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РІЗНОМАНІТНІСТЬ ВОДОРОСТЕЙ ПЛАНКТОНУ ВЗДОВЖ ГРАДІЄНТА ВИСОТИ В ПРІСНОВОДНИХ ЕКОСИСТЕМАХ РЕГІОНУ КХУМБУ, НЕПАЛ

Розуміння різноманітності та складу угруповань водоростей у різних градієнтах висоти та факторів, що на них впливають, може дати цінну інформацію про екологічні процеси, які визначають функціонування цих екосистем. Метою дослідження було встановити різноманітність і склад водоростей у регіоні Кхумбу в Непалі та оцінити їхній зв'язок із змінними середовища вздовж градієнта висоти. Альгологічний матеріал було зібрано у 13 озерах та річках на висоті від 2700 м до 5000 м і проаналізовано відносний вплив різних факторів навколишнього середовища (висоти, температури, вмісту сполук азоту і фосфору, концентрації розчиненого кисню, загальної мінералізації та електропровідності) на видове багатство водоростей. Загалом було зареєстровано 56 видів водоростей із шести класів. Серед них Bacillariophyceae були найпоширенішими. Видове багатство водоростей як збільшува-

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лось, так і зменшувалось під впливом різних фізико-хімічних факторів. Для досліджуваного регіону характерною виявилась загальна тенденція до зростання видового багатства водоростей із збільшенням висоти. Це явище можна пояснити залежним від висоти підвищенням загальної мінералізації, а також вищим вмістом сполук азоту та фосфору, зумовленим активізацією людської діяльності. З усіх змінних вміст сполук фосфору мав найсильніший вплив на видове багатство водоростей, що вказує на те, що цей чинник є найважливішим, який впливає на формування якості води. Збільшення вмісту сполук фосфору та подальше зростання видового багатства водоростей у цих високогірних прісноводних озерах і річках вказує на погіршення якості води у водоймах Гімалаїв. Оскільки кліматичні зміни та зростання інтенсивності туризму вже підвищують вразливість регіону Кхумбу, отримані дані підкреслюють важливість регулярного моніторингу досліджуваного регіону.

Ключові слова: *різноманітність водоростей, діатомові, регіон Кхумбу, фізико-хімічні властивості води, Національний парк «Сагарматха», Непал.*

Introduction

Algae are found almost in any habitat as they have unique adaptive capacities to survive in varying environmental conditions. Algae are primary producers and have the ability to absorb different organic and inorganic pollutants, including heavy metals, from the water. They play an important role in aquatic ecosystems by cycling nutrients and producing oxygen, which can help with natural water purification. They are also commonly used as bioindicators of water quality because of their sensitivity to environmental changes such as nutrient levels and pollution [41]. While algae can indicate the ecological health of aquatic systems, excessive growth, frequently caused by nutrient pollution, can result in harmful algal blooms that degrade water quality and disrupt aquatic populations.

Algal richness has been found to be correlated with a variety of environmental factors including altitude [20], pH and alkalinity [27], anthropogenic activities [38], ionic content [30], and land use pattern [2, 6]. An unimodal pattern of phytoplankton abundance, i.e., the higher the elevation, the greater the algal abundance has been found [17]. Other studies have further shown that the community structure of algae is also related to elevation [17]. However, another point of view [5] suggests that there is a difference in the linear and unimodal patterns of algal abundance, arguing that other factors could have played a much larger role than elevation. It is thought [12] that altitude alone is not a good indicator of algal species richness; instead, conductivity and the nutrient concentrations, depth, and area all play a role in determining species richness. There is now a growing consensus on the role of climate, topography, and anthropogenic activities in explaining the significant difference in algal diversity across localities.

