FLORISTIC STUDY ON CYANOPHYTA OF THREE HOT SPRINGS OF HORMOZGAN PROVINCE, IRAN

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Cyanophyta are the photosynthetic prokaryotes found in most of the ecological habitats. They are very diverse group in concept of morphology and physiology and have been an interesting group for investigation. In the present investigation thermophilic cyanophyta have been identified from three hot springs including Fariyab, Todlooye and Fotooye located in west of Hormozgan province. Sampling was based on temperature gradients. Water physicochemical characteristics analyzed because of their dramatic effects on species distribution. The collected samples from different collection sites and various temperature ranges were fixed in 4% formaldehyde to minimize quantitative and qualitative changes. Taxonomic determination was carried out using artificial keys presented in some valid publications. As a result of this study, a total of 66 species were identified including 56 species, 22 genera and 10 families from Fariyab hot spring; 29 species, 11 genera and 6 families from Fotooye hot spring and 12 species, 6 genera and 2 families from Todlooye hot spring.

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Key words: Cyanophyta; hot springs; thermophilic; Hormozgan province; Iran

مطالعه فلوریستیکی جلبکهای سبز آبی سه چشمه آب گرم در استان هرمزگان

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سیانوفیتا یا جلبکهای سبز آبی موجودات پروکاریوتی فتوسنتزی می باشند که در اغلب زیستگاههای اکولوژیکی یافت می شوند که گروهی متنوع از جنبه های مورفولوژیک و فیزیولوژیک بوده و علاقه مندی خاصی برای تحقیق در گروه را باعث گردیده است. در این تحقیق شناسایی جلبکهای سبز آبی گرما دوست در سه چشمه آب گرم فاریاب ، تودلویه و فتویه واقع شده در غرب استان هرمزگان مورد مطالعه قرار گرفت. نمونه ها از مکانهای مختلف با دما های متعدد جمع آوری گردید و بخاطر تاثیر خواص فیزیکی و شیمیایی آب چشمه ها روی تنوع گونه ای ، ویژگیهای فیزیکوشیمیایی آب چشمه های مورد نظر مورد آنالیز قرار گرفت . به منظور به حداقل رساندن تغییرات کمی و کیفی فیتوپلانکتونها ، نمونه های جمع آوری شده درمحلول فرمالین ۴٪ نگهداری شد. مطالعات تاکسونومیکی بر اساس کلیدهای معتبر بین المللی انجام پذیرفت. در این تحقیق ۶۶ گونه شناسایی شد که در چشمه آب گرم فاریاب ۵۶ گونه جلبکی متعلق به 22جنس و ۱۰ خانواده ، در چشمه آب گرم فتویه ۲۹ گونه متعلق به ۱۱ جنس و ۶ خانواده در چشمه آب گرم تودلویه و ۱۲ گونه متعلق به ۶ جنس و ۲ خانواده شناسایی شد.

INTRODUCTION

The prevail environmental condition during the origination of cyanophyta, in Precambrian included high temperature, poor amount of free oxygen, high concentration of sulfur and lots of reducing gases like methane, ammonia, and carbondioxide that shows their high tolerant ability in severe stressful environmental condition. Few organisms such as thermophile

unicellular prokaryotes belong to Archaea and Bacteria domains can withstand these hostile environments.

Highly developed cyanophyta in microbial mats of geothermal springs are common at temperature less than 74°C and the PH more than 5.0. *Synechococcus*, *Phormidium*, *Calothrix*, and *Mastigocladus* are some of the cyanophyta taxa found in thermal springs. However their distribution pattern varies with temperature ranges. Many studies have been focused on the cyanophyta of the thermal springs (Castenholz 1969 &1996; Brock 1978; Ward & al. 1987 & 1989; Sompong & al. 2005).Various names, such as cyanobacteria, blue-greens, blue-green algae, myxophyceans, cyanophytes, cyanophyceans, cyanoprokaryotes have been used for the Blue-Green Algae (BGA) which are the largest and most diverse group of photosynthetic prokaryotes (Stanier and Cohen-Bazire 1977). Iran has located on the arid belt of the world. However a small area of the northern part is affected by the temperate Mediterranean condition, but extreme severe environmental condition such as high and low temperatures, highly diverse salinity and PH ranges in inland and coastal water (Zarei-Darki 2011) make lots of diverse habitats in the country.

