Impacts of High Concentration of CO_2 on the Serum Biochemistry and Carbonic Anhydrase Enzyme Activity of Rainbow Trout, *Oncorhynchus mykiss*

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Abstract. In this study, the physiological effects that the increasing carbondioxide concentrations on rainbow trout, *Oncorhynchus mykiss* is evaluated by using serum biochemical variables and carbonic anhydrase activities. The fish were exposed for 14 days to 14 mg/L concentrations of CO_2. The serum GLU value showed a significant increase in the group exposed to CO_2 compared to the control group at days 14. Serum TRI, COL and blood CA values showed a significant decrease in the group exposed to CO_2 at day 7 compared to the control group. The TRI value a statistically significant increase in the group exposed to CO_2 at day 14 compared to the control group. In conclusion, this study results indicate that the some serum biochemical variables and blood carbonic anhydrase activity of rainbow trout significantly affected by high level of CO_2.

Keywords: *Oncorhynchus mykiss*, carbondioxide, blood, carbonic anhydrase

1 Introduction

It is now recognized that the 21st century will show a significant global warming trend induced by an increase in atmospheric greenhouse gases (Houghton et al., 2001). Carbon dioxide (CO2), one of the important green gases, has increased by 40% from pre-industrial levels from approximately 280 parts per million by volume (ppmv) in the 18th century to 390 ppmv in 2010 (IPCC 2007). Water sources are attractive sites for possible storage of CO2. Addition of CO2 to the water will result in a decrease in pH due to the bicarbonate buffer system in sea- and fresh-water. It is supposed that disposal of sufficient CO2 to stabilize atmospheric levels at twice the pre-industrial level by the end of this century would lower the pH of the entire water

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sources on average by more than 0.1 units (Caldeira and Wicket 2003). Addition of CO2 to the water will result in a decrease in pH due to the bicarbonate buffer system in seawater and freshwater. This is a large fraction of the normal variation of pH in open water sources. Research interest in CO2-driven water acidification has been centred on certain groups of calcifying water organisms, but knowledge on the possible impacts of water acidification on fish is limited.

The purpose of the present study was to evaluate the impact of high water CO2 levels (14 mg/L) in freshwater on serum biochemical variables and carbonic anhydrase activities in rainbow trout, Oncorhynchus mykiss for 14 days.

2 Material and Method

The experiment was designed in triplicate and 12 fish were placed in each experimental tank (140 L). During the experiment, the fish were exposed for 14 days to 14 mg/L concentrations of CO2 by injecting CO2 (purity 99.9%) gas by means of ceramic diffusers. Control group was not exposed to CO2. In the experiment, five fish from each aquarium on the 7th and 14th day were used for analysis. The serum biochemical variables (glucose, total protein, albumin, triglyceride and cholesterol) in the blood serum was measured according to Yılmaz et al (in press). The CO2 hydratase activity of the CA enzyme was assayed colorimetrically by using the method of Wilbur and Anderson (1976). Each value was expressed as mean ± standard error (SE) for each parameter measured. Student's t-test was used to determine the significance of differences between the exposure group and control group. The statistical analyses were carried out by using SPSS 17.0, and the significance level was considered to be 0.05.

3 Results

In the present study, results (Table 1) showed that CO2 exposed group did not show differences of Tprot, ALB and GLO values at any of the two sampling periods as compared with the control group ($P>0.05$). However, the serum GLU value showed a significant increase in the group exposed to CO2 compared to the control group at days 14 ($P<0.05$). Serum TRI, COL and blood CA values showed a significant decrease in the group exposed to CO2 at day 7 compared to the control group ($P<0.05$). The TRI value a statistically significant increase in the group exposed to CO2 at day 14 compared to the control group ($P<0.05$).

Table 1. Effect of exposure to CO_2 on serum biochemical, blood pH and carbonic anhydrase activity (EU/mg Hemoglobin) in rainbow trouts

	7 th day		14 th day	
	Control	Control+CO_2	Control	Control+CO_2
GLU (mg/dL)	58.20±4.64	50.66±4.24	64.19±2.48	80.12±5.44*
Tprot (g/dL)	3.18±0.30	2.71±0.20	2.53±0.10	3.13±0.30
ALB (g/dL)	0.60±0.05	0.52±0.04	0.59±0.03	0.56±0.06
GLO (g/dL)	2.57±0.25	2.20±0.16	1.95±0.10	2.56±0.25
TRI (mg/dL)	31.81±3.25	18.32±1.90*	23.51±1.08	46.13±3.45*
COL (mg/dL)	125.28±10.51	72.76±6.11*	132.72±3.55	138.80±9.74
Blood CA	146.69±14.41	55.90±17.12*	158.97±13.50	167.92±29.82

The asterisks in same experimental days indicate significant differences between the control and CO2 groups ($P < 0.05$).

4 Conclusion

Measurement of blood parameters can indicate the welfare status of fish (Roncarati et al 2006). The CO2 reactions within the RBC are catalyzed by carbonic anhydrase (CA) (Swenson and Maren 1987). The rapid anion exchange mechanism therefore facilitates the loading of CO2 into the blood at the tissue level and provides plasma $HCO3^-$ with access to CA during the short period that blood passes through the gills (Currie et al 1995). As a result of the study, it is identified that CO2 concentrations cause negative effects on the serum glucose, triglyceride, cholesterol and blood carbonic anhydrase activity. In conclusion, this study results indicate that the some serum biochemical variables and blood carbonic anhydrase activity of rainbow trout significantly affected by high level of CO2.

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