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Assessment of Water Quality by Bioindication of Algae and Cyanobacteria in the Peshawar Valley, Pakistan

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Abstract

In purpose to assess the water quality in Peshawar Valley, the diversity of algae and cyanobacteria were studied in 41 sites during 2018-2019. A total of 361 species-indicators of 7 Phyla were revealed. Algae and cyanobacteria in the studied sites preferred benthic and plankton-benthic lifestyle and mesotrophic waters. Indicators characterized water as moderate in temperature, medium oxygenated, low alkaline, and low saline. Algae and cyanobacteria inhabited medium-polluted and good water quality of Classes 2-3. The statistical maps were constructed for the first time to visualize the spatial distribution of diverse environmental and biological water quality variables and their relationship. The statistical maps and CCA revealed Water Temperature, Electrical Conductivity, Salinity, and Total Dissolved Solids as significant factors influenced freshwater algal and cyanobacteria communities. Statistical maps reflected an increase of dissolved substances from the foothills to the Kabul and Indus rivers' confluence. Acidification was revealed in the northeast of the valley. The bioindication results allowed us to propose that the algae and cyanobacteria communities were influenced by nutrient runoff from the surrounding foothills, agriculture, domestic and industrial effluents. The bioindication method combined with statistics can be recommended as a productive instrument for future water quality monitoring in the Peshawar Valley.

Introduction

Water is an essential component of life, but its quality has long been deteriorating due to both natural and anthropogenic activities (Uddin et al., 2021). In recent years, developing countries have faced significant problems in protecting water quality (Carvalho et al., 2011; Ortega et al., 2016). Even developed nations have been struggling to maintain or improve the status of their water quality (Abbasi & Abbasi, 2012; El-Karim, 2015). Management of water quality requires the collection and analysis of large water quality datasets that can be difficult to evaluate and synthesize (Uddin et al., 2021). Peshawar valley is an important geographic region in the upper basin of the Indus River. Water quality is a major concern here because it constantly pollutes the Indus River basin through the Kabul River (Yousafzai et al., 2010). The importance of assessing water resources in Pakistan is currently recognized as a pressing issue.

Algae and Cyanobacteria are the most diverse assemblage of organisms in an aquatic ecosystem (Wehr et al., 2015). They produce O_2 and various metabolites by utilizing CO_2, organic and inorganic pollutants, and nutrients (Bellinger & Sigee, 2015). Being susceptible to changes in the aquatic ecosystem, they have been used worldwide as bioindicators to assess water quality (Dokulil, 2003; Wehr et al., 2015). The use of algae and

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other aquatic organisms in assessing the water quality of the Peshawar Valley has been partially done previously for several points on large rivers (Ali et al., 2007; Barinova et al., 2013, 2016; Khuram et al., 2014,2017,2019; Mursaleen et al., 2018a, b). However, a complete analysis of water quality throughout the valley has never been carried out. For this purpose, we decided to create an international team of scientists from various universities in Pakistan and Israel to find the most effective methods to assess water quality in the riverine system of Pakistan, especially the Peshawar Valley. Several tools have been investigated to evaluate water quality, but bioindication through algae and cyanobacteria coupled with statistical methods are the most effective methods.

The bioindication method uses ecological characteristics (species preferences in aquatic habitats) of algae and cyanobacteria to characterize water quality (Sládeček, 1973, 1986; Barinova, 2017a). Canonical Correspondence Analysis (CCA) is a powerful and informative statistical tool for determining the major patterns of change in species composition and relating them to environmental characteristics of the habitats studied (Ter Braak & Šmilauer, 2002). Statistical maps depict the distribution of various attributes of communities in an area and help us to understand their pattern. From such maps, it is possible to infer the relation between different attributes in a region (Barinova, 2017b). Furthermore, combination of bioindication and statistics can help to reveal the trophic state and organic pollution in water, especially in developed countries where chemical analysis is expensive.

We therefore hypothesized that bioindication through algae and cyanobacteria coupled with statistical methods could be used to assess water quality in Peshawar Valley as the most effective method. Therefore, the present work was aimed at testing our hypothesis.

Materials and Methods

Description of Study Site

Peshawar Valley is the most distinct geographic region within the entire Khyber Pakhtunkhwa, Pakistan. It is located between 34°07’58’’N latitudes and 71°41’45’’E longitudes and has an altitude of about 335 m above sea level. The valley looks like an irregular ellipse with its longer axis (116 km) extending from east to west and the shorter axis (84 km) extending from north to south, girdled by the offshoots of the Hindu Kush mountains on all sides except its eastern perimeter, where the Indus River forms the natural boundary. The valley has a modified Mediterranean type of climate, having four seasons: autumn, winter,

