

# Morphological Study of Algal Species from Azad Kashmir Using Microscopic Techniques

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## Abstract

Algae have been considered as a diversified group of lower plants whose morphological characters have been used for taxonomic identification. In present research, 34 species of algae were morphologically investigated from different sites of Azad Kashmir. Collected species were divided into 2 divisions, out of them, 14 species belongs to division Cyanophyta and 20 species belongs to division Chlorophyta. All the algal species reported in present study were identified on the basis of their morphological characters up to specific level with the help of light microscopic techniques. Samples of algae were collected either by pipette or taken in polythene bags. Fresh specimens were observed under light microscope and photomicrographs have been taken. In current study it was also investigated that shape of cells, number of pyramids and the position of cells are the main distinguishing characters that differentiate one genus from other. Based on these micro-morphological characters it is easy for taxonomist to identify complex plants upto their family, genera and specific level.

**Keywords:** Azad Kashmir, Algae, Light Microscope, Morphology, Taxonomy

## 1. Introduction

Azad Jammu & Kashmir is situated at the foot hills of Himalayan region of Pakistan which covers the total area of 13,297 km<sup>2</sup> and lies between the elevation of 73–751° East longitude and 33–351° North latitude. Elevation of study area ranges between 360 m to 6325 m. The climate of Azad Kashmir is humid with hot and wet summers, a cooler and drier winter with heavy snowfall in higher elevation areas. Due to the continuous climatic changes the study area comprises of plenty of rivers, streams, lakes that contains huge biodiversity of algae (Ajaib et al., 2010).

Algae are included among those organisms which are photosynthetic due to the presence of chloroplast and has thallus shaped body. Throughout the world algae are considered as the source of nutrition for aquatic organisms as they can survive in high and low temperature. Algae are usually differentiated on the basis of their life forms, colors of their thallus and sizes. Algae lives in different forms some live together to form colonies, some are in the form of filaments, sometimes they are membranous and some are tubular. In some cases few species also have delicate membranes and complex structures, some species of genus *Sargassum* produces root, stem and leaves like structures which are similar to higher plants. Some algae contain some structures like air spaces that allow the thallus to float freely on the surface of water to absorb sunlight and prepare their food with the help of photosynthesis. However, algae exhibit different structural forms but the internal structures are having same cells having little difference in structures.

The occurrence and succession of the algal species plays an important part in the aquatic ecosystem as they are the good indicator of pollution in water (Thornton *et al.*, 1990; Patrick and Reimer, 1966). They are basic food for aquatic life (fauna and fishes). They also grow on the soil edaphically, among them, more nitrogen fixing groups are found as compared to the heterotrophic micro-organisms. Nitrogen fixing group of blue green algae, being autotrophic microbes of the photic zone, is found within the upper few millimeters in the fine soil.

Many studies have been reported previously on the biodiversity of algae from different regions of Pakistan (Aliya

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DOI

<https://doi.org/10.6084/m9.figshare.24182547#161>



et al., 2009; Saifullah and Nizamuddin, 1977; Shaikh and Shameel, 1995). Algal flora in Punjab and neighboring areas is reported by (Zarina and Shameel, 2009), Sher and Hazrat, (2012) and Salah-ud-Din et al, (2017) documented algal diversity of chlorophyta division from different regions of Khyber Pukhtunkhwa, Khuhawari et al, (2009) Bhimber Azad Jammu & Kashmir, Poonch Azad Kashmir (Ashraf et al., 2008), and Neelam river Azad Kashmir (Leghari and Leghari 2000). However, at present there is no separate documentation on the light microscopic study of morphological characters of division chlorophyta and cyanophyta of algae from Kotli Azad Kashmir. The aim of this study is to provide detailed information of structural morphological characters based on both qualitative and quantitative characters by using Light microscopy. These characters are helpful in identification of complicated species of the same division.

## 2. Materials and Methods

### 2.1 Samples Collection Sites

Algal samples were collected from 15 localities of district Kotli of Azad Jammu and Kashmir (Figure 1). The present work is the outcome of the extensive survey of the water bodies during the period of 2015 to 2016 during different months of the year to cover the variation of algal species in all the seasons of the year (Table 1). The collected samples after identification were deposited in the herbarium of Botanical Sciences Division of Pakistan Museum of Natural History, Islamabad.

