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Relationships between Nutrients and Chlorophyll a Concentration in the International Alma Gol Wetland, Iran

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Abstract

This study was conducted to find out the Relationships between Nutrients and Chlorophyll a concentration in the International Alma Gol Wetland. Water samples were collected fortnightly from five stations in the wetland. They were collected during summer and autumn. Results illustrated that there was a significant correlation between chlorophyll a and logarithm chlorophyll a with nitrate, nitrite (P<0.01) and ammonia (P<0.05) but there was no significant correlation between chlorophyll a and logarithm chlorophyll a with silica, total alkalinity, sulfate and resolve phosphorus (P>0.05).

Keywords: Nutrients; Chlorophyll a; Alma Gol; Wetland

Introduction

Wetlands are now recognized as important traits in the landscape that provide numerous useful services for people and fish and wildlife [1]. They are among the most important ecosystems on Earth and considered unique because of their hydrology and their function as ecotones between terrestrial and aquatic ecosystems [2]. These systems are known universal as biodiversity shelters and are among the most biologically various in the world [3].

Chlorophyll a is the major photosynthetic pigment in a lot of phytoplankton and a trophy index in aquatic ecosystems [4,5]. Chlorophyll a (Chl a) is often used as an estimate of algal biomass, with blooms being estimated to happen when Chl a concentrations go above 40 $\mu g \ L^{-1}$ [6]. So a methods for the estimation of the growth and development of the phytoplankton community is to perform an analysis of photosynthetic pigments, even though the content of chlorophyll in the cells changes with the availability of light [7] and thus with depth and trophic gradient [8].

Because eutrophication is defined as an aquatic ecosystems response to nutrient loading [9], the ability to identify important factors and predict subsequent algal blooms with the use of a Chl a equation could be a key lake water management tool. Both chemical and physical controls can be used to prevent or remove algae or algae byproducts from the water [6]. In particular, information about the form of nutrient-Chl relationships [10,11] has allowed lake managers to establish nutrient concentration and loading aims. Nitrogen and phosphorus are often identified limiting nutrients to algal biomass and silicon is necessary for diatom growth [12]. Nitrogen occurs in fresh water in numerous forms: dissolved nitrogen, amino acids, amines, urea, ammonium (NH $_4^+$), nitrite (NO $_2^-$), and nitrate (NO $_3^-$) [7]. In aquatic ecosystems, phosphorus (P) can be found either in particulate matter or as soluble inorganic phosphorus, orthophosphate (PO $_4^{3-}$) [13].

A review of the 1995 to 1997 biological abstracts about significant factor for algal blooms, illustrates that, of 596 articles on estuaries and nutrients, 52 % consider only nitrogen, 32 % refer to both nitrogen and phosphorus, and 16% consider only phosphorus, although the preponderance of study on N, the evidence for general N limitation of coastal systems is feeble compared to the data for general P limitation of freshwater systems [14].

There have been a small number of comprehensive analyses of the form of phosphorus-chlorophyll relationships. The phosphoruschlorophyll relationship most probably outcomes from the dependence of algal growth rates on phosphorus availability [15].

Nitrogen limitation of algal biomass seems to be more general in subtropical and tropical lakes [16-18], while phosphorus appears to be the primary limiting nutrient in temperate lakes [19]. Other nutrients, for example iron and silicate, have been reported to be limiting in some other regions [20,21]. The limiting nutrient is decided mainly by the mass equilibrium between elements such as C, N, P, and Si, and their relationship to the growth requirements of the phytoplankton [22].

This research was conducted to determine the relationships between chlorophyll a and nutrients concentrations in the Alma Gol wetland for identify important and effective nutrients on Chl a concentration. Because of Chl a identified as a major photosynthetic pigment in a lot of phytoplankton and a trophy index in aquatic ecosystems and the other hand the chlorophyll a concentration in the phytoplankton cells changes with nutrients and environmental factors so know about the effective factors on chlorophyll a concentration is very important for ecosystem management.

Materials and Methods

This study was conducted in Alma Gol international wetland, which is situated on the Turkmen steppes near the border with Turkmenistan in the Golestan Province, in north of Iran (Figure 1). Area of the wetland was 207 ha.

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Figure 1: Location of Alma Gol wetland and situation of sampling stations.

