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The Swimming Performance of Mollies and Their Interaction with Tiger Barb Fish when Exposed to Concurrent Low pH and Elevated Temperature

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Abstract: The global climate change and ocean acidification brought about by the anthropogenic release of carbon dioxide gas into the air is considered one of the greatest problems facing marine life. In this research, the interactions between two species of fish (the gold mollies and tiger barb) were investigated under two different environmental conditions, an elevated temperature of 28 °C and a low pH of 5 and a normal pH of 7 and a normal temperature of 24 °C. The mollies at pH 7 and a temperature of 24 °C exhibited scary interactions with the tiger barb. They were scared and ran fast away from the tiger barb. At the same time, the mollies at pH 5 and a temperature of 28 °C interacted normally as though both species were one species showing behavioral changes due to these two stressors (pH 5 and elevated temperature 28 °C). This could be the only research that has addressed how the kinematics and swimming interactions of two species of fish changed in response to elevated temperature and low pH.

Key words: pH 5, pH 7, temperature of 28 °C, temperature of 24 °C, gold mollies, tiger barb.

1. Introduction

The continuous anthropogenic emissions of nitric oxides, sulfur oxides, and especially carbon dioxide into the atmosphere consequently lowering the oceans' pH are among the many threats marine organisms will continue to face for many years to come. All three oxides are acidic and once they are in water bodies, they dissolve and lower the water's pH, and this, therefore, affects marine organisms in diverse ways. By taking up carbon dioxide (CO_2) from the atmosphere, the oceans slow down global climate change. But when carbon dioxide dissolves in water, this greenhouse gas triggers chemical reactions, causing the ocean to become acidic. Ocean acidification is measurable and takes place now at a faster rate and magnitude that has not yet been seen in the world in the past fifty-five million years [1, 2], a phenomenon that should be understood as one of the most serious human-caused threats to endanger our aquatic ecosystem like climate change and both are caused by CO_2 emissions. Ocean acidification is a concurrent problem with a common cause of climate change. Both are caused by increased anthropogenic release of carbon dioxide into the atmosphere (Fig. 1).

Oceans absorb all this and consequently lead to ocean acidification. In the aquatic ecosystem, the effects of this acidification (low pH) can be made intense or diminished by other factors such as competition, predation, etc. It is very clear at this point that most fish depend on their chemoreceptors for most behavioral activities like: locating food, avoiding predators, locating their home, locating mates, etc. Research studies have indicated that low-pH exposure to different species of fish impacts chemosensory receptors and thereby changes their behavior such as predator avoidance, olfactory preferences, how they perceive the environment (visual acuity) auditory responses including swimming speed,

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Fig. 1 The diagram above shows what goes on daily in both developing and industrialized countries. The ocean has absorbed approximately 30% of the anthropogenic emissions since the Industrial Revolution and as can be seen is still absorbing more.

etc. [3]. Several publications on this topic have shown that the activities of cephalopods including their defense and predatory activities were changed because of their exposure to low pH levels or elevated carbon dioxide levels [4-6]. In fishes, the most common result of elevated temperature and low pH has been altered behavior and physiology such as a decrease in antipredatory reaction, changes in olfactory responses, and loss of recognition of mates. The behavioral modifications can result in survival risks within their environments [7, 8].

So far research has been conducted on the reaction of prey to ocean acidification [9, 10] as well as predator response to the threat of ocean acidification [11]. It is now clear that both the predator and prey get affected in environments with low pH resulting in the prey losing predator avoidance. It is well understood now that some of the effects of low pH and elevated temperatures include altered ecological behavior, loss of learning, and changes in activity levels just to name a few. These altered characteristics could jeopardize the fish species in their natural environmental habitats. Could there be any difference in how the two tropical species of fish, the tiger barb (*puntigrus tetrazona*) and gold mollies (*Poecilia sphenops*) ecologically interact with each other in different environmental conditions (pH 5 and pH 7)? In this research, I investigated the ecological interaction between the gold mollies (*Poecilia sphenops*) and the tiger barb (*puntigrus tetrazona*) the fastest swimmer I have known.

