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The Impact of *Daphnia Cf. similis* on Concentration Gradient of Phytoplankton Community in Freshwater Reservoirs of Ethiopia

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Abstract: *Daphnia* is an important link of aquatic plankton communities of many freshwater ecosystems such as lakes, ponds and reservoirs, taking a central position of food webs. These freshwater ecosystems exhibit occasionally the phenomenon of phytoplankton blooming. The objective of this study is to find out the impacts of *D. cf. similis* on concentration gradients of phytoplankton community and its role in controlling phytoplankton bloom. The experiments contained five treatments with four replicates each. Out of these there was one control, which contained phytoplankton without *D. cf. similis*. The remaining four treatments contained both *D. cf. similis* and phytoplankton at different concentrations of phytoplankton. Results were statistically analyzed using Stata version 11 software. From four phytoplankton concentration gradients while they reproduced and attained high density in ¹/₄CP with low phytoplankton concentration and ¹/₈CP treatments (with small phytoplankton). Phytoplankton biomass showed a significant decrease as the density of *D. cf. similis* increased. Our findings further indicated that *D. cf. similis* can control the growth of phytoplankton if the concentration of the latter is low, but they cannot control an already existing bloom.

Key words: Phytoplankton Bloom • Treatment • Control

INTRODUCTION

The interaction of zooplankton and phytoplankton is a crucial in that it brings about alteration of the food web at top level. it is of great interest to many water quality managers. Phytoplankton and zooplankton interaction is nonlinear, as a result the zooplankton imposes two sided effects on phytoplankton which are positive (nutrient recycling brings growth stimulation) and negative (grazing mortality) [1]. When zooplankton manipulation affects phytoplankton biomass, in turn it alters phytoplankton species composition as well [2]. The bloom of phytoplankton plays important ecological role as well as contributes to the alteration of food web in the aquatic ecosystem [3]. Mostly blooms may be considered as events that are generated by the failure of zooplankton grazers in curtailing phytoplankton production [4]. Among phytoplankton community, cyanobacteria bloom in fresh water (reservoir, lakes, river etc) imposes adverse effects on health, economics, ecosystem aquatic biota and recreational services [5]. Cyanobacteria have a typical character such as buoyancy regulation ability, reduced zooplankton grazing and nitrogen fixing ability of some cyanobacteria species which helps them to become dominant in the fresh water food web [6]. The adverse impact of fresh water cyanobacteria has increased due to rising of fresh water usage, global climate change and excessive nutrient input to the fresh water in the form of nitrate and phosphorus from agricultural areas and sewage disposal from the catchment areas [7].

The cyanobacteria bloom typically and more frequently appears in the eutrophic water bodies (eutrophic lakes) and imposes adverse impact on the water quality throughout the world. During excessive

Corresponding Author: Tadesse Dejenie, Department of Biology, College of Natural and Computational Sciences, Mekelle University, Ethiopia. cyanobacteria bloom formation, different toxins (cyanotoxins) are commonly produced that cause disease and mortality in animal as well as human, such as *anatoxins, cylindrospermopsins, nodularins, saxitoxins, microcystins and lyngbyatoxins* [8]. Different bloom forming cyanobacteria also impose adverse effect on zooplanktonic organisms such as *Daphnia*. Among these bloom forming cyanobacteria *Microcystis* is most common and well known in producing toxin called hepatotoxic microcystins [5].

Ethiopia is a developing country which depends on agriculture. Many small and large reservoirs have been constructed in the country to store and supply water for agriculture. In Tigray, there are more than 70 such reservoirs constructed in the last few decades, particularly for irrigation scheme [9]. But water quality of these reservoirs is badly affected by intense algal bloom [10]. Investigations on control of phytoplankton bloom are important when considering harvesting water and water related development. The study on zooplankton interaction with phytoplankton gives an important clue on the mechanism to control the bloom forming phytoplankton, especially with *Daphnia* that has a strong grazing ability on phytoplankton. *Daphnia cf. similis* is one of such potential species.

Having better information about such interaction is relevant for critical and effective water quality management. Besides, it could be a tool for biological control to overcome the effect of toxic phytoplankton bloom on different aquatic organisms. The study of interaction between *D. cf. similis* and phytoplankton is also important to understand the mechanism of herbivore grazer (*Daphnia*) resistance against toxic phytoplankton community. Hence the aim and the objective of this research are:

- To assess the survival of *D. cf. similis* at different concentration gradients of phytoplankton
- To evaluate the association between phytoplankton biomass and *D. cf. similis*
- To examine the grazing effects of *D. cf. similis* on different phytoplankton species.

