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INTRODUCTION OF AFRICAN CATFISH (*Clarias gariepinus*) IN AQUACULTURE SYSTEM OF PAKISTAN: ITS TRANSPORTATION, ACCLIMATIZATION AND CANNIBALISM STUDY

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To increase the production capacity of the existing inland aquaculture system of Pakistan, there is a dire need for the introduction of high-value fish species with growth potential at intensified stocking densities and having export potential. The African catfish having above mentioned promising culture characteristics was tried to interpolate the local habitat of Pakistan. For this purpose four thousand African catfish at the fry stage were airlifted from Thailand to Islamabad, Pakistan. To recover the fish from transportation stress, the fish was initially kept under intensive care in circular tanks and raceways for seven days. After recovery from the stress period the fish was shifted to earthen ponds and raceways for an acclimatization period of three months. During the acclimatization period fish showed better growth in earthen ponds as compared to raceways. Negligible mortality was recorded during the process of acclimatization in raceways and earthen ponds, but low survival rate was observed in raceways (30.4%) and earthen ponds (63.4 %). This lower survival rate in spite of very low natural mortality confirmed the phenomenon of cannibalism in this fish. To minimize the process of cannibalism, sized base sorting was done after 5 days and 7 days. A significantly higher survival rate was observed in sorted groups as compared to the non-sorted group. So it was concluded from this study that African catfish were successfully transported and acclimatized in local environmental conditions of Pakistan. However the phenomenon of cannibalism existed in this catfish species that was minimized through physical grading of fish based on its size.
Keywords: African catfish, acclimatization, earthen pond, mortality rate, cannibalism.

INTRODUCTION

The aquaculture system of Pakistan is mainly extensive and in some areas semi-intensive culture is also practiced at low stocking density of low-value fish species that ultimately give low per unit fish production, estimated to be 2.5 ton per ha. The culture system of Pakistan is mainly governed by indigenous and exotic carp species (Basavaraga *et al.*, 1999). To bring the expansion and improvement in the aquaculture system of Pakistan introduction of Chinese carps (including Silver carp, Grass carp, Bighead carp and Common carp) was done initially. Although carps have a wide range of acceptability among the consumers, a large segment of the populace does not prefer these fish due to a large number of intramuscular bones in them (Mirza and Bhatti, 1999). It is need of time to move towards intensive aquaculture at high stocking densities by introducing high-value fish species with high growth potential in aquaculture and export potential (Faheem *et al*., 2019; Mahmoud *et al*., 2019). The potential candidates are catfishes like African catfish, Channel catfish, Pangasius and Sea bass. The catfishes are gaining popularity in the culture system because of their high growth, consumer preference due to good quality flesh with very few spines and high export potential (Mingkang, 2005). One of the high-value fish species American channel catfish was successfully introduced in Pakistan in 2005 (Rab *et al.*, 2007).

African catfish (*Clarias gariepinus*) has recently attracted the attention of fish culturists all over the world due to its economic advantages. African catfish has the endurance to extensive range of surroundings with high temperature and less oxygen with air-breathing ability that makes it more malleable and resilient to stress (Olaleye, 2005). It is disease resistant, highly fecund and ease of their larval production in captivity declared it commercially very important (Kestemont *et al.,* 2007). The fish species have a better survival rate (Van der Waal, 1998). Even at high stocking densities, it is found more profitable in culture (Toko *et al.,* 2007). African catfish have been successfully acclimatized in the culture system of various countries and globally well known for its culture. Several methods of culture available for *C. gariepinus* farming include still culture in the pond; flowing pond culture, re-circulated pond culture and channel production (Fourie, 2006). Indoor hatcheries were used to rear *C. gariepinus* juveniles before transferring them to grow-out ponds which have trifling water circulation. A crop of 40,000 to 100,000 kg/hectare can be reared by only 25% daily water exchange in ponds (Rouhani, 2010).However, the high rate of cannibalism in African catfish reduces the survival of African catfish during its rearing period, which is recognized in many species of catfishes. Due to the above mentioned cultural characteristics it was planned to introduce this fish in the culture system of Pakistan. The first step for the introduction of new species in an environment is its acclimatization in the local environment. So this study was planned to check various aspects of adaptability and growth performance of African catfish in culture system of Pakistan.