Cyanophyta are known to inhabit in the various extreme environments with wide range of temperature and sand salinity (Kondratyeva & al. 1984). Therefore, very diverse natural conditions of the inland waters of Iran are interesting to study the composition and distribution of blue-green algae. Some studies have been carried out on the blue-green algae of Iran (Woronichin 1925; Löffler 1959 & 1961; Hirano 1973; Wasylik 1975; Compere 1981). Cyanophyta in hot springs are largely unexplored. Hot springs are well-isolated habitats occurring as clusters in globally distant regions, and the microorganisms that inhabit in the hot springs are extra tremophiles which can survive and adapt to the severe environmental conditions (Papke & al. 2003). The hot spring mat communities in terrestrial habitats have been attractive for microbial ecologists because of their unique adaptations to the harsh environments (Brock 1967). In the most of the studies screening efforts have been done on mesophilic cyanophyta (Fish 1994) and in a little numbers of the reports screening was on thermotolerant or thermophilic genera. The present study aims to screen and culture thermophilic cyanobacteria from 3 thermal springs in Hormozgan province (Fariyab, Fotooye, Todlooye) to study their floristic composition and distribution in these hot springs.

MATERIALS AND METHODS

Three hot springs were selected in the present study. They are usually covered by cyanobacterial mats all over the year. All of these hot springs are located in western part of the Hormozgan province and characterized by highly alkaline water and high temperature. Samples collected from various temperatures were fixed in 4% formaldehyde to minimize quantitative and qualitative changes in the phytoplankton.

Due to the effects of physicochemical properties of water on the species distribution, water samples were collected from 10-30 cm and 30-50 cm depth from water edge (Masoumi & al. 2011) and analyzed to determine the chemical characteristics, including Total Hardiness (TH), Total Dissolved Solid (TDS), Alkalinity, Potassium (K^{+}), Calcium (Ca^{++}), Sodium (Na+) , Ammonium (NH4+) , Nitrite ($NO2^{-}$), Sulphate ($SO4^{-2}$), Dissolved Oxygen (DO), Chloride (Cl^{-}) and inorganic Phosphate ($PO4^{-3}$). Water temperature was measured in the field by a thermometer. Analyses were performed in the laboratory using Iran Standard 1053 method (table 1).

Cyanophyta species were studied within 24 hours after collection using light microscope (GX Australia). The mat samples temporary slides were made using glycerol 10%. The semi-permanent slides in glycerine were made and sealed with enamel paint for microscopic study. Sketches were prepared using camera lucida of Olympus binocular microscope and cells size were measured using ocular micrometer. Taxonomic identification of the species was carried out using the keys and description of Descikachary (1959), Anagnostidis and Komarek (1988 & 1990); Komarek and Anagnostidis (2005a & b). The vegetative and reproductive characters such as shape, color and size of the vegetative and apical cells and talus, presence and texture of mucilage sheath were studied as the main characters for taxonomic identification. Length and width of the cells were measured for 10 individuals of each species. Species contributing hot spring cyanobacterial mats were isolated from water samples by incubation on agar plate using BG11 media (Stanier & al. 1971). Photographs were taken using light microscope (GX Australia). Voucher specimens have been preserved in 4% formalin and deposited in department of Botany, Shahid Beheshti University, Tehran, Iran.

RESULTS

Physicochemical parameters of water

All spring waters were alkaline, transparent and colorless with strongly smelling of sulphate. Physicochemical analysis indicate that all the studied springs were classified as brackish and very hard water (TDS >5000 mg L^{-1},TH > 300 mg L^{-1}) with high density of Ca^{++}, Mg^{+} , Na^{+} , Nitrate, Ammonium,

Table 1. Physicochemical parameters in the three studied hot springs.