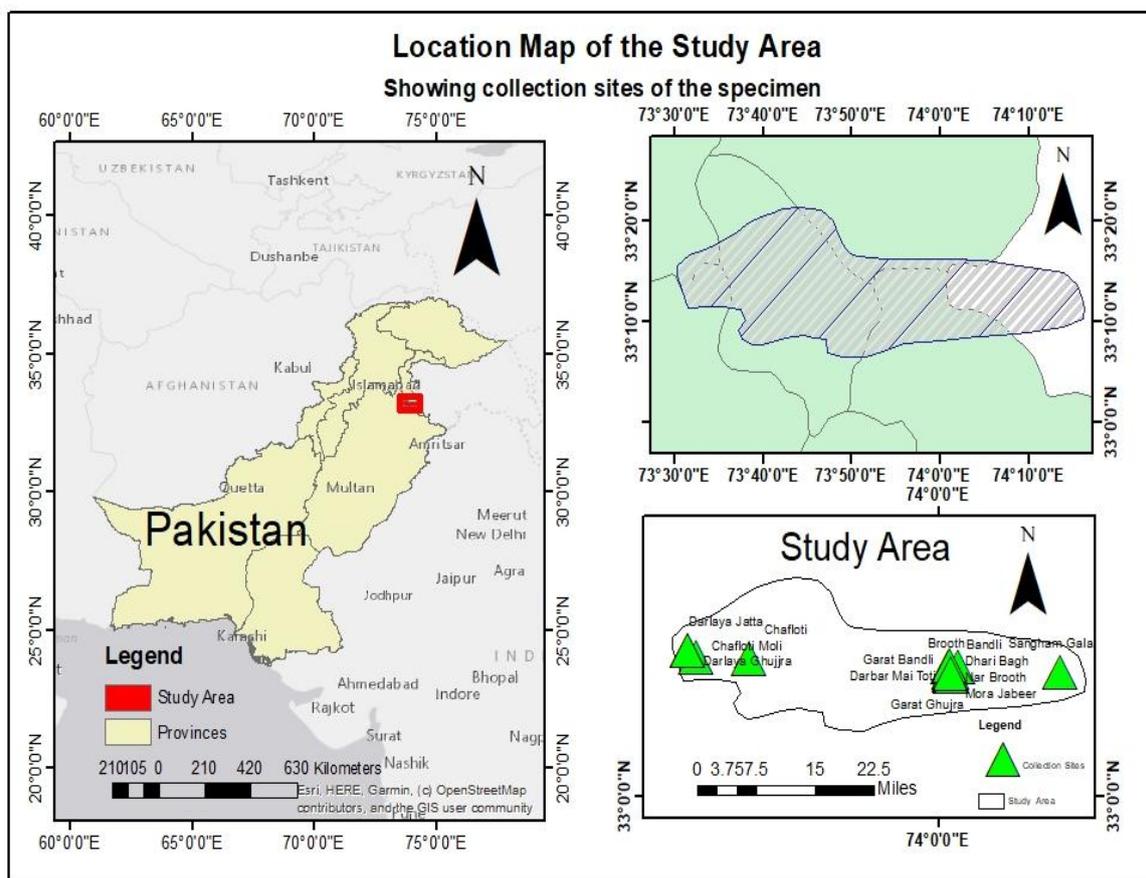


Fig. 1 Map of showing collection sites

### 2.2 Methods of Collection

Collection of algal samples has been done either with the help of pipette into bottles or with polythene bags having water. Filamentous algae were collected in bottles with the help of forceps. Blue-green algae were taken with the help of knives and also slides are used to gathered them. Green algae were collected with the help of hands. Some algal species are stick to the surface of rocks which were collected by using tooth brush and knives.

### 2.3 Preservation of Samples

The collected samples were preserved according to the standard method (APHA, 1985). Unicellular algal species were kept in 2 to 3% formalin and macro-algae in 4% formalin (Mason, 1967).

## 2.4 Analysis through Light Microscopy

One drop from each sample was taken with the help of small pipette on the slide, kept under cover slip, and examined under light microscope (BH-2 Olympus, Made in Japan). The objectives used were 10<sup>x</sup>, 20<sup>x</sup>, 40<sup>x</sup> & 100<sup>x</sup>. The eye piece (10<sup>x</sup>) was fitted with ruled ocular micrometer. The outer boundaries forming a square field were divided into 100 smaller squares by the rulings (Lund et al., 1958). The specimens were identified with the help of available literature (Tilden, 1910; Hustedt, 1930; Abdul-Majeed, 1935; Smith, 1950; Desikachary, 1959; Prescott, 1962; Siddiqi and Faridi, 1964; Patrick and Reimer, 1966; Philpote, 1967; Tiffany and Britton, 1971; Vinyard, 1979; Akiyama and Yamagishi, 1981).

## 2.5 Identification of Samples

The identification of the taxa was based on standard basis, important research publications and from thesis. For the purpose of identification light microscopy LAB 1 were used.

## 3. Results

A total 34 species were recorded from 2 divisions of algae collected from different areas of Kotli Azad Kashmir, out of them, 14 species with 4 genera and 3 families belongs to division Cyanophyta. 20 species belongs to 10 genera and 7 families of division Chlorophyta. The detail of Qualitative and Quantitative Study of Fresh Water Algae of District Kotli, Azad Kashmir is described in Table 2.