2. Method

Three aquaria A, B, and C were set up in which aquarium water was at pH 5 and temperature of 28 °C in A, while aquarium C water was at pH 6.9 and temperature set at 24 °C. Aquarium B contained 30 fish and was used as a source tank for the gold mollies (*Poecilia sphenops*) used in experimental and control and had a normal pH of 7 and a temperature of 24 °C. Buffer MES (Morpholinoethanesulfonic Acid) pH 5 and pH 6.9 were added to aquaria A and C respectively to maintain a constant pH. Two fish at a time were taken from Aquarium B and one was put in aquarium A and the other in aquarium C and left for one week to acclimate before 5 min trials could begin. Immediately after acclimation was over tiger barb (*Puntigrus tetrazona*) was introduced in A and in C and a 5-min video camera recording was turned on to record their time of interactions. This was important to avoid observed induced behavioral responses from the interactions. The recording continued up to when fifteen gold mollies in Aquarium A and fifteen gold mollies in Aquarium C were video-recorded using a video camera mounted 1 m above the aquarium. The recordings were analyzed using a computer system to digitize the movements of the gold mollies and the tiger barb (*Puntigrus tetrazona*). Parameters were established to determine changes in gold mollies' interactions with the tiger barb. The parameters were: mollies swimming towards the tiger barb, mollies swimming away from the tiger barb, mollies swimming speed, mollies swimming along with the tiger barb, and mollies swimming up and down.

3. Results

In both experiments with pH 5 and control with pH 6.9 (Fig. 2), the trials were videotaped and analyzed using a computer system to digitize the movements of

