# SOME NEW RECORDS OF OSCILLATORIAN CYANOPHYTA FROM PADDY FIELDS OF GOLESTEN PROVINCE

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*Cyanophyta* of some paddy fields of Golestan Province (North of Iran and near Caspian Sea) have been studied between autumn 2006 and summer 2007. *Lyngbya perelegans, L. rubida, L. laxespiralis, L. sordida, L. putealis, L. dendrobia, L. scotii, L. polysiphoniae, L. erugineo–coerulea, L. cryptovaginata, L. spiralis, L. nigra, L. kashyapii are new records for Golestan province and Iran. Results showed that <i>Lyngbya rubida*, in spring and winter, *L. perelegan* in summer, *L. kashyapii* and *L. dendrobia* in autumn were dominant species in all stations. Morphological characteristics of these species and some information about their ecological distribution are given.

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Key words. Cyanophyta, Iran, Golestan province, paddy field, taxonomy.

Lyngbya perelegans Lemm, L. rubida Fremy, L. laxespiralis Skuja, L. sordida Gomont, L. putealis Mont. ex Gomont, L. dendrobia Bruhl & Biswas, L. scotii Fritsch, L. polysiphoniae Fremy, L. erugineo-coerulea Gomont, L. cryptovaginata Schkorbatow, L. spiralis Geitler, L. nigra C. Ag. ex Gomont, L. kashyapii Ghose

Lyngbya rubida در فصل بهار و زمستان، L. perelegans در فصل تابستان، L. kashyapii و L. kashyapis در فصل پاییز گونههای غالب در همه ی ایستگاهها بودند. ویژگیهای آرایه شناسی این گونهها و اطلاعات پیرامون پراکنش اکولوژیکی آنها نیز ارائه میگردد.

## Introduction

*Cyanophyta* are present abundantly in rice-fields and are important in helping to maintain rice -fields fertility through nitrogen fixation, nitrogenous and non nitrogenous compounds liberation and excretion (Shokravi et al. 2007). In addition some potent bioactive compound that extracted from this group, draw clear landscape for pharmacological industries (Ghasemi et al. 2003; Soltani et al. 2006). It has been showed that many rice-fields of Asian soils contain a high density of *Cyanophyta* and over 50% of *Cyanophyta* genera that are in existence in rice-paddy fields belong to heterocystous filamentous form (Jeong–Dong & Choul–Gyun 2008). In spite of this *Cyanophyta* of paddy fields of Iran and specially Golestan province seems to be relatively unknown.

شالیزارهای ایران گزارش می گردند.

Golestan province is located between the longitudes of  $54\circ 30$  'and  $56\circ 30$  ' E and at a latitude of  $38^{\circ}15'$  N in Iran. The paddy fields were containing nearly 62000 hectares of total cultivation fields in last year. It seems

that in north paddy fields of Iran especially Golestan province, some strains of *Cyanophyta* especially is common (Shokravi et al. 2006), but there is no clear report about their morphological characterization and taxonomic situations and most of the early investigations have had physiologically or ecophysiologically approaches.

Oscillatorian Cyanophyta (Order Oscillatoriales (Bold & Wyne 1985) or Family Oscillatoriacea (John & al. 2002) are usually common members of the paddy fields. They are predominately filamentous, prokaryotic and photosynthetic Cyanophyta. It has been estimated that the number of living species of Oscillatorian Cyanophyta may be 265 (Jhon et al. 2002) and more than 24% may belong to the genus Lyngbya. Traditional identification of this genus is according to morphology of filaments, life cycle, trichome structure, and type of trichome disintegration (Anagnostidis & Komarek 1990). Until now, approximately 66 species of the blue-green algal genus Lyngbya has been recorded from marine and fresh waters as free floating or benthic forms and from terrestrial habitats like soils and artificial substrates as epidaphic, endaphic and epilithic communities (Ullmann & Budel 2001).