**Table 1** Collection sites and distribution of some Algal species from division Cyanophyta and Chlorophyta

S. No.	Name of Taxa	Collection sites	Voucher no	Coordinates	Distribution status
<b>Division: Cyanophyta</b>					
1.	<i>Aphanocapsa endophytica</i> G.M.Smith	Seri, Cross, Chafloti	SB-023	33°22.574N, 73°54.957E	Rare
2.	<i>Chroococcus limneticus</i> Lemmermann	Brotgala/ Charoi, Bandli/ Darbar, Chafloti	SB-027	33°22.535N, 73°64.702E	Rare
3.	<i>Lyngbya taylorii</i> Drouet & Strickland	Cross, Bandli/ Darbar, Chafloti	SB-018	33°19.904N, 74°02.186E	Rare
4.	<i>Merismopedia convolute</i> Brébisson ex Kützing	Brooth, Khuiratta, Chafloti	SB-036	33°20.225N, 74°01.905E	Rare
5.	<i>Merismopedia tenuissima</i> Lemmermann	Khuiratta, Seri, Bandli/ Darbar	SB-024	33°21.168N, 74°01.656E	Rare
6.	<i>Nostoc commune</i> Vaucher ex Bornet & Flahault	Draman	SB-057	33°21.247N, 74°02.038E	Very Rare
7.	<i>Oscillatoria chlorine</i> f. major Compère	Khuiratta, Seri, Brotgala/ Charoi, Khajlani	SB-012	33°21.168N, 74°01.656E	Common
8.	<i>Oscillatoria Formosa</i> Bory ex Gomont	Seri, Brotgala/ Charoi, Khajlani	SB-011	33°19.735N, 74°02.017E	Rare
9.	<i>Oscillatoria limosa</i> C.Agardh ex Gomont	Brooth, Khuiratta, Seri, Cross, Khajlani, Chafloti	SB-007	33°20.225N, 74°01.905E	Very Common
10.	<i>Oscillatoria princeps</i> Vaucher ex Gomont	Brooth, Khuiratta, Bandli/ Darbar, Chafloti	SB-046	33°20.225N, 74°01.905E	Common
11.	<i>Oscillatoria splendid</i> Greu	Khuiratta, Khajlani	SB-034	33°21.168N, 74°01.656E	Rare
12.	<i>Oscillatoria unigranulata</i> Kisselev	Seri, Cross, Bandli/ Darbar, Chafloti	SB-026	33°22.535N, 73°64.702E	Common
13.	<i>Phormidium tenue</i> Gomont	Seri, Brotgala/ Charoi, Khajlani	SB-071	33°20.280N, 74°01.940E	Rare
14.	<i>Spirulina major</i> Kützing ex Gomont	Brooth, Cross, Khajlani	SB-058	33°20.225N, 74°01.905E	Rare
<b>Division: Chlorophyta</b>					
1.	<i>Chlorella pridinosa</i> LineausKuetzing	Brooth, Brotgala/ Charoi	SB-022	33°20.225N, 74°01.905E	Rare
2.	<i>Chlorella pyrenoidosa</i> H.Chick	Seri, Brotgala/ Charoi, Chafloti	SB-044	33°22.535N, 73°64.702E	Rare
3.	<i>Cladophora glomerata</i> (Linnaeus) Kützing	Brooth, Khuiratta, Seri, Brotgala/ Charoi, Chafloti	SB-063	33°22.535N, 73°64.702E	Very Common
4.	<i>Closterium ehrenbergii</i> Meneghini ex Ralfs	Seri, Bandli/ Darbar, Chafloti	SB-079	33°19.548N, 74°02.323E	Rare
5.	<i>Closterium lancreum</i> Breb	Khuiratta, Cross, Khajlani, Chafloti	SB-029	33°22.535N, 73°64.702E	Common
6.	<i>Closterium moliferum</i> Bory	Brooth, Cross, Chafloti	SB-060	33°20.225N, 74°01.905E	Rare
7.	<i>Cosmarium leave</i> Rab	Khuiratta, Seri, Brotgala/ Charoi	SB-066	33°21.168N, 74°01.656E	Rare