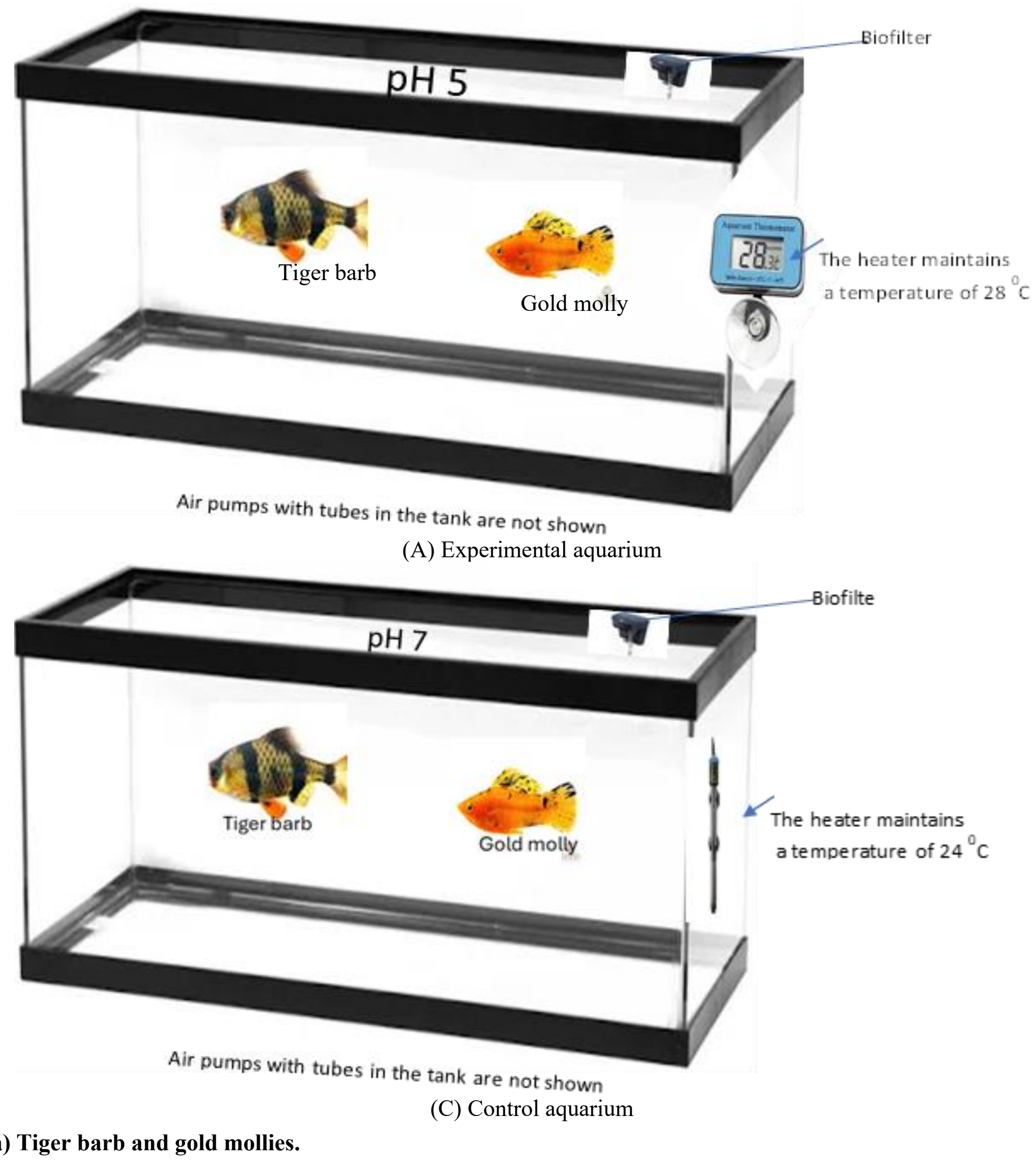

Fig. 2 (a) Tiger barb and gold mollies.

Table 1 T-test group statistics.

	Group	*N*	Mean	Std. deviation	Std. error mean
Towards tiger barb	pH 5 group	15	5.0667	3.15021	0.81338
	pH 6.9 group	15	2.9333	1.38701	0.35813
Away from tiger barb	pH 5 group	15	3.3333	1.34519	0.34733
	pH 6.9 group	15	8.5333	1.06010	0.27372
Swimming speed	pH 5 group	15	7.8000	3.05193	0.78801
	pH 6.9 group	15	14.5333	6.03403	1.55798
Swimming along tiger barb	pH 5 group	15	8.0667	5.06341	1.30737
	pH 6.9 group	15	1.0667	0.59362	0.15327
Swimming up and down	pH 5 group	15	2.8667	1.80739	0.46667
	pH 6.9 group	15	2.1333	1.18723	0.30654

the fish. The time the fish took to cover the distance on each parameter in every trial was calculated in seconds. The calculated data were statistically analyzed using Statistical Package for Social Sciences (commonly IBM SPSS) for *T*-Test group statistics (Table 1).

Here the statistical means between groups in pH 5 and groups in pH 6.9 were different. In pH 5 the gold mollies took longer to swim toward (5.0 s) the tiger barb fish than in group pH 6.9 (2.9 s). Swimming away from the tiger barb, the gold mollies in pH 5 took less time (3.3 s) to swim away from the tiger barb while the mollies in group pH 6.9 took a longer time (8.5 s). The mean swimming speed of the mollies in group pH 5 was less (7.8 s) than the mollies in group pH 6.9 (14.5). The gold mollies swam along with the tiger barb in group pH 5 for a longer time (8.06) than the swimming along with the tiger barb fish in group pH 6.9 (1.0 s). Was there any statistically significant difference?

3.1 Independent Samples Test, t-Test for Equality of Means (Table 2 below)

Table 2 Independent sample test.

		Independent samples test									
		Levene's test for equality of variances		*t*-test for equality of means						95% confidence interval of the difference	
Swimming:		*F*	Sig.	*t*	df	One-sided *p*	Two-sided *p*	Mean difference	Std. error difference	Lower	Upper
towards tiger barb	Equal variances assumed	2.084	0.160	-1.331	28	0.097	0.194	-1.20000	0.90150	-3.04663	0.64663
	Equal variances not assumed			-1.331	19.069	0.099	0.199	-1.20000	0.90150	-3.08640	0.68640
away from tiger barb	Equal variances assumed	3.022	0.093	7.402	28	<0.001	<0.001	4.66667	0.63044	3.37526	5.95807
	Equal variances not assumed			7.402	20.171	<0.001	<0.001	4.66667	0.63044	3.35230	5.98104
Speed	Equal variances assumed	4.097	0.053	3.699	27	<0.001	<0.001	6.52619	1.76428	2.90619	10.14619
	Equal variances not assumed			3.780	20.812	<0.001	0.001	6.52619	1.72659	2.93358	10.11880
along tiger barb	Equal variances assumed	12.045	0.002	-4.905	28	<0.001	<0.001	-6.66667	1.35904	-9.45053	-3.88280
	Equal variances not assumed			-4.905	14.361	<0.001	<0.001	-6.66667	1.35904	-9.57466	-3.75867
Up and down	Equal variances assumed	2.160	0.153	-1.313	28	0.100	0.200	-0.73333	0.55834	-1.87705	0.41038
	Equal variances not assumed			-1.313	24.185	0.101	0.201	-0.73333	0.55834	-1.88523	0.41856

