www.biodicon.com Biological Diversity and Conservation

ISSN 1308-8084 Online; ISSN 1308-5301 Print 9/1 (2016) 116-127

Research article/Araştırma makalesi

Seasonal monitoring of algal flora in the Pamukkale travertines and thermal springs (Denizli/Turkey)

Meltem ALTUNÖZ *[1], Olcay OBALI[1], Tahir ATICI[2], Laura ARRU[3, 4]

[1] Ankara University, Faculty of Science, Depart. of Biology, 06100, Ankara, Turkey
[2] Gazi University, Faculty of Education, Depart. Of Biology Education, 06560, Ankara, Turkey
[3] University of Modena and Reggio Emilia, Depart. of Life Science, 42122, Reggio Emilia, Italy
[4] University of Modena and Reggio Emilia, Interdepart. Research Centre Biogest-Siteia, 42122, Reggio Emilia, Italy

Abstract

In this study, the distribution and seasonal variation of the algal flora and some environmental parameters in the travertines and thermal water springs in Pamukkale-Denizli have been investigated between September 2010 and August 2011. For this purpose, a number of stations have been designated in the travertines, water channels, pools and ponds, where the algae samples were collected for the four seasons. Some of the environmental parameters such as temperature, pH, dissolved oxygen and electrical conductivity were measured in the study area in each season. The collected algae samples were identified, and their cell concentrations were calculated. The obtained data set was statistically analyzed by a series of tests in Statistical Package for Social Sciences (SPSS) software package. According to the Kruskal Wallis H test ($p < 0.05$, chi square > 5) there is a significant relationship between the water samples collected from each station and T, pH, DO. According to the Friedman test EC value has a significant relationship according to different seasons ($p < 0.05$). The highest EC value is observed in the spring, followed by summer, fall and winter, respectively. The relationship between the environmental parameters, species functional group, seasons and stations were observed by using Canonical Correspondence Analysis (CCA) software package. Negative and positive correlation and also the affinity of the species according to the environmental parameters were observed. In this study 35 genera, 57 taxa belonging to Cyanophyta (26 genera, 46 taxa), Charophyta (6 genera, 8 taxa), Chlorophyta (2 genera, 2 taxa) and Dinophyta (1 genus, 1 taxon) have been identified. *Oscillatoria limnetica* and *Cyanobacterium minervae* were the most abundant species, followed by *Chara*.sp., *Spirulina subsalsa, Chroococcua minutus*, *Anabaena* sp., *Schizothrix thermophila, Gloeocapsa* sp. and *Pseudanabaena papillaterminata*.

Key words: Thermophilic algae, Hierapolis Travertines, Phytoplankton, Benthic algae, Thermophilic Cyanophyta, Environmental parameters

---------- * ----------

Pamukkale travertenleri ve termal su kaynaklarindaki alg florasının mevsimsel izlemesi (Denizli)

Özet

Eylül 2010 - Ağustos 2011 tarihleri arasında Pamukkale–Denizli’de yapılmış olan bu çalışmada, Pamukkale travertenleri ve çevresinde bulunan sıcak su kaynaklarındaki alg florası belirlenmiş, alg türlerinin mevsimsel değişimi ve yoğunlukları araştırılmıştır. Travertenler, su kanalları, havuzlar ve göletler üzerinde dört mevsim süresince çeşitli istasyonlar belirlenerek, alg örnekleri elde edilmiş ve alg türlerinin teşhisleri yapılarak, hücre konsantrasyonları hesaplanmıştır. Çalışma alanında her bir mevsimde sıcaklık, pH, çözünmüş oksijen ve elektriksel iletkenlik gibi bazı çevresel parametreler ölçülmüştür. Elde edilen verilerin istatistiksel analizleri Statistical Package for Social Sciences (SPSS) paket programı ile gerçekleştirilmiştir. Kruskal Wallis H testine göre; T, pH, DO parametreleri ile herbir istasyondan elde edilen su örnekleri arasında, Friedman testiyle ise farklı istayonlarda ölçlülen EC değerleri arasında kayda değer bir ilişki olduğu gözlenmiştir ($p < 0.05$, ki kare > 5). En yüksek EC değeri ilkbahar mevsiminde gözlenmiştir; bunu sırasıyla yaz, sonbahar ve kış izlemiştir. Çevresel parametreler, türlerin fonksiyonel grupları, mevsimler ve istasyonlar arasındaki ilişki Canonical Correspondence Analysis (CCA) paket programı kullanılarak

---

[*] *Corresponding author* / Haberleşmeden sorumlu yazar: Tel.: +905326058539; Fax.: +905326058539; E-mail: altunozmeltem@gmail.com

belirlenmiştir. Türlerin çevresel parametrelere göre ilişkilerinin negatif ve pozitif korelasyonları, ayrıca türlerin bu parametrelere göre gösterdikleri eğilimler belirlenmiştir. Bu çalışmada Cyanophyta (26 cins, 46 tür), Charophyta (6 cins, 8 tür), Chlorophyta (2 cins, 2 tür) ve Dinophyta (1 cins, 1 tür) divizyolarına ait 35 cins, 57 tür teşhis edilmiştir. *Oscillatoria limnetica* ve *Cyanobacterium minervae* en yaygın rastlanan türlerdir ve bunları *Chara*. sp., *Spirulina subsalsa, Chroococcua minutus, Anabaena* sp., *Schizothrix thermophila,* Gloeocapsa sp. ve *Pseudanabaena papillaterminata* takip etmiştir.

Anahtar kelimeler: Termofilik algler, Pamukkale Travertenleri, Fitoplankton, Bentik algler, Termofilik Siyanofitler

1. Introduction

Algae are the unicellular organisms that contain primer photosynthetic pigment chlorophyll a and have a wide range of distribution in wetlands; for instance freshwater, salt water or brackish water. These organisms as an important source of oxygen are used by the grazers such as zooplankton in the higher steps of food chain. The organisms which are able to grow under extreme pH, salinity, temperature, nutrient concentration or radiation conditions are known as extremophilic organisms (Brock, 1978), and both prokaryotic (Cyanobacteria) and eukaryotic organisms can be found in this group (Seckbach, 2007). Hot springs represent one of the extreme habitats for these organisms since the water temperatures range from 30 °C up to boiling as 90-100 °C according to the altitude. Fumaroles can reach more than 100 °C which causes the water to be in the form of stream vapor; however, these habitats lack living organisms within (Brock, 1978). As a result of the geographical isolation and evolutionary divergence of the microorganisms in the thermophilic environments, thermophiles include endemic species due to the distribution of obligate thermophiles from rare and distant points of habitats. Biological diversity, endemism, species structure, phototrophic microvegetation of thermal algae and other photosynthetic organisms have been studied in different kinds of thermal springs (Castenholz, 1996; Kaštovský and Komárek, 2001; Kurt et al., 2013).