The Sagarmatha National Park (SNP) and its surrounding buffer zone, collectively known as the Khumbu region (Figure 1), is located between 27°30'19" and 27°06'45" N latitude and 86°30'53" to 86°99'08" E longitude. The park lies on the southern slope of the Sagarmatha (Mt. Everest) in the northeastern region of Nepal and covers an area of 1.148 square kilometers. It is characterized by rugged topography, with altitudes ranging from 2.845 meters at the Jorsalle to 8.848 meters at the summit of Mt. Everest. Approximately 80 % of the precipitation falls during the monsoon season, which lasts from June to September. The average minimum temperature recorded is in January (-7.7 °C), while the maximum recorded temperature is in August (16.2 °C). The SNP's two major lake systems are the Imja and Gokyo. The major four rivers that drain the Khumbu region from the north to the south are Dudhkoshi, Lobuche, Imja, and Bhotekoshi. The Dudhkoshi River originates from the Ngozumpa glacier and the Gokyo lake system. The Imja River flows from Lake Imja and glaciers, while the Lobuche River flows from the Khumbu glacier. The Imja River is formed when the Lobuche and Imja rivers meet below the Dingboche. There the Imja River joins the Dudhkoshi River below Phortse and is renamed the Dudhkoshi River. The Bhotekhoshi River originates in Tibet and meets the Dudhkoshi River at Larjadobhan. These major rivers also give birth to several tributaries along their course.

Construction activities, substratum eradication, deforestation, forest fires, and haphazard utilization of river water all have an impact on water quality, its volume, phytoplankton diversity loss, and ecological balance in water bodies [5, 25, 37]. This is a common global trend and is particularly high in regions with high human activities. An increasing number of tourists in the SNP, for example, has already altered its land cover pattern, trail, and overall environment [32]. The impact of tourism and overall anthropogenic pressure is particularly increasing in areas surrounding Lake Gokyo in the SNP. Hotels discharge their wastes directly or indirectly into the lake, increasing its pollution. Because the physiochemical properties of the water can determine pollution level in water bodies, understanding the nutrient content in water bodies is critical for assessing their health status [13, 31, 35]. Although several studies in recent years have explored the diversity of algae in lowland rivers and lakes, the exploration of algal diversity in high altitude rivers and lakes in Nepal and their association with different environmental factors has not yet to be made. Therefore, the goal of this study was to evaluate the distribution pattern of algae in the lake and river clusters of the Khumbu region along an elevation gradient, and also to determine the association between algal diversity and the physical and chemical characteristics of water bodies.

Material and Methods[1]

Sample collection. The samplings sites were mostly selected along the trekking routes from the Lukla to the Everest base camp, including Lake Gokyo, the Imja River and the Thame spring. Algal samples were collected from different corners of lakes and different sides of rivers between 9:00 to 12:00 am. Totally thirteen samples were collected from different water sources, including stagnant waters and running waters. Algal samples were collected by dipping plastic bottles (250 ml capacity) in water sources and preserved with formalin (with

[1] The authors would like to express their gratitude to the Department of National Parks and Wildlife Conservation for granting us permission to conduct field research in the Sagarmatha National Park (SNP). We would also like to thank the warden of the SNP for his support during the field study. We are also grateful to the field guides and local residents of the SNP for their assistance in data collection.

