# THE INFLUENCE OF STOCKING DENSITY ON GROWTH PERFORMANCE OF JUVENILE JAPANESE ORNAMENTAL CARP (KOI, *CYPRINUS CARPIO* L.)

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#### Abstract

The identification of suitable fish stocking density proves to be essential in order to elaborate a sustainable fish rearing technology. The aim of present study is to identify a proper stocking density for juvenile ornamental carp, that maximizes the productivity of this specie, in semi-intensive aquaculture production systems. Thus, two ornamental carp stocking densities were tested, in duplicate  $(V_1 - 6.3 \text{ kg/m}^3)$ , respectively  $V_2 - 7.01 \text{ kg/m}^3)$ . The results indicate a better feed conversion ratio (FCR) in case of  $V_1$ , compared to  $V_2$ . Also,  $V_1$  registered higher specific growth rate (SGR) values and a better protein efficiency ratio (PER). However, in order to maintain the technological water quality within the optimum range for rearing ornamental carp, a higher water exchange rate was applied at  $V_2$ , compared to  $V_1$ , especially in the first part of the experimental period. The FCR registered better values in the first part (1.5% feeding ratio was applied), compared to the second part of the experimental period (1% feeding ratio). However, if production maximization is required, higher ornamental carp stocking densities and feeding ratio can be applied if water quality module will be improved.

Key words: density, growth, ornamental carp.

### **INTRODUCTION**

Across the world, aquaculture is growing rapidly due to the action of two important factors: the growing demand for seafood and the low fish stocks in the world's oceans.

Japanese colored carp, called Nishikigoi in Japan (koi for short), originated from the common carp, specie raised for human consumption by the rice farmers in Niigata Prefecture. Carp were imported from China into Japan around AD 1500, although the Chinese had been raising carp for food as early as 2000 BC.

The ornamental carp, *Cyprinus carpio* L. (koi carp) is the king of pond fish, and it is currently enjoying the attention of many aquaculturists around the world. Growing valuable varieties of ornamental carp is by far the most profitable of the branches of ornamental aquaculture that can be successfully implemented in our country.

The production of the colored carp - the Japanese "nishikigoi" - presently exceeds in

monetary value the production of carp as human food. The nishikigoi as "swimming flowers" delight modern people as much as the taste of carp delighted the Romans at the beginning of carp domestication (Eugene, 1995).

Because of their large size (up to 1 meter in length), longevity (60 years or more), beautiful colours, friendly personalities and high value (some koi exemplars have sold for over a hundred thousand dollars), koi are one of the most likely fish species to be seen by veterinarians.

However, since koi carp is growth in various aquaculture production systems, their growth and physiological performance are affected by various environmental and technological conditions. Stocking density, an important technological factor, generally negatively affects fish growth and welfare (Quan et al., 2020). In aquaculture, stocking density describes the number of fish that are stocked initially per unit area; however, it is generally used to refer to the density of fish at any point of time (Eduardo et al., 2020).

Therefore, in order to be cost-efficient, aquaculture facilities must optimize their rearing technologies in terms of stocking densities. However, several types of production facilities, based on recirculating aquaculture systems (RAS) or partial RAS are forced to practice high stocking densities since they register significant high value of operational costs.

Thus, the identification of proper fish stocking densities for certain type of production systems is essential in order to maximize system economic efficiency and productivity.

### MATERIALS AND METHODS

The trials were carried out at the Aquaculture Research Centre of Food Science and Engineering Faculty during a 31 days experimental period. The aquaculture equipment was a 180 L circulating aquaculture glass tank. The experimental design was made in duplicate, by using four rearing units equipped with independent water conditioning modules. Thus, for biological, chemical and mechanical filtration, each rearing unit was connected to a Hagen AquaClear power filter (500 L/hour flow capacity), while for maintaining the oxygen concentration of technological water within optimum limits, a Resun Air Pump (1.6 L/min) was used. A daily water exchange rate of 40% was applied in order to assure optimum growth conditions for the biological material.

superior filtering module provides The biological, mechanical and chemical filtration. The area of the filter sponge and the carbon particles increases the filtering capacity. The sponges can be easily washed and reused, allowing the conservation of colonies of biological bacteria. The carbon area of the filter removes dissolved organic compounds, while the ceramic components provide a favourable environment for growing colonies of bacteria that convert ammonia and nitrites into nitrates. It should be mentioned that the sponge area washing was done with dechlorinated water, in order not to affect the colonies of bacteria.