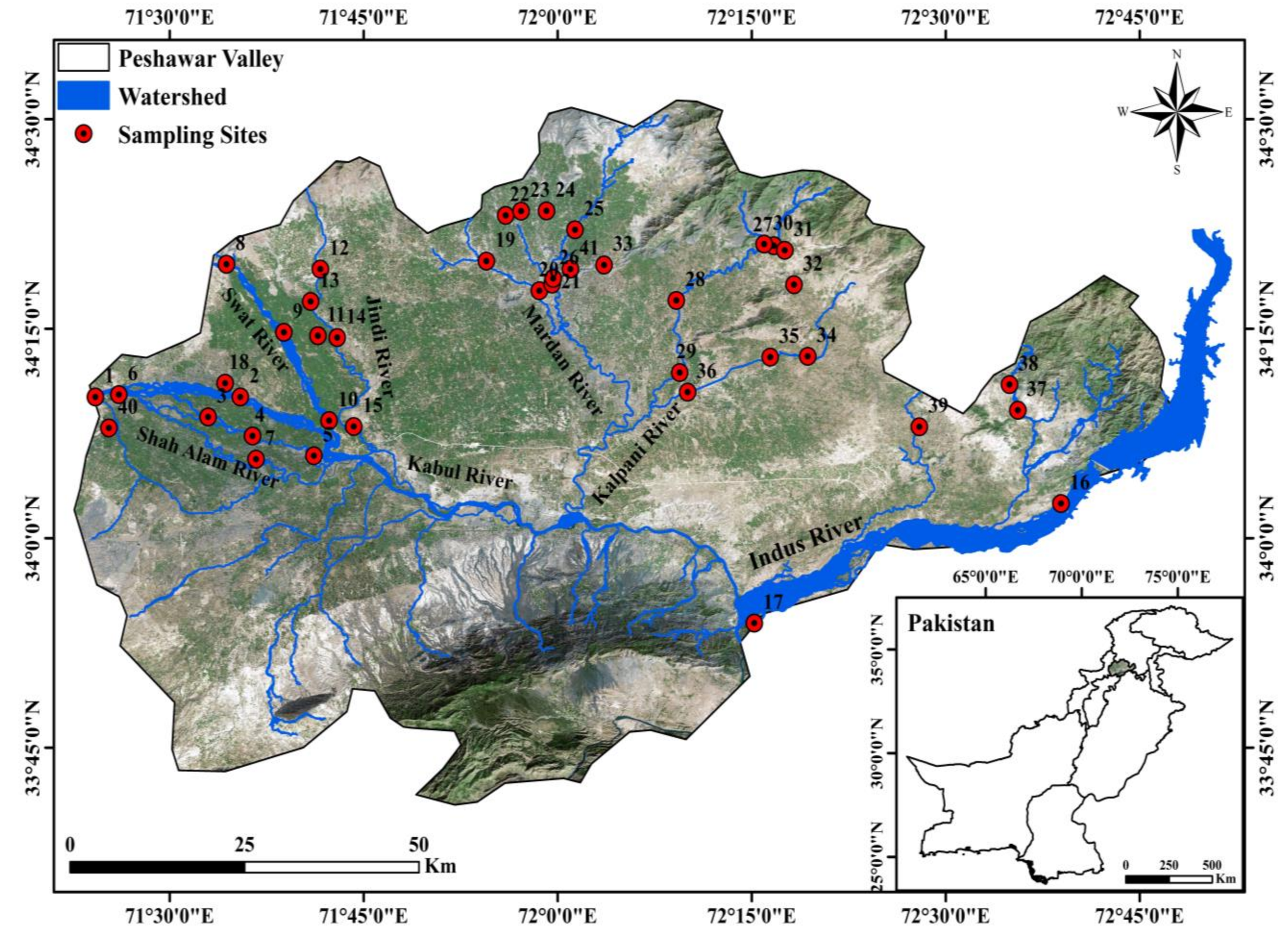

Figure 1. Sampling sites (red dots) in the Peshawar Valley in 2018-2019.

Table 1. Research sites with geospatial positions and coded name in the Peshawar Valley in 2018-2019.

No.	Site Name	Site Code	Latitude	Longitude	Altitude, m a.s.l.
1	Kabul River-Warsak	KRW	34.1686	71.4044	292
2	Kabul River-Haji Zai	KRHZ	34.1686	71.5912	279
3	Naguman River-Niyami	NRN	34.1447	71.5496	267
4	Naguman River-Naguman	NRNA	34.1221	71.6070	275
5	Naguman River-Jalabella	NRJ	34.0985	71.6860	238
6	Shah Alam River-Michni	SARM	34.1714	71.4346	289
7	Shah Alam River-Shah Alam	SARSA	34.0943	71.6114	275
8	Swat River-Munda	SRM	34.3270	71.5733	290
9	Swat River-Dildar Gharhi	SRDG	34.2457	71.6473	280
10	Swat River-Charsadda	SRC	34.1404	71.7055	247
11	Abazai River-Cheena	ARC	34.2417	71.6911	278
12	Jindi River-Tangi Harichand Road	JRTHR	34.3211	71.6943	299
13	Jindi River-Kanewar	JRK	34.2825	71.6818	280
14	Jindi River-Umarzai	JRU	34.2394	71.7160	279
15	Jindi River-Prang Majoke	JRPM	34.1334	71.7374	265
16	Indus River-Galla	IRG	34.0414	72.6488	287
17	Indus River-Attock Khurd	IRAK	33.8987	72.2535	214
18	Subhan Khwar Stream-Uchawala	SKSU	34.1850	71.5716	280
19	Jalala Stream-Jalala	JSJ	34.3307	71.9082	293
20	Jalala Stream-Mahabat Khan Koroona	JSMKK	34.2954	71.9765	263
21	Uch Khwar Stream-Umar Abad	UKSUA	34.3034	71.9930	285
22	Bama Kandah Stream-Hathian	BKSH	34.3852	71.9331	310
23	Ghargo Kandah Stream-Spalano Dheri	GKSSD	34.3904	71.9530	319
24	Lund Khwar Stream-Lund Khwar	LKSLK	34.3906	71.9857	325
25	Shamsi Dan Stream-Shamsi Dan	SDSSD	34.3682	72.0225	308
26	Shamsi Dan Stream-Said Abad	SDSSA	34.3100	71.9944	277
27	Balar Stream-Hamzakot	BSH	34.3490	72.2788	326
28	Balar Stream-Bakhshali	BSB	34.2838	72.1531	267
29	Balar Stream-Gari Kapura	BSGK	34.1976	72.1574	263
30	Pacha Tangi Stream-Cheena	PTSC	34.3510	72.2662	319
31	Dagi Stream-Hera Wand	DSHW	34.3435	72.2929	343
32	Machi Stream-Machi	MSM	34.3025	72.3047	316
33	Gadar Stream-Katlang Road	GSKR	34.3261	72.0599	298
34	Naranji Stream-Turlandi	NST	34.2171	72.3224	284
35	Naranji Stream-Adina	NSA	34.2161	72.2741	274
36	Naranji Stream-Sim Canal Road	NSSCR	34.1743	72.1676	255
37	Bada Stream-Pabaini	BSP	34.1528	72.5932	347
38	Panjman Stream-Panjman	PSP	34.1834	72.5824	362
39	Badri Stream-Mami Khel	BSMK	34.1330	72.4663	295
40	Warsak Wetland-Peshawar	WWP	34.1316	71.4217	296
41	Jamal Gharhi Wetland-Mardan	JGWM	34.3212	72.0165	310

spring, and summer with maximal precipitation of 70 mm in March and July (Government of Pakistan, 1998). Peshawar Valley has a rich hydrography, including many permanent rivers, streams, and wetlands (Figure 1). The Kabul River collects water from the perpetual and intermittent streams and rivers of the Peshawar Valley and flushes it into the Indus River near Attock (Salim et al., 1960; Government of Pakistan, 1998).

Sampling and Laboratory Processing

A total of 449 samples of algae and cyanobacteria were collected from 41 sites along the banks of 7 rivers, 16 streams, and 2 wetlands (Table 1). Unicellular, colonial, and filamentous algae ware collected by 10 ml, 20 ml, and 50 ml pipettes and by handpicking. Diatoms were collected by brushing their substrates (stones and hydrophytes) in a dish and by pipetting. Charophytes were collected by handpicking and grapnels. The samples were labeled with the site code, preserved in 5% FAA (Formaldehyde, acetic acid, and alcohol) solution, and stored in cardboard boxes to avoid spoilage (Edler & Elbrachter, 2010). Micromorphology of the non-diatomaceous specimens were examined by using the wet-mount method of Edler & Elbrachter (2010). The diatomaceous specimens were cleaned by using peroxide (H_2O_2) technique (Swift, 1967). The empty frustules were then mounted and analyzed for their micromorphology. The specimen's slides were subsequently viewed under the 10×, 20×, 40×, 60× and 100× Olympus BX-51microscope. The charophytes specimens were examined under the Olympus SZH stereo Microscope. Standard literature was followed for the taxonomic identification of the isolates (Collins, 1909; Transeau, 1951; Tiffany & Britton, 1952; Desikachary, 1959; Prescott, 1962; Krammer & Lange-

Table 2. Algae distribution across taxonomic groups in the Peshawar Valley in 2018-2019.