Parameters	Standard No.	Scale	Studied Sites		
			Fariyab	Footie	Todlooye
Temperature Range		° C	34-39 ° C	37-42 ° C	51-56 ° C
Electrical conductivity(EC)	2510B	µS/cm	31070	41320	49950
Turbidity	2130B	NTU	10.3	4.21	3.62
PH	HB 4500		6.76	6.70	6.68
Total Dissolved Solid) TDS (in180 □c)	2540 C	mg/L TDS	20800	32100	36100
OH- (mg L-1)/ compare with Phenolphthalein	2220 .B	mg/L Caco3	0	0	0
OH- (mg L-1) compare with methyl orange	2220. B	mg/L Caco3	172	154.9	148
TotalHardness (TH)	2240 .B	mg/L Caco3	1790	2420	3535
Ca++(mg L-1)	3500 .B	mg/L Ca	579.15	1112.20	1162.3
Mg++ (mg L-1)	3500-Mg. B	mg/L Mg	83.83	110.3	154.3
Na+(mg L-1)	3500-Na. B	mg/L Na	8600	9220	9320
K+(mg L-1)	3500-K .B	mg/L K	66	164.12	171.42
Fe2+(mg L-1)	3500-Fe .B	mg/L Fe	-	-	-
Mn+(mg L-1)	3500-Mn. B	mg/L Mn	-	-	-
NH4+(mg L-1)	4500-NH2.G	mg/L NH3-N	-	-	-
Cl- (mg L-1)	4500-Cl.B	mg/L Cl-	12621.6	14100.52	13208.68
F- (mg L-1)	4500-F.B	mg/L F	-	-	-
SO4 -2 (mg L -1)	4500-So42-.E	mg/L So42-	1850	3255	3570
PO 4-3 (mg L-1)	4500-P.D	mg/L Po4-	1.1	0.82	1
NO3 – (mg L -1)	4500-No3.B	mg/L No3	0.8	7.2	8.8
NO2 – (mg L -1)	4500-No2.B	mg/L No2	0.01<	0.01<	0.01<

Phosphate, Silicate and Sulfate. This study indicated increasing of pH and decreasing of some parameters such as total dissolved solid (TDS), total hardness (TH), density of Ca^{++}, Mg^{+} , Na^{+} , K^{+} and CL^{-} have increased the diversity of cyanobacteria. The highest species number was found in Fariyab (PH 6.76 , TDS 20800 mg L^{-1}, TH 1790 mg L^{-1} , Ca^{++} 579.15 mg L^{-1} , Mg^{+} 83.83 mg L^{-1}, Na^{+} 8600 mg L^{-1} , K^{+} 66 mg L^{-1}, CL 12621.6 mg L^{-1}) and lowest species number was found in Todlooye (PH 6.68 , TDS 36100 mg L^{-1}, TH 3535 mg L^{-1} , Ca^{++} 1162.3 mg L^{-1} , Mg^{+} 154.3 mg L^{-1}, Na^{+} 9320 mg L^{-1} , K^{+} 171.42 mg L^{-1}, CL 13208.68 mg L^{-1}).

Mat morphology and cyanophyta species composition

Results indicated that microbial mats in geothermal springs are predominated by cyanophyta flora. The identification of the most representative cyanophyta of the studied communities revealed a total of 66 species in three studied hot springs including 56 species of 22 genera, 10 families and 2 orders at a temperature range of 34-39°C in Fariyab, 29 species of 11 genera, 6 families and 2 orders at a temperature range of 37-42°C in Fotooye and 12 species of 6 genera, 2 families and 2 orders at a temperature range of 51-56°C in Todlooye (fig. 1, table 2). Temperature was one of the most important parameters affected cyanophyta species diversity in microbial mat of the hot springs. The studies revealed that cyanophyta diversity and complexity decreased in high temperature. The lowest species diversity was observed in Todlooye hot spring with temperature range 51- 56°C and the highest species diversity was found in Fariyab hot spring with a temperature range of 34-39°C. The relationship between temperature and the number of species was significant (fig. 2).