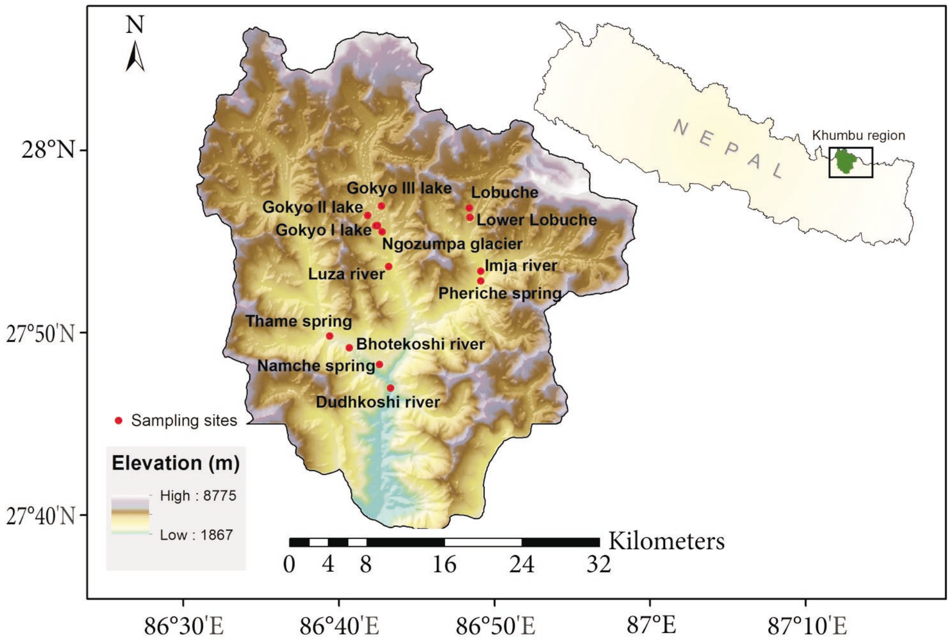

Fig 1. Map showing 13 sampling sites in the Sagarmatha National Park and surrounding buffer zone (the Khumbu region)

the final concentration of 4 %) immediately after collection. The identification of species was made with the help of a Leica binocular microscope and consulting relevant monographs e.g. [3, 9, 40, 46].

Physiochemical parameters of the water. Water temperature, pH, total dissolved solids (TDS), and conductivity were measured on site with the help of an electric kit multi-parameter probe (HANNA, HI9812-5), while dissolved oxygen (DO) was determined with the help of a meter (Ecosense ODO 200). The measurements were made during May — June.

Water samples for chemical analysis were collected from the water bodies in 125 ml acid-rinsed polythene bottles. In this case, three drops of concentrated nitric acid were used to adjust the pH of the samples below 2 (acidic medium) to preserve them. The samples were brought to Kathmandu and analyzed in the Ecology laboratory of the Central Department of Botany of the Tribhuvan University, Kathmandu. Nitrogen and phosphorus content in the lake water samples were measured using a spectrophotometer based on the colorimetric method following [43].

Data analyses. Correlation analysis was performed to identify the linear relationship among the variables representing different physical and chemical parameters of the water in the sampled sites. The Pearson's correlation coefficient was used to measure the strength of the relationship between variables. The correlation analysis was made using the «cor.test» function in R version 4.2.1. Because samples were collected from different water sources (e.g., lake, river and spring), we compared if total algal richness and the physiochemical properties varied among water sources. We calculated the mean values of each variable and compared them using one-way ANOVA followed by Tukey's HSD post-hoc test. These analyses were performed using «aov» and «TukeyHSD» functions in the R package «stats».

In order to capture the non-linear relationship (if any), generalized additive models (GAMs) were built to assess the variation in physical and chemical properties of water along an elevation gradient. Natural cubic splines with three degrees of freedom were used in the GAM model to allow for flexible modeling of the relationship between the variables. The underlying idea was to assess how the physical and chemical properties of water change along an elevation gradient. The GAM lines were plotted using «geom_smooth» function in the R package «ggplot2» with y ~ splines::ns(x,3) as the plotting formula.

To determine the impact of various physical and chemical properties of the water on algal diversity, we first combined data from all sites and conducted regression analysis with algal richness as the dependent variable and physical and chemical properties of water as independent variables, one at a time. The regression analysis was performed using the negative binomial generalized linear model. These models were built using the «glm.nb» function available in the R package «MASS». The R^2 value in these regressions was calculated using the following formula:

$$\left(1-\frac{\text{Residual deviance}}{\text{Null deviance}}\right)\times 100.$$

We also assessed the relationship between algal richness and various physical and chemical properties of the water using generalized additive models to capture more complex relationships not captured by a simpler linear model. Finally, we used random forest modeling to perform a multivariate regression. Random forest is a strong ensemble learning algorithm that is commonly used in predictive modeling and machine learning tasks. It is recognized for its flexibility, robustness, and efficiency in handling varied datasets. By computing variable significance, it provides the relative contribution of each input variable to the model's predictive performance. Random forest modeling was performed using the 'randomForest' function in the R package 'randomForest'.