No.	Phylum	Class	Order	Family	Genus	Species
1	Cyanobacteria	1	5	13	19	46
2	Bacillariophyta	3	17	29	49	124
3	Charophyta	2	3	6	13	81
4	Chlorophyta	5	11	25	44	98
5	Euglenozoa	1	1	1	2	4
6	Ochrophyta	3	4	4	4	7
7	Rhodophyta	1	1	1	1	1
Total	7	16	42	79	132	361

Bertalot, 1986, 1988, 1991a,b; Cox, 1996). To avoid the use of synonyms, the identified species were checked in (Guiry & Guiry, 2021).

Determination of Water Physicochemical Properties

The major physicochemical variables of water quality (Temperature, pH, Oxidation Reduction Potential, Electrical Conductivity, Resistivity, Total Dissolved Solids, Salinity and Dissolved Oxygen) were measured in parallel with algal sampling at each sampling station on the spot by using HANNA HI-98194 multiparameter portable water quality meter (Khuram et al., 2019).

Bioindication Analysis

The bioindication methods for a range of environment variables by the ecological preferences of the revealed algae and cyanobacteria species (Friedrich et al., 1996) and their abundance were used for analysis (Barinova, 2017a). Bioindication properties of each revealed species came from a world database (Barinova et al., 2019) compiled by us. The bioindication systems included the most effective indication of the organisms' such as substrate and nutrition type preferences, water salinity, alkalinity, organic pollution, and trophic state. The correlations between major indicative variables are given in Barinova (2017a). The saprobic index S was calculated according to Sládeček (1973, 1986) to estimate the level of organic pollution. Index values S ranges from 0 (no polluted) to 4.5 (very polluted) for the aquatic environment. All data were ranked according to the CIS countries' classification system (Barinova, 2017c) to assess the Peshawar Valley water quality.

Statistical Analysis

Canonical Correspondence Analysis (CCA) was used as a direct gradient analysis technique for the evaluation of relationships among the algal floristics and environmental data (water quality variables). The analysis was executed in CANOCO version 4.5 Program (Ter Braak & Šmilauer, 2002). Similarity was calculated by network analysis in JASP Program (Love et al., 2019). A new method of environmental mapping was carried out to reveal visual correlations between biological and environmental variables using the Statistica Program version 12.0 according to the parameter values and geospatial coordinates of each site (Barinova, 2017b).

Results

Biological Variables

A total of 41 algal communities were studied along 7 rivers, 16 streams and 2 wetlands in the Peshawar Valley. The species list of algae and cyanobacteria were comprised of 361 taxa belonged to 132 Genera, 79 Families, 42 Orders, 16 Classes of 7 Phyla (Table 2 and Appendix Table S1). The highest number of species were contributed by Bacillariophyta (124 sp.; 34.3%), followed by Chlorophyta (98 sp.; 27.1%), Charophyta (81 sp.; 22.7%), Cyanobacteria (46 sp.; 12.7%), Ochrophyta (7 sp.; 1.9%), Euglenozoa (4 sp.; 1.1%) and Rhodophyta (1 sp.; 0.3%) (Table 2). The highest number of algal species (77) were found in BSP community, while the lowest number of species (3) were observed in UKSUA community (Appendix Table S1). All revealed species were indicators of the water quality variables.

Environmental Variables

The average values of environmental variables can be seen in Table 3. The variables were varied in a narrow range which depicted that water in the studied sites was fresh, low saline, low alkaline and moderately saturated with oxygen. Peshawar Valley is a very flat place, surrounded by mountains, having an altitudinal gradient ranging from 214-362 m a.s.l. This narrow range (148 m) of altitudinal gradient had given us the possibility to construct statistical maps for this very flat area.

The distribution of major environmental variables along with altitudinal gradient as statistically generated maps can be seen in Figure 2. The relevancy of statistical mapping to the entire data gradient visualization can be seen in Figure 2a. Water temperatures in the studied water bodies were higher in the piedmont's small rivers and streams (Figure 2b). The salinity mapping showed opposite results, where most of the saline water was situated in the valley plane (Figure 2c). Water pH revealed that only one point had pH below 7 and reflected the acidification impact (Figure 2d). The highest concentrations of dissolved water oxygen were

Table 3. Averaged physicochemical variables of water in the studied sites of the Peshawar Valley with Index Saprobity S (SLA) and algal Species number in the site community in 2018-2019.