Chlorophyta, Cyanophyta and Myxophyceaecan conveniently grow in thermal waters (Ulcay et. al, 2007). Some members of the Cyanobacteria, Chlorophyta and Bacillariophyta divisions have been observed in the thermal springs of the Pamukkale Travertines throughout the previous studies (Güner, 1966; Pentecost et al., 1997). The algae play an important role as bioindicators for monitoring the ecological conditions of the aquatic habitats. Bioindicator algae species provide the information about the environmental conditions of the algae in question, which helps optimize the algal cultivation of a certain species (Shelknanloymilan et al., 2012). Furthermore, the flora and fauna of the freshwater have been observed for a long time in order to understand the bioindicator species according to their own habitats. However, thermal springs in particular provide significant hints as to the roughness of the habitat and its vicinity. Filamentous bacteria are the main microbial mat-forming organisms which include Cyanobacteria and other phototrophic bacteria that are strongly related to the mineral accumulation in their habitats (Ferris et al., 1987). Historical records of hot springs dating back to the period of the ancient Greeks and Mediterranean Romans also indicate the importance of the hot springs as an antique region; however, flora and fauna studies provide a set of data for the historical development of the relevant antique region (Florenzano et al., 2016).

Higher water temperature seems to increase algal growth to a certain degree; therefore, the number of habitats which can accommodate thermophilic organisms are expected to significantly increase in the near future. The species belonging to Cyanophyta draw specific attention on account of the increasing pollution of environment, especially by the blooms in the nutrient-rich aquatic habitats. The first organisms to have originated on Earth can be traced back to 3.5 billion years ago which are quite similar to the current members of Cyanobacteria (Whitton and Potts, 2002).

The present work deals with the search and identification of the algae flora, except diatoms, in the Pamukkale Travertines and their surroundings by taking into account the environmental parameters and the trend of algal species distribution according to the stations and seasons over one year, from 2010 to 2011.

1. Materials and methods

1.1. Study area

The “Hierapolis” (Pamukkale) is located in the southwest of Turkey, in the Aegean Region, Denizli (37°,54',34.7652" North; 29°,7',12.594" East). Hierapolis Travertines are formed by the thick lime deposits of underground water resources which are rich in calcium carbonate (Negri and Leucci, 2006). In general, the systems and biological incidents occurring in the wetlands, such as lakes, ponds or streams, should be monitored periodically to obtain the reliable data about the study area (Öztürk and Akköz, 2014). In the present study area, during the selection of the stations from where algal samples were taken; the points from which the water flows to the travertines, thermal water springs and ponds were taken into account. Station points were determined by GPS (Magellan eXplorist 610, United States). The chosen stations in the preliminary field surveys were evaluated in the same way in every season throughout the year. In this study, three stations were selected in the area (Figure 1).

Travertine Station (1st Station): At least six points from a wide range of travertine surface along the lime deposits ($CaCO_3$) were chosen to obtain samples during the study. The water channels provided for the travertines have shown differences during the year; therefore, it was not possible to obtain samples from the travertines which have not been irrigated, and no algae growth has been observed.

Pool Station (2nd Station): Human entry is prohibited in this area that consists of thermal water which hosts dense plant, plant based materials and an intense diversity of fauna compared to the other stations. However, this station is linked to the thermal springs of the Roman ruins Antique Pool (Cleopatra's Pool) which is currently a modern spa complex attracting the tourists also on account of the treatment provided by the doctor fish *Garra rufa* that is fed by dead skin cells of the human body.

Pond Station (3rd Station): It is the station, where the water flowing from the top of the travertine region through various channels is collected. The total water flowing from the top of the travertine hill consisted of the water in the pond which indicated the significance of the algae species distribution in the area.

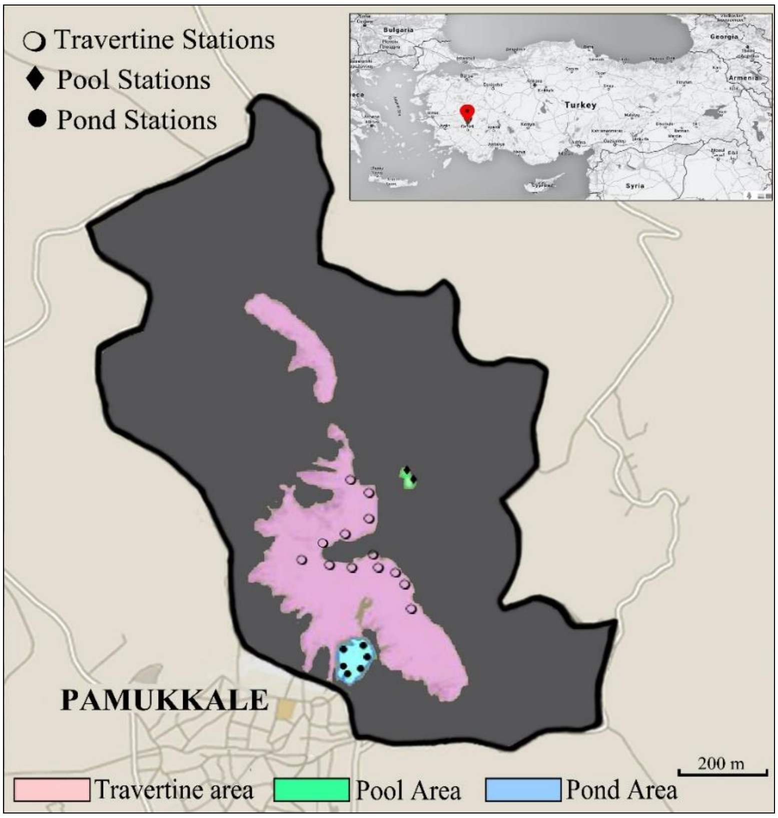


Figure 1. The map of the Pamukkale Travertines and the location of the sampling station

1.2. *Sampling methods, identification of algae species, cell counting and abundance analysis*

The samples were collected by using different methods according to the sampling stations. In the travertines, samples were scrapped by spatula. Plankton nets (55 micron, Hydrobios, Germany) were used to obtain samples from pool and pond stations. Algae samples were seasonally obtained, during one year in the fall, winter, spring and summer. The samples collected during the study were fixed using 4% formaldehyde and brought to the laboratory.

The algae samples were monitored by the light microscope (Leica DM-LS Type 020–518–500) after the needed dilutions have been prepared for the morphological species identification and cell count. Neubauer hemocytometer was used for the cell counting and calculating cell concentration (cells/ml) for each sample (Guillard, 1978).

