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Effect of Different Stocking Densities on Behavior of Common Carp (*Cyprinus carpio*) in Duhok Province, Kurdistan Region, Iraq

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Abstract— An experiment was conducted to checking up the effects of overcrowding on common carp *Cyprinus carpio* prosperity using their behavior. Nine concrete ponds were used with three replication ponds for each density (3.6 m length, 2.7 m width, 1.00 m depth and 0.80 m water level) with water volume (7.776 m³) for each pond. Number of fishes per each pond was 4 (low density), 8 (medium density) and 12 (high density), weighing 1000 g. Water volume for each fish in ponds was (1.944 m³, 0.972 m³, 0.648 m³ respectively). Fish behavior was recorded by immediate sampling with frequency and duration. Each pond was observed directly on the front of pond. Results exposed that there was a significant difference (P<0.01) of time spent feeding between treatments. In addition, a significant difference (P<0.01) was found in resting time between treatments besides there was a worthy of difference of swimming time between different stocking densities. Time spent in swimming was recorded for high stocking density fishes more than other groups significantly (P<0.01), and significantly less time spent resting. Air breathing had significantly (P<0.01) for higher stocking density behaviors were found to be more significantly; bites and attacks (P<0.05), threatening and submission (P<0.01). It's not recommend to stock less than two fishes per meter for carp at concrete ponds.

Keywords— Common carp behavior, stocking density, Aggression, overcrowding, Swimming.

I. INTRODUCTION

Farmed-fish production and shellfish has increased twice throw the thirty past years, and it seemed that increasing will be continued [1]. There is a growing concern of fish rearing welfare together with aquaculture sector growth, [2]. The major basis of animal welfare conception believes that animals are conscious, which means they are passionate creatures and have rights to have good welfare [3]. However, fish sentience is not completely awared as some scientists believe that they are emotional creatures and feel harm, whereas others do not [4,5,6]. The tipping that fish is able of both negative and positive emotions could be contributed to the launching of different strategies, for example, environmental enrichments, for promoting good welfare [7].

Welfare evaluation should include some indicators that advertise different welfare aspects, behaviour and the physiology of stress, for example. Behaviour is a key tool for the welfare of fish. On one hand, behavioural observation is clear and it is easy and fast method for observer [7]. On the other hand, it is complicated because there are some obstacles related with observing the behaviour of fish [8]. These obstacles are frequently not harmonious over time and their quantitative is difficult, observing animals require an observer having passable skills. Furthermore, the observer should be familiar with normal behaviours in order to distinguished abnormal ones because the differences between normal and abnormal behaviours are often not clear [7]. Instances of generally used behaviours as welfare indicators are feed intake, changing food expected behaviour, ventilation rate, aggression and swimming behaviour [7,9].

The above-mentioned behaviours are affected by different stocking densities that are used in fish farms are underlined as welfare concern area [2]. This is because fishes in farms are kept in higher densities than fishes in the wild. However, there are differences between species [10]. This suggests higher stocking densities may positively or negatively affect fish welfare, particularly the growth of fishes and aggressive behaviour, depending on fish species [11].

A review by [12] showed that there were more than forty studies presenting the effect of stocking densities on rainbow trout. There was, in general, no stress was caused to rainbow trout, but there were other effects, including reducing growth and feed conversion efficiency, increasing fin injuries [12]. In addition, high stocking densities decreased survival rate of Pacific Salmon [13]. High stocking densities caused heavy mortalities and stunted growth in common carps [14]. Moreover, high stocking densities increased blood cortisol levels, which leads to stress in common carp [15]. This suggests increasing stress levels may lead to increase aggression in common carp. Therefore,

This study was aimed to elucidate the effect of different stocking densities on common carp welfare using feeding behavior, air breathing, and swimming behavior, aggressive behaviors, including attacks, bites, submission and threatening.