The biological material consists in koi carp exemplars, reared by applying two stocking densities:  $V_1 - 6.3 \text{ kg/m}^3$ , respectively  $V_2 - 7.01 \text{ kg/m}^3$ , in duplicate.

The temperature, pH and dissolved oxygen (DO) were determined daily, using a pH meter WTW - pH 340, respectively a WTW Oxi 315 I temperature and DO meter.

In both experimental variants the biological material was fed with NUTRA pellets which contain fish meal, cereal and cereal by-products, oils, antioxidants (BHT). The biochemical composition of pellets is presented in Table 1.

Biochemical UΜ Concentration composition Crude protein % 54 % Crude fat 18 Cellulose % 0.6 Ash % 10 Phosphorus % 1.45 18000 Vit. A U.I./kg Vit. D3 U.I./kg 1800 Vit. E 500 mg 4.5 Cu (CuSO4) mg

Table 1. Biochemical composition of NUTRA pellets

For the calculation of feeding rate, it is necessary to know the relations between the nutritional requirements of the fish and the environmental conditions. However, it is recommended that the determination of feed quantities and their correction be based not only on the data of the feeding schedule, but also on the actual information and observations obtained during the growing period (Oprea and Georgescu, 2000).

Thus, fish were fed 3 times per day, using an average daily feeding rate of 1.25% from total fish body weight (BW), as follows: 1% BW in the first 14 days of the experimental period and 1.5% in the next 16 days, till the end of the trial. The total fish biomass was determined every 2 weeks in order to adjust the amount of administrated feed. Under growth conditions in the closed system, obtaining the maximum biomass production is essential in order to be profitable.

Thus, this requires high biomass growth rate, respectively a period of time as short as possible to reach the marketable size. To ensure this desideratum, fish are usually fed with granulated feed with high protein content. For these reasons, feed with 54% protein content was used in present experiment.

Artificial feeding is an integral part of managed fish culture practices, where the focus is on

maximizing fish production with minimum feed cost.

The analysed technological indicators were as follows: biomass gain (BG - g fish<sup>-1</sup>), relative grow rate (RGR - g g<sup>-1</sup> day<sup>-1</sup>), specific grow rate (SGR - % fish biomass day<sup>-1</sup>), feed conversion ratio (FCR) and protein efficiency ratio (PER). These technological indicators were determined by using the following formulas: (Seyyed et al., 2020; Petrea et al., 2017):

Total biomass gain: TBG = TBf - TBi
[g] where: TBf - total final fish biomass; TBi - total initial fish biomass;

2. Relative growth rate:  $RGR = ((TBf-TBi)/t)/TBi [g g^{-1} day^{-1}]$ , where: t - duration of the experiment;

3. Specific growth rate: SGR = 100 \* (ln TBf - ln TBi)/t [% fish biomass day<sup>-1</sup>];

4. Feed conversion ratio: FCR = TF/TBG, where: TF - total feed intake, TBG - total biomass gain;

5. Protein efficiency ratio: PER = TBG/(TF \* CP/100), where: CP - crude protein.

## **RESULTS AND DISCUSSIONS**

The sale of ornamental carp is, from a financial point of view, superior to that of rainbow trout. The price for ornamental carp brood is 5-6 euros for a single fish. Viewed in our country with some suspicion, in other countries, such as the Czech Republic, ornamental aquaculture brings income to the fisheries sector twice as high as the production of fish for consumption, of all species together.