Results

Species composition of alage. In total, 56 species belonging to six classes (Bacillariophyceae, Zygnematophyceae, Cyanophyceae, Chlorophyceae, Trebouxiophyceae and Euglenophyceae) were recorded across different sampling sites situated at altitudes ranging from 2777 to 5007 m (Table 1). Bacillariophyceae were found to be the dominant class in the Khumbu region. There were 21 species from Bacillariophyceae, 17 species from Zygnematophyceae, 9 species from Cyanophyceae, 6 species from Chlorophyceae, 2 species from Trebouxiophyceae and 1 species from Euglenophyceae (Table 1). The class Bacillariophyceae covered 37.5 % of the total algal distribution, followed by Zygnema-

Table 1

Algal species found at different locations in the Khumbu region

SN	Taxa	Sampling sites	Altitudinal range (m)	Habitat
	Cyanophyceae			
1	*Anabaena* sp.	8, between 2 to 3	3400—4700	Running water
2	*Gloeocapsa aeruginosa* Kützing	between 10 to 13	4300	Stagnant water
3	*Merismopedia glauca* (Ehrenberg) Kützing	7, 10, 4, between 11 to 13	3700—4300	Stagnant as well as running water
4	*Oscillatoria subbrevis* Schmidle	4	4600	Moist rocks in sloppy region
5	*Phormidium* cf. *insigne* Anagnostidis	between 1 to 2	4700	Running water
6	*Phormidium schroeteri* (Hansgirg) Anagnostidis	10	4900	Stagnant water
7	*Phormidium* sp.	3	4660	Outlet of Lake Gokyo I
8	*Planktothrix agardhii* (Gomont) Anagnostidis & Komárek	9, 10	2700—4900	Stagnant water
9	*Stigonema mamillosum* Agardh ex Bornet et Flahault	12	5000	Outlet of Lake Imja
	Bacillariophyceae			
10	*Cocconeis placentula* var. *euglypta* (Ehrenberg) Cleve	7	3700	Small pond edge of the Bhotekoshi River
11	*Craticula perrotettii* Grunow	5, 6, 7, 13	3700—4300	Running as well as stagnant water
12	*Cymbella cymbiformis* Agardh	between 2 to 3, 13	4300—4700	Stagnant as well as running water
13	*Cymbella lanceolata* Agardh	7, 10 , 13	3700—4600	Running water
14	*Denticula* sp.	between 2 to 3	4700	Running water
15	*Didymosphenia geminata* (Lyngbye) Schmidt	between 10 to 13, 8	3400—4300	Running water
16	*Eunotia lunaris* (Ehrenberg) Grunow	13	4300	Stagnant water at edge of the Lobuche River
17	*Eunotia monodon* var. *alpina* (Kützing) Grunow	5	4300	Running water
18	*Fragilaria crotonensis* Kitton	10, 13, 6	3700—4600	Running water

Table 1 (continued)

SN	Taxa	Sampling sites	Altitudinal range (m)	Habitat
19	*Fragilaria vaucheriae* (Kützing) Petersen	9, between 10 to 13, 2 to 3	2700—4700	Running water
20	*Frustulia rhomboides* (Ehrenberg) De Toni	5	4300	Running water
21	*Gomphonema sphaerophorum* Ehrenberg	between 2 to 3, 6, 7	3700—4700	Running water
22	*Hannaea arcus* (Ehrenberg) Patrick	7, 10, between 1 to 2, 2 to 3	3600—4700	Slow running water
23	*Meridion circulare* (Greville) Agardh	13	4300	Stagnant water at edge of the Lobuche River
24	*Odontidium mesodon* (Ehrenberg) Kützing	7, 9, 11,6, between 1 to 2, 2 to 3	2700—4770	Running water
25	*Pinnularia brauniana* (Grunow) Studnicka	8	3400	Running water
26	*Pinnularia viridis* (Nitzsch) Ehrenberg	6, 8, 10, 13	3400—4900	Running as well as stagnant water
27	*Stauroneis phoenicenteron* (Nitzsch) Ehrenberg	6	3700	Running water
28	*Surirella didyma* Kützing	9, between 10 to 13	2700—4300	Stagnant water
29	*Tabellaria flocculosa* (Roth) Kützing	5, 7, 13	3700—4300	Running water
30	*Ulnaria ulna* (Nitzsch) Compère	between 1 to 2, 2 to 3, 11	4300—4700	Stagnant as well as running water
	Zygnematophyceae			
31	*Actinotaenium* cf. *subglobosum* (Nordstedt) Teiling	9	2700	Stagnant water at edge of the Dudhkoshi River
32	*Closterium acerosum* Ehrenberg ex Ralfs	between 1 to 2, 2 to 3, 7, 8, 13	3400-4700	Running water
33	*Cosmarium awadhense* Prasad et Mehrotra	1, 7	3700—4660	Stagnant water
34	*Cosmarium* cf. *sublatereundatum* West et West	9	2700	Stagnant water with rocky habitat
35	*Cosmarium subspeciosum* Nordstedt	5, 13	4300	Stagnant as well as running