II. RELATED WORK

Previous studies in water bodies showed that some fishes prefer shoaling at high densities, [7]. This suggested that high densities may have positive effect on fish welfare. A research by [16] on Arctic char fish showed that low stocking densities affected the behaviour, while with medium and high densities fish had higher growth rates compared to low stocking densities. Similarly, another researcher on African catfish [17] show that positive effects were found with high densities. In addition, it was found that higher stocking densities improve African catfish's welfare, but it depends on the age of fishes, because in some ages, stocking density neither improved nor impaired fish welfare [18]. Furthermore, low stocking densities increased aggressive behaviour in 10-day-old African catfish while high densities did not affect aggression [19], whereas in other species, stocking density may cause welfare problems.

III. METHODOLOGY

Study site

The study was undertaken in the fish project of the Agricultural Engineering Sciences College, Summel, 15 Km west of Duhok, Kurdistan region of Iraq. The study was done in April, 2017.

Animals and design

Nine concrete ponds were used (low, medium and high stocking densities) and each stocking density was placed into three ponds. The dimensions of small concrete ponds are (3.6 m x 2.7 m x 1 m and 0.80 m water level. Number of fishes per each pond was 4, 8 and 12 for low, medium and high stocking densities, respectively. The adaptation period was seven days. Water temperature was checked daily using infrared thermal gun so as to avoid its effect on fish behavior recording (27.4 ±0.5°C, 26.9 ±0.4°C and 27.2 $\pm 0.5^{\circ}$ C) for low, medium and high stocking densities respectively. Diver motor was used for aeration to all ponds. Fishes brought from Suwaira fish farm in Baghdad Province, were distributed into the ponds after weighing them to prevent weight differences; the average weight of fishes was 1000 ±50 g. Fishes of age 1 year were used in this experiment. Fishes were fed daily by using standard commercial feed (energy 3925 Kcal/kg and crude protein 27.31% dry matter) with an average of 40g/fish/day gradually until the fish reach satiety. Daily feed for the low, medium and high stocking densities will be 160, 320 and 480g/day, respectively (three times daily). To maintain water quality, every two weeks a partial water exchange was performed (75%), where each treatment was maintained at a fixed level of water.

Data Collection

Fish behavior was recorded by direct observations. Immediately sampling method was used to record fish behaviors so as to measure the frequency and the duration of the behaviors. Each pond was observed directly on the front of pond. From each pond, 18 hours of direct observations were recorded with 1 minute interval. These behaviors were recorded: Foraging (feeding) behavior as duration; swimming behavior as duration; swimming time was expressed as a proportion of the total observation time. Resting time was defined as hundred minus the proportion of resting. Air breathing and aggressive behaviors were expressed depending on [7] as a frequency per unit time (Table 1).

Table	1. List	of recorded	fish	behaviors	with	their	description	ıs
		observed in	the	present exi	perim	ent.		

Behaviors	Descriptions			
Swimming	A displacement of the body, while browsing, moving, eating and air-breathing.			
Resting	Moving passively through the water or lying still at the bottom of the pond.			
Feeding	The search for and exploitation of food resources.			
Air breathing	The animal moves to the water surface and takes a gulp of air. This was checked by escaping air from the gills of the fish, when it was swimming back to the bottom of the tank.			
Aggressive behaviors				
Bites	Snapping movements of one individual towards the head of another individual			
Attacks	A fish directly attacks another fish.			
Threatening	Threatening is any behavior that signifies hostility or intent to attack another animal. Threat behavior is meant to cause the opponent to leave.			
Submission	Submission includes flight or immobility.			
	Adapted from [7] [18] and [20]			

Data analysis

All the recorded behavioral data were projected to the Microsoft Excel spreadsheet, and thus, data were prepared for statistical analysis. GenStat Software Program (17th edition, VSN International Ltd, UK, 2014) was used to analyze behavioral data. Summary statistics was obtained from Past3 software program (Paleontological Statistics, Version 3.08). Residual plots confirmed by normality test (Shapiro-Wilk test) presented that all behavioral data obtained from fishes to be nonparametric. Hence, Kruskal-Wallis test was used for data analysis followed by a two-sample nonparametric test by using Mann-Whitney U-test for post hoc comparison.

IV. RESULTS AND DISCUSSION

Feeding behavior

The effect of stocking densities on time spent feeding is shown in Figure 1. Time spent feeding was significantly (P<0.01) different between treatments. High stocking density fishes spent significantly (P<0.01) more time in feeding. In addition, medium and low stocking density groups were not significantly different.