Under natural conditions in rice fields, members of Cyanophyta are exposed to the combined influences of several factors such as pH, irradiance and dissolved inorganic carbon fluctuations, which varied both daily and over the crop cycle (Quesada & al. 1995, 1998). Growth, biochemical and physiological characteristics of Cyanophyta are influenced by environmental factors (Grossman & al. 1993; Islam & Whitton 1992; Fernández-Valiente & Leganés 1989). pH is a basic factor, which clearly affects the distribution of Cyanophyta. Most members of Cyanophyta grow in environments that are neutral to alkaline and in laboratory cultures the optimal pH ranges from 7.5 to 10. Generally, a wide range of adaptation to pH has been observed not only among different genera but also between different populations of the same species. In rice fields, the pH of floodwater varies during the day and during the growth of the crop due to the photosynthetic activity of Cyanophyta, algae and other macrophytes (Fernandez-Valiente & Leganes 1989).

The aim of this paper is to contribute to the knowledge on the algal flora of paddy fields based on morphological investigations of the *Cyanophyta* of the paddy field in Golesten province and a preliminary investigation of their abundance especially in relation to pH of the paddy fields.

#### **Materials and Methods**

Soil samples were obtained from paddy fields of different stations of Golestan province (North of Iran).

Table 1: physical- chemical properties of surface soil samples from monitoring sites in the paddy fields of Iran.

station	site	pН	conductivity
		-	$(mS/cm^{-1})$
Kordkoy	1-2-3	7.2-8.1-8.2	0.56-0.69-0.81
Gorgan	4-5-6	7.8-7.9-8.2	0.51-0.62-0.55
Aliabad	7-8-9	7.4-7.7-7.5	1.18-1.25-1.50
Azadshahr	10-11-12	7.9-8-8	0.55-0/56-0/76
Minodasht	13-14-15	6.9-7.2-7.8	1.36-1.12-0.72

Five stations were chosen in different areas of the paddy fields. The samples were taken from the surface and 2 cm depth between autumn 2006 and summer 2007. Samples were taken from flooded and nonflooded soils (Kaushik 1987). After collection, samples were brought to the laboratory for culturing, preserving and preparation for studying (Thallophyte laboratory of Isfahan University). pH and EC were measured using digital pH meter (Table 1). The samples cultured in solid BG11 medium (NaNo3, 17.65 mM, mg So4. 7H2O, 0/3mM, cacl2, 2H2O, 0/25 mM, K2HPo4.3 H20, 0/18Mm,Na2 Mg EDTA, 0/003 mM, citrate ferric ammonium, 0/02 mM Acid citric, 0/029 mM, Na2Co3, 0/188 mM , microelements lml 1)-1. The cultivation was done under illumination 1500-1600 lux, pH 7.2 and temperature  $25 \pm 2^{\circ}$ C. After colonization and isolation, samples were purified by several subculturing (Kaushik 1987). Identification of samples was carried out by usual microscopy in addition of fluorescence phase-contrast and microscopy. Determination and nomenclature accomplished according to famous keys and manuals (John & al. 2002, Anagnostidis & Komarek 1990, Prescott 1962 and Desikachary 1959). Photographs were taken with Nikon microscope, equipped with a Canon model camera. Axenity were checked daily using microscopic observations.

#### Results

A total of 17 species belonging to the genus *Lyngbya* were identified, 13 species are new records for Golestan Province and Iran. They ranged from 6-9 species in spring, 1-6 species in summer, 5-11 species in autumn and 3-6 species in winter, between stations. List of new records are given below and their photographs are presented in Fig. 1. Identified species and their distribution on the paddy field of Iran are listed in Table 2.

Divisin: Cyanophyta

Class: Cyanophyceae Order: Oscillatoriales Family: Osillatoriaceae Lyngbya perelegans Lemm

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Table 2: *Lyngbya* species and relative frequencies of occurrence on the paddy fields of Iran. Stations: 1-Aliabad. 2-Kordkoy. 3-Minodasht. 4-Azadshahr. 5-Gorgan. D=Dominant (75-100%); A=Abundant (50-75%); F=Frequent (25-50%); R=Rare (<25%).