MATERIALS AND METHODS

Transportation of African Catfish from Thailand and Its Acclimatization in Local Conditions:

Transportation, Un-packaging and Recovery**:** To start the aquaculture of the African catfish in Pakistan, the fish seed was imported from Thailand with the help of commercial importer. About 400 and 500 fish fry/bag were packed in a polythene bags filled with water and oxygen in ratio of 1:3.The polythene bags were packed in Styrofoam boxes, and two ploythene bags were kept in each Styrofoam box. The Styrofoam boxes were air lifted from Bangkok Thailand to Islamabad Pakistan. The duration of transportation was approximately 16 hours from Thailand to NARC, Islamabad Pakistan.

After transportation fish were unpacked at Aquaculture and Fisheries Program. Mortalities were recorded and water quality parameters including temperature, pH, dissolved oxygen (DO), carbon dioxide and ammonia was determined for both loading densities. Fish was initially stocked in raceways and circular tanks by slowly equalizing the water temperature of bag, circular tanks and raceways. Fish were kept in raceways for 7 days by feeding ad libitum at intensive aeration in a regular water flow throw system. Feed nutritional composition was determined as described in recent studies (Zhang *et al.*, 2015; Wang *et al.*, 2016; Niu *et al.*, 2017; Xia *et al.*, 2018a; Xia *et al.*, 2018b; Xia *et al.*, 2018c; Demir *et al.,* 2019; Raza *et al.,* 2019). Fish was offered with minced meat during this recovery period. Fish started to accept feeding after 3 days of transportation in powdered form at 35% CP level. Behavior was noted as described in recent studies (Muhammad *et al.*, 2016; Muhammad *et al.*, 2019) During this period mortality was recorded after every three hours of initial stocking on the first day, and then mortality was recorded on daily basis.

Acclimatization**:** For the acclimatization of fish in local environment, imported fish were kept in outside raceways with1500 gallons water flow through capacity and earthen ponds of size 0.04 ha in the extensive culture system. Fish were stocked at the rate of 100 fish per raceway and 1000 fish in each pond. No aeration was done during this period. The acclimatization process was continued for 90days. The feeding was done three times in a day ad-libitum. Fish was fed with locally prepared feed. For the measurement of length and weight, the fish (n=15) were captured randomly after 15 days by using hand nets and after taking required data fish were released back to their respective systems. During the process of acclimatization in raceways and ponds, the mortality of fish was recorded on daily a basis. After the acclimatization process total fish were collected for recording survival and total weight.

The growth performance was recorded in both earthen ponds and raceways by Daily weight gain, Specific growth rate (SGR) and Condition factor (C.F). For SGR and C.F, the following formulae were used.

Weight gain = Final weight-Initial weight

Survival rate (%) =

No. of fish stocked – No. of fish survived x100

SGR (%) = [(In Wf- In WI) x 100]/days

Condition factor (C.F) = $W100/L^3$

[Where W: fish weight (g), L: fish length (cm)]

Cannibalism Study**:** After acclimatization the fish, the survived fish appeared less than expected number of fish without any apparent mortality. This low fish survival was observed due to phenomenon of cannibalism. To minimize cannibalism, further investigation was done on physical grading method based upon their size for improving growth performance and survival of African catfish in raceways system. The experimental design was CRD having three treatments and three replicates. The duration of the study was three weeks. For this purpose, fish was divided into three groups.

i. Treatment 1 (T_1): Sorting interval 5 days
ii. Treatment 2 (T_2): Sorting interval 7 days
iii. Treatment 3 (T_O): without any sorting (Control group)

The 200 fish of almost same size were stocked in each raceway. Fish was graded by hand picking method. Fish were separated based upon their sizes. Fish was fed at ad libitum during the whole experimental period. Their survival and growth rate were recorded accordingly.

Water quality parameters i.e. temperature, dissolved oxygen (DO) and pH were recorded on daily basis with Limnological meter while alkalinity and hardness was measured by titration methods on weekly basis. Data was presented in the form of suitable graphics using MS excel. The significance among different parameters was checked by ANOVA (analysis of variance) and Duncan's Multiple Range Test (DMRT). The relationship among different parameters, were determined by correlation and simple regression

RESULTS AND DISCUSSION

Transportation of African Catfish from Thailand and its Acclimatization in Local Conditions

Transportation and Recovery after Unpacking: The introduction of any exotic fish species in the culture system of a country requires the successful transportation of live fish with great care and attention. In the present study about 4000 African catfish fry with an average weight of 0.200 g were transported by air from Bangkok, Thailand to Islamabad Pakistan. The total transit time from Bangkok to Islamabad was 16 hours. The fish bags were unpacked immediately after being transported at AFP, NARC Islamabad. All fish were healthy and no injured fish were seen. Mortalities and important water quality parameters were determined immediately after unpacking of bags for both packing densities (Table-1).

