

# Investigation of Seasonal Total Phenolics and Pigments in *Chara vulgaris*

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Received: February 14, 2015 Accepted: March 9, 2015

doi:10.5296/jbls.v6i2.7096 URL: <http://dx.doi.org/10.5296/jbls.v6i2.7096>

## Abstract

In this study, *Chara vulgaris* samples were taken from Gölbaşı Lake/Adiyaman at different times (October, February, April, June). Total phenolic and pigment contents of these samples were examined. It was determined that total phenolic and pigment contents (chlorophyll a, chlorophyll b, and carotenoids) showed seasonal differences. It was detected that chlorophyll a, chlorophyll b, and carotenoids were high in June, and low in April. In addition, phenolic compounds were found to be high in February, and low in April.

**Keywords:** *Chara vulgaris*, Phenolics, Photosynthetic pigments, Seasonal cycle

## 1. Introduction

Phenolic compounds are secondary metabolites produced by plants. They have various functions such as protection against pathogens and ultraviolet radiation, mechanical support, and attraction of pollinating animals (Bravo, 1998; Parr & Bolwell, 2000). Many phenolic compounds have been determined to have health benefits (Houston, 2005; Zafar et al., 2013). These compounds vary depending on environmental conditions and stress factors. In many studies, it was reported that phenolic compounds were synthesized in aquatic plants, and the amount of these compounds changed depending on the season and environmental conditions (Spencer & Ksander, 1994; Gross et al., 1996; Bauer et al., 2009).

Underwater vegetation is more exposed to seasonal changes from external factors especially in areas exposed to tides and wave motions, and in coastal regions where water loss is sudden (Whitfield et al., 2007). *Chara* is a wild genus spread over many natural waters. Species of this genus can intake nutrients very effectively owing to their rhizoids. Charophytes groups, which are known as water meadows, intake some chemicals known as nutrients (i.e. phosphate, nitrate, ammoniac etc.) and use these nutrients in photosynthesis process. If they reach adequate biomass in aquatic systems, they could affect the food chain in aquatic systems (Kufel & Kufel, 2002). *Chara* is a good hiding place for fry, a good habitat for laying eggs and important for protection of environmental balance (Denike & Geiger, 1974).

As it was suggested in numerous studies, many aquatic plants (as in *Chara* spp.) intake heavy metals in the system while intaking substances required for photosynthesis such as phosphate, nitrate, ammoniac etc. Thus, they undertake an important role by removing heavy metals in aquatic systems (Kamal et al., 2004; Narain et al., 2011; Shaikh & Bhosle, 2011). Moreover, Mahajan & Kaushal (2013) were determined that *C. vulgaris* could remove Congo red from aquatic solutions. Likewise, Shaikh & Bhosle (2011) reported that *Chara* and *Hydrilla* species could remove chromium in aquatic systems.

In aquatic habitats, a continuous macrophyte composition is essential for the system to sustain its stability by balancing its chemical composition. Contaminating factors must be removed by filtration from the environment to have usable water, which includes many nutrients such as phosphate, nitrate, ammoniac, silicium magnesium, and nitrate. For this purpose the continuity of macrophytes is obligatory in the system. Therefore, it is important to detect the changes of pigment and phenolics of *Chara*, which are depended on environmental differences that are caused by seasonal changes. To achieve this goal, seasonal changes in pigment (chlorophyll a, chlorophyll b and carotenoids) and phenolic contents of *C. vulgaris*, which was collected from Lake Gölbaşı, were determined.

## 2. Materials and Methods

*Chara vulgaris* samples were collected (in October, February, April, June) from Lake Gölbaşı/Adıyaman, Turkey between 2011-2012. Pigment contents were extracted in acetone according to the method described by De Kok & Graham (1989). Amount of pigments were determined as it was described by Lichtenthaler & Wellburn (1983). Total phenolic compounds of the samples were determined depending on folin reactive and standard gallic

acid equivalence (Slinkard & Singleton, 1977; Chandler & Dodds, 1983).

### 2.1 Statistical Analysis

The results obtained from three replications were evaluated using the SPSS 15.0 program. Duncan's test was used for significance control ( $P < 0.05$ ).

## 3. Results and Discussion

It was found that photosynthetic pigments in *C. vulgaris* varied according to season. It was determined that Chl a, Chl b, and carotenoid changes were high in June, and low in April (Figures 1, 2, 3).