The concentration (cells/ml) was calculated by using the following equations (Equation 1, 2):

$$\text{Concentration } (Cell/ml) = \frac{\text{Total Cells Counted}}{\text{Number Of Squares}} \times \text{Dilution Factor} \times 10000$$

Equation (1)

$$\text{Dilution Factor} = \frac{\text{Volume of Sample} + \text{Volume of Diluted Liquid}}{\text{Volume of Sample}}$$

Equation (2)

1.3. Data analysis

Canonical correspondence analysis (CCA) was performed to observe the relationship between the environmental parameters, species functional group, seasons and stations (CANOCO v.4.5 software). The obtained data set was analyzed in the package software SPSS 20.00 (Statistical Package for Social Sciences). Within the scope of the analysis, descriptive statistics, cross tables, bar graphics, chi square analysis, Kruskall Wallis H test, Mann Whitney U test and Friedman test were utilized. Whether the difference between the seasonal water parameters is statistically meaningful shall be established through the Friedman test in the second part of the analysis.

1.4. Environmental parameters

During the sampling period, measurements were made as to some of the environmental parameters such as temperature (T, °C), pH, dissolved oxygen (DO, mg O_2/L) and electrical conductivity (EC, µS/cm) of the water for one year. The temperature and dissolved oxygen were measured by YSI 51 model multiparameter, pH was measured by WTW340-A/SET pHmeter and EC was measured by WTW LF 92 conductivitymeter after the needed calibrations of the instruments were done in each station.

2. Results

In this study; 57 taxa, 35 genera belonging to Cyanophyta (26 genera, 46 taxa), Charophyta (6 genera, 8 taxa), Chlorophyta (2 genera, 2 taxa) and Dinophyta (1 genus, 1 taxon) were identified in the region of Pamukkale (Table 1). Identified taxa were checked on the basis of the related literature (John et. al, 2002b; Komarek and Anagnostidis, 2005; Komarek and Fott, 1983; Prescott, 1962; West, 1916; West and Fritsch, 1927) and the taxonomic information of the species were checked from the online database (http://www.algaebase.org) (Guiry and Guiry, 2015).

Table 1. The algae taxa identified in the thermal water springs in the Pamukkale Travertines and their vicinity

CYANOPHYTA	
Appendix I	**Taxa**
1	***Anabaena* sp.** Bory de Saint-Vincent ex Bornet and Flahault 1886
2	***Borzia trilocularis*** Cohn ex Gomont 1892
-	***Calothrix pilosa*** Harvey 1858
3	***Chroococcus giganteus*** West 1892
4	***Chroococcus minutus*** (Kützing) Nägeli 1849
5	***Chroococcus minutus* var. *thermalis*** Copeland 1936, Emoto and Hirose 1942, Palik 1949
6	***Chroococcus* varius** Braun 1876
7	***Coelosphaeriopsis chlamydocystis*** (Skuja) Komárek and Anagnostidis 1995
9	***Cyanobacterium minervae*** Komárek, Kopeck and Cepák 1999
10	***Cyanothece aeruginosa*** (Nägeli) Komárek 1976
11	***Eucapsis alpina*** Clements and Schantz 1909
12	***Glaucospira sp.*** Lagerheim 1892
13	***Gloeocapsa rupestris*** Kützing 1843
14	***Gloeothece dubia*** (Wartmann) Geitler 1932
-	***Gloeothece rupestris*** (Lyngbye) Bornet 1880
15	***Heteroleibleinia kuetzingii*** (Schmidle) Compère 1985
16	***Heteroleibleinia purpurascens*** (Hansgirg ex Hansgirg) Anagnostidis and Komárek 1988
-	***Jaaginema gracile*** (Böcher) Anagnostidis and Komárek 1988
17	***Jaaginema thermale*** Anagnostidis 2001
18	***Jaaginema unigranulatum*** (Biswas) Anagnostidis 2001
19	***Komvophoron constrictum*** (Szafer) Anagnostidis and Komárek 1988
20	***Komvophoron crassum*** (Vozzen) Anagnostidis and Komárek 1988
21	***Limnothrix guttulata*** (Van Goor) Umezaki and Watanabe 1994
22	***Oscillatoria limnetica*** Lemmermann 1900
23	***Oscillatoria limosa*** Agardh ex Gomont 1892
-	***Oscillatoria princeps*** Vaucher 1803
24	***Oscillatoria redeckei*** Goor 1918
25	***Oscillatoria simplicissima*** Gomont 1892

Table 1. The algae taxa identified in the thermal water springs in the Pamukkale Travertines and their vicinity (continuation of the table)

CYANOPHYTA	
Appendix I	**Taxa**
-	***Oscillatoria tenuis*** Agardh 1813
26	***Phormidium laminosum*** Gomont 1892
27	***Pseudanabaena limnetica*** (Lemmermann) Komárek 1974
28	***Pseudanabaena papillaterminata*** (Kiselev) Kukk 1959
29	***Pseudanabaena* sp.** Lauterborn 1915
30	***Rivularia sp.*** C.Agardh ex Boryanet and Flahault 1886
32	***Schizothrix fragilis*** (Kützing) Gomont 1892
31	***Schizothrix thermophila*** Copeland 1936
33	***Schizothrix tinctoria*** (C.Agardh) ex Gomont 1892
34	***Scytonema arcangelii*** Bornet and Flahault 1886
Appendix II	**Taxa**
1	***Spirulina major*** Kützing 1843
2	***Spirulina* robusta** H.Welsh 1965
3	***Spirulina subsalsa*** Oersted 1842
4	***Spirulina tenerrima*** Kützing ex Gomont 1892
5	***Stichosiphon willei*** (Gardner) Komárek and Anagnostidis 1995
-	***Symploca thermalis*** Kützing ex Gomont 1892
-	***Synechococcus aeruginosus*** Nägeli 1849
6	***Synechocystis. sp.*** Sauvageau 1892
CHAROPHYTA	
Appendix II	**Taxa**
9	***Chara sp.*** Linnaeus 1753
8	***Cosmarium laeve*** Rabenhorst 1868
-	***Cosmarium sp.*** Corda ex Ralfs 1848
12	***Gonatozygon monotaenium*** De Bary 1856
11	***Gonatozygon sp.*** de Bary 1858
13	***Mesotaenium sp.*** Nägeli 1849
15	***Spirogyra sp.*** Link 1820
16	***Spirotaenia sp.*** Brébisson 1848
DINOPHYTA	
Appendix II	**Taxa**
14	***Peridiniopsis thompsonii*** (Thompson) Bourrelly 1968
CHLOROPHYTA	
Appendix II	**Taxa**
8	***Protococcus viridis*** (Agardh) Silva 1980
7	***Tetraedron regulare*** Kützing 1845

1.5. Species distribution according to the stations, seasons and environmental parameters

Due to the fact that there are three different kinds of stations in the present study, algae species, environmental parameters and seasons were monitored according to the stations. Canonical correspondence analysis (CCA) data were used to observe triplot of ordination diagram under the Monte Carlo test (Figure 2 - 4). Length of environmental vector indicates its importance to the ordination. Direction of the vector indicates its correlation with each of the axes. Angles between vectors indicate the correlation between the environmental variables themselves.