Site	Temp., °C	pH	ORP, mV	EC, µS cm^{-1}	RES, Ohm-cm	TDS, ppm	Salinity, PSU	D.O., mg L^{-1}	SLA	No of Species
1. KRW	15.84	8.2	67.4	435	1786	197	0.19	3.34	1.44	58
2. KRHZ	14.90	8.4	88.6	468	2158	240	0.24	4.16	1.50	42
3. NRN	11.58	8.3	147.3	471	2123	236	0.23	3.12	1.64	15
4. NRNA	11.94	8.3	28.0	468	2137	234	0.23	3.82	1.22	21
5. NRJ	14.07	8.4	86.3	438	2283	219	0.21	4.42	1.59	9
6. SARM	18.61	8.3	101.3	437	2288	218	0.21	3.48	1.51	40
7. SARSA	11.66	8.4	-50.8	474	2110	237	0.23	2.27	1.61	21
8. SRM	13.90	8.7	94.9	243	4115	122	0.12	4.34	1.91	14
9. SRDG	15.28	7.8	24.6	525	1905	262	0.26	3.91	1.47	17
10. SRC	16.44	8.4	74.5	308	3247	154	0.15	4.01	1.61	22
11. ARC	17.44	7.9	63.1	469	2132	234	0.23	3.83	1.69	45
12. JRTHR	16.42	8.5	68.2	582	1718	291	0.28	2.93	1.43	66
13. JRK	16.09	8.5	99.1	531	1883	265	0.26	4.39	1.46	28
14. JRU	15.24	8.1	67.0	565	1770	283	0.28	4.15	1.38	18
15. JRPM	14.74	8.2	82.4	549	1821	274	0.27	4.29	1.79	19
16. IRG	16.54	8.4	131.4	311	3215	156	0.15	3.44	1.73	72
17. IRAK	10.32	8.3	120.1	643	1555	321	0.32	3.84	1.10	7
18. SKSU	18.80	8.0	99.3	430	1753	176	0.17	4.02	1.29	25
19. JSJ	15.71	5.6	236.2	586	2152	297	0.29	3.34	1.66	14
20. JSMKK	14.79	8.3	63.3	554	1805	277	0.27	4.79	1.72	11
21. UKSUA	15.03	8.3	78.8	532	1880	266	0.26	4.66	1.10	3
22. BKSH	18.19	8.2	106.5	547	1828	274	0.27	4.59	1.58	6
23. GKSSD	16.93	7.8	124.5	581	1721	291	0.28	4.52	1.83	10
24. LKSLK	17.89	8.2	105.8	258	2175	315	0.31	4.46	1.60	25
25. SDSSD	15.25	8.5	101.2	296	2256	326	0.32	6.20	1.57	9
26. SDSSA	14.86	8.4	76.4	477	2096	239	0.23	5.09	1.46	14
27. BSH	19.37	7.7	85.7	557	1795	278	0.27	3.98	1.23	4
28. BSB	17.53	8.8	75.0	506	1976	253	0.25	8.09	1.47	16
29. BSGK	14.49	8.4	70.2	552	1812	276	0.27	4.59	1.84	10
30. PTSC	18.50	7.9	60.9	487	2053	244	0.24	3.82	1.74	13
31. DSHW	15.28	8.2	16.5	238	4202	119	0.11	2.66	2.01	48
32. MSM	18.53	7.9	78.7	402	2488	201	0.19	3.82	1.68	33
33. GSKR	18.22	8.1	87.7	579	1727	289	0.28	4.11	1.50	12
34. NST	14.66	8.0	71.5	468	2137	234	0.23	3.52	1.25	7
35. NSA	14.38	8.0	87.4	501	1996	250	0.24	3.46	1.20	6
36. NSSCR	12.28	8.4	71.1	639	1565	320	0.31	3.75	1.41	7
37. BSP	10.86	9.6	65.6	203	4926	102	0.10	4.34	1.65	77
38. PSP	11.99	8.8	63.5	269	3717	135	0.13	4.19	1.54	43
39. BSMK	16.33	8.1	54.2	509	1965	255	0.25	3.58	1.74	32
40. WWP	18.86	8.9	28.5	513	1949	256	0.25	2.22	1.46	42
41. JGWM	16.62	9.3	74.9	257	3891	128	0.12	7.02	1.47	14

found in a few piedmont habitats (Figure 2e). Only one point near the inflow of the Kabul River to the valley showed the influence of anoxia on aquatic organisms (Figure 2f).

Bioindication Analysis

Bioindication analysis showed that most of the algae and cyanobacteria species preferred benthic and planktonic-benthic habitats with moderate temperature, low streaming waters with medium enrichments of oxygen, low saline, and low alkaline water. More than half of the species were indicators of organic pollution, which preferred class 3 of water quality, moderate trophic level and autotrophic, tolerated an elevated concentration of organically bound nitrogen (Appendix Table S2).

Statistical Analysis

Correlation Analysis

The species-indicators content and taxonomic data was compared in the studied communities of the Peshawar Valley to reveal the similarity and main influencing factors (Figure 3). The richest communities with maximum species were grouped in cluster 2. So, cluster 2 (Figure 3) united communities dominated by charophytes (*Chara contraria* A. Braun ex Kützing, *C. globularis* Thuiller, *C. vulgaris* Linnaeus), cyanobacteria (*Oscillatoria princeps* Vaucher ex Gomont), along with diatoms (*Ulnaria acus* (Kützing) Aboal, *U. ulna* (Nitzsch) Compère) and green (*Tetradesmus incrassatulus* (Bohlin) M.J.Wynne, *Scenedesmus quadricauda* (Turpin) Brébisson, *Pseudopediastrum boryanum* (Turpin) E.

