

## Evaluation of Hatchability from Various Intraspecific Crosses of Carp and Effect of Frequencies of Feeding on Stocking Density of Common Carp Fry

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### Abstract

Common carp (*Cyprinus carpio*) germplasm were sourced from Bauchi and Ibadan aimed at studying their hatchability from the various intraspecific crosses and effect of frequencies of feeding on stocking density of Carp fry. Both male and female were injected with Ovaprim at 0.5ml/kg body weight intramuscularly at a single dose. The number of eggs in 1 gram mass was determined, Fertilization rate and percentage of hatchability were calculated. One hundred and eighty (180) fry of *Cyprinus carpio* were randomly selected and distributed in nine bowls of 50 litres at three stocking densities with replicates for each treatment. The frequencies of feeding were in these order; Ten (10) fry per bowl of 50 litres were fed twice a day, Twenty (20) fry per bowl of 50 litres were fed four times a day, Thirty (30) fry per bowl of 50 litres were fed six times a day, respectively for 8 weeks with 30gm of 0.2mm Coppens starter diet. The Ib♀ x Ba♂ cross had the highest hatchability though highest fertilization rate was recorded by Ba♀ X Ba♂ cross. The various crosses had above 50% hatchability, though percentage hatchability differed significantly at ( $P < 0.05$ ). While the growth pattern of the different stocking densities at different feeding frequencies differed significantly ( $p < 0.05$ ). The intra specific crossing of different sexes from the two sources gave a higher yield as can be seen with the IB♀ x BA♂. Whereas carp fry stocked at a lower stocking density with a fewer feeding frequency had best growth.

**Key words:** Common carp (*Cyprinus carpio*), intra specific crosses, hatchability, feeding frequencies, stocking density.



## Introduction

The success of any fish farming operation depends on the availability of ready supply of fish larvae for on growing to market size (Rottman *et al.*, 2003). According to Mittlemark and Kapuscinski (2006), Brooders should be fast growing, disease free and sexually ripe. Brzuska (2003a) reported that heavier females yielded eggs of heavier weights expressed in grammes. As females are collected from the brood fish ponds, it is advisable to disinfect the fish with a 50 -150ppm formalin bath for three hours before they are brought into the hatchery. This precaution is taken to prevent pathogens from being transmitted to eggs and larvae. The report by Hill *et al.*, (2009) revealed average success rates of 50% ovulation, 54% spermiation and 1.3% mortality were recorded after injection of different species with Ovaprim. Also, Ovaprim has been used successfully for hypophysation in different families of fish like Cyprinidae (Hill *et al.*, 2009), characidae and cobitiidae (Yanog *et al.*, 2009).

Fertilization, hatching and larvae survival are vital for successful culture of the African catfishes (Haniffa *et al.*, 2000) and this has been investigated earlier (Ataguba *et al.*, 2009). Theoretically, apart from biological factors, physical and chemical parameters are known to affect egg development. For example, temperature is known to be the main environmental factor governing fish egg development (Blaxter, 1992). Temperature affects certain morphological features, hatching rate and larval behaviour. In earlier studies, temperature influenced egg development and hatching in *O.niloticus* (Bhujel *et al.*, 2000), *Tilapia zilli* (Omotosho, 1998) Common carp, *Cyprinus carpio* (El-Gamal, 2009), and cod, *Gadus morhua* L (Page and Frank, 1989; Geffen *et al.*, 2006).

Egg counting in fish can only be achieved if the eggs are degummed because of the adhesive nature of the eggs when they are freshly stripped, to enhance degumming, 15g of sodium sulfite is weighed and dissolved in 100 ml of distilled water making up the concentration stock solution. The stock solution is diluted with 900ml of hatchery water (Khan *et al.*, 1986).