Table 1. Post transport mortality and water quality parameters of African catfish packed in polythene bags using two different loading densities.

Parameters	400fish/bag	500fish/bag
Mortality (%)	0.5±0.01b	1.4±0.01a
Ave Temperature (^{O}C)	23.5±0.50b	24.2±0.48a
pH	7.0±0.01a	6.4±0.03b
DO(mg/l)	5.90±0.02a	5.30±0.04b
Ammonia-N (mg/L)	4.1±0.05b	5.3±0.07a
Carbon dioxide (mg/L)	11.2±0.02b	13.1±0.03a

Different letters indicate significant differences between groups ($p<0.05$)

A negligible (≤1%) fish mortality was observed at both packing densities. These mortalities were within the permissible limits during the transportation of live fish. The average water temperature after the unpacking of fish bags was 23.5 and 24.1 ^{0}C in packing density of 400 and 500 fish/bag, respectively which is considered suitable for the survival of African catfish (Khattab *et al.*, 2000; Otubusin, 2001). The concentration of dissolved oxygen in water was 5.90 and 5.30 mg/l at both packing densities. Enough amount of dissolved oxygen in water indicated that oxygen is not the limiting factor at the packing of 4:1 ratio (Tubulin, 2001). Average values of free CO_2 and pH ranged from 11.2-13.1mg/L and 6.4-7.0 for density 1(400 fish/bag) and density 2(500 fish/bag) respectively. Accumulation of carbon dioxide originated from fish and bacterial respiration lowered the pH of water (Swann, 2012). The amount of free ammonia in water is most important factor. The concentration of free ammonia during the transportation of *Clarias gariepinus* was 4.10 and 5.30 in both packing densities. Free carbon dioxide and free ammonia were directly related to packing densities, while dissolved oxygen and pH are inversely related to the packing densities. According to Alikunhi (1954) the water quality of container in which fish fry are kept is adversely affected by overcrowding in the container. Survival of African catfish even at high values of ammonia could be attributed to its hardy nature as extremely high tolerance of ammonia up to 6.5mg/L in *Clarias gariepinus* was described (Olleremann, 1995).

Another greatest challenge after the live transportation of exotic fish species for the introduction purpose is the recovery of fish after arrival at destination. In the present study the fish was initially released in raceways and circular tanks to recover from stress. Post- release mortality was initially higher, and then it becomes stabilized gradually as shown in Fig. 1. More death rates could be due to the higher ratio of stress in the newly introduced environment. Even within a short duration of Transport, carrying fish are often bared to numerous stressors. Poor quality of water (Weirich and Tomasso, 1991; Carmichael *et al.*, 1992), tank confinement (Davis and Parker, 1986), more packing densities (Piper *et al.*, 1982), physical handling (Maule *et al.*, 1988; Cech *et al.*, 1996) and conditioning fish to a new environment (Carmichael *et al.*, 1984; Brick and Cech, 2002) are main stressors related with transporting.

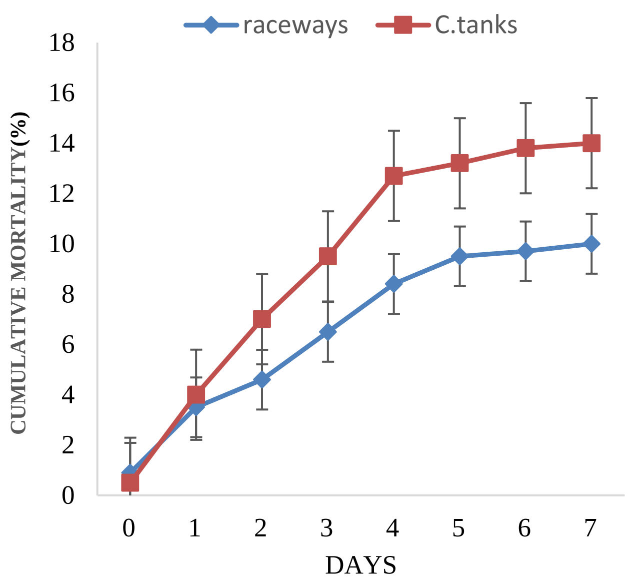

Figure 1. Post release mortality in African catfish during recovery phase after transportation at Aquaculture and Fisheries, NARC.