1.5.1. Travertine station

According to the CCA analysis in travertine station, *B .trilocularis, P. laminosum, C. aeruginosa, C. pilosa, C. giganteus, S. fragilis, C. varius, S .arcangelii, C. laeve, S. major, S. aeruginosus* species were only observed in the fall season; *K. constrictum, Spirotaenia* sp., *H. purpurascens, S. thermophila* in the winter; *Glaucospira* sp., *G. dubia, L. guttulata, Pseudanabaena* sp. in the spring and *S. robusta* only in the summer (Figure 2).

The length of the environmental vector indicates the importance to the variation. Accordingly, temperature is not significantly meaningful for this station; however, EC, DO and pH variables appear to be important, respectively. *Glaucospira* sp., *G. dubia, L. guttulata, Pseudanabaena* sp. species have the highest affinity to EC. In the travertine station EC and DO variables have meaningful effects on species composition. The species close to origin of the axes are the common species in the station in question, such as *Chara*.sp., *S. subsalsa, C. minutus, O. limnetica, C. minervae* and *Anabaena* sp.

Gloeocapsa sp., *J. thernalis, P. limnetica, K. constrictum, H. pupurascens, Spirotaenium* sp. and *S. thermophila* similarly have the highest value of affinity to temperature variable. During the fall, diversity of the species was observed to be highest compared to the other seasons. Moreover, dissolved oxygen has the highest value in travertine station, meanwhile *Glaucospira* sp., *Pseudanabaena* sp., *G. dubia, L. guttulata* have the highest affinity to the EC in the fall season (Figure 2).

1.5.2. Pool station

S. major, S. willei, S. arcangelii, S. aeruginosus and *O. tenuis* species were observed only in the winter and the species in this season have positive correlation with EC parameter and negative correlation with water temperature. *Chara* sp., *C. pilosa, J. gracile* and *C. minutus* were only observed in the fall; accordingly, the species here have the highest affinity to DO, and negative correlation with EC. The species close to origin of the axes are the species that are common in the station in question, such as *Anabaena* sp., *O. limnetica, C. minervae, S. thermophila, S. subsalsa* and *P. papillaterminata* (Figure 3).

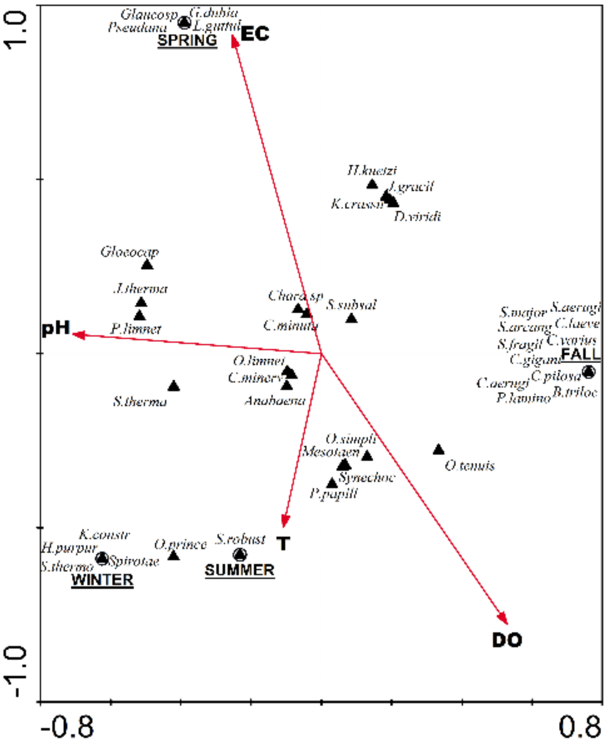


Figure 2. Diagram representing the CCA triplot frequency of every season, species and environmental factors in the travertine station

T: Temperature, **EC:** Electrical Conductivity, **DO:** Dissolved Oxygen

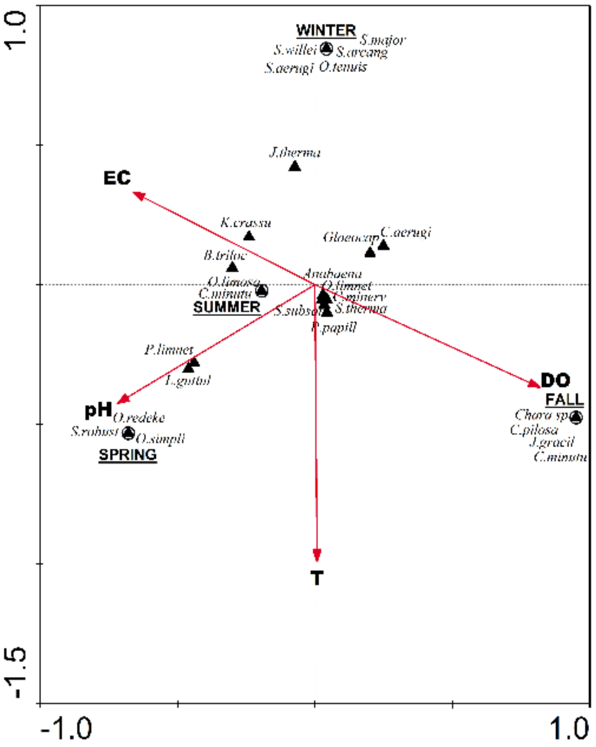


Figure 3. Diagram representing the CCA triplot frequency of every season, species and environmental factors in the pool station