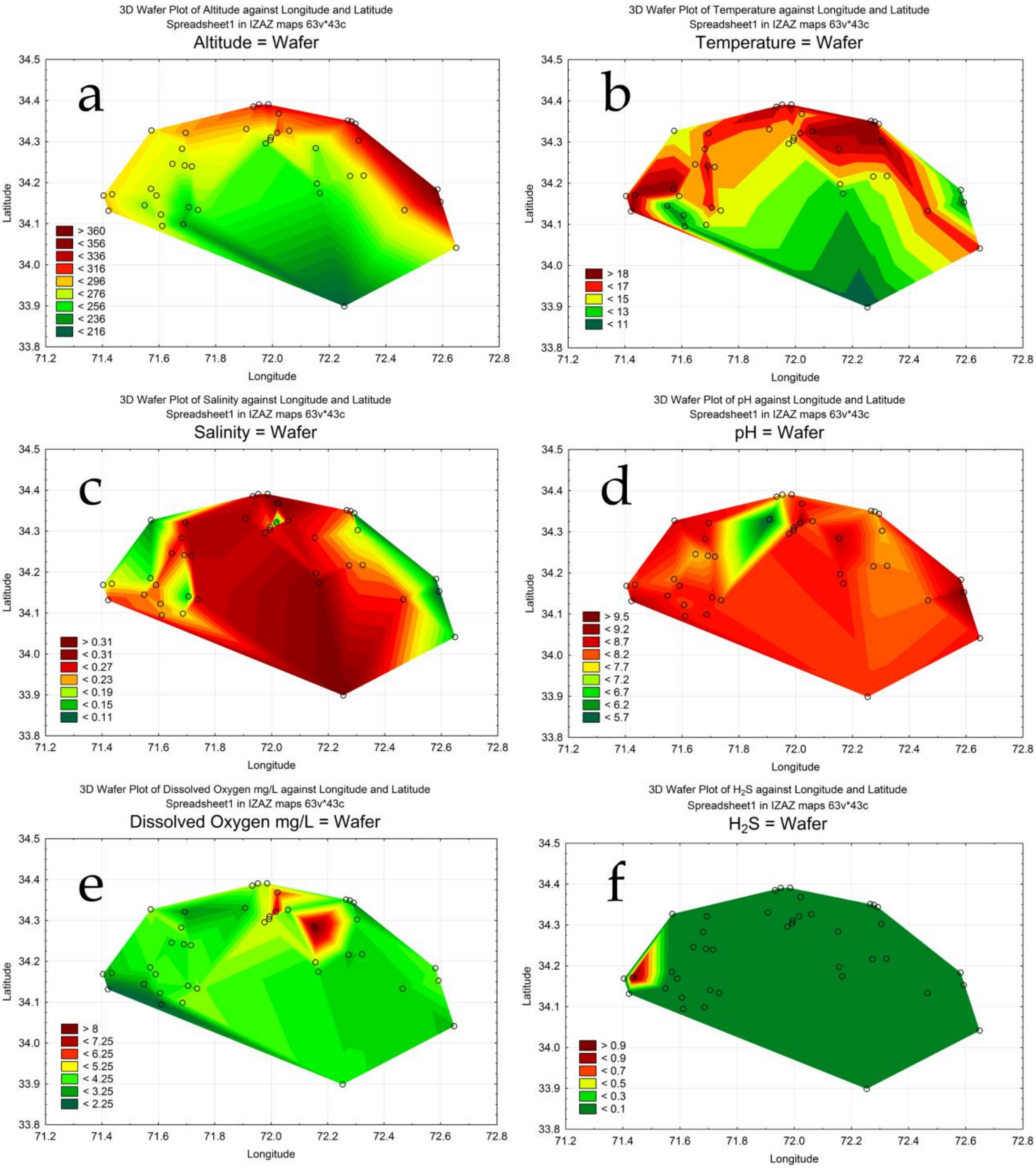

Figure 2. Statistical maps of spatial distribution of the environmental variables at the sampling sites in the Peshawar Valley in 2018-2019. a – site altitude; b – water temperature; c – water salinity; d – water pH; e – Dissolved oxygen concentration; f – H_2S bioindication.

Hegewald, *Hydrodictyon reticulatum* (Linnaeus) Bory) with rare algae *Cymbella kappii* (Cholnoky) Cholnoky, *Oedogonium pisanum* Wittrock ex Hirn, and *Rhizoclonium tortuosum* (Dillwyn) Kützing, whose ecology is poorly developed yet. Dominant bioindicators characterized the environment as moderately oxygen-enriched, slightly alkaline, fresh, and moderately polluted with organic substances, quality classes 2-3 with a developed autotrophic fouling community (Appendix Table S1).

Cluster 1 (Figure 3) was composed of communities dominated by charophytic (*Chara contraria* A.Braun ex Kützing, *C. vulgaris* Linnaeus, *C. globularis* Thuiller, *Spirogyra* sp., *Zygnema* sp.), green (*Cladophora glomerata* (Linnaeus) Kützing), and diatoms (*Surirella librile* (Ehrenberg) Ehrenberg) algae, which characterized the environment as medium oxygen-rich, alkaline, fresh, quality classes 2-3, from oligotrophic to eutrophic with a developed community of benthic autotrophic species (Appendix Table S1).

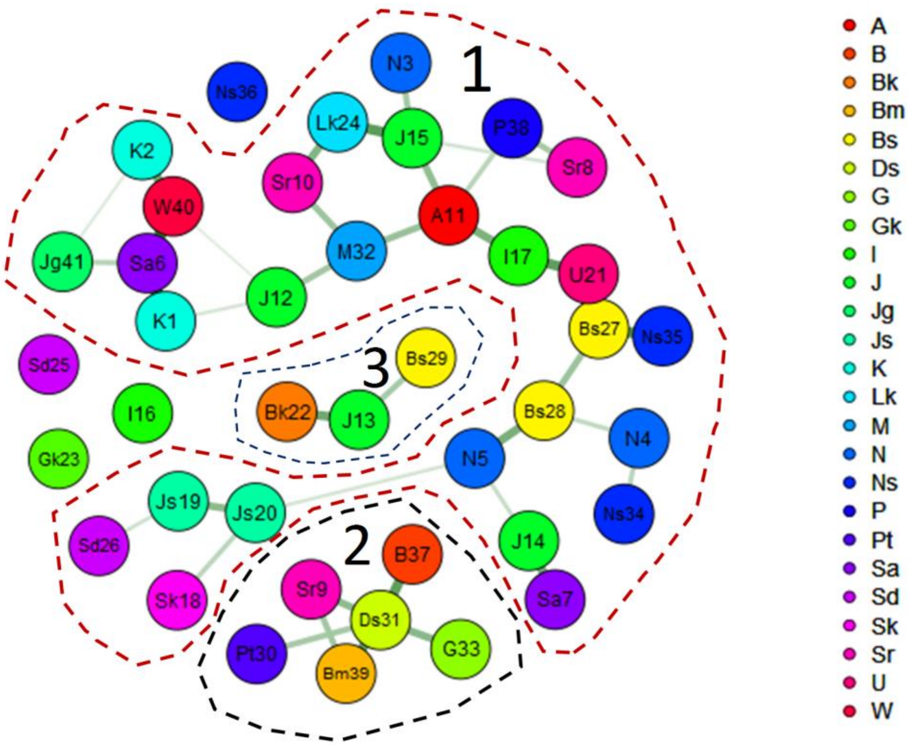

Figure 3. JASP Network graph of 50% similarity (significant only, p<0.05) of algae and cyanobacteria community at the sampling sites in the Peshawar Valley in 2018-2019. Numbers of corner are the same as sites in Table 1. The sites of the same watershed toned by the same color in the legend. The line thickness reflects the similarity level. Similarity clusters marked by dashed line.

Cluster 3 contained communities that were mostly distinct from one another, while the remaining communities were grouped in Cluster 1, which had a moderate degree of similarity. For cluster 3 (Figure 3), charophyte algae *Cosmarium granatum* Brébisson ex Ralfs, *Zygnema oveidanum* Transeau, *Chara vulgaris* Linnaeus, *C. globularis* Thuiller, *Spirogyra* sp., *Mougeotia* sp., as well as green alga *Cladophora glomerata* var. *crassior* (C. Agardh) C. Hoek were dominated. They formed permanent communities, which low changing in time. The dominant species-indicators characterized the environment as sufficiently oxygen-rich, fresh, mesotrophic, 2 classes of water quality (Appendix Table S1).

As a whole, dominated species of clusters were mostly benthic, pH-indifferent, autotrophic, preferred to grow in moderate temperature, medium oxygenated mesotrophic waters with low salinity and Class 2-3 of water quality. The numbers of corners on Figure 3 are the same as sites in Table 1. In the legend, sites in the same watershed are toned with the same color. The line thickness reflects the similarity level. Similarity clusters are marked by dashed line.

Statistical Mapping

Statistical maps clarified the distribution of most important groups of environmental indicators such as salinity, trophic state, as well as nutrition type of algae and cyanobacteria species on the Peshawar Valley. Figure 4a showed the distribution of halophiles that were mostly presented in the communities near rivers inputs to the valley. Mesohalobes that preferred brackish waters have two peaks in the center and the east part of the valley where agricultural press is higher (Figure 4b). Meso- to eutrophic condition indicators revealed two parts of the valley related to the Swat River input as well as the Indus River dam in the eastern part of valley (Figure 4c). In contrary, the hypertrophic condition was related to high agricultural part of the valley (Figure 4d). Autotrophic algae were mostly growing in the Indus River after dam (Figure 4e) whereas heterotrophs developed in the same communities where mesohalobes and hypertrophic species were dominated (Figure 4f). Bioindication of anoxia shows only onepoint water quality near the inflow of Kabul River to the valley influenced the aquatic organisms (Figure 2f) that correlated to high water temperature (Figure 2b) and low salinity (Figure 2c).