Table 1 (continued)

SN	Taxa	Sampling sites	Altitudinal range (m)	Habitat
36	*Cylindrocystis brebissonii* (Ralfs) De Bary	5	4300	Running water
37	*Cylindrocystis* sp.	8	3400	Running water
38	*Euastrum coralloides* var. *trigibberum* Lagerheim	5	4300	Running water
39	*Euastrum oblongum* Greville ex Ralfs	13	4300	Stagnant water
40	*Hyalotheca dissiliens* Brébisson ex Ralfs	13	4300	Stagnant water
41	*Mougeotia* sp.	4, 5, 13 between 2 to 3	4300—4700	Moist steep rocks, stagnant as well as running water
42	*Netrium digitus* (Brébisson ex Ralfs) Itzigsohn & Rothe	5	4300	Running water
43	*Penium cylindrus* Brébisson ex Ralfs	13	4300	Stagnant water
44	*Spirogyra* cf. *amplectens* Skuja	1, 4, 8, 9	2700—4660	Rocky sloppy region, running as well as stagnant water
45	*Staurastrum* sp.	5, 8, 9, between 2 to 3, 10 to 13	2700—4700	Running as well as stagnant water
46	*Staurodesmus mordax* var. *nudus* (Turner) Compère	4	4600	Moist sloppy rocks
47	*Zygnema* sp.	4, 10, between 1 to 2	4700—4900	Moist rocky habitat as well as running water
	Trebouxiophyceae			
48	*Botryococcus* cf. *braunii* Kützing	between 10 to 13	4300	Stagnant water
49	*Chlorella vulgaris* Beijerinck	between 2 to 3	4650	Slow running water
	Chlorophyceae			
50	*Bulbochaete* sp.	3	4660	From outlet of Lake Gokyo I
51	*Desmodesmus communis* (Hegewald) Hegewald	4, 13	4300—4600	Sloppy moist rocky region and running water

Table 1 (continued)

SN	Taxa	Sampling sites	Altitudinal range (m)	Habitat
52	*Oedogonium* sp.	between 10 to 13, 3	4300—4660	Stagnant water
53	*Pediastrum duplex* Meyen	between 10 to 13	4300	Stagnant water
54	*Sphaerocystis schroeteri* Chodat	between 10 to 13	4300	Stagnant water
55	*Tetradesmus obliquus* (Turpin) Wynne	8, 11, 13	3400—4300	Running water, stagnant water
	Euglenophyceae			
56	*Phacus* sp.	8	3400	Running water

Note. 1 — Lake Gokyo III; 2 — Lake Gokyo II; 3 — Lake Gokyo I; 4 — the Ngozumpa glacier; 5 — the Luza River; 6 — the Thame Spring; 7 — the Bhotekoshi River; 8 — the Namche Spring; 9 — the Dudhkoshi River; 10 — the Lobuche River; 11 — the Lower Lobuche River; 12 — the Imja River; 13 — the Pheriche spring.

tophyceae (30.4 %), Cyanophyceae (16.0 %), Chlorophyceae (10.7 %), Trebouxiophyceae (3.6 %), and Euglenophyceae (1.8 %).