Species	Spring				Summer					Autumn					Winter					
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
L. perelegans	R	R	I	-	F	-	R	F	F	Α	I	I	R	R	-	R	-	-	-	-
L. rubida	F	R	R	F	Α	R	I	-	Α	Α	F	F	R	I	-	R	R	F	F	F
L. sordida	-	I	F	-	F	R	R	-	I	I	F	I	-	F	-	R	-	F	-	-
L. kuetzingii	-	I	I	-	-	-	I	-	I	I	I	I	-	I	-	-	-	-	-	-
L. kashyapii	F	I	R	F	-	-	R		I	I	I	F	R	R	F	R	-	-	-	R
L. erugineo-coerulea	-	F	I	F	-	-	R	-	R	I	I	R	-	I	R	-	R	-	R	R
L. lemnetica	-	I	R	R	-	-	I	-	I	I	I	I	-	I	-	-	-	-	-	-
L. nigra	-	I	I	R	-	-	I	-	I	I	I	I	-	I	-	-	-	-	-	-
L. aestuarii	-	I	I	-	-	-	R	-	R	I	I	F	-	F	-	-	R	-	-	-
L. spiralis	-	I	I	-	-	-	I	-	I	I	I	F	R	F	F	-	-	-	-	-
L. dendrobia	R	R	R	-	-	-	I	-	I	R	I	R	-	R	F	-	-	R	R	R
L. laxespiralis	-	I	I	R	F	-	I	-	R	I	R	F	R	F	R	-	-	-	R	-
L. contorta	R	I	I	R	-	R	I	-	I	I	R	I	-	R	-	-	R	-	-	R
L., scotii	-	I	I	-	-	R	I	-	I	R	I	R	-	R	R	-	-	-	-	-
L. polysiphoniae	-	R	I	-	R	I	I	-	1	1	I	-	-	R	R	-	-	-	R	-
L. cryptovaginata	-	-	-	-	R	-	R	-	-	-	-	R	R	-	R	-	-	-	R	-
L. putealis	F	R	-	-	R	-	-	-	-	-	-	F	-	F	F	-	-	-	-	R

Filaments straight or curved, pale green, 2-2.5  $\mu$  broad; sheath very thin, hyaline; trichomes 2  $\mu$  broad, not constricted at the cross walls; cells 7-8  $\mu$  long and 1.9-2  $\mu$  broad, granulate; end cell rounded, not capitate, not calyptrate.

# L. rubida Fremy

Thallus more of less expanded; filaments straight, 5-6  $\mu$  broad, loosely intricate; sheath very thin, not lamellate, at first colourless, later reddish; trichome brown, not constricted at the cross wall; cross wall not visible, not granulated; cells granulate and rounded; calyptra absent.

# L. laxespiralis Skuja

Filaments inter mixed with algae, sometimes irregularly bent of more or less straight 11  $\mu$  broad; sheath thin, smooth, hyaline, 1.5-2  $\mu$  broad; trichomes 9  $\mu$  broad, not constricted at the cross walls; cells quadrate and shorter than broad, granulate; cross well not granulated; end cell not attenuated, rounded.

# L. sordida Gomont

Thallus caespitose; filaments more of less straight, 10-12  $\mu$  broad; sheath smooth, 1.5-2  $\mu$  broad; trichomes dull green, when dried dark violet, distinctly constricted at the cross wall, not granulated, 9-10  $\mu$ broad; cells 3-4  $\mu$  long and 8.5-9.5  $\mu$  broad, granulate; end cell rounded with calyptras, in another specimen end cell rostrate. (Fig.1n).

### L. putealis Mont. ex Gomont

Filaments curved or nearly straight, 12-14  $\mu$  broad; sheath thin; trichome dull green, 11-13  $\mu$  broad, at the

cross wall constricted, slightly attenuated at the apex; cells 3-4  $\mu$  long and 10 -12 $\mu$   $\mu$  broad, not granulated; end cell rounded.