Statistical maps of pollution indication revealed that communities near the most watered areas, such as the Kabul and Swat rivers, as well as the area before the Indus River dam, had the highest number of species-indicators of Class 4 (polluted waters) water quality (Figure 5a). A map of the Index saprobity S value distribution (Figure 5b) revealed that most polluted areas were near piedmonts, which are represented by small streams.

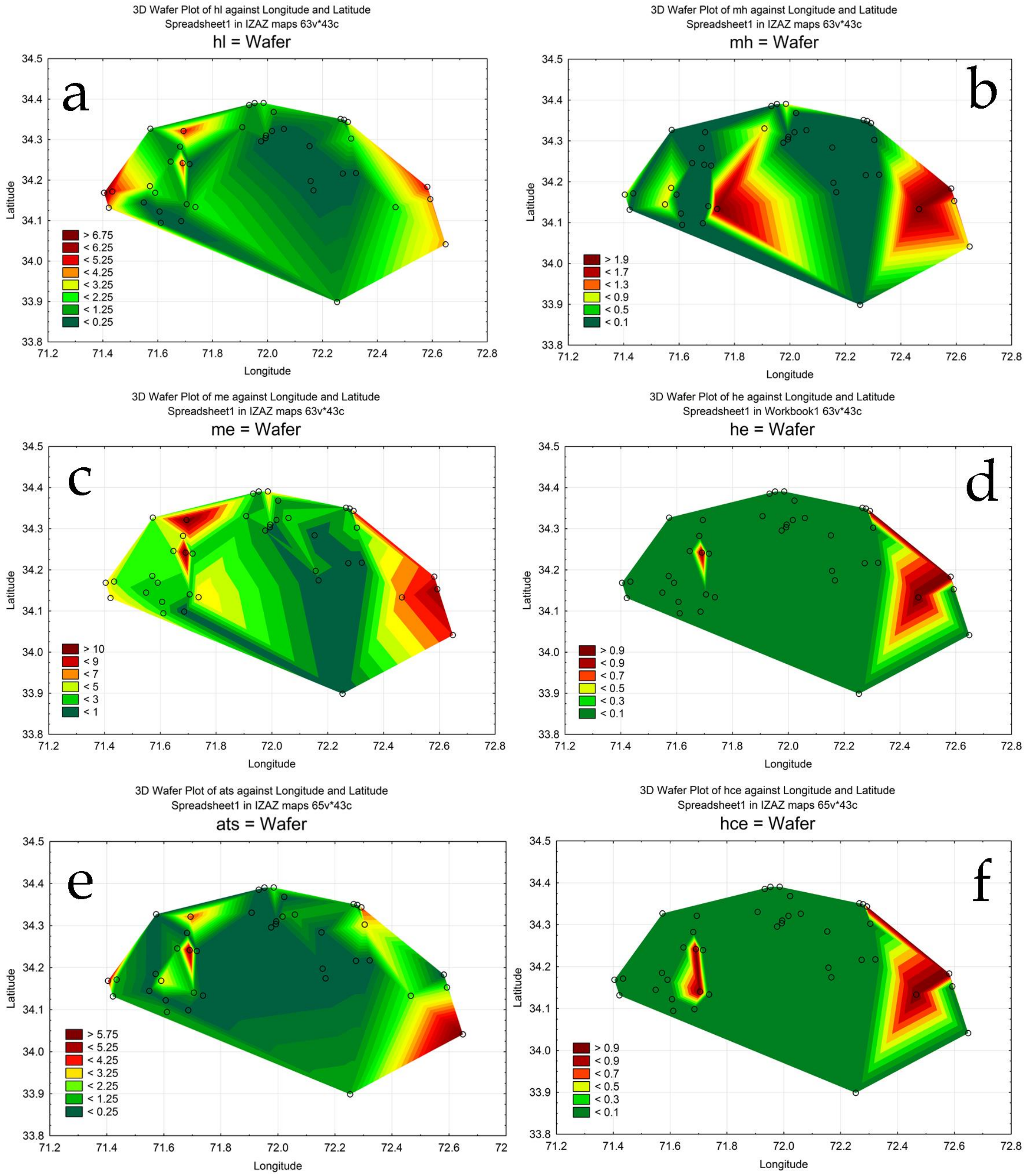

Figure 4. Statistical maps spatial distribution of the indicator species of algae and cyanobacteria at the sampling sites in the Peshawar Valley in 2018-2019. Salinity indicators: a – halophiles (hl); b – mesohalobes (mh); Trophic state indicators: c – meso-eutrophic (me); d – hypereutrophic (he); Nutrition type indicators: e – autotrophic (ats); f – facultative heterotrophic (hce).

At the final step of bioindication, we constructed the water quality map by following the EU method. The method is realized when the watercourse is marked by color related to the water quality Class defined by the Index saprobity S. Figure 6 showed that water in small tributaries and the upper reaches of rivers that fed into the valley was classified as Class 3. However, the lower parts of the rivers demonstrated self-purification and are mostly related to Class 2.

Species-Environment Relationships

The algae and cyanobacteria species represented by 7 Phyla along with eight water quality variables were subjected to CCA analysis to evaluate the algal communities' responses to environmental variables. CCA analysis (Figure 7) showed that overall electrical conductivity, temperature, salinity, and total dissolved solids influenced Rhodophyta species and most of the communities, while dissolved oxygen had weakly influenced Bacillariophyta and Chlorophyta species and only three communities. Cyanobacteria and Charophyta species and a few communities were influenced by any the water quality variables. However, Euglenozoa species were influenced by pH and resistivity in some communities.

Discussion

Algae are primarily aquatic organisms and occur in all surface water bodies in every biome across the globe. Their diversity is mostly affected by the physical, chemical, and biological parameters of the water body in which they live. These factors also regulate the distribution, survival, and occurrence of algae (Wehr et al., 2015). The current study revealed 361 algal species-indicators of water quality in the water bodies of the Peshawar valley, which is 46.70% of the total algal flora of Pakistan (Khuram et al., 2019).

Different algal species have different ecological preferences in aquatic habitats. They are very susceptible to environmental changes, which makes them excellent indicators of water quality (Maestre et al., 2012).