The identification of suitable fish stocking density proves to be essential in order to elaborate a sustainable fish rearing technology.

The growth of numerous varieties of koi carp may be a profitable activity if the eco-biological requirements are properly known. In Romania, koi carp is starting to be more appreciated, being preferred by aquarium entrepreneurs due to good growth rate and high sell price. However, maintaining water quality parameters within an optimal range is essential for obtaining the maximum growth performance, associated to a certain fish rearing technology. Fish being poikilotherms, has the levels of their metabolism directly affected by the ambient temperature which can alter their levels of routine metabolism, food utilization and growth (Aleksander et al., 2010).

Different environmental factors play an important role in the growth and survival of fish. Temperature is probably the most important abiotic factor affecting life (Brett, 1979).

The activities of feeding, digestion and conversion of food are strongly influenced by the ambient temperature, which is ultimately reflected in the variation of the growth rate. Temperature affects the rate of food digestion by influencing the activity of digestive enzymes (Scerbina and Kazlauskiene, 1971).

The optimum temperature required for growth and other physiological activities varies greatly depending on the species. Each species has an optimum temperature for growth which is probably determined by the optimum temperature for growth which is probably determined by the optimum temperature for the specific activity of the enzymes (Davis and Parker, 1990).

The ornamental carp tolerates a fairly large temperature range, between 3-32°C, but the optimal temperature is 23-27°C.

At the optimum temperature, fish grow faster, efficiently convert food and are relatively more resistant to disease.

Thus, for present study, the technological water temperature registered values within optimum limits (20-23.5°C) for koi carp growth, with no significant differences (p>0.05) recorded between the experimental variants.

In both experimental variants, the fish showed an active feeding behaviour and a good state of health.

The pH registered values between 6.1-8.2 pH values. However, the pH registered an up/down trend, most probably due to daily water exchange rate (40%). Thus, the added water had a pH which varied between 7.8–8.5 upH (fig. 1, 2), generating therefore high pH values during the experimental period, an advantage for the technological water biological filtration process. The dissolved oxygen (DO) concentration in water registered mostly values within 4.4 and 6.5 mg L<sup>-1</sup>, with significant decreases after the feed administration (Figures 1 and 2).

However, it can be observed that DO values are lower in  $V_2$  experimental variant, compared to  $V_1$ , fact that can be explained by the applied stocking density, since a high stocking density will generate more metabolic fish wastes (Figures 1 and 2).



Figure 1. The dynamics of pH and DO for  $V_1$  experimental variant

Stocking density is a major factor determining fish production and farm profitability because it directly influences fish survival, growth, behaviour, health, water quality and feeding (Vincent & Neill, 2019).



Figure 2. The dynamics of pH and DO for  $V_2$  experimental variant

Increased stocking density causes stress and water quality deterioration. Stressful conditions lead to cortisol release, which chronically threaten the fish health and welfare and cause disease outbreak and fish loss; thus, it is necessary to find methods to suppress such a stress (Seyyed et al., 2020).

Technological performance plays an important role in the operation of a recirculation system, depending on a multitude of factors including meeting the nutritional requirements of the crop species and maximizing feeding efficiency, and optimizing the feed conversion coefficient. The capacity of a recirculation system is not determined by the volume of water contained, which has the role of ensuring only a minimum dilution of the residues produced, or the density of fish fillings, but by the ability of aeration and filtration systems to maintain water quality in the optimal range during the most intense feeding.

Usually, the measure of the intensity of a closed growth system is expressed by the population density, i.e. the amount of biomass per unit volume (Cristea et al., 2002).

Popular density, expressed in this way, is not however, the most suggestive measure of expressing the production capacity of a closed production system.

The performance of a system is assessed, first of all, depending on the level of feeding intensity that can be sustained by the system, a level expressed as a percentage of the crop biomass.

The time required for the fish to grow to marketable size is also an important criterion for assessing the performance of such a system. Reducing the time required to obtain a marketable size of fish requires the administration of an optimal level of food.

