoutsoglou *et al*., 2000) [49].

2.1 Tank Background Colour effect on Hatching of *O. niloticus* eggs

Hatching is the mechanical and enzymatic process, of breaking of the egg shell (chorion) and release of larvae (Hogsetle, 1999) [18]. Compared to hatching in stagnant waters, hatching in running water is retarded due to washing out of the hatching enzymes (Stanley, 2003) [42]. Therefore, the technique of incubation in stagnant water is slightly preferable. Survival of fry is the determination of the number of fry that remain after completing their yolk sac after hatching (Chinen *et al.*, 2005) [8]. The main composition characteristic of light is its spectral composition. When the light hits water, light rays pass to different depths depending on their wavelengths, absorption and diffusion and also other factors like the water properties, components and the organisms that exist in the water. This has necessitated most fish species to develop good colour sight and sensitivity to colour and coloured light (Volpato and Barreto, 2001) [46]. Light in its many forms such as light intensity, light colour and photoperiod are known to affect fish life cycle. All this aspects of light affect the fish at least one of its different stages of development and life though most are crucial through the entire fish's life cycle (Papoutsoglou *et al*., 2005) [48].

UV light has been used in hatching of eggs because of its ability to destroy the bacteria on the surfaces of the egg shells (Stanley, 2003) [42]. Naturally provided by sunshine, UV light can also be obtained artificially at 254nm and at this level it can kill most of the microorganism ranging from moulds, yeast, bacteria, viruses and fungi (Hogsetle, 1999) [18]. Eggs which have been exposed to UV light treatment have been known to have very low numbers of aerobic microbial counts (Hogsetle, 1999) [18]. Under UV illumination, the cuticle emits an orange- red fluorescence, which can be evaluated based on its presence, color, intensity and distribution of the fluorescence. UV light can be used to identify and remove eggs that have shell surface defects not visible under white light (Stanley, 2003) [42].

UV light can prevent the unnecessary use of formaldehyde on eggs assumed to be clean, especially the harmful effects of formaldehyde, which is widely used today, on embryo will be prevented (Barahona-Fernandes, 1979) [2]. Hence therefore the florescent light which provides UV light can be used to reduce the cases of egg spoilage thereby enhancing their hatchability (Stanley, 2003) [42]. Ultraviolet light application is a fast and easy method that is used at the evaluation of the cuticle characteristics of eggs large in number without damaging the embryo. It helps to evaluate the cuticle characteristics of eggs large in number before they are put in the hatchery. (Stanley, 2003) [42]. According to a study done by Primavera *et al.* (1992) [34], on the effect of light color on maturation in intact and eyestalk-ablated Penaeus monodon using green, blue and white fluorescent lights, with natural light as a control discovered unablated females under green light produced the highest number of spawns (5), total number of eggs.

2.2 Survival of *O. niloticus* fry

According to Strand *et al.* (2007) [43] it is important to be able to understand the performance and behavior of O. niloticus to be able to optimize the cultivation and production. This is because much has been done on the fish in its natural environment, however the natural environment differs from the artificial environment and this may affect the performance of the fish in the different aspects such as its health, feeding, growth. Environmental colour is one of the factors present in the artificial environment that may affect the performance of the fish (Volpato *et al.*, 2001) [46]. Light colour and intensity affect the fish's ability to detect its food, the Food Conversion Ratio (FCR), as well as the feeding success (Levine &MacNichol, 1982) [23]. These factors directly impact on fish performance like reproduction, growth and survival (Henne & Watanabe, 2003) [15, 16].

Rotland *et al.* (2003) did a study that concluded light intensity and the tank background colour maybe agents that induce stress to the fish which in turn may in turn affect the the fish aggressiveness (Papoutsoglou *et al.*, 2005) [48], their activities (Mesa & Shreck, 1989) [29] and swim performances as well as their utilization of the habitats available to them (Shreck *et al.*, 1997). Levine & Macnichol (1982) [23] noted that fish will behave and perform better in colour like blue, green and infrared which are the common colours in their surrounding natural environments while Nicol (1963) [31] observed that the fish will repond selectively to these colours.