T: Temperature, **EC:** Electrical Conductivity, **DO:** Dissolved Oxygen

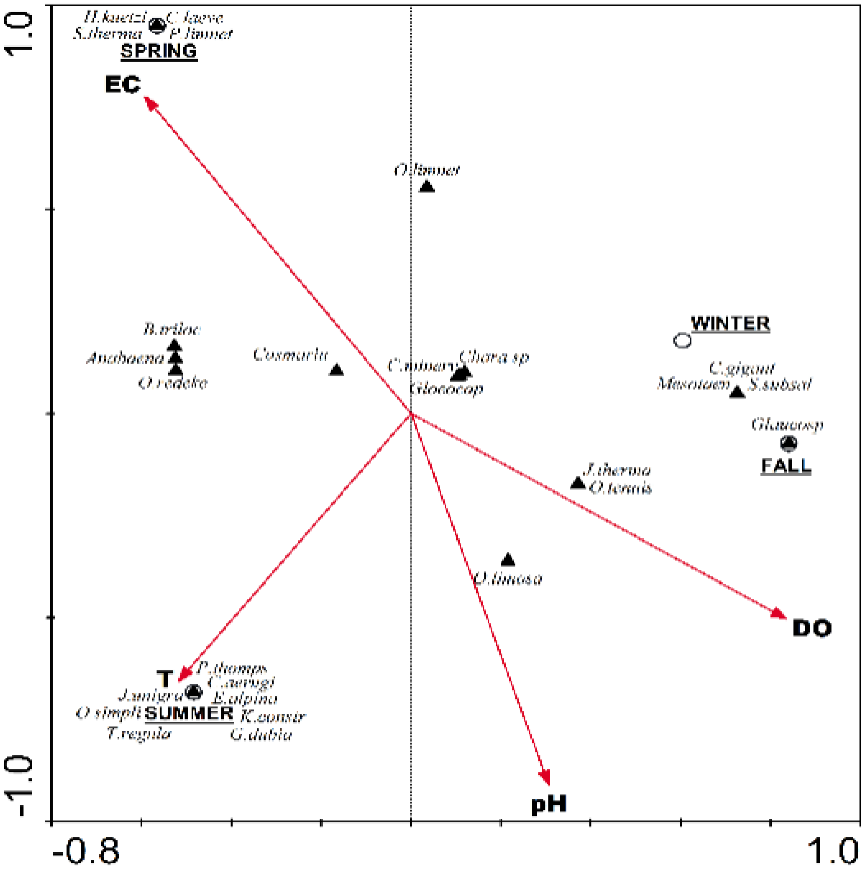


Figure 4. Diagram representing the CCA triplot frequency of every season, species and environmental factors in the pond station

T: Temperature, **EC:** Electrical Conductivity, **DO:** Dissolved Oxygen

1.5.3. Pond station

H. kuetzingii, S. thermalis, C. laeve and *P. limnetica* are the species which are only observed in the spring; accordingly, these species have the highest affinity to EC and negative correlation with the other environmental variables. *Glaucospira* sp. is observed only in the fall, which means this species has positive correlation with DO and pH, negative correlation with temperature and electrical conductivity. *P. thompsonii, C. aeruginosa, E. alpina, K. constrictum, G. dubia, T. regulare, O. simplicissima, J. unigranulatum* species were observed only in the summer; accordingly, they have a significant positive correlation with water temperature. Most abundant species observed in the pond station were *C. minervae*, *Chara* sp. and *Gloeocapsa* sp. (Figure 4).

1.6. *Data Analysis of Environmental Parameters*

Statistical significance between the water samples obtained from different stations in different seasons in relation to environmental parameters such as T, pH, EC and DO were analyzed by package software SPSS 20.00 in the following tests.

There is a significant relationship between the water samples collected from each station and T, pH, DO according to the Kruskal Wallis H test ($p < 0.05$, chi square > 5). EC values of the water samples collected from different stations do not have any significant relationship ($p > 0.05$, chi square < 5). The Mann Whitney U test was performed to determine the paired comparison with respect to the differences between the water parameter of the stations mentioned in Table 2.

According to the Mann Whitney U test, it is observed that T value of pool station is higher than that of the pond station. Moreover, pH value of the travertine station is higher than that of the pool station, and pH value of the pond station is higher than those of the travertine and the pool stations. Meanwhile DO value obtained from pond station is higher than the travertine and pool station (Table 3).

Table 2. The Comparisons with respect to stations using chi square test and statistical significance (p)

Parameter	*Station*	*N*	*Mean Rank*	*Chi Square*	*p*
T(ºC)	Travertine	4	6.75	6.060	0.048
	Pool	4	9.50		
	Pond	4	3.25		
pH	Travertine	4	6.50	9.881	0.007
	Pool	4	2.50		
	Pond	4	10.50		
EC	Travertine	4	5.50	1.846	0.397
	Pool	4	8.50		
	Pond	4	5.50		
DO	Travertine	4	6.25	9.302	0.010
	Pool	4	2.75		
	Pond	4	10.50		

Table 3. The comparisons between the stations using Mann Whitney U test and statistical significance (p)

Parameter		*Station*	*N*	*Mean Rank*	*MWU*	*p*
T (ºC)	***1st Comparison***	Pool	4	6.50	0.000	0.020
		Pond	4	2.50		
pH	***1st Comparison***	Travertine	4	6.50	0.000	0.020
		Pool	4	2.50		
	2nd Comparison	Travertine	4	2.50	0.000	0.020
		Pond	4	6.50		
	3rd Comparison	Pool	4	2.50	0.000	0.020
		Pond	4	6.50		
DO	***1st Comparison***	Travertine	4	2.50	0.000	0.020
		Pond	4	6.50		
	2nd Comparison	Pool	4	2.50	0.000	0.020
		Pond	4	6.50		

Significant difference is not observed between the seasons and T, pH, DO values according to the Friedman test ($p > 0.05$), except for EC value, it has a significant relationship according to different seasons ($p < 0.05$). The highest EC value is observed in the spring, followed by summer, fall and winter, respectively (Table 4).

3. Conclusions and discussion

The study area is a geothermal region that has extreme conditions in comparison to the other freshwater habitats depending on water temperature. In the travertine and pool stations, minimum and maximum temperatures of the water were measured as 30.5 °C and 35.6 °C, respectively. In the summer, the water temperature is observed to be higher than the other seasons; which is estimated to stem from the higher atmospheric temperature in the summer. In the spring maximum and minimum atmospheric temperatures are 12°C and 25 °C, followed by the maximum and minimum atmospheric temperatures of 20°C and 34 °C in the summer (World Weather Online), which explains the effect of atmospheric temperature on the water temperature during one year.

In a biogeographical habitat, the relation between the environmental parameters such as pH, temperature and the electrical conductivity indicates the distribution of the thermophilic Cyanobacteria (John et. al, 2002a). The most important groups of algal colonization in the non-acidic thermal waters are also known as Cyanobacteria. Filamentous Cyanobacteria members without heterocyst are typically found in the active travertines. Oscillatoria, Phormidium and Schizothrix genera are important members of Cyanobacteria among the algal flora of travertines (Pentecost, 2005). In the present study, 6 species belonging to Oscillatoria genus were observed quite abundantly in all stations while less species and lesser abundance were observed in terms of the other 2 genera. *Rivularia* sp. and *Calothrix* sp. that were observed rarely in the study area are referred to as the species with heterocyst that is found in the hot water springs (Pentecost, 2005).

