

Effects of Different Substrates on Growth and Survival of Labeo victorianus (Pisces: Cyprinidae, Boulenger 1901) Fry towards Its **Conservation along the Mara Basin**

Paul Sagwe Orina¹, Lucy Wangari Ikanya², Robin Abell³, Leonard Akwany³, Mercy Chepkirui¹, Rasowo Joseph⁴

¹Kenya Marine & Fisheries Research Institute (KMFRI), Kegati Aquaculture Centre, Kisii, Kenya ²Faculty of Agriculture, Department of Animal Sciences, Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya

³Conservation International, 2011 Crystal Drive, Arlington, USA

⁴Technical University of Mombasa, Mombasa, Kenya

Email: paulorina@gmail.com

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Abstract

The study evaluated the role of different substrates on water quality, growth and survival of Labeo victorianus fry. Random sampling was used to select 225 fry, stocked at 100/m³ and fed on shell free artemia[®] (40% - 60% crude protein). Each treatment was carried out in triplicates using gravel and sand as substrates and water media without substrate (control). Growth and survival of the fry was monitored over 4 weeks period at a hatchery set-up. Physicochemical parameters were measured weekly while mortality was documented daily. Substrate type had an effect on water quality, growth and survival of fry. Dissolved Oxygen, growth and survival was higher in tanks under gravel substrate than sand and control while mortality was higher (22%) for sand ($p \le 0.05$) than gravel and sand treatments. Fry in gravel substrate had the best growth response (1.77 \pm 0.92 g) while control and sand recorded 1.57 \pm 0.90 g and 1.45 \pm 0.97 g respectively. In contrast, growth was homogenous during the first 2 weeks of the trial but gravel substrate fry significantly out-grew sand and control in the remaining 3 weeks. However, fry in sand and control experienced compensatory growth towards the end of the study. Therefore, gravel substrate enhances L. victorianus performance and is recommended in the introduction of the species into aquaculture and restocking back to the wild.

Keywords

Aquaculture, Water Quality, Fry, Treatment

1. Introduction

The Mara river basin covers a surface area of $13,325 \text{ km}^2$, with approximately 65 and 35 percent located in Kenya and Tanzania respectively. From its source in the Mau Escarpment in Kenya, the river flows for about 400 km and drains into Lake Victoria, the second largest freshwater lake in the world [1]. The basin forms part of the most important river ecosystems in East Africa traversing through the wildlife rich terrestrial Maasai Mara and Serengeti ecosystems recently declared the 8th wonder of the world informed by the unique wildebeest migration. Despite this global and regional significance, the river remains threatened by destruction of its source forest cover (Mau Forest), poor farming practices, high population growth rate and climate change [2]. Consequently, the seasonally fluctuating Mara river water levels have had far reaching aquatic ecological effects including fish distribution and biodiversity. Floods have become more common and large parts of the Tanzanian's Mara wetlands have become more permanent instead of temporary wetlands [3]. These anthropogenic and environmental changes have affected the Mara river ecosystem service and significantly contributed to experienced decline of commercially important indigenous species such as Labeo victorianus.

L. victorianus (Pisces: Cyprinid) locally known as Ningu, is the only labine species in Lake Victoria and its catchment in Kenya [4]. It is an omnivore, consuming predominantly mixed diets of detritus consisting mainly of plant material [5] [6]. It is known to occupy shallow inshore waters and affluent rivers of L. Victoria. It was previously the most popular indigenous species in Lake Victoria until late 1950s when the population drastically declined to near absence at fish landing areas [7]. L. victorianus spend most of their life span in the open lake, but migrates upstream to spawn in flooded vegetated areas beside both permanent and temporary rivers and streams [8]. This predictable upstream migratory habits of breeding populations coupled with the fish's delicacy have over time contributed to intensified application of fish traps by local communities along the rivers resulting in a considerable decline of the fish [7] [9] just like the native tilapiine fishes (Oreochromis esculentus and Oreochromis variabilis) which suffered a decline from early 1970s to uncontrolled fishing effort and selective fishing [10] [11]. L. victorianus population decline in the L. Victoria basin has been closely associated with Nile perch (Lates niloticus) predatory behavior, overfishing, inter-species competition especially by introduced Nile tilapia (Oreochromis niloticus), eutrophication and the use of illegal fishing gears [12] [13]. Currently L. victorianus is on the International Union for Conservation of Nature (IUCN) 2018 red list of endangered fish species for L. Victoria [14]. Recent efforts to conserve the species have included transportation, captive breeding and culture [15] [16].