Acclimatization: After keeping the fish in an intensive environment for recovery from transportation stresses it was distributed in two different culture systems for acclimatization. During the first day, 1%, mortality occurred in raceways and 4% mortality was observed in earthen ponds within the first eighth hours after stocking the fish. The higher death rate was recorded in the fishes as a result of handling in raceways and stress of the newly introduced environment of earthen ponds. When working with catfish fry in small earthen ponds, Bindu (2006) reported to had major mortalities that arose during the nurturing stage when catfish fry was stocked in earthen ponds. Present mortality recorded in this study is due to handling stress.

Fortnightly growth trend during acclimatization for 90 days of African catfish fed with feed (35% Crude Protein) at ad-libtum in raceways and earthen ponds is given in Fig 2. The fortnightly growth trend showed gradual increase in weight gain in raceways and earthen ponds. Total weight in earthen ponds is significantly higher than cemented raceways. The better growth in earthen ponds are in agreement as reported by the other authors (Akinwole *et al., 2016*; Marimuthu, *et al.*, 2010; Nahar *et al.*, 2000). The better growth in earthen ponds may be due to the higher water temperature (27-30°C) as compared to the temperature of raceways. The natural food in ponds may contribute to the significantly better growth rate in earthen ponds as compared to the race ways. Present results were similar to that reported earlier by Rab *et al.* (2007) who described substantial growth in summer within earthen ponds with sufficient supply of natural prey items. The natural feeding resulted in increased growth rate in earthen ponds with nearly 8-9% (Bosworth and Wolters, 2005). Our results are in contrast to the finding of some other authors reporting more gain in weight under raceways (Marimuthu, *et al.*, 2010; Nahar *et al.*, 2000). The quality of water can easily be managed and controlled in outdoor and indoor tanks, as compared to earthen ponds (Keremah and Esquire, 2014). Some other studies have also reported the farming of *C. gariepinus* in rigorous and extensive type culture systems in both raceways and earthen ponds (Keremah and Esquire, 2014).

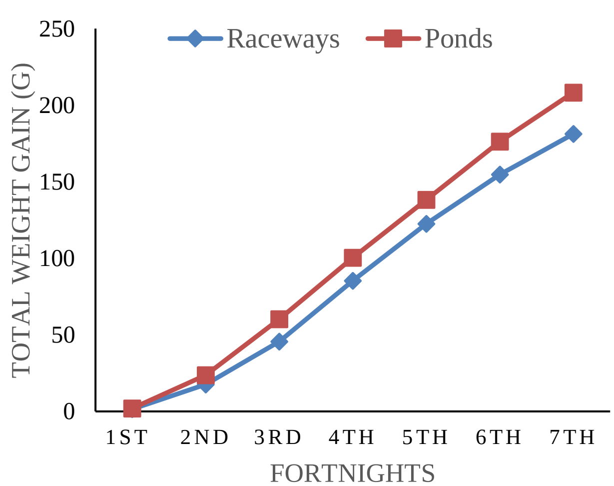

Figure 2. Line graph showing growth of African catfish during acclimatization (June- August 2015) fed on imported floating feed in raceways and earthen ponds.

The performance of African catfish in raceways and earthen ponds during acclimatization period of 90 days is given in Table 2. Significantly higher weight gains and specific growth rate was observed in earthen ponds as compared to raceways. African catfish were successfully acclimatized in the present study in both type of culture systems without aeration. Fish survived even in very low water exchange in raceway culture. As compared to other species, catfish have the ability to grow at even severe culture conditions without requirement of pond ventilation or higher water exchange (Huisman and Richter, 1987). The exclusive potential of survival also lies in its air breathing ability, omnivorous type feeding habit and its forbearance to adversative water quality conditions (Nguyen and Pongthanapanich, 2016).

Table 2. Growth performance of African catfish in raceways and earthen ponds during acclimatization period of 90 days.

Parameters	Raceways	Earthen ponds
Mortality (during first 8 hours)	1.00 %	4.00 %
Initial weight (g)	1.9±0.45a	2.0±0.61a
Final weight (g)	181.2±2.61b	208.3±2.88a
Weight gain(g)	179.3±1.25b	206.3±1.08a
Specific growth rate	4.9±0.07b	5.2±0.03a
Survival fish after 90days	30.4±0.04b	63.4±0.07a

Value are mean ± S.D.; Difference was significant (P<0.05).