Table 4. The comparisons with respect to seasons using chi square test and statistical significance (p)

	Season	***Mean Rank***	***Chi Square***	***p***
T(C^0)	Fall	2.83	3.000	0.392
	Winter	1.67		
	Spring	2.17		
	Summer	3.33		
pH	Fall	2.50	0.103	0.991
	Winter	2.50		
	Spring	2.33		
	Summer	2.67		
EC	Fall	2.00	9.000	0.029
	Winter	1.00		
	Spring	4.00		
	Summer	3.00		
DO	Fall	4.00	7.552	0.056
	Winter	2.83		
	Spring	1.50		
	Summer	1.67		

Single celled Cyanobacteria species are known to be more termophilic as *Synechococcus* sp. (Seckbach, 2007). This genus has been mentioned as an important species which can be found in the different thermal regions all over the world. *Cyanobacterium minervae* (basionym; *Synechococcus minervae* Copeland 1936) is the most abundant species in the study area with 34.2 % percentage in the pool, travertine and pond stations (Figure 2-4).

The species *O. princeps* was only observed in the travertine station which is already known as a facultative thermophile cyanobacterium found in both freshwater resources and the hot springs (Debnath et. al, 2009). *O. limnetica,* which has a wide range of temperature affinity, was also observed in all stations in this study. This species is known to be able to use sulfide as an electron donor source in photosynthesis. Sulfide content is an environmental parameter which allows the distribution of the species among the stations (Ward and Castenholz, 2002).

There are a few thermophile species of Chlorophyta which mostly belong to Charales, Chaetophorales and Zygnematales (Pentecost, 2005). In the study area, the pond station is a quite shallow lake covered by *Chara* sp. in the bottom which causes it to be regarded as “the Chara Lake”. This macro algae has rhizoidal structures that enable them to hold and spread on the sediment surface in the bottom of the lake. Chara lakes usually lack nutrients; these sediments are mostly blackened and rich in hydrogen sulphur. These lakes have alkali character, and the presence of Charophyta members are due to their ability to use bicarbonate ions in alkaline lakes of this character (Round, 1984). During the preliminary field survey in the Pamukkale Region, in October 2009, it was seen that the lake had been discharged to purify the area while *Chara* sp. assemblages and black mud were coming out from the bottom of the lake.

Dissolved oxygen that is measured in the pool station has the minimum value among all the stations. It is estimated that the way of thermal water source flowing to this station emerges from the underground water source which reduces dissolved oxygen in this station with the telluric effect. It is presumed that the reason why the dissolved oxygen ratio is higher in the pond station is the circulation of the lake and its purification in the previous year. The water flowing from the travertines is being gathered in this lake and afterwards, it leaves through the water channels which appears to provide the circulation in the lake. During the fall season dissolved oxygen is maximum compared to other seasons and in the spring electrical conductivity has the highest value; therefore, DO value is minimum. The EC of the water mainly means the concentration of dissolved solid materials in the water, meanwhile dissolved oxygen refers to amount of dissolved oxygen in the water. When the amount of solid matter increases in the water, the amount of dissolved oxygen decreases. As a consequence of this inverse proportion, DO is minimum while EC is maximum in the spring season.

Spirulina sp. is a common species found in hot water springs. In our study this genus is represented together with *Symploca thermalis*, as an epiphytic algae species, which shows the importance of epiphytic algae presence in the study area and in the thermal water springs in general. Almost all of the diversity and evenness indices depend on the relative abundance of the species. Abundance of different species in a sample, or species estimated in a community provide us with rare species or abundant species in this area. By virtue of the CCA analysis and SPSS analysis, it has been possible in this study to monitor the species distribution according to the environmental variations and to identify the significance of the comparison between the samples. Environmental factors and species frequency are quite important to understand the ecology of the aquatic habitats.

In this study, algae species have been identified and interpreted by using various techniques, which are important for the algal flora of thermal springs all over the world. The results showed us the presence of both common and rare thermal algal species and their correlation according to the environmental conditions. Algal flora plays a significant role in many fields ranging from potable water supply to the diversity of all living organisms.

Another aim of this research was to prospect into whether the tarnishing of the Pamukkale Travertines is based on algae growth which occurred in the recent years. As a result, the fact that the tarnishing of the travertines increases particularly during the periods in which travertines are not irrigated leads to the thought that the tarnishing does not stem from algae. During the field surveys, tarnishing was observed at the travertines' contact with air since dried travertine regions became white again when they were irrigated. However, in order to shed light on this issue, more detailed and long-term studies are required.

Acknowledgements

We would like to express our warmest thanks to Mert Elverici for his participation in the field surveys carried out in the Pamukkale Region. We are appreciated to Aytekin Altunoz and Sinem Özen for being with us during the preliminary studies of this research project. We are also thankful to the Pamukkale Protection of Special Area Agency Directorate for supporting this study, especially in the field surveys. This research was supported by The Scientific and Technological Research Council of Turkey (TUBITAK) under the project account no. 110Y201.

References

Brock, T. D. 1978. Thermophilic microorganisms and life at high temperatures: Springer-Verlag.

Castenholz, R. W. 1996. Endemism and biodiversity of thermophilic Cyanobacteria. Nova Hedwigia Beiheft, 112: 33-47.

Debnath, M., Mandal, N. C., Ray, S. 2009. The Study of cyanobacterial flora from geothermal springs of Bakreswar, West Bengal, India. Algae. 24/4: 185-193.

Ferris, F. G., Fyfe, W. S., Beveridge, T. J. 1987. Manganese oxide deposition in a hot spring microbial mat. Geomicrobiology Journal. 5: 33-42.

Florenzano, A., Mercuri, A. M., Altunoz, M., and Garcea, E. A. A. Palynological evidence of cultural and environmental connections in Sudanese Nubia during the Early and Middle Holocene. Quaternary International. doi: http:// dx. doi.org/10.1016/ j.quaint.2016.01.001.

Guillard, R. R. L. 1978. Counting slides. In Phytoplankton Manual-Monographs on Oceanographic Methodology France: UNESCO.

Guiry, M .D., Guiry, G. M. 2015. AlgaeBase. World-wide electronic publication, National University of Ireland, Galway. http://www.algaebase.org [16 December 2015].

Güner, H. 1966. Pamukkale termal suyunun mikroflorasi: Die Mikroflora der Thermalwasser von Pamukkale (Vol. 31). İzmir: Scientific Reports of the Faculty of Science, Ege University.

John, D. M., Whitton, B. A., Brook, A. J. 2002a. The freshwater algal flora of the British Isles : an identification guide to freshwater and terrestrial algae. Cambridge, UK: Cambridge University Press.