There was a statistically significant difference between gold mollies' swimming toward tiger barb in group pH 5 and group pH 6.9, with a *p*-value of 0.001. There was a statistically significant difference between gold mollies in group pH 5 and group pH 6.9 swimming away from the tiger barb fish with a *p*-value of 0.001. There was a statistically significant difference in swimming speed between the mollies in group pH 5 and those mollies in group pH 6.9, with a *p*-value of 0.001. Again, there was a statistically significant difference between the mollies swimming along with the tiger barb in group pH 5 and group pH 6.9 with a *p*-value of 0.001. There was no statistically significant difference in swimming up and down between the mollies in group pH 5 and the group pH 6.9 with a *p*-value of 0.200.

4. Discussion and Conclusion

In fish, the most noted effects of low pH and elevated temperature are altered behavior (changes in olfactory and auditory preferences, changes in activity levels, loss of learning and visual acuity, and reduced anti-predatory response, just to mention a few) [6, 8, 9]. The behavioral changes would translate into risks that jeopardize their survival in their natural habitats [10, 11]. The elevated temperature affects many of the physiological aspects of the fish, the fish's aerobic respiration, metabolic rate [12, 13], reduced cardiac output [14], decreased muscle ability to swim [7, 16], and sensory functions [17] and interactions between species of fish [19-22] are some of the consequences.

In this research, the interactions between these two species of fish were affected by the low pH of 5 and elevated temperature of 28 °C.

Mollies swimming towards the tiger barb: In the pH 6.9 group the mollies could not swim towards the tiger barb for a longer time maybe they were scared (tiger barb having stripes and swimming faster) but the group of mollies in pH 5 swam towards the tiger barb longer meaning they lost the capacity to be afraid or scared of the tiger barb fish.

Mollies swimming away from the tiger barb: For the mollies that were in pH 5 the mean seconds were only 3.3, which means the mollies did not swim far away from the tiger barb, they were within the vicinity. But for the fish in pH 6.9 the mean seconds the mollies swam were 8.5, they must have been running away from the presence of the tiger barb.

Mollies swimming speed: The mollies in pH 5 mean swimming speed was 7.8 s while the mollies in pH 6.9 average swimming speed was 14.5 s, indicating that mollies in pH 5 experienced decreased muscle ability to swim (muscle fatigue) due to being in acidified water and elevated temperature and could not swim faster than the mollies in pH 6.9.

Mollies swimming along with the tiger barb: The mollies in pH 5 swam with the tiger barb and were not scared for an average mean second of 8.06, while the mollies in pH 6.9 swimming with the tiger barb for an average of 1.0 s, must have been scared.

The whole thrust here is that the mollies that were in pH 5 must have lost the olfactory responsiveness and anti-predatory responses towards the tiger barb and interacted well with the tiger barb, while the mollies under pH 6.9 had all their responses to their environment and responded to the presence of the tiger barb (another species) with a normal interaction.

These interactions of different species of fish to environmental stressors can be quite complicated because of behavioral subtleties that are hard to detect and may not easily be captured on a video camera. As a result, that may portray an inadequate comprehension of the long-term response to climate change in a complex ecological environment [23, 24]. The mollies that were at pH 6.9 and 24 °C exhibited scary interactions with the tiger barb of being scared and running fast away while the mollies at pH 5 and a temperature of 28 °C interacted normally as though both species were one species showing behavioral changes. This could be the only research that has addressed how the kinematics and swimming interactions of two species of fish changed in response to elevated temperature and low pH.

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