The fish growth performance parameters registered at both experimental variants are presented (Table 2 and Table 3).

The average specific growth rate indicates a superior fish production at V1 (6.3 kg/m<sup>3</sup>), compared to V2 (7.01 kg/m<sup>3</sup>) experimental variant (Tables 2 and 3). Also, from the perspective of feeding strategy efficiency, the average food conversion ratio (FCR) indicates better values for V1 (6.3 kg/m<sup>3</sup>) experimental variant, compared to V2 (7.01 kg/m<sup>3</sup>) (Figure 3).

Table 2. The growth performance parameters for V<sub>1</sub> experimental variant

Indicator	Recorded
	values
Experimental period (days)	31
Total administrated feed quantity (g)	510
Total biomass gain (g)	435
Survival (%)	100
FCR	1.17
RGR (g g-1 day-1)	0.012
SGR (%/day)	1.06
PER	1.28

The protein efficiency ratio (PER) has registered higher values at V1 ( $6.3 \text{ kg/m}^3$ ) experimental

variant, compared to V2 (7.01 kg/m<sup>3</sup>), revealing the ability of fish organism to utilize proteins, which positively affects growth rate.

Table 3. The growth performance parameters for V<sub>2</sub> experimental variant

Indicator	Recorded values
Experimental period (days)	31
Total administrated feed quantity (g)	375
Total biomass gain (g)	183
Survival (%)	100
FCR	2.04
RGR (g g-1 day-1)	0.009
SGR (%/day)	0.43
PER	1.13



Figure 3. The FCR for both variants

The relative growth rate registered significant better values at  $V_1$  (6.3 kg/m<sup>3</sup>), compared to  $V_2$  (7.01 kg/m<sup>3</sup>) (Figure 4).



Figure 4. The SGR for both variants

In accordance with our findings, Hussain et al., (2014) have reported that the SGR of Koi carp cultured in intensive systems was strongly reduced with increasing density of fish from 1.4 kg m<sup>3</sup> to 2.1 kg m<sup>3</sup>, respectively to 2.8 kg m<sup>3</sup>. Also, according to Hussain et al. (2014) findings, the mortality of Koi carp increased (from 0% to 2%) when the density of fish increased from 2.1 kg m<sup>3</sup> to 2.8 kg m<sup>3</sup>.

Also, according to Bahremand and Soleimanirad, 2017, a stocking density more than 150 fish per  $m^3$ , leads to growth reduction and incidence of stressful conditions in koi carp biomass. Also, same study he pointed out that the optimal stocking density, was 150 fish per  $m^3$ .

Considering the modern aquaculture sector, different types of aquaculture systems such as recirculating aquaculture systems (RAS), aquaponics and biofloc systems were established to conserve and minimize the water usage and to optimize the production. In a RAS, wastewater is reused after appropriate treatment and conditioning. It is one of the most intriguing strategies for intensifying aquaculture production while simultaneously reducing wastes (Nuwansi et al., 2019).

#### CONCLUSIONS

It can be concluded that stocking density of koi carp had a major impact on fish growth performance parameters (FCR, SGR, RGR, PER), since better values were registered at  $V_1$ experimental variant.

Therefore, it can be stated that, in present research, during the analyzed koi carp development stage, a better cost efficiency as well as production maximization is registered if applying a  $6.3 \text{ kg/m}^3$  stocking density. However, if water conditioning units which are integrated in the production system are upgraded, higher ornamental carp stocking densities may be possible to be applied, therefore improving the production technology and its performance.

### ACKNOWLEDGEMENTS

This work was supported by Center for Scientific Research "Romania Center for Modeling of Recirculating Aquaculture Systems" - (MoRAS), from University Dunărea de Jos, of Galați which was established as a result of the implementation of the POSCCE project 622 / 11.03.2014.