Fish growth is influenced by feed availability and intake, genetics, age and size, environment and nutrition. Of these factors, feed intake is perhaps the principal factor affecting growth rate of fish (Li *et al.*, 2004). Feed management in terms of optimization of feeding rate and frequency has become one of the crucial areas of research in the field of aquaculture. Over feeding and left over feed reduce the water quality (Ng, 2002) while inadequate food supply has

direct impact on production cost (Mihelakakis *et al.*, 2002). Different species of Fish have been shown to have different optimum feeding frequencies, for instance young salmon can feed continuously for 15 hours a day (Shearer, 1994). Diet and stocking densities are two major factors in aquaculture influencing growth, welfare and health (Ellis *et al.*, 2002, Alcorn *et al.*, 2003). Feed costs make up a large part of total fish production costs. The optimization of growth rates and feed efficiency depends on the quantity of feed delivered, feeding method and feeding frequency, quality and composition of the diet (Gelineu *et al.*, 1988; Yang *et al.*, 2007, Erundu *et al.*, 2006). In aquaculture, stocking density directly influences survival, growth, behaviour, water quality and feeding. In aquaculture, stocking density is the concentration which fish are stocked into a system (Gomes *et al.*, 2006, de Oliveira *et al.*, 2012). Generally, increases in stocking density results in direct increase on the stress condition causing a reduction in growth rate and feed utilization (Sharma and Chakrabarti 1998). On the other hand, in very low densities fishes may not form shoals and may feel unprotected. Consequently, identifying the optimum stocking density for a species is a critical factor not only to enable efficient management and to maximize production and profitability but also for optimum husbandry practices (Leatherland and Cho 1985; Kristansen *et al.*, 2004; Rowland *et al.*, 2006). Various efforts have been made to determine appropriate stocking densities to circumvent this limitation but until now recommended stocking densities vary considerably (Russell *et al.*, 2008). With limited land resources and high competition with agriculture for land, there is need for urgent refining these recommendations so that appropriate stocking densities are adopted in time to minimize production deficiencies per unit space available.

Growth, survival and yield effects of stocking densities on aquaculture are well known for a diversity of species (Zhu *et al.*, 2011, Garr *et al.*, 2011, Khatune- Jannat *et al.*, 2012) and seem to impact production differently. Both growth performance and survival rate, for instance, tend to be higher in lower stocking densities in the African catfish *Clarias gariepinus* (Hecht *et al.*, 1996). In the Thai climbing perch, *Anabas testudineas* (Khatune-Jannat *et al.*, 2013) in Amur sturgeon, *Acipeinser schrenckir* (Zhu *et al.*, 2011), but only survival is higher under same conditions in *Oreochromis spp* (Ridha 2005). In some cases, such an advantage of lower stocking densities is either non-existent, as is the case in channel catfish (Southworth, *et al.*, 2009) in *Oreochromis*

*niloticus* (Osofero *et al.*, 2009) Although they may promote for food and negatively influence reproductive success via reduced fecundity and egg quality (Tave, 1986), high stocking densities may sometimes have no effect on mortality rates and may actually enhance total fish yield (Abou *et al.*, 2007, Gokcet and Sorphea *et al.*, 2007; Pouey *et al.*, 2011 and Pouey *et al.*, 2011; Khatune – Jannat 2012). Where land costs, freshwater, manpower and other facilities are limiting it may be more profitable to adopt higher stocking densities (Ridha, 2005). The rearing conditions adopted for a specific aquaculture programme will therefore be a compromise between biological and economical requirements of the chosen species

### Aims

The first aspect of the study attempted to evaluate the hatchability of the off springs of the same species through intra specific crosses from two different sources. While the second study attempted to determine the effect of feeding frequencies at different stocking levels of common Carp fry.

### Materials and method

#### Study Area

The studies were under taken at Benue State fish hatchery, Ministry of Agriculture, located along Beach Road Makurdi. Makurdi, the Benue State capital (Nigeria) is located on the following geographical co-ordinates 7°44'0"North, 8°32'0" East. The carp fry used in this study were hatched in the Benue state hatchery.

Both male and female broodstocks to be crossed were injected with ovaprim at 0.5ml/kg. The hormonal administration was a single dose and intramuscularly administered above the lateral line, a little distance from the head. After 12 hours of injection, a quantity of eggs were stripped and one gram weight of the eggs was weighed. Degumming of the eggs was achieved by dissolving 15g of sodium sulfite in a 100ml of distilled water and diluting it in 900ml hatchery water the solution was then poured on the eggs, this process allowed for the eggs to degum to enhance counting of the eggs. This process was repeated three times to arrive at counting average of 820 eggs in one gram of carp eggs.