8.	<i>Cosmarium occidentals</i> Gerffy	Khuiratta, Seri, Brotgala/ Charoi	SB-084	33°21.168N, 74°01.656E	Rare
9.	<i>Cosmarium phaseolus</i> var. <i>psedostigmasum</i> Forster	Khuiratta, Seri, Brotgala/ Charoi	SB-091	33°21.168N, 74°01.656E	Rare
10.	<i>Cosmarium punctatum</i> Lemmerman	Brooth, Seri, Bandli/ Darbar, Khajlani	SB-059	33°20.225N, 74°01.905E	Common
11.	<i>Cosmarium tetrachodrum</i> Skuja	Khuiratta, Seri, Brotgala/ Charoi	SB-061	33°21.168N, 74°01.656E	Rare
12.	<i>Hormidium gubtile</i> Kuetzing	Brooth, Seri, Bandli/ Darbar, Chafloti	SB-085	33°21.168N, 74°01.656E	Common
13.	<i>Oedogonium epiphyticum</i> Skvortsov	Brooth, Seri, Khajlani, Chafloti	SB-098	33°21.168N, 74°01.656E	Common
14.	<i>Oedogonium sphaericoinconspicm</i> Silva	Brotgala/ Charoi, Bandli/ Darbar Khajlani, Chafloti	SB-070	33°20.280N, 74°01.940E	Common
15.	<i>Scenedesmus denticulatus</i> Lagerheim	Khuiratta, Seri	SB-013	33°21.168N, 74°01.656E	Rare
16.	<i>Spirogyra porticalis</i> Muceller	Khuiratta, Seri, Bandli/ Darbar, Khajlani, Chafloti	SB-021	33°21.168N, 74°01.656E	Very Common
17.	<i>Spirogyra</i> sp.	Brooth, Cross, Brotgala/ Charoi, Khajlani, Chafloti	SB-039	33°20.225N, 74°01.905E	Very Common
18.	<i>Spirogyra</i> sp.	Brooth, Khuiratta, Cross, Brotgala/ Charoi, Khajlani, Chafloti	SB-048	33°20.225N, 74°01.905E	Dominant
19.	<i>Tetraëdron muticum</i> (A.Braun) Hansgirg	Brooth, Khuiratta, Bandli/ Darbar, Khajlani, Chafloti	SB-062	33°20.225N, 74°01.905E	Very Common
20.	<i>Zygnema pacnitum</i> Vauch	Bandli/ Darbar, Khajlani, Chafloti	SB-099	33°19.904N, 74°02.186E	Rare

**Table 2** Quantitative and qualitative morphological characters of some algal species collected from different localities of district Kotli Azad Jammu and Kashmir

S. No		Length of cell in $\mu\text{m}$	Width of cells In $\mu\text{m}$	Surface of cells	Shape of semi cells	Colour of thallus	Shape of apex
<b>Division: Cyanophyta</b>							
1.	<i>Aphanocapsa endophytica</i>	4.2-6.5	12-18	Smooth	Oval to rod-like	Bluegreen or olive-green	Round
2.	<i>Chroococcus limneticus</i>	4.4-6.7	2.2-2.9	Smooth	Hemispherical	Dark green	Round
3.	<i>Lyngbya taylorii</i>	2.8-3.9	1.6-2.7	Granulate	Quadrate	Dark blue or green	Slightly rounded
4.	<i>Merismopedia convoluta</i>	1.6-2.1	2.2-2.9	Smooth	Oblong	Green with slightly bluish shade	Rounded
5.	<i>Merismopedia tenuissima</i>	3.6-5.3	1.1-2.8	Smooth	Hemispherical	Light blue green	Rounded
6.	<i>Nostoc commune</i>	14.2-18.9	4.8-6.3	Smooth	Spherical	Bluish-green	Heterocyst presentspherical to oval
7.	<i>Oscillatoria chlorina</i>	13.2-15.1	4.4-6.8	Granulate	Cylindrical	Red to purple-violet	Rounded
8.	<i>Oscillatoria formosa</i>	4.3-6.9	2.6 - 5.8	Slightly granulate	Cylindrical	Blue- green	Bluntly rounded
9.	<i>Oscillatoria limosa</i>	11 – 22	2.2 – 5.1	Granulate	Semi-circular	Blue green to brown or olive	Rounded
10.	<i>Oscillatoria princeps</i>	16 - 60	3.5 – 7.0	Granulate	Cylindrical	blue – green to brownish, reddish or violet	Hemispherical
11.	<i>Oscillatoria splendid</i>	12.1-15.5	3.9-6.2	Granulate	Cylindrical	Blue green	Tapering apical cells
12.	<i>Oscillatoria unigranulata</i>	4.5-10.9	2.6-5.8	Slightly granulate	Cylindrical	Blue or bright green	Rounded
13.	<i>Phormidium tenue</i>	2.3-5.9	10.6-12.1	Smooth	Cylindrical	Blue-green	Rounded-truncate
14.	<i>Spirulina major</i>	2.8-4.6	1.1-2	Smooth	Spirally coiled	Blue-green	Rounded
<b>Division: Chlorophyta</b>							
1.	<i>Chlorella pridinosa</i>	5.2 – 12.2	3.2-4.1	Granulate	Globose or rarely ellipsoid	Dark to light green	Rounded
2.	<i>Chlorella pyrenoidosa</i>	5.2-6	3.1 – 4	Granulate	Globose	Light green	Rounded
3.	<i>Cladophora glomerata</i>	45-150	150-360	Smooth	Cylindrical	Olive green	Rounded