## L. dendrobia Bruhl & Biswas

# Syn.: Porphyrosiphon notarii Kutz

Filament long and flexible, 10  $\mu$  broad; sheath usually thin, smooth, hyaline, 1.5  $\mu$  broad; trichome 9  $\mu$  broad, at the cross wall not constricted, granulated; cells 5-6  $\mu$  broad and 7-8  $\mu$  long, in various shades of brown, not granulate; end cell rounded.

# *L. scotii* Fritsch

Filament straight, 4-4.5  $\mu$  broad; sheath smooth, hyaline, lamellate, 2  $\mu$  broad; trichome 2.5  $\mu$  broad, attenuated at the end, at the cross wall not constricted; end cell prominently conical; cells as long as broad, granulate, no calyptra.

## L. polysiphoniae Fremy

Filament straight or curved, single, 11  $\mu$  broad; sheath thin, colorless; trichome 10  $\mu$  broad, at the cross wall constricted, not attenuated at the apex; cells 4-5  $\mu$  long and 2 times as long as broad, not granulated; cross wall visible; end cell convex, rounded.

# L. kashyapii Ghose

Filament straight, more or less curved, 2.5-3  $\mu$  broad; sheath very thin, pink or purple, smooth; trichome 2.5  $\mu$  broad, at the cross wall not constricted; cross wall not visible; cells granulated; end cell in some of the specimens rounded and not attenuated, and in the other specimens bent and a little attenuated.

L. erugineo-coerulea Gomont

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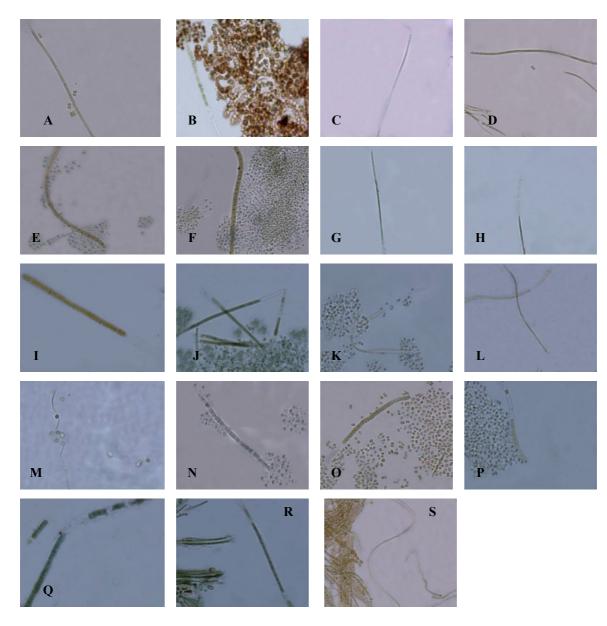


Fig. 1: a: Lyngbya kuetzingii, b: L. polysiphoniae, c,d: L. contorta, e: L. dendrobia, f: L. laxespiralis, g, h: L. scotii, i: L. cryptovaginata, j: L. aerugineo-coerulea, k: L. kashyapii, l; L. perelegans, m: L. limnetica, n,o: L. sordida, p: L. rubida, q: L. putealis, r: L. nigra, s: L. spiralis.

Filament single or flexuous, 10  $\mu$  broad; sheath thin, smooth, not lamellate; trichome 8-9.5  $\mu$  broad, at the cross wall not constricted; cells often granulated, end not attenuated, no calyptra.

# L. cryptovaginata Schkorbatow

Filament single, straight, 8  $\mu$  broad; sheath thin, not lamellated, 2  $\mu$  broad; trichome 6  $\mu$  broad, at the cross wall constricted; cells nearly quadrate, 2  $\mu$  long, 5-6.5  $\mu$  broad, not granulate; end cell attenuated, rounded. *L. nigra* C. Ag. ex Gomont

Filament straight, 8-9  $\mu$  broad; sheath thin, not lamellate; trichome green-brown, 8.5  $\mu$  broad, not constricted at the cross wall, not granulated; cells 3  $\mu$  long and 7-7.5  $\mu$  broad, ends attenuated; end cell conical.