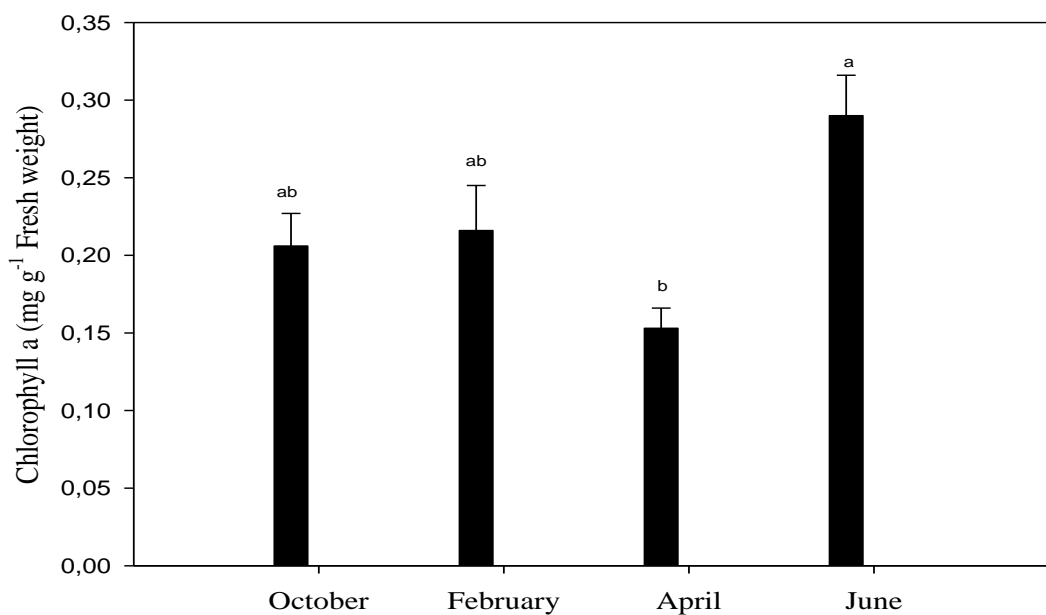


Fig. 1. Seasonal chlorophyll a changes in *Chara vulgaris*

(Data followed with different letters are significantly different from each other ( $p < 0.05$ ) according to Duncan's test).

In studies on macrophyte species, Pilon & Santamaría (2001) detected that chlorophyll concentrations change according to species and months. When *Callitriche obtusangula* is evaluated in terms of total chlorophyll concentration, it was reported as a species which alters seasonally, and chlorophyll concentrations were found to be highest in March and July, and lowest in May. Anton & Putt (1988) determined the photosynthetic productivity of *Chara vulgaris* L., which was maximum for submersed macrophytes in early summer. As described in the studies above, in this study pigment levels were also found to be high in early summer (June) (Figures 1, 2, 3).

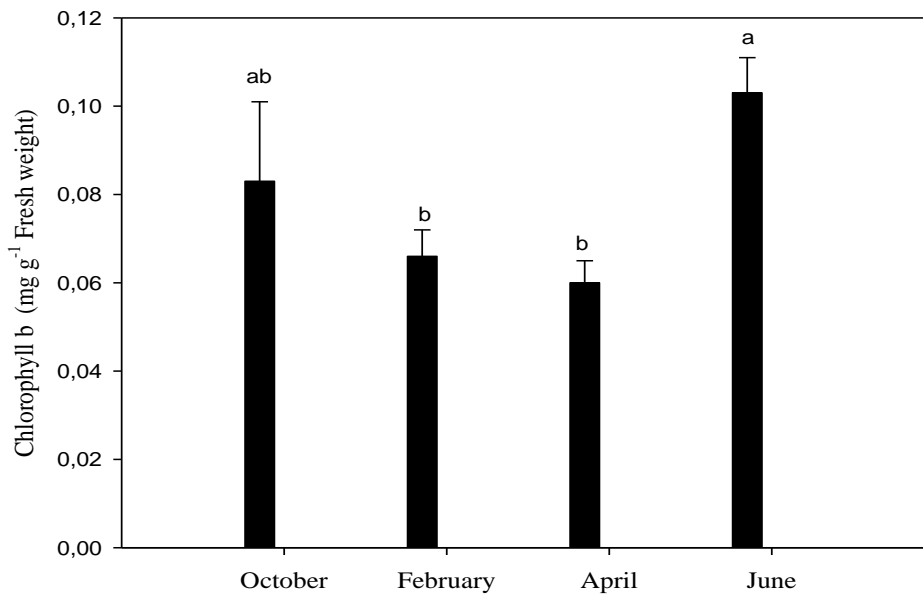


Fig. 2. Seasonal chlorophyll b changes in *Chara vulgaris*

(Data followed with different letters are significantly different from each other ( $p < 0.05$ ) according to Duncan's test).