DISCUSSION

As several reports indicate, one of the most important parameters for cyanoprokaryota species diversity in hot springs is temperature. Therefore thermal gradients can determine the occurrence of the species in thermal springs. Most of the cyanobacterial species were recorded in the temperature range 35–40°C, followed by 40–50°C, 50–60°C and 60–70°C. Diversity and complexity of cyanobacteria species decrease with increasing temperature (Ferris & al. 1996a & b; Ward & al. 1998). Results of this study also support the previous studies. Maximum species diversity was observed in Fariyab (34-39°C), and minimum species diversity in Todlooye (51-56°C). Skimisdottir & al. (2000) showed that in thermal gradients from 50°C to 75°C, the layered mats are characterized by the presence of unicellular forms such as Synechococcus. The cyanobacterial mats occurring

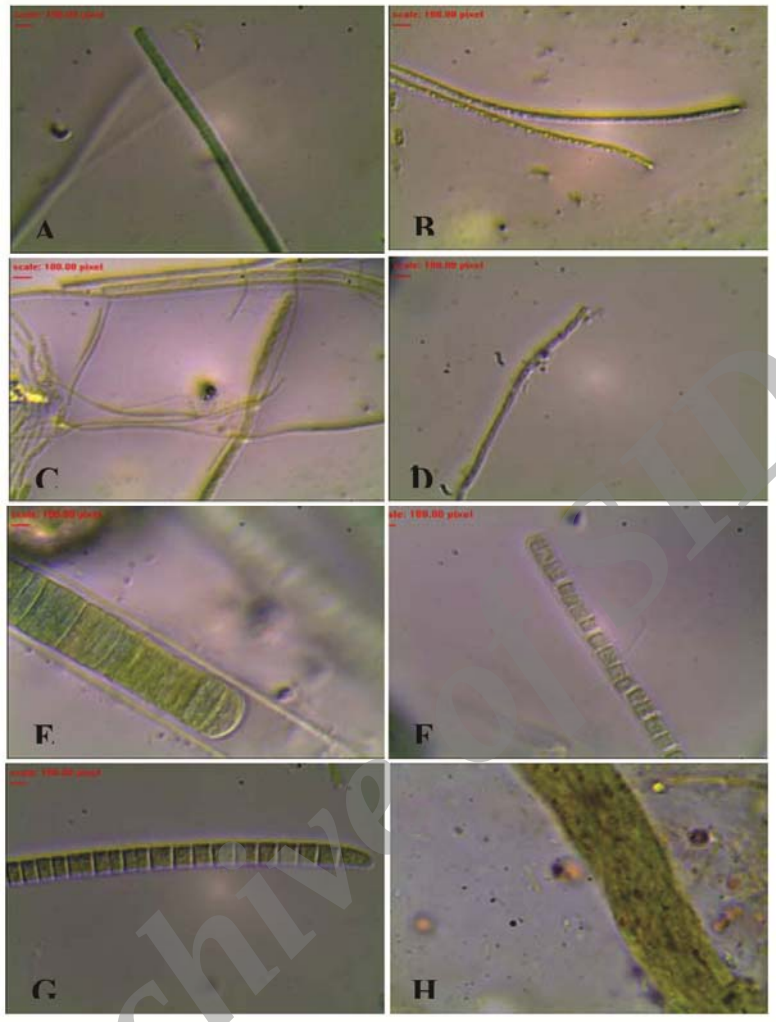

Fig. 1. A, *Jaaginema pseudogeminatum*; B, *Limnothrix mirabilis*; C, *Pseudanabeana limnetica*; D, *Pseudanabeana catantea*; E, *Lyngbya major*; F, *Phormidium diguetii*; G, *Phormidium formosum*; H, *Tricocoleous sociatus*.

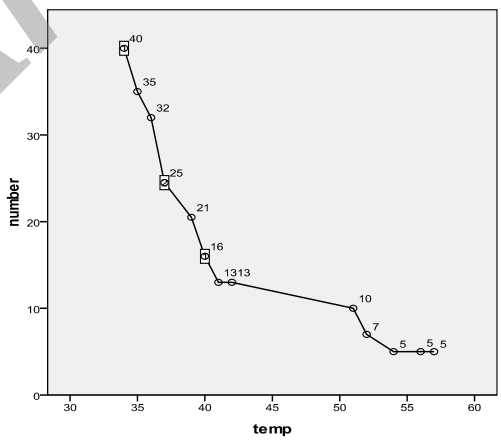

Fig. 2. Relationship between the temperature and species number in studied hot springs.

Table 2. List of Cyanoprokaryota species (Oscilatoriales Order) in studied hot springs.