John, D. M., Whitton, B. A., Brook, A. J. 2002b. The freshwater algal flora of the British Isles: An identification guide to freshwater and terrestrial algae. Cambridge: Cambridge University Press.

Kaštovský, J., Komárek, J. 2001. Phototrophic microvegetation of thermal springs in Karlovy Vary, Czech Republic. Nova Hedwigia, Beihefte. 123: 107–119.

Komarek, J., Anagnostidis, K. 2005. Cyanoprokaryota, part 2. Oscillatoriales (B. Büdel, G. Gartner, L. Krienitz, M. Schagerl Eds. Vol. Band 19). Jena: Gustav Fischer.

Komarek, J., Fott, B. 1983. Chlorophyceae (Grünalgen) Ordnung: Chlorococcales. Das phytoplankton des Süsswassers (Vol. 7).

Kurt L., Ozbey, B.G., Kurt, F., Ozdeniz, E., Bolukbasi, A. 2013. Serpentine Flora of Turkey. Biodicon. 6/1: 134-152.

Negri, S., Leucci, G. 2006. Geophysical investigation of the temple of Apollo (Hierapolis, Turkey). Journal of Archaeological Science. 33: 1505-1513.

Öztürk, B.Y., Akköz, C. 2014. Investigation of water quality of Apa dam lake (Çumra - Konya) and according to the evalution of PCA. Biodicon. 7/2: 136-147.

Pentecost, A. 2005. *Travertine*. Berlin: Springer.

Pentecost, A., Bayari, S., Yesertener, C. 1997. Phototrophic microorganisms of the Pamukkale travertine, Turkey: their distribution and influence on travertine deposition. Geomicrobiology Journal, 14/4: 269-283.

Prescott, G. W. 1962. Algae of the western Great Lakes area, with an illustrated key to the genera of desmids and freshwater diatoms. Dubuque, Iowa: W. C. Brown Co.

Round, F. E. 1984. The ecology of the algae. Cambridge Cambridge Universty Press.

Seckbach, J. 2007. Algae and cyanobacteria in extreme environments. Hamburg, Germany: Springer.

Shelknanloymilan, L., Atici, T., Obali, O. 2012. Removal of nitrogen and phosphate by using Chlorella vulgaris on synthetic and organic materials waste water. Biodicon. 5/2: 89-94.

Ulcay, S. O., Kurt, T., Taskin, O., Ozturk, M. 2007. Distribution of thermal Cyanophyceae (Blue-Green algae species in Dikili (İzmir) thermal water. Turkish Marine Life Journal, 5/8: 371-378.

Ward, D. M., Castenholz, R. W. 2002. Cyanobacteria in geothermal habitats. The Ecology of Cyanobacteria their Diversity in time and space: Springer.

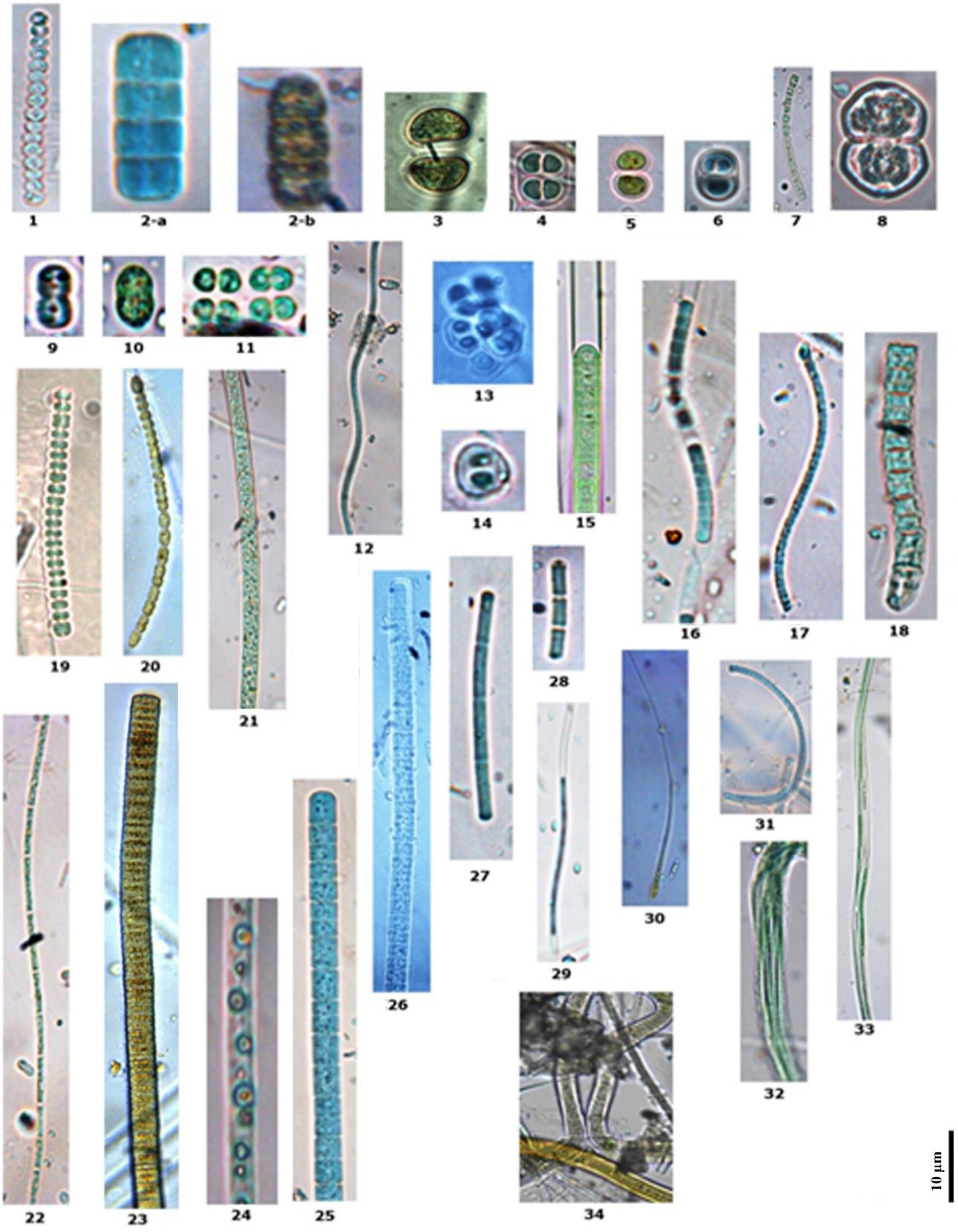
West, G. S. 1916. Algae I. Cambridge: Cambridge University Press.