Captive breeding has been cited as the most immediate solution to reducing fishing pressure and ensuring recovery of wild stock [17] [18]. Breeding and feeding experiments have demonstrated promising species conservation results

[9]. L. victorianus has high aquaculture potential having demonstrated ability to cope with captive conditions, artificial feed acceptability, artificial and semiartificial spawning with appreciable growth rates [15] [16] [19]. Globally, aquaria fish rearing is the most popular comprising of large and diverse freshwater fish, marine fish, corals and macroinvertebrates [20]. However, this is more on ornamental marine and freshwater fish as opposed to food fish which is high grown in ponds and cages. Substrate covers the bottom of aquarium and acts as the host for beneficial bacteria and serves as a good source of natural nutrients, enhances water pH and increases the buffering capacity of water as well as shelter. They also help in filtration, mimicking of the natural environment and enhancing anesthetic appearance of the aquarium. Among the substrates commonly used include pebbles, soil, gravel, sand, aragonite, stones, vermiculate, crushed corals, marbles and clay [21]. The choice of the substrate depends on the culture unit size, water quality effects and associated fish species behavior. However, although studies have been done on L. victorianus demonstrating its natural environment substrate preference [22], limited studies exist on the effects of different substrates used in captivity. Therefore, this study attempted to assess the effects different substrates on growth performance, water quality and survival of L. victorianus fry. Further to this, the study explored the possibility of reviving wild populations through wild broodstock breeding and restocking putting into consideration past experiences [23].

2. Materials and Methods

Wild L. victorianus broodstock were caught from Mara Rive (01°03'00"S, 035°14'00"E) located in the Southern Rift Valley in Kenya (Figure 1) using an electro fisher, sexed and only gravid females and ripe males retained for ex-situ management according to [15]. They were packed in 3 L capacity polythene bags filled with river water and ammonia toxicity managed using sodium chloride at 2 g/l [24]. The fish carrying bags with a third air allowance area were aerated using oxygen meter to 100% saturation ready for transportation according to [15]. On arrival at KMFRI Sagana aquaculture Center, they were conditioned and eventually stocked in holding tanks containing clear river Ragati inflow water. The broodstock was acclimatized for three days and were fed using KMFRI formulated feeds (30% crude protein) at 3% body weight [25]. Each gravid female was induced to accelerate egg maturation and eventual spawning by injecting Ovaprim® 0.2 mL/Kg of fish followed by pairing at a ratio of 2 males to 1 female in a glass aquarium at a regulated temperature of 28°C. Broodstock were removed from the aquarium after 12 hours upon spawning and egg fertilization. Hatched larvae were monitored daily until they absorbed all the yolk and started exogenous feeding at day 4. A 4 weeks trial using complete random design with three replicates aquarium containing gravel, sand and water (control) as substrates were used. A total of 225 fry with average body weight of 0.28 \pm 1 g were used at a stocking density of 25 fingerling/20L aquarium. Feeding was done