4.	<i>Closterium ehrenbergii</i>	60 – 145	285 – 720	Smooth	Sickle-shaped	Yellow to brown	Obtusely rounded
5.	<i>Closterium lancreum</i>	2.2 – 5.1	25 – 100	Smooth	Sickle-shaped	Green	Obtusely rounded
6.	<i>Closterium moliferum</i>	30 – 68	188 – 420	Smooth	Sickle-shaped	Colorless	Obtusely rounded
7.	<i>Cosmarium leave</i>	11 – 20	221.0 – 37.5	Smooth	Elliptic	Green	Strongly rounded
8.	<i>Cosmarium occidentals</i>	65-68	42-45	Smooth	Pyramidal	Brownish green	Rounded-truncate
9.	<i>Cosmarium phaseolus</i> var. <i>Pseudostigmatum</i>	37–45	32–37	Punctate	Sub-circular	Pale green	Circular
10.	<i>Cosmarium punctulatum</i>	27-38	17-19	Granulate, punctate	Oblong-trapeziform	Light green	Broadly truncate
11.	<i>Cosmarium tetrachodrum</i>	17–22.8	10–19	Smooth	Truncate-pyramidal	Dark green	Broadly truncated
12.	<i>Hormidium gubtile</i>	2.1-2.7	3.2-4.1	Smooth	Spherical	Green	Rounded
13.	<i>Oedogonium epiphyticum</i>	14.4-19.9	26.3-40.6	Smooth	Rectangular	Light green	Rounded
14.	<i>Oedogonium sphaericoinconspicm</i>	12.2-21.6	21.2-50.5	Smooth	Cylindrical	Olive green	Rounded
15.	<i>Scenedesmus denticulatus</i>	5.1–11.3	7.2– 15.6	Granulate	Ellipsoid	Bright green	Rounded
16.	<i>Spirogyra porticalis</i>	110.2-160.6	26.23-28.6	Smooth	Cylindrical	Dark green	Rounded
17.	<i>Tetraëdron muticum</i>	22.4-30.2	4.5-8.9	Smooth	Triangular with the sides slightly concave and angles broadly rounded to truncate.	Olive green	Rounded
18.	<i>Zygnema pachitum</i>	35.2-50.8	32.1-39.3	Smooth	Cylindrical	Pale green or yellowish green	Slightly rounded

### 3.1 Division Cyanophyta

In present study 14 species of division Cyanophyta have been morphologically observed with the help of light microscopy. The genus *Oscillatoria* contains 3 species, 2 species of genus *Merismopedia* and 1 species of each genus *Aphanocapsa* and *Nostoc*. Some species of this division have the tendency to grow in high temperature i.e. up to 84°C. While the species of genus *Phormidium* can survive in cold spring, hot sulphur spring, high and low saline water, marine water, etc.

#### *Oscillatoria limosa*

Cells 11 – 22 × 2 – 5 µm, granulate at cross – wall ; trichomes usually straight, dark blue green to brown or olive – green, not constricted at cross – walls, with broadly rounded ends cells, with slightly thickened outer membrane, becoming blackish blue – green to brown in aggregates.

#### *Oscillatoria prices*

Cells 16 - 60 × 3.5 – 7.0 µm, with granulose protoplasm, not granulate at cross – walls; trichomes blue – green to brownish, reddish or violet, not constricted at cross- walls, generally broadly, terminally light and abruptly constricted and bent; end cells broadly rounded, lightly capitates, without calyptas; generally in masses, sometimes single; plant mass dark blue – green .