However, the decreased in the number of fish was recorded in both cemented raceways and earthen ponds during the acclimatization process although apparent natural mortality was very low. This was due to cannibalism that is reported in African catfish and many other fish species. The cannibalism was reported as one of the major problems hindering survival in *C. gariepinus*, Tilapia, *H. longifilis* fry and fingerlings (Royle, 2001). The present outcomes were supported by different earlier studies (Baras, 1999; Yalcin *et al*, 2002; Dadebo, 2009). According to Naumowicz *et al.* (2017), more than 30 fish families have been reported to exhibit cannibalism phenomenon. Cannibalism is highly detrimental and disadvantageous for most of these fish species which are valuable contributors of aquaculture. Some wild populations of African catfish were also reported to have cannibalism (Corbet, 1961; Bruton, 1979). The cannibalistic cohort of species has also been reported in larval and juvenile sibling (Aboul-Ela *et al.*, 1973). This decrease was more obvious in raceways as compared to earthen ponds (P<0.05). Ponds provide significantly higher survival rate to African catfish as they have more space to reduce interaction between individual fish and they also provide place to smaller fish to avoid attacks by larger fish. The rate and extent of cannibalism in African catfish larvae and juveniles is affected by numerous environmental factors (availability of food& prey, feeding frequency, size variation, stocking density, refuge, light and feed distribution (Reddy *et al.*, 1995).

It was observed that cannibalism was high during the initial period of acclimatization then gradually moves to decline. Natural mortality also found reduced with the passage of time, ultimately it diminishes and survival rate improves as shown in Fig. 3 and 4. A higher rate of cannibalism was observed in early weeks or months during the uneven growth of organisms (David *et al.*, 2010). Cannibalism is thus eased by heterogeneity of the size in fish. The smallest fish are consumed by the larger ones same as happens in control group in present study Bara *et al.* (1999) and Haylor, (1989). Two types of cannibalism including 'tail-first' and 'head-first' are defined by Hecht and Appelbaum (1988) in culture of *C. gariepinus* who reported the start of cannibalism from 8 mm to about 80 mm. Hafidh and Ali (2004) also reported that cannibalism is mainly causing the mortality in African catfish and it became more prominent in species that are not fed properly (2-4%). According to Mukayi *et al.*, (2013) chemical substances that were produced from the bruised skin surface of African catfish elicited cannibalistic behavior in other fish present near the injured fish are also accountable for cannibalistic behavior. They commend larval rearing of African catfish in dark or dim conditions to increase survival rates in catfish.

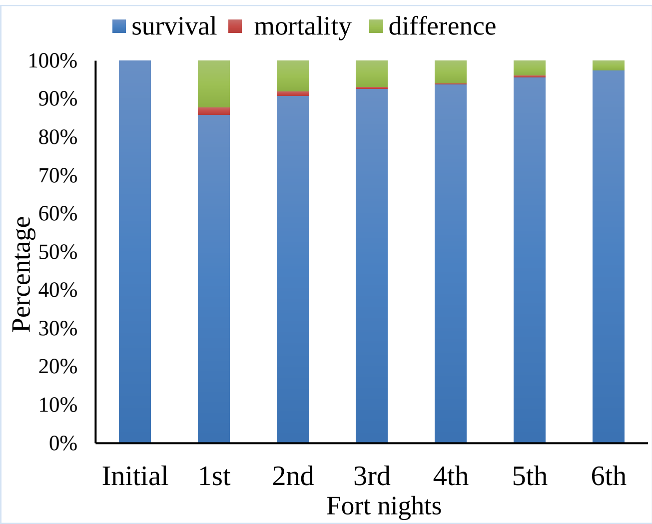

Figure 3. Difference in African catfish survival in earthen ponds during acclimatization.

