Effects of Sodium Bicarbonate on Anaesthesia of Common Carp (*Cyprinus carpio* L., 1758) Juveniles

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Received 21 March 2008
Accepted 03 December 2008

Abstract

In current study, anaesthetic effects of sodium bicarbonate ($NaHCO_3$) in 6 concentrations were determined in common carp (*Cyprinus carpio*) juveniles (7.70±0.4 g and 8.38±0.8 cm) kept in two different pH levels and at 23°C water temperature. Its effective concentrations were 1000 and 600 mg L^{-1} in pH 6.5 and 7.7, respectively. While stage of anaesthesia and recovery time increased, induction time decreased; opercular rate firstly increased but than slowly decreased with increasing the concentration of anaesthetic in both pH. No mortality was observed in the study. Applications of $NaHCO_3$ in high pH levels, seems to be suitable on anaesthesia of common carp.

Keywords: *Cyprinus carpio*, sodium bicarbonate, induction time, recovery time, opercular rate, pH, water temperature

Introduction

Sodium bicarbonate ($NaHCO_3$) known as baking soda, is white in color, dissolved in water easily and gives carbon dioxide when dissolved in water. Carbon dioxide gas is listed for anaesthetic purposes in cold, cool and warm water fishes and has been used primarily to sedate fish during transport or to allow handling of large numbers of fish (Bowser, 2001). It was firstly described as a fish anaesthetic by Fish (1942). It is safe for human and there is no banning or restrictions in its using (Summerfeld and Smith, 1990; Çetinkaya and Şahin, 2005). It might also be only partly effective at immobilizing fish, slow acting, and lethal after repeated exposures (Marking and Meyer, 1985). Carbon dioxide gas is soluble in water. The gas is bubbled in the water. It is introduced into the water either directly through an air stone or indirectly by addition of sodium bicarbonate as a source of carbon dioxide. When $NaHCO_3$ is dissolved in water, it slowly releases carbon dioxide gas (Prince *et al.,* 1995; Bowser, 2001).

While this is somewhat effective in immobilizing fish, deep anesthesia is difficult to achieve (Prince *et al*., 1995). The authors reported that anesthesia stage 4 of 5 informed by Keene *et al.* (1998) could be achieved in adult sockeye salmon *Oncorhynchus nerka* within about 6 minutes; however, it was noted that the procedure should be tested to determine appropriate concentrations on nonsalmonids.

Booke *et al.* (1978) studied the effects of $NaHCO_3$ on common carp juveniles at only low temperature (10ºC). They are acclimated the fish to low water temperature in the laboratory during 10 days before treatment for this application.

In fisheries researches and aquaculture, some applications such as weighing or transporting the fishes from pond to pond should be carried out in a short time. Optimum water temperature for growth of *Cyprinus carpio* is around 25°C (Juriaan *et al*., 2003). The decreasing of water temperature may cause stress on fishes. So the determining the effects of anaesthetics appearing on the fishes at their natural ambient water temperature is important.

Main aim of the current study was to determine the effective anaesthetic concentrations of sodium bicarbonate as an anaesthetic in different water pH conditions and at an its optimal water temperature on a nonsalmonid fresh water fish, common carp (*Cyprinus carpio* L, 1758) juveniles.

Materials and Methods

Common carp (7.70±0.4 g and 8.38±0.8 cm) was obtained from Fisheries Department, Regional Directorate of State Hydraulic Works (Adana). The experiment was carried out in glass aquaria (5 L) in laboratory of Fisheries Faculty of University of Cukurova. All of the fish were starved for 24 h prior to experiment (Weyl *et al*., 1996).

Experiment was performed out at 23ºC water temperature and in triplicate. Common carp generally prefers slightly acidic or alkali ambience and its optimal pH range is 6.5-9.0 (Tekelioğlu, 2000; FAO, 2008). For this reason, two water resources having different pH level were used in this experiment; one of them was tap water (natural pH level 7.7) used in the laboratory and the other was adjusted (to 6.5 pH) by adding hydrochloric acid (HCl) (37%), 12 normality (Merck) to this water. Sodium bicarbonate ($NaHCO_3$) (extra pure, Merck) in 6 concentrations (0 (control), 200, 400, 600, 800, 1000 mg L^{-1} for 6.5 pH,

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and 0 (control), 400, 600, 1000, 1400, 2000 mg L^{-1} for 7.7 pH) was added into each aquarium and water was mixed. The concentrations were established by taking into consideration of the results obtained from previous studies (Booke *et al.*, 1978; Prince *et al.*, 1995). Because $NaHCO_3$ alters pH, pH level of the water in each aquarium was measured, when fish was stocked. Measurements of pH and temperature of the water was carried out by pH meter (Toledo mark).

After adding the anaesthetic agent, five fish was stocked into the each aquarium. When fish reached anaesthesia, induction time, anaesthesia stage, opercular rate and mortality of the fish were noted. It is generally accepted that there are five stages of anaesthesia in fish (Coyle *et al.*, 2004): however, in aquaculture applications, stages were informed in different number and feature in previous studies. In this study, fish was observed for four different anaesthesia stages shown below and modified from anaesthesia stages identified by Abbas *et al.* (2006):

1. Tranquility period (slow swimming and slight increase in opercular rate),
2. Excitation period (unrest voluntary swimming, still possible increase in opercular rate high reaction to external stimuli),
3. Light anaesthesia level (turning to one side, still reaction to external stimuli, high opercular rate loss of co-ordination excrement discharge)
4. Deep anaesthesia (lying on one side without movement, opercular movement very high (up to 200 min^{-1}) in some of the fish and very low (9-17 min L^{-1}) in others increase in excrement discharge high reaction to external stimuli in fish with a high opercular rate, and no reactions to external stimuli in those with a slow opercular rate).