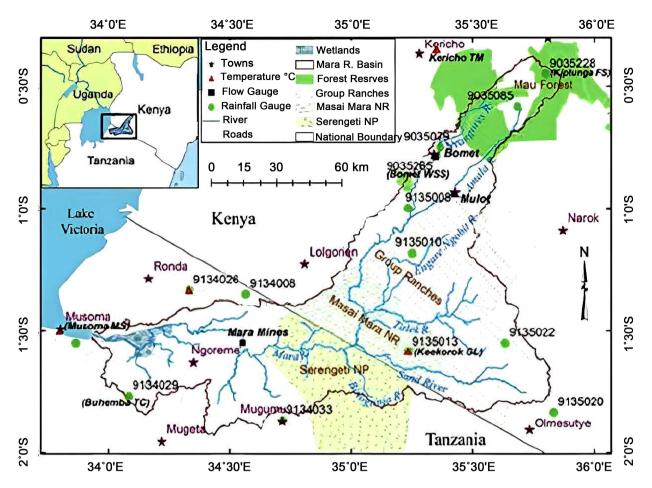


Figure 1. Mara river location, major land use and distribution (Source: [26]).

three times a day (9.00 am, 1.00 pm and 5.00 pm) with equal amount of artemia^{*} *ad libitum.* The water in the aquarium was exchanged once every week. This was done by siphoning out 50% of the water and filling with equal volume (all with same temperature). Weight and length were measured bi-weekly using electronic balance (readability 0.0001 mg) and a transparent measuring board to the nearest 0.1 mm respectively. Water quality parameters were measured weekly using Hanna Multi-parameter HI 9829 while mortality was recorded daily before feeding.

Data Analyses

Data collected was keyed in an Ms Excel spread sheet on Windows 2019 and presented as mean \pm standard error. The data was then tested for normality using QQ plots followed by one-way analysis of variance (ANOVA) to compare means of water quality, growth performance and survival. Values with ($p \le 0.05$) were considered significant. In case of any significant difference, Tukey's HSD multiple comparisons test was done to evaluate specific differences in water quality, growth performance and survival among the treatments. All analyses were done using Statistical Package for Social Sciences (SPSS) package (Version 20, SPSS Inc., Chicago, USA).

3. Results and Discussion

3.1. Effects of Substrate on Water Quality and Survival

Water quality parameters were within the acceptable ranges for the growth of L. victorianus [27], though insignificant variations were observed during the first week of experiment. Temperature ranged between 22.34°C to 22.67°C with no significant difference (p > 0.05) in the three treatments whose results similar to previous studies [28] on Labeo rohita fingerlings cultured using different diets containing biofloc and commercial feed pellets. According to [27] L. victorianus thrived well in temperature range of between 22.34°C to 22.67°C a range within earlier findings (18.3 °C to 37.8 °C). Dissolved oxygen (DO) ranged between 5.91 mg/L and 7.49 mg/L, gravel substrate recorded the lowest DO a trend that was associated with algal build-up on the gravel substrate. Further, gravel acted as a home for beneficial bacteria thus respiration and decomposition of uneaten feed and fecal waste beneath gravel led to a lower DO value and vice versa for media without substrate (control). Generally, cyprinids can tolerate low DO of 3 mg/L [29]. Even though, the nature of gravel substrate allowed water to pass through freely hence it had sufficient DO to support L. victorianus. Water pH was alkaline ranging between 8.63 and 8.92 with no significant difference (p > 0.05) among treatments. This reflected good productivity; calcium presence and high capacity for cationic exchange [30] thus helping to maintain the alkalinity of water. According to [31] pH level of 6.7 to 9.5 is generally recommended for fish culture. Gravel substrate recorded high survival rate of up to 80%, while sand substrate unlike gravel and media without substrate (control) posted high (10%) mortality the first week of trial, resulting to 78% survival rate which is closely associated with the increase in salinity, total dissolved solids (TDS) and conductivity that affect oxygen-carrying capacity of water. Similar studies on Labeo rohita have documented high survival at different levels of sugarcane baggase substrate compared to control [32].