MATERIALS AND METHODS

Sample Collection: The sample of *D. cf. similis* was collected from Gereb Awso, Tigray by using $64\mu m$ zooplankton net. *Daphnia cf. similis* was separated from the sample and transferred into a plastic jar by using

pipette which has a large opening at its tip. Phytoplankton species were collected using $30\mu m$ mesh size phytoplankton net from Gereb Beati in Tigray.

Laboratory Procedures

Cultural Techniques: Experimental animal viz. *Daphnia cf. similis* was cultured in a 250ml plastic jar containing filtered reservoir water. As food *Scenedesmes* was provided after culturing them separately by adding nitrate and phosphate. *Scenedesmes* is known as good and quality food for *Daphnia*. At every two days interval the water was changed and fresh water was added by filtering it using 30µm mesh size net and *Scenedesmes* was added as a food.

Experimental Design: It consisted of four treatments and one control with four replicates in 2 liter jars. The treatments included different concentration gradients of phytoplankton and 30 individuals of Daphnia cf. similis each. It comprised 500ml concentrated phytoplankton (CP) with 1000ml water, 250ml half concentrated phytoplankton (1/2CP) and 1250 ml water, 125ml quarter phytoplankton concentration (1/4CP) with 1375 ml water, 62.5ml phytoplankton (1/8CP) with 1437.5ml water and the one control group (ND) with 500ml phytoplankton, 1000ml water and no D. cf. similis. The total biomass was calculated during all the experiments. The water for all treatments was collected from the dam and filtered with 30µm phytoplankton net. The experiments were conducted in the laboratory. Observations were made for a period of 18 days.

Measurement and Sample Analysis: Temperature and dissolved oxygen were measured using Thermometer and oxygen meter respectively; chlorophyll a and turbidity were measured using flurometer. *Daphnia cf. similis* was directly counted following Minalu [11], at 3 day interval. Ten ml sample of phytoplankton was taken with a pipette, preserved in lugol's solution and was counted using inverted microscope and the phytoplankton biomass was estimated from the cell volume according to the method recommended by Irigoien *et al.* [3]. Necessary measurements of phytoplankton were taken during the time of counting by following to geometric shapes like sphere, cylinder and cone [12].

RESULTS

Efficiency of D. *cf. Similis to Control* Phytoplankton: Multivariate regression measure revealed the negative

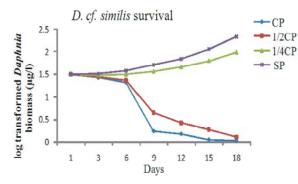


Fig. 1: The relative survivals of *D. cf. similis* at different concentration gradient of phytoplankton. Note: CP= high phytoplankton concentration, SP=

with small phytoplankton

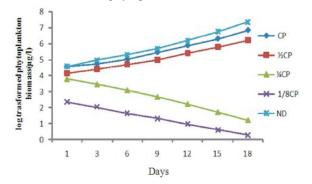


Fig. 2: Phytoplankton biomass in the presence and absence of *D. cf. similis* across the treatment through time.

Note: CP= high phytoplankton concentration, ND=No *Daphnia*

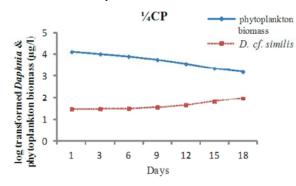


Fig. 3: Interaction between *D. cf. similis* and phytoplankton biomass at ¹/₄CP

Note: ¹/₄CP= quarter concentrated phytoplankton

effect of phytoplankton biomass on *D. cf. similis* across the treatment along time (F= 29.66, P= 0.00). The paired t-test also showed negative correlation between *D. cf. similis* and phytoplankton biomass across

the treatment along time (P< 0.05). Concentration gradient had effect on reproduction and growth of *D. cf. similis* that is; at higher concentration phytoplankton (CP) *D. cf. similis* were not able to survive up to the end of the experiment (Fig. 1). Similarly, in the half concentrated phytoplankton ($\frac{1}{2}$ CP) most of the *D. cf. similis* were dead. While in low concentrated phytoplankton ($\frac{1}{4}$ CP) treatment *D. cf. similis* managed to survive and increased in density up to the end of the experiment. *Daphnia cf. similis* populations were able to reproduce and grow and attain very high biomass in $\frac{1}{4}$ CP (with small phytoplankton). In $\frac{1}{4}$ CP treatment with low concentration of phytoplankton, *D. cf. similis* could grow and reproduce, but it was not to the extent as that of $\frac{1}{4}$ CP treatment.

At high phytoplankton concentration (CP) treatment Phytoplankton biomass grows abundantly at the end of the experiment. Similarly, the phytoplankton biomass in half concentrated phytoplankton showed an increase in biomass. While in low concentrated phytoplankton ($^{1}/_{4}CP$) treatment the phytoplankton biomass decreased (Fig. 2). Liner regression was conducted to show how the presence of *Daphnia* affects the abundance increase of phytoplankton biomass. The result was statically significant at (F= 533.2; P= 0.002).