West, G. S. and Fritsch, F. E. 1927. A treatise on the British freshwater algae. Cambridge: Cambridge University Press.

Whitton, B. A. and Potts, M. 2002. The ecology of cyanobacteria: their diversity in time and space (B. A. Whitton, M. Potts Eds.). Dordrecht: Kluwer Academic Publishers.

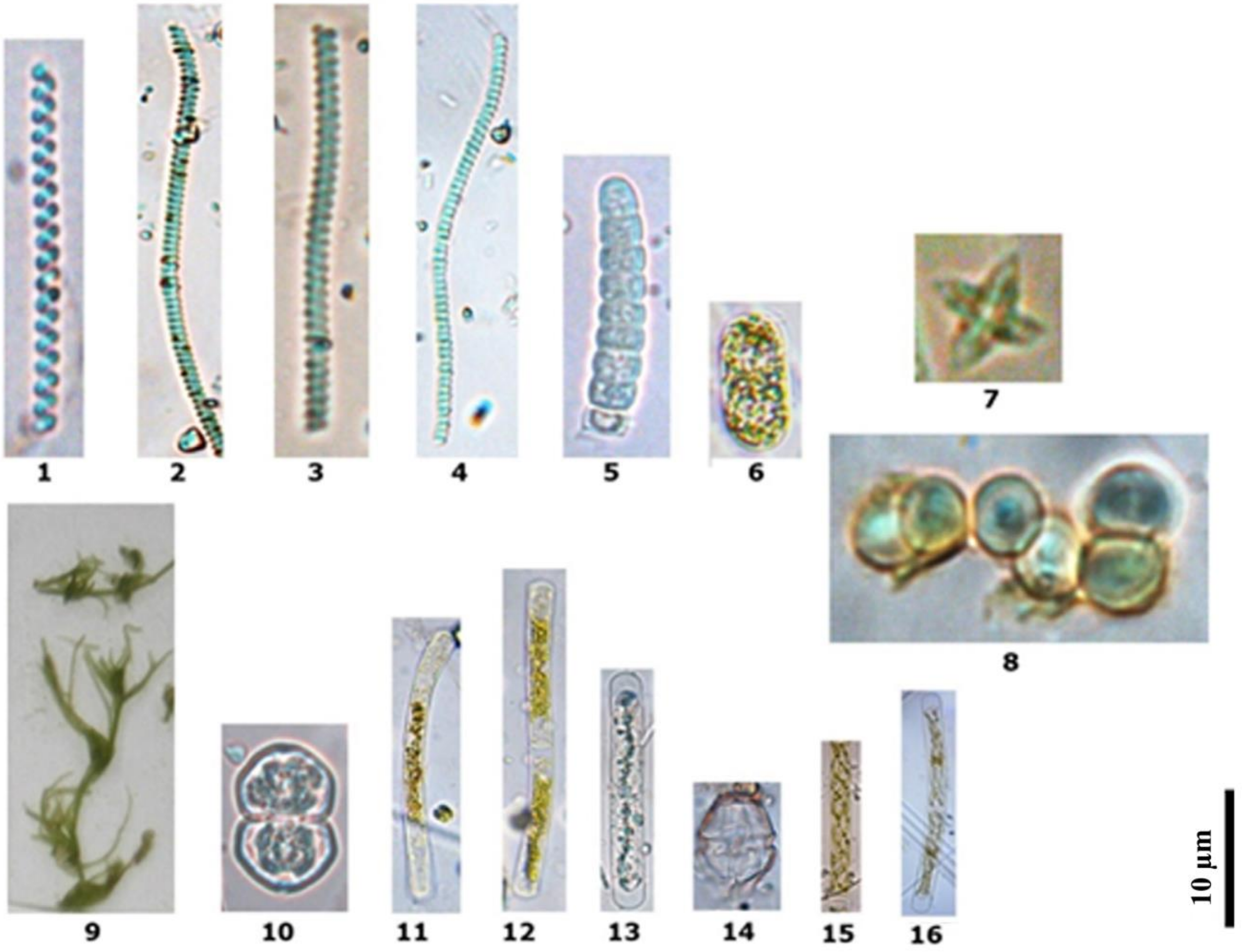
World Weather Online: http://www.worldweatheronline.com [Accessed March 2016].

APPENDIX I - Cyanophyta



1. *Anabaena* sp., ***2.*** *Borzia trilocularis, 3.* *Chroococcus giganteus,* ***4.*** *Chroococcus minutus,* ***5.*** *Chroococcus minutus var. thermalis,* ***6.*** *Chroococcus varius,* ***7.*** *Coelosphaeriopsis chlamydocystis,* ***8.*** *Cosmarium laeve,* ***9.*** *Cyanobacterium minervae,* ***10.*** *Cyanothece* sp., ***11.*** *Eucapsis alpina,* ***12.*** *Glaucospira* sp., ***13.*** *Gloeocapsa rupestris,* ***14.*** *Gloeothece dubia,* ***15.*** *Heteroleibleinia kuetzingii,* ***16.*** *Heteroleibleinia purpurascens,* ***17.*** *Jaaginema thermale,* ***18.*** *Jaaginema unigranulatum,* ***19.*** *Komvophoron constrictum,* ***20***. *Komvophoron crassum,* ***21.*** *Limnothrix guttulata,* ***22.*** *Oscillatoria limnetica,* ***23.*** *Oscillatoria limosa,* ***24.*** *Oscillatoria redeckei,* ***25.*** *Oscillatoria simplicissima,* ***26.*** *Phormidium laminosum,* ***27.*** *Pseudoanabaena limnetica,* ***28.*** *Pseudanabaena papillaterminata,* ***29.*** *Pseudanabaena* sp., ***30.*** *Rivularia* sp., ***31.*** *Schizothrix thermophila,* ***32.*** *Schizothrix fragilis,* ***33.*** *Schizothrix tinctoria,* ***34.*** *Scytonema arcangelii* (Scale 10µm)

APPENDIX II - Cyanophyta, Charophyta and Chlorophyta



1. *Spirulina major,* **2.** *Spirulina robusta,* **3.** *Spirulina subsalsa,* **4.** *Spirulina tenerrima,* **5.** *Stichosiphon willei,* **6.** *Synechocystis* sp., **7.** *Tetraedron regulare,* **8.** *Protococcus viridis,* **9.** *Chara* sp., **10.** *Cosmarium laeve,* **11.** *Gonatozygon* sp., **12.** *Gonatozygon monotaenium.,* **13.** *Mesotaenium* sp., **14.** *Peridiniopsis thompsonii,* **15.** *Spirogyra* sp., **16.** *Spirotaenia* sp. (Scale 10µm – except *Chara* sp.)

(Received for publication 15 April 2015; The date of publication 15 April 2016)