3.2. Effect of Different Substrates on Growth of Labeo victorianus Fry

The growth curve in animals is sigmoid in shape [33]. In the present study, fry reared under all substrates within aquaria set-up exhibited normal sigmoid growth pattern, however between week 2 and week 3 control and sand lagged behind on weight gain respectively compared to gravel. The continuous lagging behind of sand substrate fry may be associated with delay in coping with high TDS levels (Table 1). However, fry reared in water and sand substrate experienced compensatory growth towards the end of the study and indicator of *L. victorianus* ability to adjust at early development stages to prevailing environmental conditions. Previous studies on different substrate have indicated that organisms reared on water (control) have the lowest survival and growth performance whereas biodegradable substrate enhanced growth [34] [35] [36] [37]

Water quality parameters	Control	Gravel substrate	Sand substrate
Conductivity (ppm)	63.29 ± 0.01^{a}	73 ± 0.02^{b}	$94.42 \pm 0.02^{\circ}$
Salinity (psu)	$0.03\pm0.00^{\mathrm{a}}$	0.03 ± 0.00^{a}	0.03 ± 0.01^{a}
Dissolved Oxygen (Mg/l)	7.49 ± 0.01^{a}	5.91 ± 0.01^{b}	$6.9\pm0.05^{\mathrm{a}}$
Temperature (°C)	$22.67\pm0.10^{\rm a}$	22.45 ± 0.10^{a}	$22.34\pm0.10^{\rm a}$
Total Dissolved Solids (TDS)	$31.88\pm0.12^{\text{a}}$	$37 \pm 0.10^{\mathrm{b}}$	$47.67\pm0.13^{\rm c}$
pH	$8.92\pm0.01^{\text{a}}$	8.63 ± 0.01^{a}	8.69 ± 0.01^{a}
Survival	77%	80%	78%

Table 1. Water quality parameters throughout the experimental period.

Values are reported as mean \pm S.E.M. Means identified by different alphabetical letters in the rows were statistically significant (p < 0.05) as determined by analysis of variance and Turkey's comparison of mean values.

[38]. Substrates used with different crayfish species *Pacifactacus leniusculus* [39] *Cherax quadricarinatus* [40], *Cambarellus patzcuarensis* [41], *Procambarus clarkia* [42] reported that substrates mimicking natural habitat situation had the best performance effect. In this study, fry in gravel substrate demonstrated the best growth response recording (1.77 \pm 0.92 g), while control and sand recorded 1.57 \pm 0.90 g and 1.45 \pm 0.97 g respectively (**Figure 2**). This indicated that *L. victorianus* cultured in gravel substrates attained higher body weight since gravel mimicked the natural substrate at Mara basin river rocky breeding grounds.

The surface area of gravel in the tanks provided an additional surface area for the growth of algae and phytoplankton which act as natural food for fry [43]. Gravel also provided algae growth ground and shield from over-foraging, this thus provided room for algae recovery and sufficient supply to fry reared in gravel substrate thus their enhanced growth compared to sand and water substrate. Therefore, the growth of fish can be related to the abundance of natural food. Gravel also act as a home for beneficial bacteria [30] which helped in the breakdown of fecal waste and excess feed, turning them into nitrates (high salinity and total dissolved solids) that the algae fed on hence increased the energy and nutrient transfer efficiencies of the fry [44].

Intensified illegal, unreported and unregulated fishing in the Lake Victoria basin has contributed to diminishing fish stocks including *L. victorianus* among other indigenous fish species. Captive breeding and restocking has been reported as one of the ways to maintain genetic diversity and fitness within the population [45]. Across the globe, previous studies for various species have reported successful restocking including that of Nile tilapia (*Oreochromis niloticus*) in L. Naivasha, Kenya in 2011, Brown trout (*Salmon trutta* L.) in River Ume and Vindelalven, Sweden [46], and Salmonids in Latvia. This study helps in conservation and eventual curbing of depletion of *L. victorianus* in Mara basin ecosystem, home to several families of sedentary and migratory genetic material of *L. victorianus* and other indigenous species. Further the findings provide greater opportunity for culture of the species, expansion of cultured species, reduced