In low concentrated phytoplankton treatment ($\frac{1}{4}$ CP) *D. cf. similis* showed reproduction and growth, while the biomass of phytoplankton reduced (Fig. 3). Besides, the performance of *D. cf. similis* was excellent in SP (with small phytoplankton). On the other hand the phytoplankton biomass reduced in $\frac{1}{6}$ CP as the *D. cf. similis* reproduced and grew at the end of the experiment compared to the other treatments. Multivariate regression analysis showed negative effect of *D. cf. similis* on phytoplankton biomass at $\frac{1}{4}$ CP treatment (F= 21.47, P= 0.00)

Effects of Phytoplankton Biomass on *D. cf. similis*: *Daphnia cf. similis* was not successful at higher phytoplankton concentration (CP) that is, all *D. cf. similis* died at the end of the experiment, while the phytoplankton grow abundantly and their biomass continue increasing and reach very high (Fig. 4). Similarly in the second treatment half concentrated phytoplankton ($\frac{1}{2}$ CP) also showed negative influence on the growth of *D. cf. similis* that is, *D. cf. similis* highly decreased, but its phytoplankton biomass remains very high. Regression measure also revealed the strong negative effect of phytoplankton biomass on survival of *D. cf. similis* (F=44.42, P= 0.001).

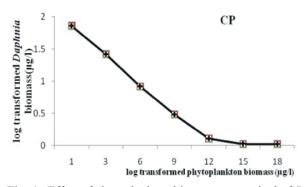


Fig. 4: Effect of phytoplankton biomass on survival of *D*. *cf. similis* at CP



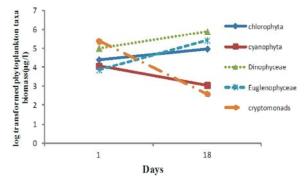


Fig. 5: The effects of *D. cf. similis* grazing on phytoplankton composition at ¹/₄CP treatment.

In low concentrated phytoplankton $(\frac{1}{4}CP)$ cyanophyta and cryptomonads did not show growth, rather their biomass was very low and their growth was almost controlled by D. cf. similis (Fig. 5). Here the cyanophyta group includes the larger size phytoplankton (Anabaena, Aphanizomenon, Microcystis and Pseudoanabaena) and at the same time a group that can produce toxic substance. On the other hand, in such treatment D. cf. similis grow and reproduce. Regression measure showed the negative effect of D. cf. similis on cyanophyta (F= 16.44, P= 0.01). Regression measure also revealed the negative effect of D. cf. similis on cryptomonads (F=97.52, P=0.00).

DISCUSSION

In high phytoplankton concentration (CP) *D. cf.* similis were not able to survive, that is, all *Daphnia* died before the end of the experiment. The reason might be *Daphnia* ingested more readily colonial or filamentous cyanobacteria and were thereby more strongly affected by the toxic chemical produced by those cyanobacteria [13]. Similarly, DeMott *et al.* [14] and Boersma & Vijverberg [15] reported that *Daphnia* population declined or died at higher concentration due to the colonial form of *Microcystis's* digestibility, toxicity and low fatty acid composition. At high phytoplankton concentration (CP) treatment cyanophyta (*Microcystis* and *Anabaena*) were dominant and all the *Daphnia* were dead. This increased mortality of *Daphnia* might be because of the toxins produced by *Microcystis and Anabaena* [15].

The result of this study clearly showed how a gradient of phytoplankton concentration affects the ability of Daphnia to suppress cyanophyta (Microcystis). This is in agreement with the majority of studies that reported the effect of Microcystis on Daphnia (Lurling [16] and Ghadouani et al. [17]). The accumulation of toxic cyanobacteria such as Microcystis and its effect on D. cf. similis was correlated with the concentration gradient of the treatment a long time. At high phytoplankton concentration treatment (CP) in which its concentration was higher than the natural system more *Microcystis* got accumulated and subsequently Daphnia died. This finding is in agreement with the work of Ghadouani et al. [18] who observed the mortality of zooplankton species in the presence of dissolved microcystins at high concentration experiment.

There is a controversy on the result of the interaction between D. cf. similis and phytoplankton such as cyanobacteria [19]. However, the result of this study showed two important points: 1) D. cf. similis grow and reproduce at low phytoplankton concentration and 2) D. cf. similis died in the treatment that has higher phytoplankton concentration. These observations have been also reported by Sarnelle [20] who conducted enclosure experiment using Daphnia pulicaria and cyanobacteria at different initial concentrations. Probably the reason is as the concentration of the secondary metabolites produced by toxic cyanobacteria (like Microcystis and Anabaena), such as poly unsaturated fatty acid or protease inhibitors increase, which might lead to the death of Daphnia [21]. The dominance of cyanophyta at high concentrated phytoplankton had a strong effect on feeding of D. cf. similis by reducing the collection and ingestion of more nutritious resources [22]. As a result in high phytoplankton concentration D. cf. similis might not be able to filer the available food which in turn leads to starvation. Therefore the death of D. cf. similis at high phytoplankton concentration might be due to starvation.