#### *Oscillatoria splendida*

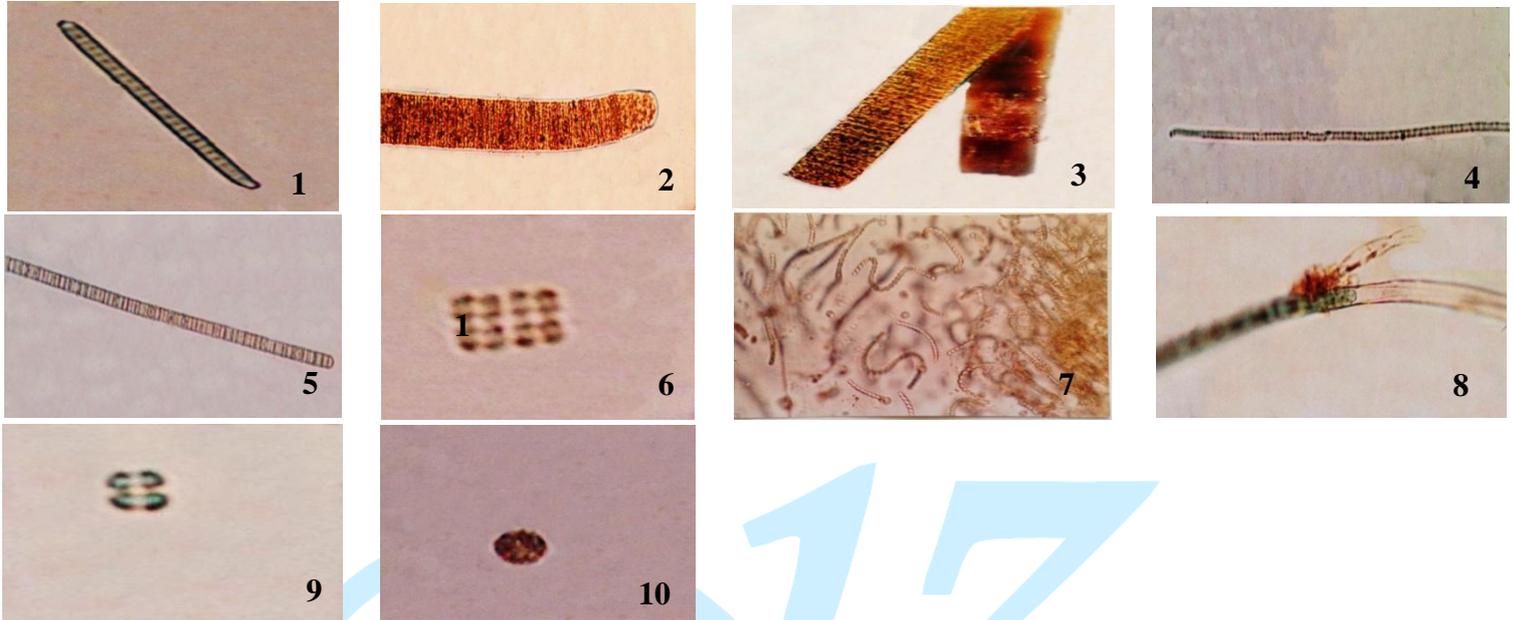
Cells 2 – 3 × 3 – 9 µm, homogeneous, blue green; trichomes straight or bent, not constricted but somewhat granulate at cross – wall, gradually tapering, at the apex, end cell capitata, nearly round, forming dark blue – green thin masses, or scattered.

#### Genus *Aphanocapsa*

Cells spherical, appearing ellipsoid and nearly hemispherical in division; colonies many celled, attached to substrate, usually microscopic at maturity; gelatinous matrix homogeneous.

### Genus *Nostoc*

Members of this genus are found in the form of spherical colonies; thallus exhibit trichomes like structure which are twisted in appearance and enclosed in a matrix which is yellowish brown in color. Size of the cell ranges 3.2-4.1×2.7-4-3 μm. Surface of the thallus contains viscous gel that spreads throughout the body and in some species trichomes are embedded in this gel. Shape of the cell is spherical and olive green in colour. Cells of the members of this genus contains heterocysts which are either present in the center or at the ends of the trichomes. Heterocysts are usually barrel-shaped or in some species spherical shape is also found.



**Fig. 2** Light microscopic photographs of members of division Cyanophyta

(1). *Oscillatoria* sp, (2). *Oscillatoria princeps*, (3) *Oscillatoria limosa*, (4) *Oscillatoria splendida*, (5) *Oscillatoria spseudgeminates* var. *unigranulates*, (6) *Mevismopedia tenuissima*, (7) *Nostoc camuna*, (8) *Lyngbya taylorii*, (9) *Chroococcus limneticus*, (10) *Aphanocapsa endophytica*

### 3.2 Division Chlorophyta

In present study 20 species belong to 10 genera, in which maximum number of species as 5 were found in *Cosmarium*, 3 species in the genus *Closterium*, *Spyrogira*, 2 species of each genus *Chlorella*, *Oedogonium* and 1 species of each genus *Scenedesmus*, *Tetraedron*, *Cladophora*, *Hormidium*, *Zygnema*.

#### Genus *Spirogyra*

Simple filaments with cylindrical cell, 0.5-30.0 μm; diameter long ends wall plane, colligate, semi-replicate, replicate; chromatophores 1-16, straight or spirally arranged parietal ribbon-like bodies embedded in the cytoplasm, each with several to many more or less distinct pyrenoid; nucleus centrally placed supported by many strand of cytoplasm which terminate nearby pyrenoid.

#### Genus *Chlorella*

Members of this genus are small in size, shape of these cells is globose or sometimes ellipsoid in appearance, aggregated or solitary having thin layer of mucous membrane is found. In some species only a single chromatophore has been observed which is parietal and usually cup like appearance, in many species pyrenoid is usually absent.

#### Genus *Oedogonium*

The members of this genus have single filaments which are not branched and attached to substratum with the help of holdfast, cell are uni-nucleated, cylindrical in appearance, or some cases undulate or capitellate nodulose cells are also observed; reticulate chromatophore are present, pyrenoids are present at large intersections of reticulum; cells present at the base are with holdfast; terminal cells are obtuse in shape.