In our analysis, bioindication revealed the major water properties such as alkaline, low saline, and low- to moderate organically polluted habitats with temperate temperatures and moderately saturated by oxygen. Community comparison revealed three groups that

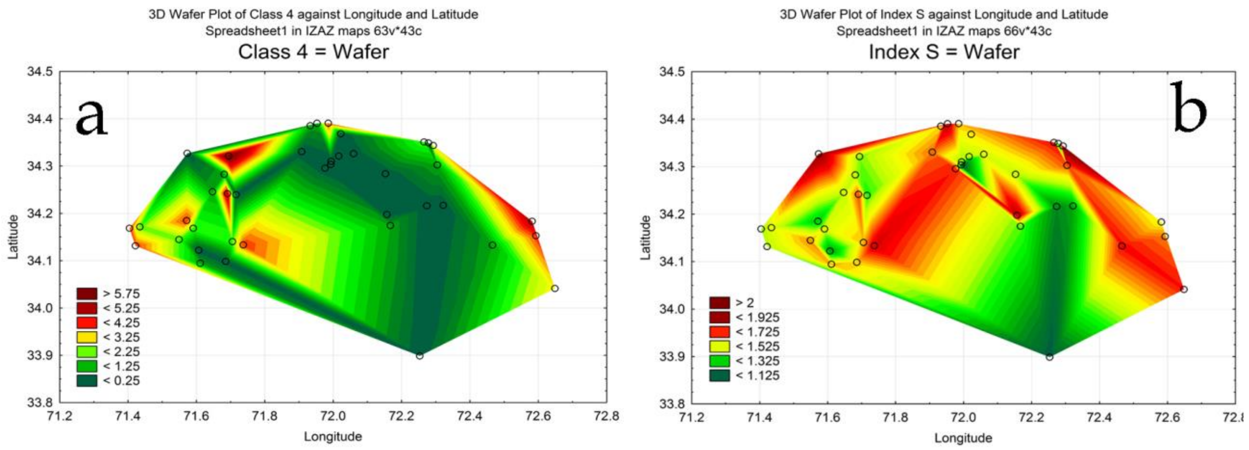

Figure 5. Spatial distribution of the indicator species of algae and cyanobacteria over coordinates of the sampling sites in the Peshawar Valley in 2018-2019. a – Class 4 indicators map; c – Index saprobity S map.

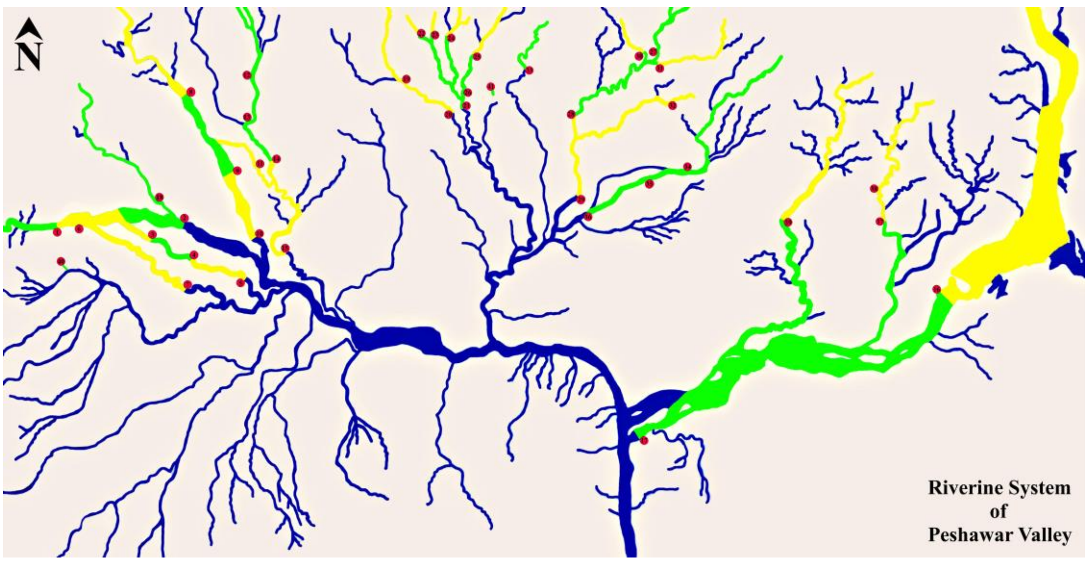

Figure 6. Water quality map on the base of Index saprobity S value in the sampling sites of the Peshawar Valley in 2018-2019 with EU color code: green – Class 2, yellow – Class 3.

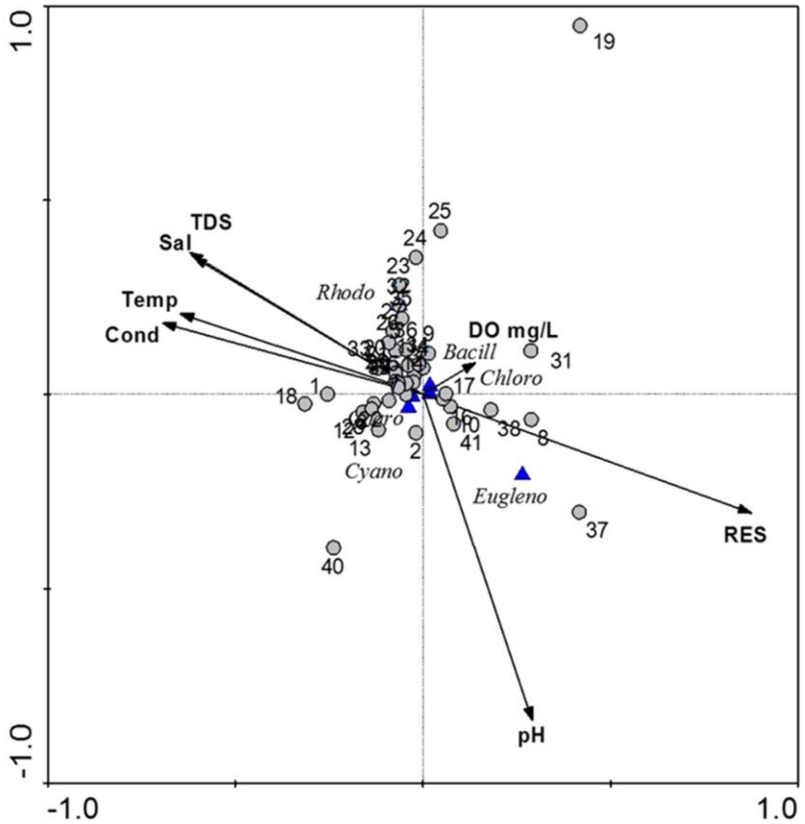

Figure 7. Canonical Correspondence Analysis (CCA) triplot of environmental variables and species richness in algal and cyanobacteria Phyla relationships on the sampling sites of the Peshawar Valley in 2018-2019. Eigenvalue 0.029; p=0.0740.

related to the trophic state of water as a major factor that regulated species content. The trophic state is related to algal productivity, which is directly linked with the water quality (Barinova, 2017a,c). Our results revealed that in the Peshawar Valley, most sites were inhabited by mesotrophic indicators, but in a few sites, the oligotrophic species were dominated in the community, while some sites were defined as a eutrophic. The level of trophicity is determined by the concentration of nutrients, which, therefore, is a determining factor in developing the algae assemblage in the Upper Indus region (Ali et al., 2007; Barinova et al, 2013; Mursaleen et al., 2018a, b).