Taxa	Fariyab					Fotooye					Todlooye				
	27º 25′ 57/81″ N 54º 16′ 44/78″ E					27º 19′ 59/35″ N 54º 24′ 24/70″ E					27º 17′ 34/46″ N 55º 46′ 16/22″ E				
	34 ºC	35 ºC	36 ºC	37 ºC	39 ºC	37 ºC	39 ºC	40 ºC	41 ºC	42 ºC	51 ºC	52 ºC	54 ºC	56 ºC	57 ºC
Jaaginema angustissimum (W. et G.S. West) Anagnostidis et Komárek1988	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Jaaginema metaphyticum (Komárek in Anagnostidis et Komárek1988)	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Jaaginema pseudogeminatum (Schmid) Anagnostidis et Komárek1988	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
**Jaaginema thermale* Anagnostidis 2001				+	+										
Jaaginema subtilissimum (Kützing ex De Toni) Anagnostidis et Komárek 1988	+	+													
Jaaginemaa geminatum (Meneghini ex Gomont) Anagnostidis et Komárek 1988	+	+				+	+	+	+	+	+	+	+	+	+
Jaaginema quadripunctulatum (Brühl et Biswas) Anagnostidis et Komárek 1988											+	+	+	+	+
Limnothrix mirabilis (B□cher) Anagnostidis 2001	+		+												
Limnothrix redekei (Van Goor) Meffert 1988	+	+	+	+	+										
Pseudanabaena mucicola (Naumann et Huber-Pestalozzi) Schwabe1964	+														
Pseudanabaena limnetica (Lemmermann) Komárek 1974											+	+			
Pseudanabaena catenata Lauterborn 1915	+	+	+	+											
Pseudanabaena frigid (Fritsch) Anagnostidis 2001	+	+													
Geitlerinema amphibium (Agardh ex Gomont) Anagnostidis 1989	+	+	+	+		+	+	+			+	+	+	+	
Geitlerinema thermale Anagnostidis 2001													+	+	
Geitlerinema jasorvense (Vouk) Anagnostidis 1989				+											
Spirulina major Kützing ex Gomont 1892	+	+													
Spirulina meneghiniana Zanardini ex Gomont 1892	+	+	+												
Spirulina subtilissima Kützing ex Gomont 1892	+	+	+	+	+	+	+	+	+	+	+				
Spirulina tenerrima Kützing ex Gomont 1892	+	+	+	+	+	+	+	+	+	+	+				
Planktolyngbya limnetica (Lemmermann) Komárk ová-Legnerová et Cronberg 1992	+	+	+	+	+										
Leptolyngbya foveolarum (Rabenhorst ex Gomont) Anagnostidis et Komárek 1988	+	+													
Lyngbya martensiana Meneghini ex Gomont 1892	+	+	+	+	+	+	+	+	+	+					

Table 2. Continued.

Taxa	Fariyab					Fotooye					Todlooye				
	27º 25′ 57/81″ N 54º 16′ 44/78″ E					27º 19′ 59/35″ N 54º 24′ 24/70″ E					27º 17′ 34/46″ N 55º 46′ 16/22″ E				
	34 º C	35 º C	36 º C	37 º C	39 º C	37 º C	39 º C	40 º C	41 º C	42 º C	51 º C	52 º C	54 º C	56 º C	57 º C
*Lyngbya nigra*Agardh ex Gomont 1892			+	+	+										
Lyngbya hieronymusii Lemmermann 1905	+	+	+												
Lyngbya thermalis Kützing ex Gomont 1892				+	+			+	+	+					
Lyngbya aestuarii Liebman ex Gomont 1892	+	+				+	+								
Lyngbya maior Meneghini ex Gomont 1892	+		+												
Oscillatoria limosa Agardh ex Gomont 1892	+	+				+	+								
Oscillatoria tenuis Agardh ex Gomont 1892	+	+	+	+	+	+	+	+	+	+					
Oscillatoria subbrevis Schmidle 1901	+	+				+	+								
Phormidium lucidum Kützing ex Gomont 1892	+														
Phormidium ambiguum Gomont ex Gomont 1892	+	+	+	+	+	+	+	+	+	+	+				
Phormidium tergestinum (Kützing) Anagnostidis et Komarek1988					+	+									
Phormidium diguetii (Gomont) Anagnostidis et Komárek 1988	+	+	+	+											
Phormidium chalybeum (Mertens ex Gomont) Anagnostidis et Komárek 1988	+	+		+											
Phormidium formosum (Bory ex Gomont) Anagnostidis et Komárek 1988			+	+	+										
Phormidium pachydermaticum Frëmy 1930	+	+													
Phormidium ornatum (Kützing ex Gomont) Anagnostidis et Komárek 1988			+	+	+										
Komvophoron minutum (Skuia) Anagnostidis et Komárek 1988				+											
Gomontiella marthae Claus 1960	+														
Trichocoleus sociatus (W. et G.S. West) Anagnostidis 2001			+												
Aphanocapsa thermalis Brügger 1863				+	+			+	+	+					
Aphanocapsa elachista W. et G.S. West 1894	+	+	+	+	+	+	+								
Aphanocapsa salina Voronichin 1929					+										
Aphanocapsa delicatissima W. et G.S. West 1912	+	+	+	+	+										
Synechocystis thermalis Copeland 1936				+	+										
Synechocystis aquatilis Sauvagea 1892	+	+	+			+	+	+	+	+	+	+			
Synechocystis minuscule Voronichin 1926	+	+	+												