At lower concentration of phytoplankton ($\frac{1}{4}$ CP) D. cf. similis survival rate was high that is, D. cf. similis were able to graze and control the phytoplankton biomass. This can be justified to the fact that different phytoplankton taxa such as cyanophyta are among the complementary food items of Daphnia [23]. There are also supporting evidence indicating that small cryptomonads are the most vulnerable taxa to grazing and consumption by Daphnia [24]. This further indicated that D. cf. similis was able to feed and resulted in decrease of phytoplankton biomass and at the same time D. cf. similis to grow and reproduce in the treatment that has low phytoplankton concentration (¹/₄CP). This is in agreement with the finding of Muylaert et al. [25] who provided considerable evidence that the growth of Daphnia population has led to the reduction of phytoplankton biomass through grazing.

The *D. cf. similis* that were introduced at lower concentration of phytoplankton (¹/₄CP) showed an increase in biomass and created negative impact on the population growth of cyanophyta (such as *Microcystis*) and cryptomonads (*Cryptomonas*). This finding is in agreement with the work of Chen *et al.* [26] that reported a reduction of *Microcystis* population as a result of introducing of *Daphnia*, with the work of Thys *et al.* [24] that reported a reduction of cryptomonads in the presence of *D. galeata.*

The study showed that present at high phytoplankton concentration the phytoplankton biomass increased abundantly as the D. cf. similis died. This seems due to the absence of grazing by D. cf. similis, because D. cf. similis were dead before the end of the experiment [4]. Phytoplankton biomass decreased in low concentrated phytoplankton as D. cf. similis reproduced, grew and increased in density, which might be due to grazing pressure of Daphnia [25].

In the low concentration (¹/₄CP) treatment dinophyceae increased and grew abundantly as *D. cf. similis* reproduce and grow. This might be as a result of the fact that the selection of food by *Daphnia* depends on the shape, size and diameter of the food particle [24] and the production of toxin by *Peridinium* that helps it to get competitive advantage over other phytoplankton by inhibiting the growth of other phytoplankton [27]. In the present study at low concentration level of phytoplankton (¹/₄CP), *Peridinium* toxicity did not affect the survival of *D. cf. similis*, that is, *D. cf. similis* reproduced and grew well in the presence of high biomass of *Peridinium*. This condition was also reported by Rengefors & Legrand [27] who conducted laboratory experiment on the effect of *Peridinium aciculiferum* toxicity *on D. galeata*.

Chlorophyta (such as Staurastrum and Cosmarium) increased as D. cf. similis increased at low phytoplankton concentration (1/4CP), which might be due to the large size of Staurastrum and Cosmarium which cause ingestion problem in Daphnia [28]. The other reason for chlorophyta increase in low concentrated phytoplankton might be due to its resistance to the grazing of D. cf. similis through colony formation [29]. While cryptomonads highly decreased as the D. cf. similis increased, which might be due to the fact that cryptomonads with its weak and easily broken cell wall are the most preferred feed for Daphnia [24]. In high phytoplankton concentration (CP) treatment cryptomonads increased as all D. cf. similis died before the end of the experiment and they were free from being grazed by D. cf. similis. The reason for this might be due to the fact that cryptomonads were more preferred by Daphnia [24]. Similarly cyanophyta also increased abundantly in CP as all D. cf. similis died before the end of the experiment and they were free from being grazed by D. cf. similis. This seems to be due to the reason that larger Daphnia have a broader diet and so may have been better able to consume colony-forming cyanobacteria [20].

The present study revealed that, D. cf. similis had a strong top-down effect on cyanophyta and cryptomonads in which it decreased as D. cf. similis increased. On the other hand, dinophyceae, euglenophyceae and chlorophyta increased with D. cf. similis. The reason for this might be that phytoplankton taxa are differentially grazed upon by Daphnia and that there are abundant larger phytoplankton species that are outside the range filtered by Daphnia [30]. In high concentrated phytoplankton cyanophyta were dominant over other phytoplankton taxa. Chlorophyta, dinophyceae and euglenophyceae decreased as cyanophyta increased. This might be due to the fact that cyanophyta formed scum and suppressed the growth of other phytoplankton taxa [31, 32].

CONCLUSION

The results of this study showed that *D. cf. similis* can suppress phytoplankton only if the concentration of phytoplankton is low, but *D. cf. similis* cannot control phytoplankton once bloom is formed.

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