Bulletin UASVM, Veterinary Medicine 68(1)/2011
pISSN 1843-5270; eISSN 1843-5378

Assessment of the Risk Level for Carp Welfare Based on the Water Quality in RAS

Renata RELIC[1)], Milos CIRIC[2)], Marijana VUCINIC[3)], Brana RADENKOVIC-DAMNJANOVIC[3)], Vesna POLEKSIC[1)], Zoran MARKOVIC[1)]

[1)] University of Belgrade, Faculty of agriculture, Nemanjina 6, Belgrade-Zemun, Serbia
rrelic@agrif.bg.ac.rs
[2)] Institute of Chemistry, Technology and Metallurgy, Karnegijeva 4, Belgrade, Serbia
[3)] University of Belgrade, Faculty of veterinary medicine, Bulevar oslobodjenja 18, Belgrade, Serbia

Abstract. In this paper results of the risk level assessment of carp welfare (*Cyprinus carpio*) in recirculating aquaculture system (RAS) are presented. We used different quantitative nutrition levels (3% and 4% of feed) and examined changes of the water quality in tanks during first, second and third month of the experiment. The risk assessment is based on carp needs regarding optimal ranges of water temperature, dissolved oxygen (DO), and suggested upper level of unionized ammonia (NH_3-N) content. According to the results, average values of the water temperature and DO were in optimal range. The average content of NH_3-N was above recommended level. Water quality was deteriorated along with duration of experiment. In the first period i.e. first month of the experiment welfare risk in both groups is at the lowest level and the highest in the last period. The total score is higher in group fed with 4% of feed then in group which received 3% of feed.

Keywords: carp, water quality, welfare, risk assessment

INTRODUCTION

For the welfare of farmed fish, the quality of the water is central. It is a primary environmental consideration, with the potential to markedly affect health. Inappropriate levels of water quality parameters affect physiology, growth rate and efficiency, cause pathological changes and organ damage and, in severe cases, cause mortality. The sub-lethal effects of poor water quality are also commonly linked to increased disease susceptibility (MacIntyre *et al*., 2008).

Considering that poor animal health has direct impacts on reducing production and increasing costs, maintaining good water quality conditions is of primary importance in any type of aquaculture system, especially in recirculating aquaculture system (RAS). The most important disadvantage in RAS is the deterioration of the water quality (Isla Molleda, 2008). The water quality in RAS depends on different factors most importantly the source, the level of recirculation, the species being cultured and the waste water treatment process within the system (Losordo *et al*., 1999). Most water quality problems experienced in RAS were associated with low dissolved oxygen and high fish waste metabolite concentrations in the culture water (Sanni and Forsberg, 1996). Waste metabolites production of concern include total ammonia nitrogen (TAN), unionized ammonia (NH_3-N), nitrite (NO_2-N), nitrate (NO_3-N), dissolved carbon dioxide (CO2), suspended solids (SS), and non-biodegradable organic matter. Knowing values of certain physical, chemical and biological properties of the water in RAS allows estimation of the system functioning and making necessary corrections.

The use of data about water quality in fish welfare assessment can be different. Mostly it is a scoring system based on results of multivariate analysis as in paper by Turnbull *et al*. (2005). However, in some publications for assessment of farm animal welfare (including fish) terminology and methodology of the food risk assessment are used (Müller-Graf, 2007; EFSA, 2008; EFSA, 2010).

Risk assessment is a systematic, scientific-based process to estimate the magnitude of exposure to a hazard and includes 4 steps: hazard identification, hazard characterization, exposure assessment and risk characterization. A hazard in animal welfare risk assessment is a factor with a potential to cause a negative animal welfare effect (adverse effect) and a risk is a function of the probability of occurrence and the consequences of occurrence.

Applying of this method in carp production is described by EFSA (2008). We tried to use the method in practical conditions in the Center for Fishery and Applied Hydrobiology. The Center belongs to the Faculty of Agriculture University of Belgrade and it is a leading institution in Serbia for research in the field of cyprinid fish breeding, nutrition, and fish stress. Activities of the Center are aimed at improvement of fish rearing conditions in ponds and tanks according to international standards and scientific data. The welfare assessment is integrated with current experiments in the Center, as is described in papers by Hristov *et al*. (2009) and Relić *et al*. (2010a,b,c). The aim of this paper is to present a part of results from researches conducted in the Center.

MATERIALS AND METHODS

This examination is conveyed in total 96 days, divided in three periods (each is 32 days long) and in tanks which belongs to the recirculating aquaculture system (RAS). Total 240 fingerlings of carp (*Cyprinus carpio*) were distributed in two groups and fed with 3% or 4% of commercial extruded feed (38% of proteins and 12% fat, Veterinarski zavod Subotica, Serbia) in relation to ichthyomass. The average individual body mass was 6,43 ± 0,02 g, and the fish density in tanks was 2,14 kg/m^3.