Figure 1: Effect of different stocking densities on foraging behavior (time spent feeding) of common carp, expressed as Mean \pm SEM per minute. Different letters mean that there is a significant difference (P<0.01). P1 is low, P2 is medium and P3 is high stocking density.

Swimming and Resting Behavior

Figure 2 shows the effect of three stocking densities on swimming and resting time of common carps. A significant difference (P<0.01) was found in resting time between treatments as well as there was a considerable difference of swimming time (P<0.01) between different stocking densities. High stocking density fishes spent more time in swimming than other groups significantly (P<0.01), and significantly (P<0.01) less time spent resting.



Figure 2: Effect of different stocking densities on the proportion of time spent resting and swimming of common carp, expressed as % of total observation time. P1 is low, P2 is medium and P3 is high stocking density.

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Swimming was in highest level with high stocking density over the observation time (60 minutes). It was considerably higher (P<0.01) than other groups (Figure 3). In addition, there was no significant difference between medium and low stocking density groups.



Figure 3: Effect of stocking density on time spent swimming throughout the mean observation time (60 minutes) of common carp.

Resting was in lowest level with high stocking density over the mean observation time (60 minutes). It was significantly (P<0.01) lower than other groups (Figure 4). No significant difference was found between medium and low stocking density groups.



Figure 4: Effect of stocking density on time spent resting throughout the observation time (60 minutes) of common carp. P1 is low stocking density, P2 is medium and P3 is high stocking density.

Aggression and Breathing Behaviors

Air breathing and aggressive behaviors are shown in Table 2. Air breathing was significantly (P<0.01) different between different stocking densities. The higher stocking density had more air breathing than other stocking densities. All aggressive behaviors were significantly different; bites and attacks (P<0.05), and threatening and submission (P<0.01). Significantly more aggressive behaviors were found in higher stocking densities.

Table 2: Effect of three stocking densities on the frequency of aggressive behaviors and air breathing (Mean±SEM) per minute.

Behaviors		St	P-			
(mear	n±SEM)	P1	P2	P3	value	
Air breathing		0.003 ±0.002 ^a	$0.033 \\ \pm 0.008^{b}$	0.214 ±0.027 ^c	0.05	
	Bites	0.003 ± 0.002^{a}	0.011 ± 0.005^{ab}	0.022 ±0.007 ^b	0.05	
Aggressive	Attacks	0.019 ± 0.006^{a}	0.044 ± 0.010^{ab}	$\begin{array}{c} 0.050 \\ \pm 0.011^{b} \end{array}$	0.05	
behaviors	Threatening	0.025 ± 0.008^{a}	0.072 ±0.013 ^{bc}	0.106 ±0.017 ^c	0.05	
	Submission	0.019 ± 0.008^{a}	0.050 ± 0.010^{bc}	0.078 ±0.015 ^c	0.05	

Different letters in each row mean that there is a significant difference (P<0.05). P1 is low stocking density, P2 is medium stocking density and P3 is high stocking density.

Discussion

It was obvious that high stocking density fishes spent more their time in feeding and more swimming and less resting. However, more air breathing and aggressive behaviors were found in high stocking density group.

Feeding Behavior

Foraging or feeding is defined as searching for and exploiting the resources of food [21]. In fishes, feeding behavior has been documented with regards to daily feed intake, the feeding latency, the self-feeders activation and total time of feeding [22] in this experiment; feeding rate was used as time spent feeding.

Fluctuations in feed consumption of fishes have been frequently used as an indicator of welfare reduction. Different stressors have been revealed to reduce the intake of feed in various species of fishes. Instances of stressors are changes in the quality of water in Atlantic salmon, Nile tilapia and sea bass [23, 24] stocking density such as in rainbow trout [12], vaccination and anaesthetization in Atlantic salmon [25] dominance hierarchies' establishment in Arctic char [26]. In addition, social structure changes of the groups, for instance, in African catfish [7, 27]. Furthermore, cleaning protocols, for exampletanks brushing and emptying can affect negatively on self-feeding of sea bass [28].