After the anaesthesia, fish was removed from anaesthetic added water and transferred to clean water aquarium. Recovery time was recorded and fish was maintained there for 48h in order to observe possible mortality.

Results

Anaesthetic effects of $NaHCO_3$ in different concentrations and pH values at 23°C on common carp were shown in Table 1.

After stocking into the treatment aquaria, fish began to swim towards to aquarium walls and became hyperactive in first one or two minutes in each group. Then, fish gathered in together and began to enter the anaesthesia. An increase in the concentration of $NaHCO_3$ increased pH level of the water, slightly (Table 1).

In pH 6.5, fish reached from 1 to 3 of the anaesthesia stage; however, only half of the fish entered the 3rd stage of anaesthesia in the highest concentration. Only the 3rd stage of anaesthesia could be seen in all concentrations except first concentration of pH 7.7.

Induction time decreased with increasing of the concentration of $NaHCO_3$ in both pH treatments; to average 7 in pH 6.5 but to average 4 in pH 7.7. Recovery time increased with an increase in the concentration in both water resources. Opercular rate firstly increased and then slowly decreased with increasing the concentration of anaesthetic in both pH levels (Table 1).

No mortality was recorded in any concentration of $NaHCO_3$ during the application and post recovery period.

Discussion

It is suggested that pH-controlled carbon dioxide release from the sodium bicarbonate caused the anesthetic response (Booke *et al.*, 1978). Thus, pH level of the water after addition of $NaHCO_3$ generally was adjusted with HCl (Booke *et al.,* 1978) and sulphuric acid (H_2SO_4) (Hseu *et al.,* 1995) and glacial acetic acid (CH_3COOH) **(**Quinlan, 1997). In this study, HCl was used.

The result that the fish become hyperactive in one or two minutes is similar with the result informed for brook trout (*Salvelinus fontinalis*) by Quinlan (1997).

Booke *et al.* (1978) implied that $NaHCO_3$ at combination 642 mg L^{-1}, pH 6.5 (10ºC) on common carp was the most effective for causing the fish to cease locomotion and slow opercular rate, but to retain reflex response to pressure on the caudal fin.

In the present study (23ºC), the best results were seen in the concentrations of 1,000 mg L^{-1} and 600 mg L^{-1} for the water resources which pH levels were 6.5 and 7.7, respectively before adding $NaHCO_3$. Concentration of $NaHCO_3$ to reach the stage of anaesthesia 3 in pH 6.5 was higher (1000 mg L^{-1}) than results of species (rainbow trout (*Oncorhynchus mykiss*), brook trout (*Salvelinus fontinalis*) and common carp (*Cyprinus carpio*)) in study of Booke *et al*. (1978).

With increasing of the water temperature, solubility of $NaHCO_3$ increases (Anonymous, 2006a). However, any increment in temperature slightly decreases solubility and CO_2 level of water (Cirik and Cirik, 1991; Wedemeyer, 1996; Göksu, 2003). So, more $NaHCO_3$ (1000 mg L^{-1}) was used to anaesthetize common carp in this study than that of Booke *et al.* (1978).

In contrast to Booke *et al.* (1978), stages of anaesthesia were higher in the same concentrations of pH 7.7 than those in pH 6.5 in this study. This can indicate that high pH level of the water is more suitable to anaesthetize the fish with $NaHCO_3$ in this temperature. Already, $NaHCO_3$ dissolved more in high pH level; additionally $NaHCO_3$ increases the water pH level (Anonymous, 2006b) as seen in Table 1 of this study.

Table 1. Anaesthetic effects of $NaHCO_3$ (mg L^{-1}) at 23°C in different pH levels on common carp

pH Level of the Water Before Adding $NaHCO_3$	Concentrations of Anaesthetic (mg/L)	pH Level of the Water After Adding $NaHCO_3$	Stage of anaesthesia	Induction time (min)	Recovery time (min)	Opercular rate (per/min)
	0 (Control)		-	-	-	78±3.6
6.5	200	7.00	1	9	1-2	125±6.5
	400	7.11	1	8	1-2	130±3.8
	600	7.25	2	7.5	3-4	111±4.2
	800	7.38	2	7	9	102±6.0
	1000	7.47	2-3	7	10	105±4.3
	0 (Control)	-	-	-	2	78±3.1
7.7	400	8.19	2	7	5-6	85±2.0
	600	8.27	3	7	5-6-7	113±4.3
	1000	8.40	3	5	10	100±4.7
	1400	8.47	3	5	11	85±3.2
	2000	8.53	3	4	14	72±1.7

Induction time, recovery times and opercular rate of the fish can vary depending on concentration of anaesthetic (Booke *et al*., 1978; Hseu, 1995; Yanar and Kumlu, 2001). In $NaHCO_3$ application, induction time for common carp changed between 4-12 min; recovery time was 15 min for 3rd stage of anaesthesia (Booke *et al*., 1978). Induction times for this stage in all concentrations of present study were in harmonious with the result of the authors but recovery times were shorter.

Consequently, this study highlighted that high concentration of $NaHCO_3$ is needed in order to anaesthetize common carp juveniles at higher temperatures including optimal water temperature for growth. Additionally, $NaHCO_3$ applications at high pH level can be a good alternative to other anaesthetics.

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