In order to ascertain the percentage of hatching 1g of eggs were counted to consist of 820 eggs, then one gram of eggs were fertilized and replicated in three bowls. Upon hatching, the hatchlings were counted. On this basis hatchability was determined.

Mathematically represented as;

$$\% \text{Hatching} = \frac{n}{820} \times 100$$

Where: n=number of hatchlings

### Determination of Fertilization Rate

One (1) gm of egg mass that was not fertilized was observed to determine fertilization rate. The time taken for these control eggs to become blurred (dead eggs) was noted, after which the clear appearing eggs in the incubation tanks were termed as fertilized, since a number of eggs in one gram mass had being established earlier.

Fertilization rate was determined using the method described by Ella (1987) a 300mm long glass tube with a diameter of 2.5mm was dipped into the egg mass while keeping the upper end closed with the thumb. The thumb was lifted and eggs were allowed to fill the tube. Representative sample were taken from the surface, middle, and bottom of the fertilized egg mass. The glass tube was raised up against light and the total number of good and bad eggs were counted. The good (fertilized) eggs were Transparent and their contents were clear as against the blurred and whitish appearance of bad (unfertilized) eggs. This was then used to estimate the number of bad eggs in each cross using simple proportion.

$$\% \text{ Fertilization} = \frac{N-b}{X} \times 100$$

Where (N) represent the sample of spawned eggs (b) number of bad eggs is obtained from the relation below.

$$B = \frac{Y}{X} \times N$$

Where (X) is the total number of eggs in the three representative sample and (Y) is the number of bad eggs counted, hence number of good eggs (g) is denoted using;

Total number of larvae=300x change in water level with all larvae. Change in water level with 300 larvae. Fertilization and hatching rates of different treatment were analyzed using one -way ANOVA followed by using Least significant difference to determine differences among the means.

While in the second study, One hundred and eighty (180) fry of *Cyprinus carpio* were randomly selected and distributed into nine bowls of 50 litres at three stocking densities with replicates for each treatment. The frequencies of feeding were in these order; Ten (10) fry per bowl of 50 litres were fed twice a day in replicates, Twenty (20) fry per bowl of 50 litres were fed four times a day in replicates, Thirty (30) fry per bowl of 50 litres were fed six times a day in replicates respectively for 8 weeks with 30gm of 0.2mm Coppens starter diet

During the experimental period, the

following water quality parameters were measured, Temperature, Total dissolved solutes, Dissolved Oxygen and PH. At the end of the trials the surviving fish were sampled, weighed (gm) with the use of a sensitive weighing balance to determine the mean initial weight, mean final weight and measured in (mm) with the use of a measuring rule to determine the mean initial length and mean final length and the number that survived were counted and evaluated based on Survival rate, Growth rate, Specific growth rate (SGR) and Protein Efficiency Ratio (PER).

#### Protein Efficiency Ratio (PER)

$$PER = \frac{\text{Weight Gain}}{\text{Protein Intake}} \quad \text{-----}(6)$$

#### Specific Growth Rate (SGR)

$$SGR = \frac{\text{Log}W_2 - \text{Log}W_1}{T} \times 100 \quad \text{-----}(7)$$

Where: T = Time

LogW<sub>1</sub> = Initial weight of fish

LogW<sub>2</sub> = Final weight of fish

The study was subjected to Analysis of

Variance (ANOVA), and Least Significant Difference (LSD) was used to separate the means, where the difference between two means was equal or greater than LSD, a superscript was given, to indicate the difference. Confidence limit of 5% was adopted in the computations.