#### REFERENCES

Aleksander, I., Radoslav, D., Jelena, B., Tibor, K., Branislav, M., Svetlana, L., Tamara, G. C., Marija, V. (2010). FCR and growth rate of common carp *Cyprinus carpio* Linnaeus 1758 at different water temperatures. *Proceedings of Eco-Conference Ecological Movement of Novi Sad*, 273-280.

- Bahremand, M., Soleimanirad, A. (2017). Effects of stocking density on growth performance, immune and stress responses in koi carp (*Cyprinus carpio* var. Koi (Linnaeus, 1758)). *Journal of Aquatic Ecology*, 6 (4), 10-20.
- Brett, J. R. (1979). *Environmental factors and growth. In Fish physiology.* New York, USA: Academic Press Publishing House.
- Cristea, V., Grecu, I., Ceapa, C. (2002). Engineering of recirculating systems in aquaculture. Bucharest, RO: Didactic and Pedagogical Publishing House.
- Davis, K. B., Parker, N. C. (1990). Physiological stress in striped bass: effect of acclimation temperature. *Aquaculture*, 91, 349 358.
- Eduardo, K., Battistia, A., Rabaiolib, J., Uczayc, F., Jonas, S., Rafael, L. (2020). Effect of stocking density on growth, hematological and biochemical parameters and antioxidant status of silver catfish (*Rhamdia quelen*) cultured in a biofloc system. *Aquaculture*, 524, 1-6.
- Eugene, K. B. (1995). Origin and domestication of the wild carp, Cyprinus carpio: from Roman gourmets to the swimming flowers. *Aquaculture*, 129 (1-4), 3-48.
- Hussain, T, Verma, A. K, Tiwari, V. K, Prakash, C., Rathore, G., Shete, A. P., Nuwansi, K. K. T. (2014). Optimizing koi carp, *Cyprinus carpio* var. koi (Linnaeus, 1758), stocking density and nutrient recycling with spinach in an aquaponic system. *Journal of the World Aquaculture Society*, 45(6), 652– 661.
- Nuwansi, K. K. T., Verma, A. K., Rathore, G., Chandra, P., Chandrakant, M. H., Prabhath, G. P. W. A. (2019).

Utilization of phytoremediated aquaculture wastewater for production of koi carp (*Cyprinus carpio* var. koi) and gotukola (*Centella asiatica*) in an Aquaponics. *Aquaculture*, 507, 361–369.

- Oprea, L., Georgescu, R. (2000). Nutrition and feeding of *fish*. Bucharest, RO: Technica Publishing House.
- Petrea, S. M., Mogodan, A., Metaxa, I., Plăcintă, S., Vasile, M. A., Huian, G. (2017). A comparative study on the evaluation of cyprinids growth performance in IMTA systems. *AACL Bioflux*, 10(1), 87 – 102.
- Quan, Y., Liang, G., Bao-Suo, L., Hua, Y., Guo, K. C. Z., Nan, Z., Shi, G., Jiang, D., Chang, Z. (2019). Effects of stocking density on the growth performance, serum biochemistry, muscle composition and HSP70 gene expression of juvenile golden pompano *Trachinotus ovatus* (Linnaeus, 1758). *Aquaculture*, 518, 1-8.
- Scerbina, M. A., Kazlauskiene, O. P. (1971). Temperature regime of water and digestibility of nutrient by Cyprinus carpio L. *Gidrobiologicheskii Zhurnal*, 7(3), 49-53.
- Seyyed, M., Hoseinia, A., Taheri, M., Melika, G., Esmaeil, P., Yousef, I., Abdolvahhab, K. (2020). Effects of dietary tryptophan supplementation and stocking density on growth performance and stress responses in rainbow trout (*Oncorhynchus mykiss*). *Aquaculture*, 519, 1-8.
- Vincent, O., Neill, J. G. (2019). The effect of stocking density on profitability of African catfish (*Clarias* gariepinus) culture in extensive pond systems. Aquaculture, 507, 385-392.