### **Genus *Scenedesmus***

Species of this genus exhibit oblong, ellipsoid and circular shapes of cells and are usually present as a multiple of 2; sometimes cells laterally in contact or rarely; connected by short process, in 1 or 2 rows (usually with quadrate arrangement); surface of the cell wall is either smooth or sometimes it is ornamented, having lateral or terminal teeth or spines, single chromatophore is observed, parietal in position, occupying the larger portion of cell, single pyrenoid has been observed.

#### *Scenedesmus dentiatulus* Lagerheim

Length of cells is measured to be 5.2 to 11.1  $\mu\text{m}$  whereas the width of the cells is observed to be 7.3 to 15.2  $\mu\text{m}$ , shape of the cells are mostly ovoid to ellipsoid, whereas these cells are including in straight or sub-alternating series, length of the spines along the poles are measured to be 1.1 to 4.2  $\mu\text{m}$  which are too short.

### **Genus *Tetraedron***

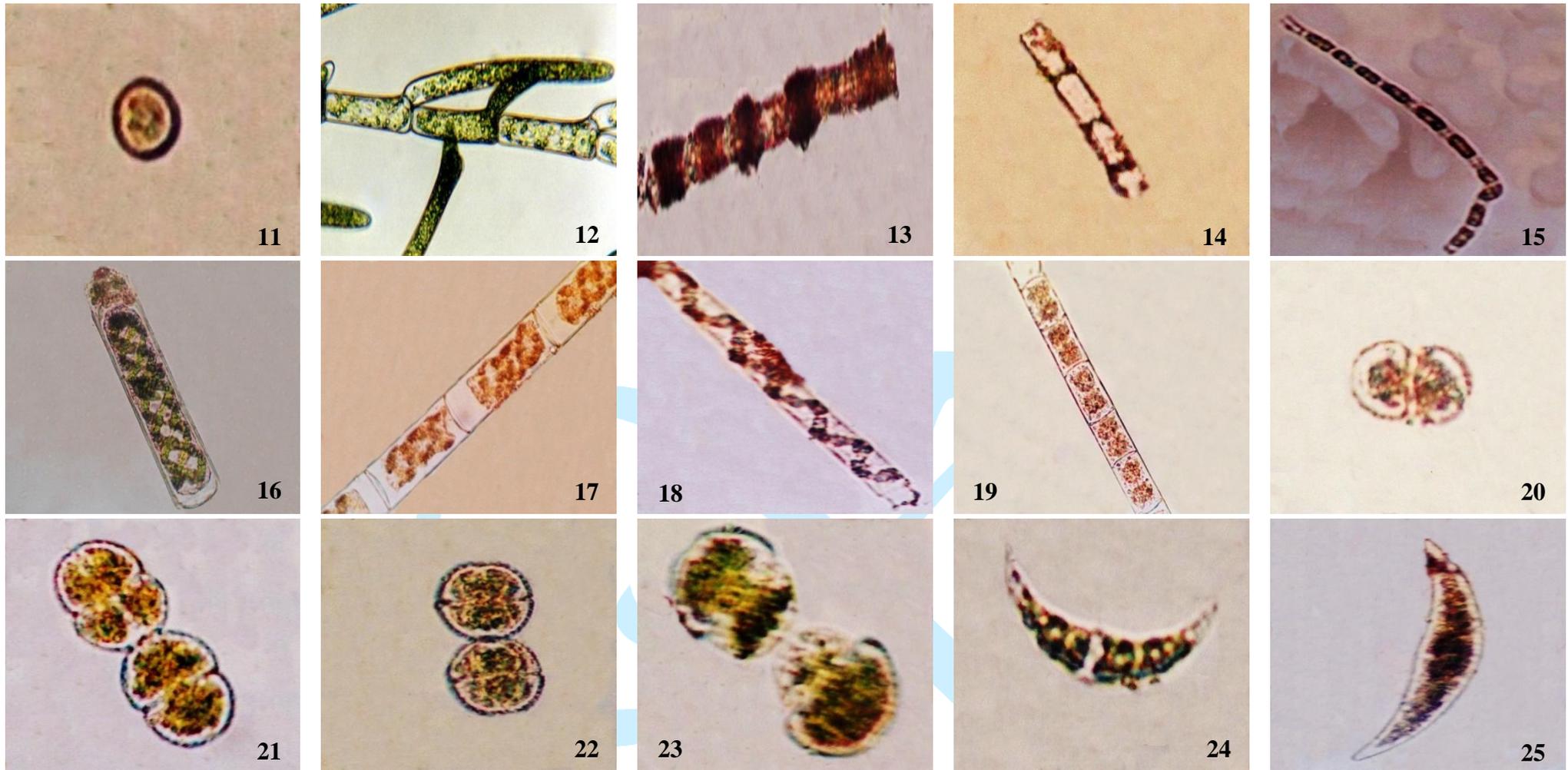
Cells of the members of this genus are solitary in position, pyramidal or triangular in symmetry, 4 to 5 angles are present which are simple in appearance and slightly extended, each angle consist of stout spines. In some species, base of the spines are inflated and they are much longer than the size of the cell. Each cell contains 1 pyrenoid and 1 chloroplast.