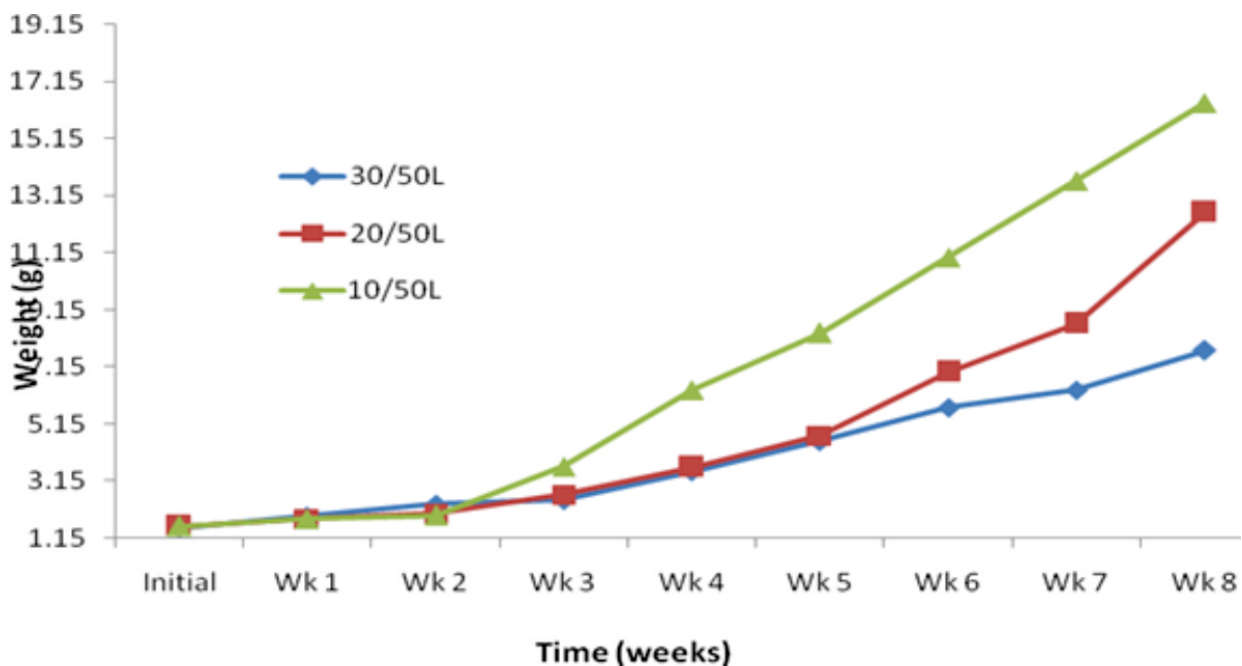
#### Results

Table 1 shows that Ba♀ X Ba♂ had fertilization rate of 85.5±0.23 and Ib♀ X Ib♂ had fertilization rate of 79.6±0.3 while Ib♀ X Ba♂ had fertilization rate of 78.30±0.55 in terms of the hatchability Ib♀ X Ba♂ had a value of 440.32±5.32 and Ib♀ X Ib♂ had hatchability value of 390.23±0.32 and Ba♀ X Ba♂ recorded 380.45±0.47 as hatchability value. This shows that the highest percentage of hatchability was recorded by Ib♀ X Ba♂ with a value of 53.65±0.15. Ib♀ X Ib♂ recorded 47.56±0.03 as percentage of hatchability while Ba♀ X Ba♂ had percentage hatchability of 46.30±0.45. The means differ significantly (p<0.05).

**Table 1:** Hatchability of the Different Crosses of Carp obtained from Bauchi and Ibadan

Parameters	BA♀ x BA♂	IB♀ x BA♂	♀ IB x IB♂
Spawned eggs (no.)	820.64 ± 0.26	824.43 ± 0.39	818.13 ± 2.33
Fertilization %	85.5 ± 0.23 <sup>a</sup>	78.30 ± 0.55 <sup>b</sup>	79.67 ± 0.32 <sup>b</sup>
Hatchlings (no.)	380.45 ± 0.47 <sup>b</sup>	440.32 ± 5.32 <sup>a</sup>	390.23 ± 0.32 <sup>b</sup>
Hatchability%	46.30 ± 0.45 <sup>b</sup>	53.65 ± 0.15 <sup>a</sup>	47.56 ± 0.03 <sup>b</sup>

Means on the same row separated by superscripts differ significantly (p<0.05)



**Fig. 1:** Growth Patterns of the Progenies of *C. carpio* (Ib♀ X Ba♂ Cross) at Different Stocking Densities.



The growth pattern of the different stocking densities at different feeding frequencies indicated that the first one week showed uniform growth pattern, with the thirty (30) fry at six times feeding frequency showing slight increase in the growth pattern between weeks one and two, while the twenty (20) fry fed four times feeding frequency had a steady rise in the growth pattern from week two to week eight, similarly ten (10) fry fed two times feeding frequency had uniform growth

pattern up to the second week then rose steadily to week eight. Though the twenty (20) fry fed four times and the thirty (30) fry fed six times showed similarity in the growth pattern between weeks three and five. With the twenty (20) fry fed four times and thirty (30) fry fed six times a day having a steady increase in the growth pattern between weeks five and eight. However, the ten (10) fry fed two times performed best within the same period.