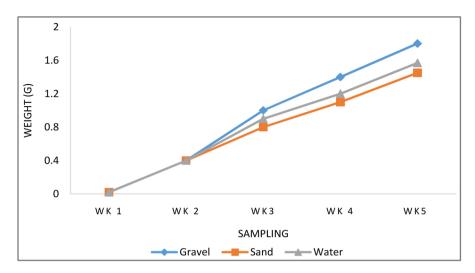


Figure 2. Mean weight of *L. victorianus* throughout the experimental period.

fishing pressure and enriched food security and nutrition to the fast growing human population enlightened on fish consumption health benefits.

3.3. Mara River Basin Recent Alteration

The basin activities have altered the two major rivers ecosystem and thus affecting L. victorianus potamodromatic behavior during breeding with eventual listing of the fish under the IUCN red list of endangered species (Figure 3 and Figure 4). The Mara catchment has in the recent past undergone significant alteration courtesy of deforestation of Mau forest, intensified land reclamation for livestock and crop framing, human settlement and intensification of hospitality industry activities (Figure 5). Mau forest has been reduced from 380,000 hectares to 285,000 hectares over the last 15 years. This has significantly affected the rains and in effect the prolonged low water levels for Mara and associated river basins. The long-term effects on the fishery and associated aquatic ecosystem services can never be underestimated. These effects are likely to be felt in all the 12 rivers emanating from the Mau Forest catchment with far reaching ecological distribution and diversity effects (Figure 6). The effects are likely to interfere with the documented breeding and aggregatory (01°13.425'S, 035°02.177'E; 01°12.794'S, 035°02.502'E; 01°13.266'S, 035°01.098'E) spots along the Mara river [9].

4. Conclusion and Recommendations

The successful captive propagation of *L. victorianus* in this study confirms previous studies' findings and thus provides a great window of opportunity to mass culture the species for introduction to aquaculture as well as restocking the wild to assist recovery of stocks. The study findings further confirm that in aquaculture production, a gravel lining of aquaria or any other fish rearing unit will create a favorable aquatic environment for optimal growth and survival of *L. victorianus* fry. The changes in water quality observed, although minimal, imply



Figure 3. Mara river anthropogenic land scape.

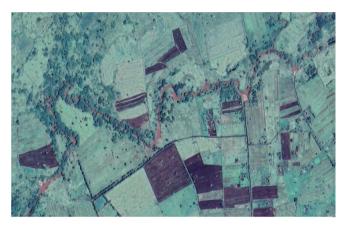


Figure 4. Migori river within Mara basin with agricultural activities intensification.

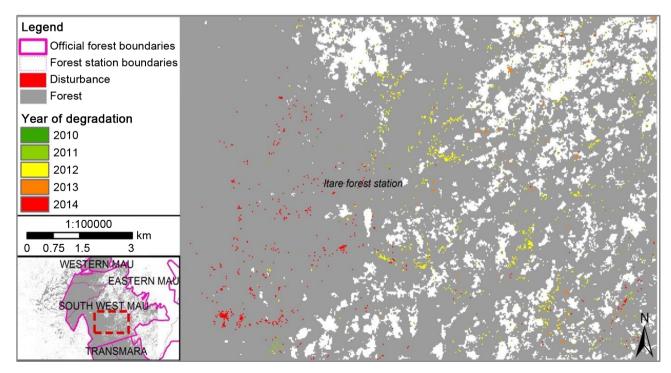


Figure 5. Mau degradation spots (Bewernick, 2016).



Figure 6. Low water levels in Mara river.

that for successful *L. victorianus* culture, fish farmers should apply high levels of water quality maintenance to improve growth performance and survival of *L. victorianus*. Furthermore, there is a need for policy on restocking and conservation program in Kenya and the Lake Victoria basin systems shared resources with neighboring countries to restore the population of *L. victorianus* in the ecosystem.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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