Time spent feeding was significantly increased with increasing stocking density in the present study. Similar results were previously found by [7]. They found that time spent feeding was significantly (P<0.05) increased of heavy weight fishes than low weight fishes both before and during feeding. In addition, they found that heavy weight fisheswere faster in consuming their feed [7].

Swimming and Resting

In the present experiment, high stocking density had higher percentage of time spent swimming and lower percentage of time spent resting. The findings of this study are in agreement with previous results of [7] who found that swimming activity of juvenile African catfish for heavy weight fishes was significantly (P<0.01) higher and resting was significantly (P<0.05) lower than low weight fishes. [29] showed that high stocking density increased the activity of swimming, compared to medium and low stocking densities in Atlantic halibut Hippoglossushippoglossus Fishes, such as rainbow trout Oncorhynchus mykiss, are able to increase their swimming time in response to an increase in stocking density [30]. Similar results were found in the present study (Figure 2). [20] Found that Arctic char Salvelinus alpines spent significantly more time spent swimming at high stocking densities than fishes placed at the lower densities. Therefore, the present study confirms the previous results of [20].

Air Breathing and Aggression

Air breathing can be increased as a result of some stressors or procedures of aquaculture. The main factors that affect ventilation activity are carbon dioxide (CO₂) and the dissolved oxygen (O₂) levels in the water with air breathing being contrarily linked to the oxygenation of water. Ventilation frequency increases when O₂ levels are not enough to fish needs. Contrarily, the ventilation frequency is decreased when oxygen is surplus [22, 31]It was found in this experiment that increasing density significantly (P<0.01) increased air breathing. Similar results were obtained by [18] that air breathing significantly (P<0.05) increased with increasing stocking densities of juvenile African catfish *Clarias gariepinus*..

There are several factors that affect aggressiveness of fishes. For instance, according to ecological studies, aggressive behavior is increased with increasing temperature above 27°C [32] High temperature degrees increased the injuries of dorsal fins, which in turn, indicating increased aggressiveness [33]. In addition, the intensity of light has also been recommended to affect aggression. However, no evidence was found by [34] that the intensity of light affects the condition of dorsal fin, as an indicator of aggressive behavior. In addition, the aggression frequency is related in reverse to the availability of food [12]. Several factors related to feeding affect aggression, specifically ration, feeding distribution in time and space, and composition of feeding [33, 34, 35, 36]. Moreover, stocking density and fish size can also affect aggressive behavior. The majority of aggressive acts are initiated by larger fishes [12]. However, in Atlantic salmon, aggressive behavior was decreased with the presence of few larger fishes [37].

[38] Found that aggression rates were increased when fishes placed at higher stocking densities. [39] Did not found that aggressive acts decreased at high stocking density in *Salmo gairdneri*. In addition, [40] showed that increasing stocking density increased the duration and frequency of aggression in *S. gairdneri*. Similar results were found in the present experiment. Therefore, results of this study agree with previous findings. Similarly, [20] revealed that Arctic char *Salvelinus alpinus* had significantly fewer aggressive acts at high stocking densities than at lower stocking densities.

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The present experiment results are in agreement with [20]. This suggests that increasing aggression with high densities of Common carp may increase mortality rate as previously found by [41], However, feeding activity and proportion of their time spent swimming increased with increasing stocking densities.

On the other hand, in 10 day-old African catfish *Clarias* gariepinus, aggressive behavior was increased with lower stocking densities, while high stocking densities had no effect on aggressiveness (Kaiser et al., 1995). Therefore, results ofthe present study do not agree with previous findings of [42].

V. CONCLUSIONAND FUTURE SCOPE

It can be concluded that stocking density has a considerable effect on common carp behaviour. High stocking density increased percentage of time spent swimming and decreased percentage of time spent resting. In addition, high densities increased time spent feeding of common carp. compared to lower densities. On the other hand, high density significantly increased aggressive behaviour. However, mortality rate was not measured in this experiment. It was previously stated that it is increased as stocking densities increasing and therefore, no concrete conclusion can be made from this work but low stocking density is not recommended. To recommend high densities, further research is required to use the effect of different stocking densities on common carp behaviour, growth and mortality rate. In addition, more research is needed to confirm this work.

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