**Table 1:** Effect of Feeding Frequencies on Stocking Density of *C. carpio* (Ib♀ x Ba♂)

Parameters	30/50L at 6 X ff	20/50L at 4 X ff	10/50L at 2X ff
MIW (g)	1.52 ± 0.02	1.52 ± 0.01	1.53 ± 0.02
MFW (g)	7.76 ± 0.38 <sup>b</sup>	12.57 ± 1.81 <sup>ab</sup>	16.39 ± 1.61 <sup>a</sup>
MWG (g)	6.29 ± 0.36 <sup>b</sup>	11.01 ± 1.82 <sup>ab</sup>	14.84 ± 1.61 <sup>a</sup>
MIL(mm)	12.03 ± 0.03	12.03 ± 0.03	12.07 ± 0.09
MFL(mm)	30.33 ± 0.33 <sup>b</sup>	36.68 ± 1.67 <sup>a</sup>	39.00 ± 2.08 <sup>a</sup>
MLG (mm)	18.30 ± 0.35 <sup>b</sup>	24.63 ± 1.63 <sup>a</sup>	26.93 ± 2.08 <sup>a</sup>
Growth rate	0.11 ± 0.001 <sup>b</sup>	0.20 ± 0.03 <sup>ab</sup>	0.26 ± 0.03 <sup>a</sup>
SGR	2.89 ± 0.07 <sup>b</sup>	3.68 ± 0.26 <sup>a</sup>	4.19 ± 0.17 <sup>a</sup>
PER	0.12 ± 0.001 <sup>b</sup>	0.22 ± 0.04 <sup>ab</sup>	0.30 ± 0.03 <sup>a</sup>
Survival (%)	73.00 ± 0.33	77.15 ± 1.20	76.40 ± 1.07

Mean in the same row with different superscripts differ significantly (P<0.05)

## Discussion

The study revealed that Ibadan ♀ x ♂ Bauchi cross had the highest percentage hatchability of 53.65 ± 0.15 and the Ibadan ♀ x ♂ Ibadan had 47.56 ± 0.03 while Bauchi ♀ x ♂ Bauchi had 46.30 ± 0.45. It then holds that the percentage hatchability of 53.65 attained from the Ibadan ♀ x ♂ Bauchi cross met with the report of (Viveen *et al.*, 1986) that in general the mean percentage hatching of eggs should be between 50-80%, for a successful hatching or breeding activity. Fish egg hatchability could be attributed to several factors ranging from the intensity, stage of maturity of the parent stock this is further buttressed by Blaxter, (1992) who stated that theoretically, apart from biological factors, physical and chemical parameters are known to affect egg development, for example temperature is known to be the main environmental factor governing fish egg development. Temperature affects certain morphological features, hatching rate and larval behavior. In earlier studies temperature influenced development and hatching in *Tilapia zilli* (Omosho, 1998, *O. niloticus* (Bhujel *et al.*, 2000, Cod, *Gadus morhua* (Page and Frank, 1989, Geffen *et al.*, 2006) and Common carp, *Cyprinus carpio* (El-Gamal, 2009).

While the second study revealed that three levels of stocking densities at three different

feeding frequencies were involved, in which thirty (30) fry fed six times a day, twenty (20) fry fed at four times a day and ten (10) fry fed twice a day with water flowing through in a 50 litre container in replicates. The 10/50litre at two times feeding frequency had mean weight gain of 14.84g, the 20/50 litre at four times feeding frequency had mean weight gain of 11.01g while the 30/50litre fed six times a day had mean weight gain of 6.29g. The mean length gain followed the same trend as the 10/50litre two times feeding frequency had mean length gain of 26.93mm, 20/50litre four times feeding frequency had a value of 0.20 and the 30/50litre at six times feeding frequency had a value of 0.11. The results obtained in this study are in line with earlier work done by Sharma and Chakrabarti 1998 that increases in stocking density results in direct increase on the stress condition causing a reduction in growth rate and feed utilization. Growth, survival and yield effects of stocking densities on aquaculture are well known for a diversity of species (Zhu *et al.*, 2011, Garr *et al.*, 2011, Khatune- Jannat *et al.*, 2012,) and seem to impact production differently. Both growth performance and survival rate, for instance, tend to be higher in lower stocking densities in the African catfish *Clarias gariepinus* (Hecht *et al.*, 1996). In the Thai climbing perch, *Anabas*