Although not much has been done to comprehend the effects of tank background colour on the biology of fish, it is certain that these tanks background colours affect the fish in terms of its colour attractiveness, its reaction to fright, reproduction, growth as well as survival Levine & MacnNichol (1982) [23]. (Tamazout *et al.*, 2000). Owing to the wider range of colours that are available in the environment, fish will respond discriminately and differently to the different environmental lights (Appelbaum & Kamler, 2000) [1] and hence the need to investigate effects of different tank colours on the hatching of eggs and survival of tilapia fry (Rad *et al.*, 2006) [35].

Rad *et al.*, 2006 [35] showed a significant difference only during the fingerling stage. Fish are very sensitive to light, and although thresholds needed for reproduction and maturation are not yet known (Bromage *et al.*, 2001) [5], in general it has been found that high intensity light levels are required for growth optimization (Boeuf and Le Bail, 1999). In most studies fluorescent lamps are used for light related experiments, resulting in what humans perceive as white light but this may not necessarily be the case because in natural fish habitat, wavelength of light penetrating water varies greatly, fish vision and spectrum perception are strongly adapted to each species natural habitat and living ethology (Head & Malison, 2000) [14]. Downing *et al.* (2000) [9] noted that the survival rate of haddock larvae *(Mellanogrammus aeglefinus L.)* is higher environments with blue and green light. Green light accelerates the growth rate of silver carp larvae *(Hypophthalmichthys molitrix Val.)* and young carp *(Cyprinus carpio L.)* fish species (Ruchin 2004) [37]. Tank colour also affects the success of larval swim bladder inflation (Martin-Robichaud & Peterson, 1998) [27, 28]. Background colour effects on a fish's life are species-dependent phenomenon which may be related to specific habitat characteristics (Volpato and Barreto, 2001) [46]. Tank background colour has also been found to affect skin pigmentation (Karakatsouli *et al.*, 2007) [20], reproduction and nest building behaviour (Volpato *et al.*, 2004) [47]. Survival and growth of fish culture under different background colours have been studied in many fish species and background colour was found to have a degree of influence egg hatchability as well as survival rate of fish fry.

Staffan (2004) [41] also noted that in the green and the blue coloured tanks the fish moved more freely because they were more free and comfortable in these environments. In an experiment done by Mairesse *et al.* (2005) [25] using the sea horse fish, the fish that were kept in the dark tanks (black coloured tanks) were found to be more pale on the skin and less active an indication that the different light colours and intensity directly or indirectly the survival and colour changes of the juvenile sea horses and other fish. Skin pigmentation of the fish is affected by the effect of light on the sympathetic nervous system. According to Van der Salm *et al.* (2005) [45], a fish especially under stress induced by colour is affected and regulated by the Melanophore Stimulating Hormone (MSH). This was also observed by Strand *et al.* (2007) [43] also observed the same as he noticed differences in the skin pigmentation of the fish reared under the black and the yellow tanks.

$$\text{Hatchability} = \frac{\text{Number of eggs hatched (Fry)}}{\text{Number of eggs incubated (fertilized)}}$$

$$\text{\%Egg Hatching} = \frac{\text{Number of whitish broken eggs x 100}}{\text{Number of eggs fertilized}}$$

$$\text{Survival} = \frac{\text{Initial Number of Fry (with Yolk after hatching)}}{\text{Final Number of Fry (After Yolk Disappears)}}$$

3. Materials and Methods

3.1 Study Area

The experiment was carried out at the research laboratory of the Department of Fisheries Management, University of Eldoret, Uasin Gishu County, Kenya. The Fisheries hatchery of University of Eldoret is situated along Eldoret- Iten- ziwa road, 9km from Eldoret town. Eldoret is situated at 0^0 35'N and 35^0 12' E and its altitude is 2140m above the sea level. It has an average temperature range of between 17^0C and 24^0C and an annual rainfall of 1100mm p.a.

3.2 Source of Eggs
Ten Brooders with mean weight of 500g were obtained from the fish farm of University of Eldoret. The female brooders obtained from the farm were checked for presence of fertilized eggs in their mouth. The eggs were then extracted and preserved in formalin during transportation to the hatchery where they were be incubated in the different experimental set ups.

3.3 Experimental Set up and Incubation of the eggs
The set up consisted of two replicates of the four experimental units which are the red, blue, green and the transparent with no colour on it acting as the control in the experiment. In each 55 fertilized tilapia eggs were incubated in tanks. Eggs were being placed gently in the water and were to be monitored for the spoilt eggs which were separated from the unspoiled eggs. These eggs were then left to hatch and then separation of hatched larvae from the unhatched eggs was done and the numbers noted. The hatched larvae were also being observed till the entire York sac was used up and the survival rates for each experimental set up recorded.