Field sampling

During summer to autumn in 2011, water samples were collected fortnightly from five sampling stations to determine nutrients and chlorophyll-a concentration. Site sampled showed in Figure 1 by white stars. Stations chose with navigate location and regards water depth, macrophyts populations, wind direction, input and output of wetland water. All sampling conducted between 10 to 1 o clocks. The water temperature during sampling was 21.10 ± 9.59 centigrade. Usually the weather in summer was sunny and in autumn was rainy and windy. Samples gave from Secchi disk depth by P.V.C. tube. At each sampling station, one liter water collected to determine chlorophyll a and one liter for measured nutrient concentrations. Water samples were light-protected and transferred to laboratory at 4°C.

Chlorophyll a determination

For determination chlorophyll a concentration, samples were shaken and certain volume of water (based on water color) was filtered using a vacuum pomp and GF/F filter. Thereafter, filter was pulverized with 90% acetone in a mortar. The resulting mixture was centrifuged for 10 min. (3000 rpm) and supernatant was poured into a glass cavetti. The optical density was read at 630, 647, 664 and 750 nm. Chlorophyll a concentrations were calculated according to Jeffrey and Humphrey [23].

Nutrient determination

In this research we measured ammonia, nitrite, nitrate, silica, sulfate, total alkalinity and resolve phosphorus by especial tablets and photometer set with Wagtech specific recipe and method.

Statistical analyses

Data were analyzed by statistical software SPSS v. 18 and Microsoft office Excel 2007. Data were subjected by correlate and Bivariate test to find significant relationship between chlorophyll a and nutrient concentration and Duncan's test to find significant difference between nutrients concentration during sampling. P<0.05 considered to be significantly different.

Results

Results illustrated that there was a negative and significant relationships between chlorophyll a and logarithm chlorophyll a with nitrate, nitrite (P<0.01) and ammonia (P<0.05) but there was no

significant correlation between chlorophyll a and logarithm chlorophyll a with silica, total alkalinity, sulfate and resolve phosphorus (P>0.05) (Table 2). Chlorophyll-a ranged between 4.38 to 156.55 mg/m³, sulfate ranged between 138 to 190 mg/l, total alkalinity ranged between 80 to 280 mg/l, silica ranged between 3.80 to 35.00 mg/l, phosphate ranged between 0.02 to 3.70 mg/l, ammonia ranged between 0.10 to 11.90 mg/l, nitrate ranged between 0.01 to 2.75 mg/l and nitrite ranged between 0.01 to 0.39 mg/l. We illustrated the average of these factors and significant differences between months in table 1.

Regression line for Log Chl a:NO₂, Log Chl a:NO₃, Log Chl a:NH₃, Log Chl a:SO₄, Log Chl a:SiO₂, Log Chl a:PO₄, Log Chl a:CaCO₃ relations was showed in Figure 2 and explained below.

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y=-0.003x+2.118 R<sup>2</sup>=0.012, r=-0.105
                                           Log Chl a:SO<sub>4</sub>
                    R<sup>2</sup>=0.161, r=-0.654
                                          Log Chl a:NO,
y=-1.888x+1.758
                                           Log Chl a:SiO,
y=0.004x+1.482
                    R^2=0.002, r=0.124
y=-0.289x+1.706
                    R^2=0.261, r=-0.701 Log Chl a:NO,
y=-0.046x+1.688
                    R<sup>2</sup>=0.110, r=-0.457
                                           Log Chl a:NH<sub>2</sub>
                                           Log Chl a:PO,
y=0.017x+1.519
                    R^2=0.000, r=0.005
y=0.001x+1.392
                                           Log Chl a:CaCO,
                    R^2=0.014, r=0.158
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Discussion

The significant correlation between chlorophyll a and nitrite, nitrate and ammonia was found in this research, as the amount of chlorophyll a was high the amounts of nitrite, nitrate and ammonia were in the lowest, but chlorophyll a was not affected by silica, total alkalinity, sulfate and resolve phosphorus.

Power relationships between phosphorus, chlorophyll, and water clarity have been observed for freshwater systems in the world [24,25]. The strong relationship between chlorophyll and phosphorus established by Sakamoto [24] in numerous Japanese lakes forms appropriate testable hypothesis: chlorophyll is both a useful and an easy estimator of phytoplankton standing crop and is now more generally used than cell number or cell volume.