*testudineas* (Khatune-Jannat *et al.*, 2013) in Amur sturgeon, *Acipenser schrenckii* (Zhu *et al.*, 2011), but only survival is higher under same conditions in *Oreochromis spp* (Ridha 2005).

### Conclusions

The intra specific crossing of different sexes from the two sources tend to give a higher yield as can be seen with the Ib♀ X Ba♂. Whereas in the other study, Carp fry stocked at a lower stocking density with a fewer feeding frequency had best growth.

### Acknowledgement

The author is grateful to West Africa Agricultural Productivity Programme (WAAPP) that funded the Carp project under the competitive Agricultural Research grant Scheme won by Prof. S.G. Solomon.

### References

- Abou, Y., Fiogbe, E.D, Micha, J.C (2007). Effects of stocking density on growth, yield and profitability of farming Nile tilapia, *Oreochromis niloticus* fed azolla diet in earthen ponds. *Aquaculture Research* 38: 595-604.
- Alcorn, S.W., Pascho, R.J., Murray, A.L., Shearer, K.D., (2003). Effects of ration level on immune functions in Chinook Salmon (*Oncorhynchus tshawytscha*) *Aquaculture* 2003: 217(1-4):529-545.
- Ataguba, G.A., Annune, P.A and Ogbe F.G. (2009). Induced breeding and early growth of progeny from crosses between two African clarid fishes *Clarias gariepinus* (Burchell) and *Heterobranchus longifilis* under hatchery conditions *Journal of Applied Biosciences* 14 755-760 from <http://www.bioscienceselewa.org/JABS/2009?14/pdf>.
- Bhujel, C.R, Luang K, Thani, P., (2000). A Review of Strategies for the Management of Nile Tilapia (*Oreochromis Niloticus*) Broodfish in Seed Production Systems, Especially Hapa-Based Systems. Thailand pp 90 -92.
- Blaxter, J.H.S., (1992). The effect of temperature on larval fishes. *Netherlands Journal of Zoology*. 42:336-357.
- Brzuska, E. (2003a). Artificial propagation of Cat fish. On line accessed 15<sup>th</sup> September 20 15. *Czech Journal of Animal science* 48: (4)152-153.
- De Oliveira, E.G., Pinheiro, A.B., De Oliveira, V.Q., Da Silva Junior, A.R.M., De Motaes, M.G., Rocha, I.R.C.B., De Sousa, R.R., Costa, F.H.F, (2012) . Effect of stocking density on the performance of Juvenile pirarucu (*Arapaima gigas*) in cages. *Aquaculture* 2012; 370-371(0) 96-101.
- El-Gamal, A.E., (2009). Effect of Temperature on Hatching and Larval Development and Mucin Secretion in Common Carp, *Cyprinus carpio* (Linnaeus, 1758). *Global Veterinaria* 3(2):80-90.
- Ella M.O (1987). Simple calculations in Fish farming. Wusen press Ltd Calabar, Nigeria pp1-64.
- Ellis, T., North, B., Scott A.P., Bromage, N.R., Porter, M., Gadd D., (2002). The relationships between stocking density and welfare in farmed rainbow trout. *Journal of Fisheries Biology* 2002:61(3);493-531.
- Erondu, E.S., Bekibela, D., Gbulubo, A.T. (2006). Optimum crude protein requirement of catfish (*Chrysichthys nigrodigitatus*) *Journal of Fisheries international* 1: 40-43.
- Garr, A. L., Lopez. H., Pierce R. Davis. M. (2011) .The Effect of stocking density and diet on the growth and survival of cultured Florida apple snails *Pomacea paludosa*. *Aquaculture*, 311(2011) pp 139-145.
- Geffen A.J., Fox C.J., Nash, R.D.M., (2006). Temperature-dependent development rates of cod *Gadus morhua* eggs. *Journal of Fisheries Biology*. 69:1060- 1080. Doi:10.1111/j.1095-8649.2006.01181.x
- Gelineau, A., Corraze, G., Boujard, T. (1988). Effects of restricted ration, time- restricted access and reward level on voluntary food intake, growth and growth heterogeneity of rainbow trout (*Oncorhynchus mykiss*) fed on demand with self feeders. *Aquaculture* 167(3-4) 247-258.
- Gockcek, C.K., Akyurt, I. (2007). The Effect of stocking density on yield, growth and feed efficiency of himri Barbel (*Barbus luteus*) nursed in cages. *Aquaculture*, 311:139-145.
- Gommes, L.C, Chagas, E.C, Martins-Junior H., Roubach R, Ono E.A, de Puala Lourenco J.N (2006). Cage culture of tambaqui (*Colossoma macropomum*) in central Amazon flood plain lake. *Aquaculture* :253(1-4) 274-384. Doi 10.1016/ *Journal of Aquaculture*:08.020.
- Haniffa, M.A., Merlin, R.T. and Francis T. (2000). Induced spawning of the striped mullet *Channa stratus* pituitary extracts, human chorionic gonadotropin Luteinizing hormone/releasing hormone analogue and ovaprim *Acta ichthyologica piscivorus* 30:53.6.
- Hecht, T. Oelmann L., Verheest L. (1996). Perspectives on Clarrid catfish culture in