3.4 Statistical analysis
Data on hatchability of *O. niloticus* eggs and survival of *O. niloticus* fry generated was subjected to a non- parametric test, Mann Whitney for comparison of medians of the hatchability and survival. All that was done by using Minitab 14 statistical software.

4. Results
4.1Hatchability of *O. niloticus* Eggs
The fertilized eggs that were incubated for hatching were observed to remove spoiled eggs so that they don't contaminate the other viable eggs. In terms of the spoiled eggs per treatment the average spoilage did not have any significant difference ($p > 0.05$) in the different tanks with the recording the lowest 8±0.71 eggs in the blue tank and the highest being 10±0.71 eggs in the control tanks and 9±0.00 eggs in the red and the green tanks. Hence the eggs available for hatching were between 85% (47±0.71 eggs) for the blue tank, 84% (46±0.00 eggs) for the red and the green tanks while the least was in the control that stood at 82% (45±0.71 eggs).

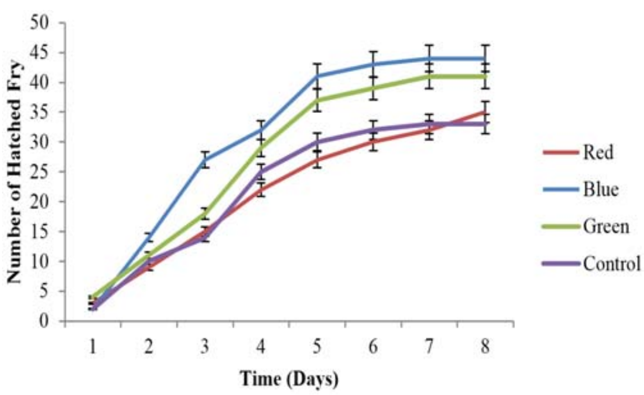

Fig 1: Trend of hatching of the fertilized O. niloticus eggs

From Figure 1 above hatching trends were rising as the days progressed. Initially the hatching was high depicted by the steepness of the graphs. By the fifth day the hatching had stabilized and by the eighth day there was no more hatching. Blue Tank recorded the highest hatching of 93.62±6.05 % (44±4.07 eggs), followed by green tank that recorded hatchability of 89.13±4.87% (41±1.95 eggs), the other two tanks recorded hatchability of 74.47±5.49% (35±2.30 eggs) for the red tank and 71.74±7.42% (23±4.07)for the control tank. However there was no significant difference ($p > 0.05$) in the hatching of the O. niloticus eggs.

4.2 Survival of fry

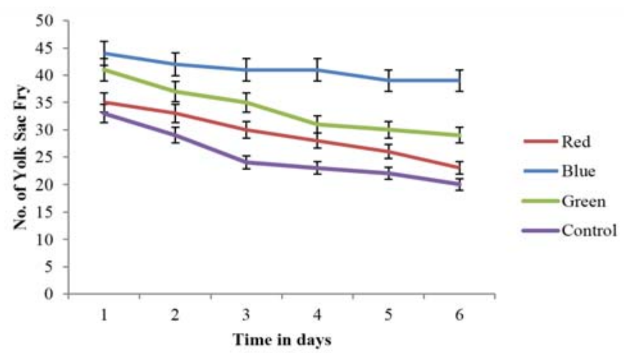

Fig 2: Graph of trend of survival of Yolk Sac Fry reared in glass tanks of different background colours

From the trends in Figure 2 it is evident that the survival of the yolk sac fry in the different tanks varied. The survival rates were significant ($p < 0.05$). Slope of the blue tank was the least steep indicating the lowest mortalities at with survival of 88.64±6.76%. The slope of the green tank is relatively steeper than the blue tanks with survival at 80.49±1.00%. Mortalities in the red and control tanks were slightly higher with survival of 71.43±5.41% and 75.75±2.35% respectively.