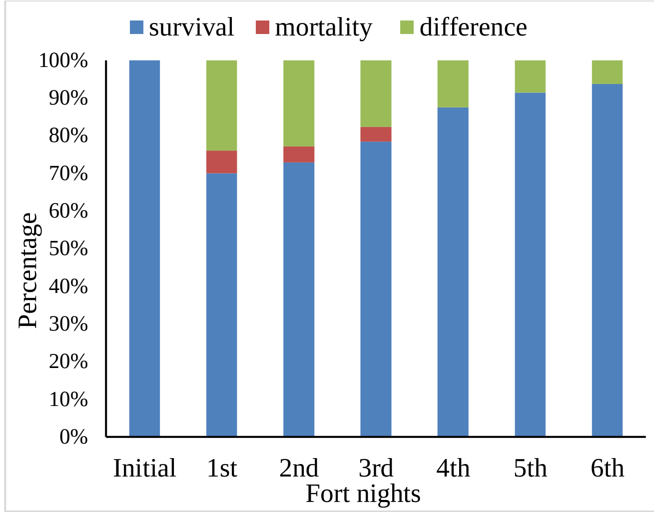

Figure 4. Difference in African catfish survival in raceways during acclimatization.

The average temperature recorded during the experimental period in outdoor concrete raceways and earthen ponds was 27.00± 0.7ºC and 30.9± 1.00ºC, respectively. The range of different limnological features recorded in raceways and earthen ponds are given in Table 3. Different studies have shown the maximum rate of growth of *C. gariepinus* at the temperature of 28-30°C and 24-29°C (Mollah, 1984; Henken *et al.*, 1986). The ranges of dissolved oxygen values recorded in the present study were 5.4-7.8 mg/1. The range of DO reported earlier was 6.7-8.3 ppm, considered suitable for fish production (Lakshmanan *et al.*, 1967). The pH ranges recorded in the present study were similar to that described earlier (Dewan *et al.*, 1991). Proper grading according to size is also used to decrease size discrepancy of many other cultivated species of fish (Popper *et al.*, 1992; Kamstra, 1993).

Table 3. Water quality parameters recorded in raceways and earthen ponds.

Parameters	Earthen ponds	Raceways
Temperature °C	30.9±1.00	27.0±0.7
DO(mg/l)	5.4±0.40	8.7±1.3
pH	7.2±0.30	145.2±0.4
Alkalinity(mg/l)	168.3±7.63	157.0±3.4
Hardness(mg/l)	130.0±10.0	136.2±5.8

Control of Cannibalism in African Catfish through Physical Grading: Physical grading is an important tool to control the phenomenon of cannibalism. The Table 4 showed that there was a significant difference in the survival rate of African catfish at different treatments. The fish which was sorted after five days interval (T1) has maximum survival rate (87.7%) followed by the fish sorted after seven days interval (T2) having survival rate of 85.2% and control

Table 4. Survival rate, natural mortality and cannibalism-induced mortality of African catfish.

Group	T1(sorting interval 5 days)	T2 (Sorting interval 7 days)	T3 (without sorting)
Number of fish stocked	200	200	200
Final number of fish harvested	175.5±25.3b	17.50±28.7a	23.20±4.57c
Natural mortality (No.)	9.4±0.62a	8.90±0.79a	10.70±0.54a
Cannibalism induced mortality (No.)	15.1±0.74c	20.60±0.52b	166.10±0.67a
Total survival (%)	87.7±1.65a	85.25±1.73a	11.60±0.25b
Average Final weight(g)	44.6±1.00c	46.63±0.65b	78.56±7.34a
Weight of total harvest(g)	7827.3±0.95a	7950.40±0.87a	1822.59±0.65b

group (11.6 %) respectively. The results of this study are in line with the findings of Mollah *et al.* (1999) who reported that grading is useful for improving survival in African catfish. The maximum cannibalism was observed in control group (unsorted group) followed by T2 (7 days sorting interval) and T1 (5 days sorting interval). The highest rate of cannibalism in the control group was due to the size difference of fish (Baras and Jobling, 2002). The cannibalism can be significantly lowered by sorting of individuals (Szczepkowski *et al.*,2011). Natural mortality was non- significantly different among in all the three treatments. African catfish are hardy and are able to thrive in harsh environmental conditions; they are less prone to natural mortalities (Bruton, 1979; Uys, 1989). The average weight gain was maximum in control group (non-sorted group) followed by T_2 (7 days sorting) and T_1(5 days sorting) while the total weight of harvest was maximum in the fish which was sorted after 5 days interval followed by T_2(sorted after 7 days) and control (unsorted). An improved rate of growth was also observed by some researchers after the removal of dominant individuals from systems (Jobling and Reinsnes, 1986).

It was concluded that the exotic African catfish was successfully acclimatized in local environment of Islamabad. Low survival rate due to cannibalism at early stage was a major problem for its culture. The issue of low survival due to cannibalism at initial stages can be improved by sorting the fish at 5 days interval.

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