We revealed the prevalence of autotrophic taxa in the studied sites. Almost many communities' indicators demonstrated a low to moderate level of dissolved organic matter. The index saprobity S values were fluctuating near the boundary of Class 2 and Class 3 of water quality with prevalence of Class 3 indicators (Appendix Table S2) as revealed in the Upper Indus tributaries (Ali et al., 2007; Barinova et al., 2013; Mursaleen et al., 2018a,b). Calculated indices of saprobity and the list of revealed species indicators clearly demonstrated a dynamic and high trophicity of the studied area. Studied waters were classified as medium eutrophic (beta-mesosaprobes), but the saprobity index values showed the water quality Class 2 and 3 describing not only moderate organic pollution but also the intensive self-purification processes. Comparing our findings to those of other studies in the Peshawar Valley, we concluded that we obtained a similar assessment of the state of the environment, as well as including bioindication and pollution indices within the framework of the WFD (Guidance, 2002; Poikane et al., 2016). CCA showed that Electrical Conductivity, Temperature, Salinity, pH were major influencing factors for communities in Peshawar Valley as revealed by Khuram et al. (2017) in their studies.

Since our study's goal was a comprehensive study on water quality assessment by algae and cyanobacteria in the Peshawar Valley, we tried to use new, up to now unused statistical methods, such as statistical mapping (Barinova, 2017b). The method's applicability was verified by constructing a statistical map of the distribution of altitude of sampling sites. This was a key point in our study that gave a positive result because the Peshawar Valley is a flat plain between the mountains and since the number of research points is forty-one. In our case, statistical maps helped visualize the distribution of various parameters of algal communities and their environmental variables on the surface of the earth (Skorobogatova et al., 2019). Comparison of our maps can be doing to recent environmental mapping constructions (Barinova, 2017b; Krupa et al., 2018,2019; Skorobogatova et al., 2019), which were primarily related to lentic water bodies (Barinova, 2017b) and then to lotic water body catchment areas (Barinova, 2017b; Krupa et al., 2019). The following conditions served as the foundation for this study in terms of constructing ecological maps using statistical methods:

(1) the leveled surface of the Peshawar Valley (214 to 362 meters), where samples were taken on all types of water bodies at the same time, and (2) a dense network of sampling stations (41 sampling stations). Thus, the distribution of all the identified environmental parameters and algal communities along the well-limited surface of the Peshawar Valley was mapped for the first time.

Bioindication characteristics on the map reveal zones of increased indicator value in the areas of the Kabul and Swat rivers at the places of their entry into the valley and the Indus River in the northeast. The maps help to identify acidification in the north of the valley. This may be the result of anthropogenic impact on the aquatic communities of the Peshawar Valley. The distribution map of the Saprobity Index S (Figs. 5b, 6) reflects the diversity of the effects of organic pollution, which is associated with the influx of pollution from large rivers and has a local character associated with economic activities in the valley. In any case, the map of Index S shows that aquatic ecosystems utilize organic pollution in the direction southeast to the confluence of the Kabul and Indus rivers. The fact that the pollution is of a local toxic nature is shown by the map of the distribution of heterotrophy indicators with a high number in the east of the valley and partially at the confluence of the Swat and Kabul rivers, where economic activity is most active. So far, we have limited material to compare our statistical mapping results with those of other river basin communities since this statistical method was first applied here in the Peshawar Valley. Our earlier experience with statistical mapping concerned the oil-producing region in Northern Siberia, where there was no geographical boundary of the landscape (Skorobogatova et al., 2019), and the delta of a large river (Barinova et al., 2021).

Conclusions

Our study of environmental variables of water quality demonstrated a narrow range of variation that does not allow us to reveal some specific points or variables in the valley. It encourages the use of bioindication methods for assessing water quality. As a result, studies of algae and cyanobacteria diversity in 41 aquatic habitats of the Peshawar Valley revealed large species richness of 361 species considered to 7 Phyla. The water quality indicators were mostly benthic and plankton-benthic inhabitants, and characterized water as moderate in temperature, low moving, neutral in pH, low saline, mesotrophic, Class 2-3, good to moderate water quality. CCA analysis defined water temperature, electrical conductivity, salinity, and TDS as main factors influencing the algal communities. Statistical distribution maps showed an increase in water saturation by dissolved substances from the foothills to the Kabul and Indus rivers' confluence that cannot be revealed by other methods. Moreover, bioindication characteristics on the distribution maps revealed zones of increased low water quality indicators in the areas of the Kabul and Swat rivers at the places of their entry into the valley and the Indus River in the northeast. Therefore, the algal communities in Peshawar valley were influenced by nutrient runoff from the surrounding foothills, agriculture, and dumping domestic and industrial effluents in the valley's water bodies.

Thus, we confirmed our hypothesis about the effectiveness of bioindication by algae and cyanobacteria communities and statistical mapping to identify the impact on the Peshawar Valley waters, which was impossible to do only using chemical data that had a narrow amplitude. The use of statistical maps as an assessment model helped to identify the range of water quality in the valley and ranked environmental factors in terms of their importance for the algae community and the direction of impact. Because statistical mapping helps to reveal visible correlations between environmental and biological variables, further application of the bioindicator method and statistics can help to assess the temporal and spatial dynamic of trophic relationships in the aquatic ecosystems of the Peshawar Valley for improving water quality monitoring in the future.

Supplementary Materials: Appendix Table S1: Diversity of algae and cyanobacteria in the sites of the Peshawar Valley with species-specific ecological preferences and abundance scores in the Peshawar Valley; Appendix Table S2: Summary of species number in taxonomic Phyla and indicator taxa number in the studied sites of the Peshawar Valley.

Author Contribution

I.K.: Conceptualization, methodology, validation formal analysis, investigation, data curation, writing—original draft preparation, writing—review and editing. N.A.: Conceptualization, data curation, supervision. C.N.S.: Validation, writing—original draft preparation, writing—review and editing. S.B.: Conceptualization, methodology, software, validation, formal analysis, data curation, writing—original draft preparation, writing—review and editing, visualization, supervision.

All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

The authors declare that they have no known competing financial or non-financial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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