- Africa. *Aquatic living Resource* 9:197-206.
- Hill, J.O., Coates, D.E, Whitebread, A.M., Clem, R.L., Robertson, M.J., Pengelly, B.c., (2009) Seasonal changes in pasture quality and diet selection and their relationship with weight gain of steers grazing tropical grass and grass –legume pastures in North Australia. *Animal production science* 49(11)983-993. [www.feedipedia.org/node/14684](http://www.feedipedia.org/node/14684)
- Khan, H.A., Gupta, S.D., Reddy P.V.G.K., and Saboo, S.K., (1986). Use of milk, Urea, Sodiamsulphite and human urine for degumming fertilized eggs of Common carp (*Cyprinus carpio* .L.) *Aquacultura Hungarica* (szarvas) 5;47-54.
- KhatuneJannat, M., Rahman M.M., Bashir M.A., Hassan M.N, Ahamed F. (2012)Effects of stocking density on survival, growth and production of Thai climbing perch (*Anabastudineus*) under fed ponds. *Sains Malaysia* 41(2012) pp1205-1210.
- Kristiansen, T.S., Ferno, A., Holm, J.C., Privitera, L., Bakke S., Fosseedengen, J.E (2004) Swimming behavior as indicator of low growth rate and impaired welfare in Atlantic halibut (*Hippoglossu shippoglossus* L) reared at three stocking densities. *Aquaculture* 230(1-4); 137-151.
- Leatherland, J.F, Cho C.Y (1985). Effect of rearing density on thyroid and internal gland activity and plasma and hepatic metabolite levels in rain bow trout (*Salmo gairdneri*) Richardson. *Journal of Fisheries Biology* 1985, 27(5): 583-592.
- Li, K.M, Gottman, H., Chapman, G., Funge-Smith, S. (2004). Culture of fish in rice fields. Food and Agriculture Organization. [www.FAO.org/3/9-90823e.pdf](http://www.FAO.org/3/9-90823e.pdf).
- Mihelakaks, A.H., Azaddine, A., Mohamed, I., Ali, A.A.,(2002). Broodstock feeding effects on spawning performance (fertility, eggs, and larvae quality of the Mediterranean red (Pagrus pagrus Linnaeus 1758 during two years. <https://www.researchgate.net/--/242232072>.
- Mittlemark, P. and Kapuscinski, C.E.(2006). Induced Reproduction in fish [Http://www.seagrant.umn.edu/Aqua/induce .Html](http://www.seagrant.umn.edu/Aqua/induce.Html)
- Ng, W.K., (2002). Potential of Palm oil utilization in Aquaculture feeds. *Asia Pacific Journal on Nutrition*. 5473 – 5476.
- Omosho, J.S., (1988). The Development of Eggs and Larvae in *Tilapia zilli* (Gervais). *Nigerian Journal of Science*. 22(1,2):24-28.
- Osofero, S. A., Otubusin S. O., Daramola, J. A (2009). The effect of stocking density on tilapia (*Oreochromis niloticus* Linnaeus 1757) growth and survival in bamboo net cages trial. *African Journal of Biotechnology* 8: pp 1322-1325.
- Page, F.H, Frank. K.T., (1989). Spawning time and egg stage duration in Northwest Atlantic haddock (*Melanogrammus aeglefinus*) stocks with emphasis on Georges and Browns Bank. *Canadian Journal of Fisheries Aquatic Science*. 46(1): 68–81.
- Pouey, J. L.O.F., Piedras S.R.N, Rocha C. B. Tavares R. A., Santos J.D.M (2011). Production performance of silver catfish, *Rhamdia quelen*, Juveniles stocked at different densities. *Ars veterinaria*, 27: pp 241-24
- Ridha, M. T (2005). Comparative study of growth performance of three strains of Nile tilapia *Oreochromis niloticus* at two stocking densities. *Aquatic Research* 37: 172-179.
- Ridha, M. T (2005). Comparative study of growth performance of three strains of Nile tilapia *Oreochromis niloticus* at two stocking densities. *Aquatic Research* 37: 172-179.
- Rottman, R.W., Graves J.S., Watson C. and Yanog R.P.E (2003). Culture Techniques of Moina. University of Florida, IFAS Extension, Circular 1054.
- Rowland, K.J.W, Smith, S.D.A., Cowden, K.L., Purcell, S.W. (2006). Effects of stocking density on the performance of the Australian fresh water silver perch (*Bidyanus bidyanus*) in cages. *Aquaculture*: 253(1-40) 301-308.
- Russel, A.J.M, Grotz P.A. Kriesemer S.K., Pems D.E (2008) Country case study; development and status of freshwater Aquaculture in Malawi. World Fish center studies and reviews vii +55pp.
- Sharma, J.G., Chakrabarti, R., (1998). Effect of different of different stocking densities on survival and growth of grass carp (*Ctenopharyngodon idella*) larvae using a recirculatory culture system. *Journal of applied Aquaculture* 8:(3) 79-83.
- Shearer, K., (1994). Effects of different feeding frequencies on fish growth. K SU Aquaculture. [www.ksoaquaculture.org/pdfs /.../hstb%20feeding%20frequencies%2000pdf](http://www.ksoaquaculture.org/pdfs/.../hstb%20feeding%20frequencies%2000pdf).
- Southworth, B.E, Engle C.R, Ruebush K. (2009). The effect of under stocking of channel Cat fish stockers in multiple batch production. *Journal of Applied aquaculture*, 21: 21-30.
- Tave, D (1986). Genetics for Fish Hatchery