The quality of the water is monitored during the whole period, whereby sampling of the water and direct measures were done at the same time in the morning (8 AM), before food distribution. Water temperature and dissolved oxygen (DO) have been measured directly in tanks twice a week and by appropriate probe (MULTI 340i/SET, WTW, Germany). Values of unionized ammonia (NH_3-N) in the water samples were measured twice a month by spectrophotometer (PhotoLab® 6100 VIS, WTW, Germany).

Water quality parameters are selected according to their significance for fingerling welfare, and hazards are defined by EFSA (2008). However, longtime shortage of natural food as important welfare hazard was not taken into account in these experimental conditions. The risk score for each hazard was calculated using the formula:

Risk score = (severity/4)*(duration of effect)*(proportion of the population affected/5)*(frequency of hazard/5)*100

The data were processed using descriptive statistics and standard parametric (ANOVA, LSD) or nonparametrics tests (Kruskal-Wallis ANOVA, Mann-Whitney U test), depending on the character of data.

RESULTS AND DISCUSSIONS

In tables 1 and 2 average values of water quality parameters through three periods are presented:

Tab.1.

Descriptive statistic for the water quality parameters in group fed 3% of feed in relation to ichthyomass

Parameters	Periods	Mean*	SD**	Min	Max	Cv**
Water temperature (°C)	I	23.11 ^{A}	1.52	20.06	24.70	6.57
	II	20.41 ^{B}	2.19	16.30	24.10	10.73
	III	20.98 ^{B}	1.25	18.80	24.00	5.94
Dissolved oxygen (mg/l)	I	3.88 ^{aA}	0.42	3.47	5.35	10.75
	II	4.73 ^{b}	0.43	4.09	5.53	9.03
	III	5.84 ^{aB}	2.04	2.02	9.00	34.89
Unionized ammonia (mg/l)	I	0.29 ^{ab}	0.04	0.26	0.33	12.30
	II	0.27 ^{a}	0.17	0.10	0.53	65.17
	III	0.45 ^{b}	0.27	0.04	0.98	59.19

* a, b – significant difference (p < 0,05) between values marked by different letters; A, B – very significant
difference (p < 0,01) between values marked by different letters; There is no significant difference between values
marked by the same letter (p > 0,05)
**SD-standard deviation; Cv- coefficient of variation (%)

Table 1 shows that average values of the water temperature were in optimal range for carp according to Schreckenbach (2002). The highest average temperature was in the first period (23.11 ± 1.52°C) and the difference between first and other periods is significant (P < 0.01). The lowest average water temperature was in the second period (20.41 ± 2.19°C), with more frequent variations of values comparing to other periods and the lowest single temperature value (16.30°C). This temperature is not detrimental on welfare per se but could decrease food intake and metabolic processes in carp as well as temperatures less than 20°C i.e. temperature out of optimal range (Johansen *et al*., 2006; Szumiec, 1984). Rapid temperature change rather than absolute limits have more serious negative effect on welfare, as is described by EFSA (2008).

Amount of oxygen dissolved in the water was in increase, and in first period (3.88 ± 0.42 mg/l) is significantly lower (P < 0.01) than in third period (5.84 ± 2.04 mg/l). Difference between the second and other periods is significant (P < 0.05). Average values of DO were satisfactory according to Poli (2009), with greatest variation in the third period. In that period minimal value which can negatively affect carp welfare was measured (2.02 mg/l).

Average content of unionized ammonia was also in increase, and differences are significant (P < 0.05) between values in second (0.27 ± 0.17 mg/l) and third period (0.45 ± 0.27 mg/l). NH_3-N had a great variability in second and third period of examination. All average and the most of single values are above suggested for carp, according to Svobodova *et al.* (1993). The upper limit for these authors is 0.05 mg/l of NH_3-N although Poli (2009) cited higher value as a boundary (i.e. 0.1 mg/l). Ammonia concentrations in one system may affect fish health while the same concentration in another system may have no affect (Meade, 1985). However, due to well-known toxicity of unionized ammonia and its' potentially harmful effects on fish we took lower recommended value as a limit.

Group fed with 4% of food has the similar results which are presented in the following table:

Tab.2.

Descriptive statistics for the water quality parameters in group fed 4% of feed in relation to ichthyomass

Parameters	Periods	Mean*	SD**	Min	Max	Cv**
Water temperature (°C)	I	23.06 [A]	1.40	20.07	24.80	6.07
	II	21.05 [B]	2.38	16.60	24.60	11.33
	III	20.94 [B]	1.42	18.60	24.20	6.76
Dissolved oxygen (mg/l)	I	3.82 [a]	0.26	3.43	4.59	6.72
	II	4.19 [a]	0.43	3.65	5.33	10.24
	III	4.28 [a]	1.94	0.93	8.32	45.36
Unionized ammonia (mg/l)	I	0.26 [a]	0.02	0.25	0.28	6.70
	II	0.26 [a]	0.12	0.10	0.38	46.72
	III	0.40 [a]	0.21	0.08	0.76	51.50

* a, b – significant difference (p < 0,05) between values marked by different letters; A, B – very significant
difference (p < 0,01) between values marked by different letters; There is no significant difference between values
marked by the same letter (p > 0,05)
**SD-standard deviation; Cv- coefficient of variation (%)

Average values of the water temperature were also in optimal range in this group: the lowest in the third period (20.94 ± 1.42°C), and the highest in the first period (23.06 ± 1.40°C). The difference between first and other periods is significant (P < 0.01) and the most frequent changes of the temperature were in the second period. As well as in previous group, the lowest value which can affect carp activities (16.60°C) is registered here.