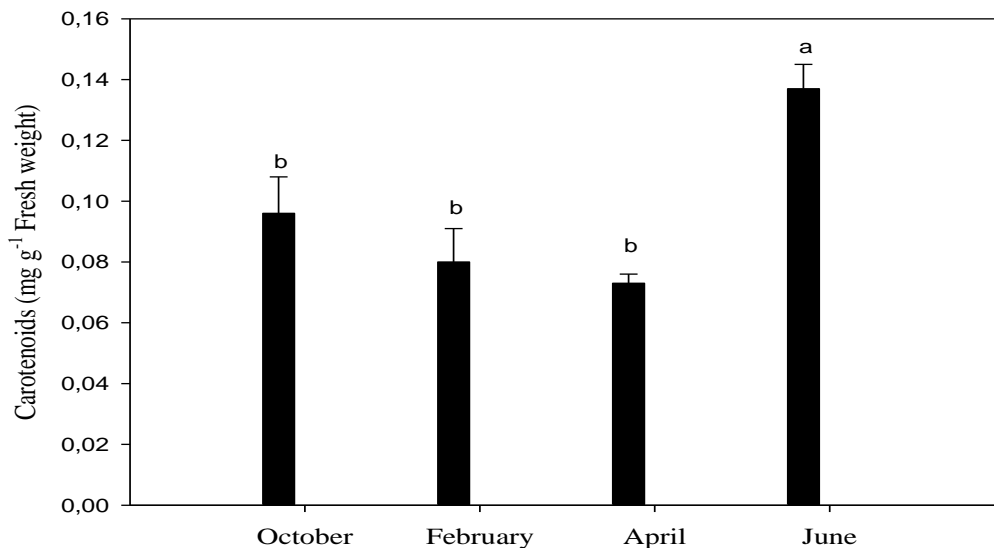


Fig. 3. Seasonal carotenoid changes in *Chara vulgaris*

(Data followed with different letters are significantly different from each other ( $p < 0.05$ ) according to Duncan's test).

In addition, it was detected that phenolic compounds change depending on months. It was determined that phenolic compounds were at highest concentration in February ( $0.454 \mu\text{g mg}^{-1}$ ), and at lowest concentration in April ( $0.158 \mu\text{g mg}^{-1}$ ). It was also detected that phenolic levels in June are close to that are in February ( $0.440 \mu\text{g mg}^{-1}$ ) (Figure 4). It was determined that phenolic compound production of plants changes depending on biotic and abiotic factors. Also it was shown that the differences in phenolic compound may occur depending on plant species, genetic characteristics and plant type (Smolders et al., 2000; Boege, 2005; Cronin & Lodge, 2003; Li et al., 2010). In another study, it was determined that phenolic compounds in aquatic plants and filamentous algae change depending on plant organ and season but doesn't change depending on the age of the plant (Pip, 1992). Our findings are in agreement with the study described above, which suggested that phenolics changed depending on the season.

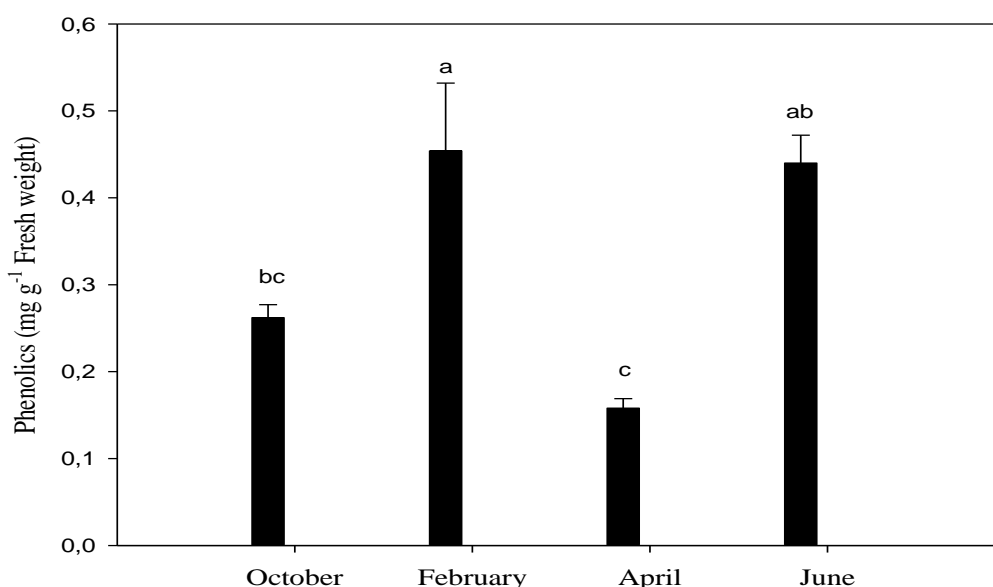


Fig. 4. Seasonal total phenolic compounds in *Chara vulgaris*

(Data followed with different letters are significantly different from each other ( $p < 0.05$ ) according to Duncan's test).

#### 4. Conclusion

Determining of seasonal photosynthetic pigments and phenolics synthesized for defense in *Chara vulgaris* may provide information regarding to the continuity and metabolic responses of this species depending on environmental conditions that may change in aquatic systems.

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