Table 2. Continued.

Taxa	Fariyab 27º 25′ 57/81″ N 54º 16′ 44/78″ E					Fotooye 27º 19′ 59/35″ N 54º 24′ 24/70″ E					Todlooye 27º 17′ 34/46″ N 55º 46′ 16/22″ E				
	34 ºC	35 ºC	36 ºC	37 ºC	39 ºC	37 ºC	39 ºC	40 ºC	41 ºC	42 ºC	51 ºC	52 ºC	54 ºC	56 ºC	57 ºC
Chroococcus minutes (Kützing) Nägeli 1849						+	+	+	+	+					
Chroococcus turgidus (Kützing) Nägeli 1849	+	+	+			+	+	+	+	+					
Chroococcus globosus (Elenkin) Hindák 1978	+	+	+			+	+	+	+	+					
Chroococcus membraninus (Meneghini) Nägeli 1849	+	+				+	+								
Chroococcus limneticus Lemmermann 1898									+						
Chroococcus minor (Kützing) Nägeli 1849									+						
Chroococcus thermalis (Meneghini)Nägeli 1849										+					
Chroococcus tenax (Kirchner)Hieronymus 1849									+						
Chroococcus macrococcus (Kützing) Rabenhorst 1865						+	+	+							
Synechococcus elongates (Nägeli) Nägeli 1849						+	+	+							
Cyanobacterium minervae (Copeland) Komárek et al 1999					+	+	+	+							
Eucapsis starmachii Komárek et Hindák 1989	+	+	+												
Microsystis sp.	+	+	+												
Aphanothece minutissima (W. Weat) Komárko vá-Legnerová et Cronberg 1994	+	+													
Aphanothece nidulans Richter in Wittrock &Nordstedt 1884	+	+													
Cyanosarcina thermalis (Hindák) Kováčik 1998	+	+													
Cyanosarcina chroococcoides (Geitler) Kováčik 1988	+	+													

at the lower end of thermophily (40-50°C) are often dominated by filamentous cyanobacteria like *Phormidium, Oscillatoria, Pseudanabaena, Calothrix* and *Fischerella* (Ward & Castenholz 2002; Sompong & al. 2005). However, Norris & al. (2002) reported that cyanobacteria such as *Synechococcus* also co-occur with other unicellular and filamentous forms at lower temperature. In the present study, *Jaaginema* has been found in all studied hot springs and in all temperature gradient and *Synechococcus* found in 39-42 °C with other unicellular and filamentous forms. Some studies also focus on the role of PH and other parameters on the species distribution in the communities of cyanobacterial mats below 60 °C (Ward & Castenholz 2002; Sompong & al. 2005). Results of this study showed that increasing of PH and alkalinity and decreasing of other factors such as TDS, TH, density of Ca^{++}, Mg^{+}, Na^{+} K^{+} and CL^{-} are important factors affecting species diversity and the diversity in cyanobacterial species is affected by micro-chemical characteristics of the spring waters.

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