### **Genus *Cladophora***

Members of this genus are usually found in the form of filaments which are having profuse lateral branches, which are attached by rhizoidal cells or they can freely float on the surface of water as intertwined masses of unknown shares; single chromatophore is present whose surface pattern is reticulate or discoid, several pyrenoids are present in these species.

#### *Cladophora glumerrata* (L) Kuetzing

Length of cells is observed to be 45.2 to 150.1  $\mu\text{m}$  whereas the width of cells are measured as 300.5 to 1000.2  $\mu\text{m}$ ; size of the branches were also measured which is 35.3 to 60.7  $\mu\text{m}$  in length and 150.1 to 360  $\mu\text{m}$  in width; branches are often in the form of clusters. Length of the whole plant is measured to be 5.2 to 10.1 cm while in some species the length increases upto 1m; shape of the filament is Y shaped; color of the whole plant is light green sometimes dark green colour has also been observed.



**Fig. 3** Light microscopic photographs of members of division Chlorophyta: (11) *Chlorella pridinosa*, (12) *Cladophora glomerata*, (13) *Oedogonium epieticam*, (14) *Oedogonium sphaericoin* var. *conspicuum*, (15) *Hormidium gubtile*, (16) *Spirogyra* sp, (17) *Spirogyra* sp, (18) *Spirogyra porticulus*, (19) *Zygnema pacnitum*, (20) *Casmarium leave*, (21) *Casmarium phaseolus* var. *pseudostigmosum*, (22) *Casmarium occidentale*, (23) *Casmerium tetrachondrum* (peronatum), (24) *Closterium Incuruum*, (25) *Closterium moniliferum*

#### 4. Discussions

In the current research, various sites of District Kotli Azad Jammu & Kashmir were visited for the purpose of collecting freshwater algae. A total of 34 species belonging to 2 divisions were collected from various sites of study area. Division chlorophyta is the most dominant among the collection as it comprises of 20 species, 10 genera and 7 families. Light microscopic techniques were used to observe the micro-morphological characters of algae which will help to identify different species of algae. The classification of algae was investigated in detail according to taxonomic classification of Shameel, (2001).

Among division Chlorophyta class Chlorophyceae (*Cladophora*) and Siphonocladophyceae (*Oedogonium*) comprises of single genus while class Zygnemophyceae contains 2 genus and 4 species, genus *Spirogyra* 3 species and genus *Zygnema* with single species all these are identified from study area of Khajlani Charoi and Bandli of District Kotli. Kashif Ali et al., (2015) also reported the identical species of Scendesmaceae and Oedogoniaceae of algae from Shah Alam and Bara River in Peshawar similar was observed during present study. Pooja, (2014) reported from India that algal species of *Spirulina* and *Chlorella* as important source of food and having high medicinal values similar species were identified during the present study from Khuiratta, Bandli and Charoi of District Kotli. During present survey family Oscillatoraceae includes 4 genera and 4 species, genus *Phormidium*, *Aphanocaspe*, *Chroococcus*, and *Lyngbia* were reported among Class Chrocophyceae 1 genera and 2 species, genus *Merismopedia* (2spp). Class Nostocophyceae included 3 genera and 8 species, Genus *Nostoc* (1ssp), *Oscillatoria* (6spp), and *Spirulina* (1spp) were identified from the study area Bandli, Mora, Daraman, Khajlani and Seri Village of Kotli. (Tariq Ali et al., 2010) reported *Phacus*, and *Euglena*, from Punjab Province of Pakistan Lahore, Rawalpindi, Sargodha, Sheikhpura and Sialkot.

In present study from Class Euglenaphyceae 2 genera *Euglena* (2ssp), and *Phascus* (1ssp) were identified from Brooth and Dari Sudhana of district Kotli. From study area the Class Nostocophyceae with 3 genera and (8spp) were reported from Khuiratta. During present study out of 32 genus and 59 species including *Lyngbya*, *Chlorella*, *Spirogyra*, *Nostoc*, *Oscillatoria*, *Nitzschia*, *Synedra*, and *Phormidium* were reported. Among these species *Diatoms* and *Spirogyra* species identified were used for biodiesel production as previously reported by Kim et al., (2012).

#### 5. Conclusion

Micro-morphology plays a significant role in systematics study of algae. The present study clarifies the importance of light microscopy for the correct identification and delimitation of algal species based on morphological characters. This study contributes to taxonomic aspects and subsequent knowledge on the algal species. This is the first microscopic investigation that describes the morphology of 34 species of algae from District Kotli Azad Jammu & Kashmir.

#### Funding Information

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

#### Declaration of Conflict

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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