# L. spiralis Geitler

Filament entirely for major part spirally coiled, distance between two spirals 70-80  $\mu$ ; sheath delicate and not lamellate; trichome 2.5  $\mu$  broad, at the cross wall not

constricted and not granulate, end not attenuated, rounded.

# Discussion

Collectively our knowledge about Cyanophyta of Golestan province is limited. However, until now, a few reports have been published with the highest degree of consideration on stigonematalean species (Sepehri & al. 2003, Norouzi & al. 2004). About oscillatorian strain, the dominant of our knowledge is considerably narrower. A few species of Oscillatoria has been recorded by Siahbalaie & al. (2008) and Shokravi & al. (unpublished data). About species of Lyngbya unfortunately we have no report yet. Morphological versatility and the narrowing border between such genera and others like Planktothrix Anagnostidis & Komarek, Phormidium Kutzing ex Gomont and rarely Oscillatoria Vaucher ex Gomont and Plectonema Thuret ex Gomont may be the main reason. However in our study on the algal flora in paddy fields the members of Cyanophyta formed the majority with a ratio of 75 %, this can be explained by the variety of habitats or may have been the result of physical, chemical or geographical differences between paddy fields. There were 17 taxa in total, four species has been reported from the lakes and rivers for Iran. L. contorta (Zarei-Darki 2004), L. kuetzingii (Compere 1981). L. limnetica (Compere 1981, Hirano 1973, Afsharzadeh 2003, Zarei-Darki 2004) and L. aestuerii (Zarei-Darki 2004). Rest of the species are first records for paddy fields of Iran. The flora of algae in paddy fields varied between stations and months. The number of species at each site ranged from 3 to 11 with maximum at Kordkoy, Gorgan and Azadshahr stations, a minimum at Minodasht and Aliabad stations. Dominant species were in all stations L. rubida in spring and winter, L. perelegan in summer, L. kashyapii and L. dendrobia in autumn. Many of *Lyngbya* species are useful indicators in paddy field *L*. borgerti Lemm and L. hieronmusii Lemm, L. kuetzingiana Kirchner were not observed in this area. However, L. scotii, L. aerugineo-coerulea, L. limnetica are also characteristic species in the paddy field and this study. Results of this survey show that morphological variation of some Cyanophyta such as Lyngbya in paddy field is very high, that need to improve identification keys. However, based on the results it may be possible to draw a relatively primitive picture of the morphological and taxonomical situation of Cvanophta especially Lyngbya species in paddy fields of North of Iran.

In this study, we can attribute to some characteristics of *Lyngbaya* species in paddy fields of Iran. Absence of thick and visible sheath, absence of

calyptra, absence of constricted and granulated cross wall, are noticeable characters from morphological point of view (exception 3-4 samples), that can be useful for identification *Lyngbaya* species.

They are common inhabitants of aquatic and terrestrial surfaces. The widespread distribution of Cyanophyta indicates that they can cope with a wide spectrum of global environmental stresses such as temperature, pH, desiccation, etc. Salt stress and pH are from the most limiting factors on the growth and productivity of microorganisms. The main ecological factors of pH, EC affected the development of the algal flora in paddy fields (Whitton & Potts 2000). Lower acidities and higher EC may simply have a delaying effect on the development of the algal flora (Broady 1984). Electric conductivity values ranged from 0.51 to 1.50 ms.cm<sup>-1</sup>. A maximmum value of EC was observed at site 8, 9 and 13 in Aliabad and Minodasht. The pH values ranged from 6, 9, 8.2. The pH values at site 3, 6 and 11 were higher than values at the other sites. Lowest pH and highest EC were measured at Minodasht and Aliabad stations, as they are near the forest but Azadshahr, Gorgan and Kordkoy had lower EC and highest pH, these stations are near the agricultural fields. Thus, factors pH and EC affect on the Cyanophyta variations in the paddy fields. Our result is in agreement with Hickman (1978).

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