Amount of DO was slightly increased, and differences between periods are not significant (P > 0.05). Average value is lowest in the first period (3.82 ± 0.26 mg/l) and highest in the third period (4.28 ± 1.94 mg/l). Average values of DO were in the optimal range, with greatest variation in the third period. The lowest measured value (0.93 mg/l) is very unfavorable although carp can tolerate it (Albers *et al.*, 1983). Welfare issues arise before lethal limits are reached (EFSA, 2008).

Periods statistically differ not in average content of NH_3-N (P > 0.05) despite slight increase in the third period (0.40 ± 0.21 mg/l) compared to the first and second period (respectively, 0.26 ± 0.02 and 0.26 ± 0.12 mg/l). In this group the higher variability of NH_3-N in the third period is present. The average and single values are above those which are suggested for carp, according to Svobodova *et al.* (1993).

Applied treatment with different quantity of feed showed significant influence on the quantity of DO (P < 0.01) and no influence on the water temperature and NH_3-N content (P > 0.05). All three parameters were significantly influenced by period (P < 0.01). For example, water temperature in tanks is related to the temperature of inlet water and seasonal fall of air temperature because the water and/or the hall were not heated. Furthermore, changes in the temperature affected solubility of oxygen in the water (EFSA, 2008) and oxygen is consumed in the process of organic matter decomposition (Marković and Tutundžić, 2003).With the progress in duration of this study organic matter gradually accumulated. It was reflected in increasing of NH_3-N content in the water.

According to data (Albers *et al.*, 1983; Sollid *et al.*, 2003; Svobodova *et al.*, 1993) carp and other Cyprinids can survive even in the water of poor quality. Levels of the potential negative influence of the water quality on carp welfare in our experiment are presented in the table 3.

Tab.3.

Results of welfare risk analysis

Hazard	Description of the hazard	Final risk score depending on food quantity and duration of the exposure					
		3% of food			4% of food		
		day 1-32	day 32-64	day 64 -96	day 1-32	day 32-64	day 64 -96
Water oxygen content	too low	0	0	16	0	0	24
Water temperature	too low	0	54	30	0	48	36
Unionized ammonia content	too high	132	264	352	132	264	396
Water temperature	rapid change	16	16	12	8	16	12
Water temperature	too high	0	0	0	0	0	0
Total		148	334	410	140	328	468
Total for all 96 days		892			936		

Results from table 3 showed changes in water quality along with duration of experiment. These data are in accordance with our results of analysis of variance and results of Isla Molleda (2008). In the first period welfare risk for both groups was at the lowest level in contrast to the last 32 days of the experiment when the risk was the highest. Total score was higher in group fed 4% of food (936) then in group which received 3% of feed in relation to ichthyomass (892).

In rearing systems much more factors than those from Table 3 can influence carp welfare, but selected parameters have the most serious effects and can cause mortality of fish. This method of the risk assessment does not include interactions between the hazards. The scoring system allows hazards to be ranked; however, the scores need to be interpreted with caution. Higher score for one hazard does not mean higher importance of that hazard comparing to others (EFSA, 2008). This is supported by the fact that fish tolerate constant concentrations of ammonia better than fluctuating levels (Thurston *et al.*, 1981).

Assessment of the welfare risk should point out the weaknesses in the rearing system for the same life stage, and in different periods (EFSA, 2008; Relić *et al.*, 2010b,c) which is applicable in our research. For example, high values of NH_3-N (in Tab. 1 and 2) and high score for this parameter (in Tab. 3) indicate the capacity and stability of the system under given conditions of rearing and nutrition. This can be useful for further use of the system in terms of appropriate technological procedures application.

CONCLUSIONS

- Presented data have shown that conditions in RAS during 96 days were optimal for carp considering average values of the water temperature and DO. The average values and the most single values of NH_3-N were above recommended level.
- Based on results of the welfare risk analysis water quality was gradually deteriorated. The total score is higher in group fed with 4% of feed then in group which received 3% of feed. The welfare risk level is the lowest for both groups in the first month and the highest in the third month of study. This indicates that system under given conditions of fish density and level of nutrition can operate for one month without compromising carp welfare.

ACKNOWLEDGEMENTS

This paper is financed by project of the Ministry of Science and Technological development of Republic of Serbia TR 31075.

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