Smith [26] achieved the most comprehensive analysis of phosphorus-chlorophyll relationships to date, and was the first to illustrate that information of both TP and TN concentrations could improve predictions of algal biomass. A hypothesis that fits more closely with the classic Liebigian paradigm [27-30] would consider other nutrients that might limit algal growth at high levels of ambient phosphorus. It would predict that chlorophyll should rise linearly until some other quantity (such as nitrogen, silicon, molybdenum and light) becomes limiting. Despite the fact that nonlinear relationships between TP and chlorophyll between lakes are possible [31,32], there have been little quantitative examinations on their shape or form.

Hoyer et al. [33] suggested that phosphorus accounts for more variance in chlorophyll than in nitrogen in nearshore coastal Florida waters and fresh water. While nitrogen had been shown to limit algal populations in both systems [34,35], the data presented suggested that phosphorus was the primary nutrient limiting algal populations amongst the 300 nearshore coastal locations sampled. They recognized, though, that some of these 300 stations may at some times be limited by nitrogen. Canfield [32] and Brown et al. [25] have formerly affirmed that phosphorus is the primary limiting nutrient in Florida lakes, and when the total nitrogen to total phosphorus ratio moves below 10, nitrogen may become limiting. It is normally assumed, that nitrogen is the primary limiting nutrient for phytoplankton production in most coastal waters [35].

Brown et al. [25] explained that TP alone accounted for a significant

| Variables | August | September | October | November | December |
|-------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| NO ₂ | 0.14 ± 0.1 ^{ab} | 0.18 ± 0.1 ^a | 0.08 ± 0.02b | 0.11 ± 0.03 ^{ab} | 0.06 ± 0.02b |
| NO ₃ | 0.60 ± 0.58b | 1.34 ± 0.95° | 0.18 ± 0.03b | 0.47 ± 0.32b | 0.33 ± 0.08b |
| NH ₃ | 4.72 ± 3.50 ^a | 4.78 ± 3.88 ^a | 1.90 ± 0.69 ^a | 5.00 ± 2.32° | 3.56 ± 0.92a |
| PO ₄ | 0.50 ± 1.13 ^a | 0.14 ± 0.06° | 0.12 ± 0.09 ^a | 0.11 ± 0.07 ^a | 0.11 ± 0.08 ^a |
| SiO ₂ | 10.15 ± 6.28 ^a | 9.67 ± 1.94° | 9.36 ± 1.24 ^a | 12.27 ± 8.18 ^a | 7.16 ± 1.51 ^a |
| CaCO ₃ | 153.00 ± 67.59 ^a | 88.50 ± 5.80 ^b | 94.00 ± 6.52 ^b | 108.50 ± 13.75 ^b | 113.00 ± 10.37 ^b |
| SO ₄ | 160.30 ± 16.53 ^b | 184.00 ± 3.16 ^a | 185.00 ± 6.12a | 176.50 ± 3.37 ^a | 177.00 ± 2.74 ^a |
| Chl a | 52.46 ± 20.79b | 23.03 ± 14.59 ^{cd} | 136.83 ± 15.80 ^a | 42.08 ± 21.67bc | 11.03 ± 3.82d |

Different letters show significant difference between columns (P<0.05).

a and b show significant differences between nutrient concentration in different months.

Table 1: Mean ± SD of nutrients concentration during sampling in Alma Gol wetland.

| | Chl a | Log Chl a | NO ₂ | NO ₃ | NH ₃ | PO ₄ | SiO ₂ | CaCO ₃ | SO ₄ |
|-------------------|----------|-----------|-----------------|-----------------|-----------------|-----------------|------------------|-------------------|-----------------|
| Chl a | 1 | | | | | | | | |
| Log Chl a | 0.889** | 1 | | | | | | | |
| NO ₂ | -0.486** | -0.654** | 1 | | | | | | |
| NO ₃ | -0.489** | -0.701** | 0.895** | 1 | | | | | |
| NH ₃ | -0.444* | -0.457* | 0.482** | 0.535** | 1 | | | | |
| PO ₄ | -0.038 | 0.005 | -0.005 | -0.120 | -0.013 | 1 | | | |
| SiO ₂ | 0.061 | 0.124 | 0.186 | 0.092 | -0.043 | -0.160 | 1 | | |
| CaCO ₃ | 0.043 | 0.158 | -0.220 | -0.365* | -0.117 | 0.249 | -0.312 | 1 | |
| SO ₄ | 0.055 | -0.105 | 0.227 | 0.283 | 0.027 | -0.373* | 0.159 | -0.763** | 1 |

^{**}Correlation is significant at the 0.01 level (2-tailed).

Table 2: Nutrients and chlorophyll a relationship in Alma Gol wetland.