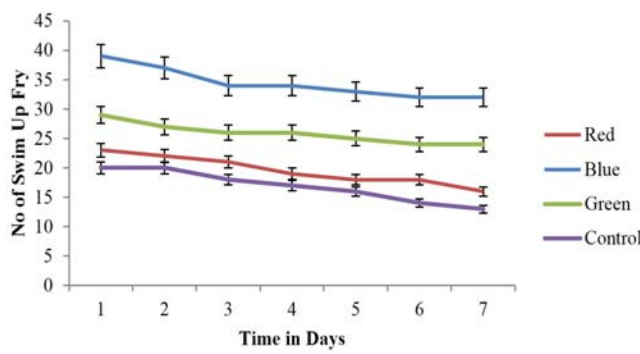

Fig 3: Graph of trend of survival of Swim up Fry reared in glass tanks of different background colours

Trend of the survival of swim up fry as depicted by Figure 3 gives slopes that are gentle, almost flat indicating very low mortalities at the development of the fry. Mortalities of the swim up fry are least with most of the swim up fry surviving to the fingerling stage.

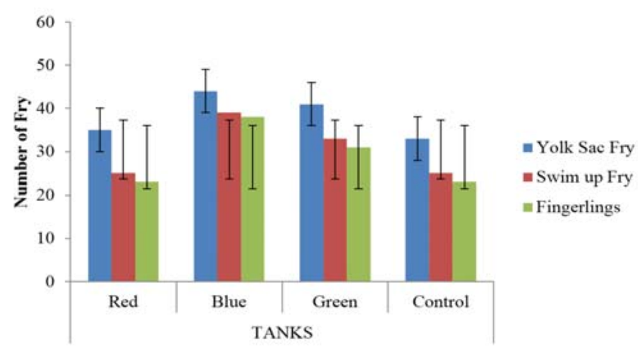

Fig 4: Comparison of the survivals at the different fry stages of fry reared in glass tanks with different background colours

In comparing the losses between the different treatments as highlighted in Figure 4, the highest losses were recorded in the red and the control tanks respectively. However notable in all the tanks is that the losses are significant ($p < 0.05$) in the yolk sac fry stage compared to the swim up fry stage. Losses of the swim up fry stage are almost equal in all the treatments and insignificant ($p > 0.05$) although the same cannot be reported for the yolk sac fry which has significant differences between the different tanks.

5. Discussion

Blue environmental color influenced fish reproductive behavior, although the mechanisms involved are not completely understood. The higher hatchability behavior observed in groups under blue and green light strongly suggests that this light stimulates reproduction in Nile tilapia. Since white light is the light background (fluorescent light) for laboratory studies all around the world, we used this as the control condition. Thus, under the conditions used hatchability was not expected and the higher frequency of reproduction in the blue, green and red light environment might represent improvement in reproduction (Jentoft *et al.*, 2006) [19].

A study done by Volpato and Barreto (2001) [46] on the effect of light color on stress response in *O. niloticus* species showed that blue light prevents an increase in cortisol during stress. This effect and the known suppressive effect of stress on reproduction support the notion that blue environmental color might improve reproduction and hatchability in this species. White color is the usual illumination (fluorescent light) in laboratory studies hence used as a control color in the analysis of the effects of blue, green and red lights on reproduction in Nile tilapia.

The mechanisms of the effect of color on hatchability are still unknown. However, stress may be involved because it is a process that suppresses reproduction and that is prevented by blue environmental color in Nile tilapia.

Survival is high in the blue and green tanks compared to the other tanks. Results reported by Volpato *et al.* (2004) [47] who indicated that color may affect different biological systems. Fish demonstrate a preference for blue and green color as they represent most closely their natural habitat (Levine & MacNichol, 1982) [23] this explains why they were notably less aggressive in the blue color groups. The aggressiveness observed in red adapted fish against each other was mostly a reflection of the stress induced by red color. In a similar experiment by Staffan (2004) [41], the fish moved freely in tanks with green and blue background, but they did not seem to show similar preference to other colors. Blue color exposure causes least significant change in the enzyme activity hence enhances the survival of the fry. However the results suggest that blue light may prevent the increase of stress-induced Acetylecholinesterase (that accelerates physiological activity) in the Nile tilapia which enhances the fry survival while the red color may increase stress induced Acetylecholinesterase hence the high mortalities (Sabri *et al.*, 2012) [38].

6. Conclusion

The results show that blue color lightening or background in aquaculture techniques can be used to maximize the growth rates of juvenile tilapia, reduce their stressful behaviors, and minimize their mortality to the lowest rate. This application can therefore be used to improve the culture performance of tilapia, and increase the productivity of fish in the aquaculture industry. On the contrary, exposing fish to red color and darkness does worsen the growth conditions and fish productivity and should be avoided in order to increase aquaculture fish output.

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