- Managers. Avi publishing company, Westport, Connecticut (1986).
- Viveen W.J.A.R., Riditer, J.J.C., Oordtrot, P.G., Janseen, W.I. and Huisman, E.A. (1986) Practical Manual for the Culture of *Clarias gariepinus* Joint Publication of : Director-General International cooperation of the Ministry of foreign affairs, the Hague, the Netherlands. Department of Fish culture and fisheries of the Agricultural University of wageningen, The Netherlands Research Group for comparative Endocom, Department of Zoology, University of Utrecht, the Netherlands. pp 50 - 60.
- Yang, S.D., Lin, T.S., Peng, H.K (2007). Influence of dietary protein level on growth performance, a carcass composition and liver lipid classes of juvenile, *Spinibarbus hollandi (Oshima)* *Aquatic Resources* 2003: 34(8) 661-666.
- Yanog, F.A., Teles-Oliva, A. (2008). Nutrition and Health of Aquaculture. Fish-www. vesc. b./cursos/posgraduacac/./nutritionrition and health of aquaculture – fish.pdf.
- Zhu, Y.J., Yang D. G, Chen J.W., Yi, J. F., Liv W.C (2011). An *evaluation* of stocking density in the cage culture efficiency of Amur sturgeon, *Acipenserschrenckii* *Journal of Applied Ichthyology*, 27 pp. 545-549.