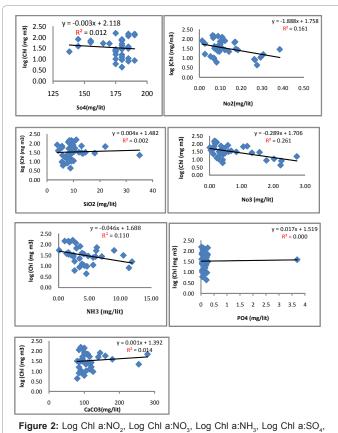


Figure 2: Log Chl a:NO₂, Log Chl a:NO₃, Log Chl a:NH₃, Log Chl a:SO₄, Log Chl a:SiO₂, Log Chl a:PO₄, Log Chl a:CaCO₃ relations for Alma Gol wetland

amount of the variance (R_2 =0.76) of observed Chl measurements and TN alone accounted for less variance of observed Chl measurements

 $(R_2=0.46)$, but a multivariate model using both TP and TN also accounted for a significant amount of the observed variance $(R_2=0.78)$. The coefficient of determination values for TP-Chl and the multivariate nutrient-Chl, were similar $(R_2=0.76$ against 0.78), suggesting that Chl concentrations can be predicted rationally well using TP alone. The coefficient of determination for the relationship between TP and Chl for both equations was positive and significant.

Redfield [36] expressed that phosphorus (P) has been considered a key limiting nutrient in marine systems. Furthermore, P controls of phytoplankton biomass in numerous freshwater systems and similarities in phytoplankton physiology and nutrient requirements in coastal and freshwater systems [12] make P control of coastal systems naturally appealing. Nevertheless, following Ryther and Dunstan's [37] influential work, nitrogen (N) is generally seen as the limiting nutrient in coastal systems and has received the bulk of research interest. Meeuwig et al. [14] illustrated that the relation between Chl and TN is marginally stronger than that between Chl and TP suggests that TN, rather than TP, limits estuarine Chl. The average TN: TP ratio of 4.5 also supports the argument for TN as the key limiting nutrient in estuaries in Prince Edward Island in Canada. However, the relative strength of these patterns and, the TN: TP ratio cannot be used to conclude which nutrient is limiting phytoplankton biomass, the low yield of Chl per unit nutrient points to the importance of other factors such as herbivory and turbidity, and potentially to indirect control by iron, in determining phytoplankton biomass.

Canfield [32] developed a Chl a equation using samples from 223 Florida lakes, 27% of which were considered N-limited. Because of the long growing season in Florida, these samples were taken during August 1979 to September 1980. Chl a showed significant correlations with both TP (r=0.79, p<0.01) and total N (TN) (r=0.87, p<0.01).

The TN relationship was obvious when plotting Chl a concentrations versus TP. Total P concentrations over 100 μ g L⁻¹, which for most lakes

^{*}Correlation is significant at the 0.05 level (2-tailed)

yielded TN/TP ratios fewer than 10, produced a gradient change of the TP/Chl a linear relationship. This suggested that P is the limiting nutrient lower TP levels of 100 μ g L⁻¹, and that N is the limiting nutrient higher than that concentration. Researchers have shown that there is often a strong correlation between total phosphorus (TP) and algal biomass [10,24,38,39]. This suggests that P may be the element controlling algal growth. Though, lakes surrounded by rich phosphate deposits and P-containing soils may be N-limited.

Wu and Chou [22] indicated that both the concentration of chlorophyll a and phytoplankton biomass have shorter Euclidean distances to silicate, nitrate, biochemical oxygen demand, and temperature, than to phosphate, nitrite, ammonium, or physical factors such as conductivity, pH, and dissolved oxygen, suggesting that phytoplankton are associated with silicate, nitrate, biochemical oxygen demand and temperature. These results supported the hypothesis that nutrients such as silicate and nitrate play a more important role in regulating phytoplankton in subtropical eutrophic estuary of Taiwan than do other factors.

In conclusion, results illustrated that there was a negative and significant relationships between chlorophyll a and logarithm chlorophyll a with nitrate, nitrite (P<0.01) and ammonia (P<0.05) but there was no significant correlation between chlorophyll a and logarithm chlorophyll a with silica, total alkalinity, sulfate and resolve phosphorus (P>0.05) in this research. Some research supported the result of this study and some of them were against. Although we could not find any relationship between Chl a and P it can be, because of measured resolve phosphorus instead of total phosphorus.

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