Correlation among different physiochemical properties of the water. Bivariate correlations showed high congruence among the variables representing physiochemical water parameters. For example, nitrogen content had a negative relationship with pH, phosphorus content, dissolved oxygen, and temperature, and a positive relationship with total dissolved solids and conductivity (Figure 2). Total dissolved solids and conductivity had the highest correlation coefficient ($r = 0.93$) followed by that between phosphorus content and pH ($r = 0.73$).

Variation in algal richness and physiochemical properties of the water across three ecosystems. The ANOVA and Tukey's HSD tests revealed no significant differences ($p>0.05$) in the mean values of algal richness and other physiochemical properties of the water across three ecosystems (Figure 3).

Variation in physiochemical properties of water across elevation. The result of generalized additive model showed that the total nitrogen, total phosphorus, total dissolved solids and conductivity increased with increasing elevation, while temperature and dissolved oxygen decreased with increasing elevation (Figure 4). While some variables showed near linear relationship (dissolved oxygen), most other variables exhibited a hump-shaped or non-linear relationship.

Influence of elevation and physiochemical properties of the water on algal richness. Along an elevation gradient, the highest number of algal species was recorded from Lake Gokyo I (4661 m), whereas in the Imja River (4165 m), only one species of Cyanophyceae was recorded (Table 2). The algal diversity

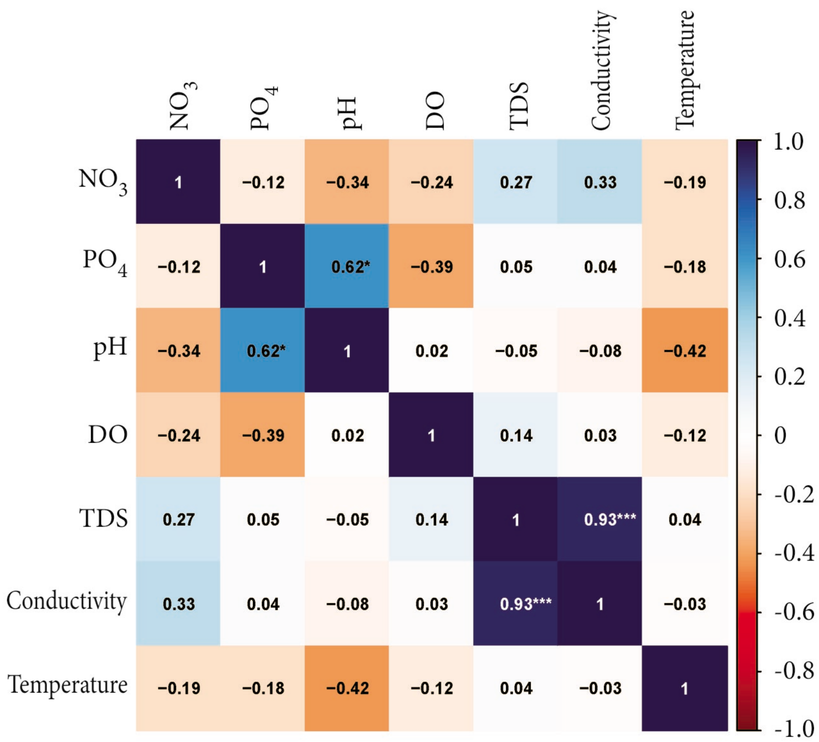

Fig 2. Correlation among the variables representing different physical and chemical properties of the water. Significant correlations are indicated with an asterisk (*), where the number of asterisks denotes varying levels of significance. *** p<0.001, * p<0.05

showed a hump-shaped relationship with the elevation (Figure 5), with species richness peaking at around 4500 m, after which the algal diversity decreased. Species richness exhibited both increasing and decreasing trends in relation to various physiochemical properties of the water. For example, species richness increased with the increasing value of total phosphorus and pH. Both total dissolved solids and conductivity displayed a bimodal pattern, exhibiting fluctuations where algal diversity initially decreased, then increased, and subsequently decreased again (Figure 5).

The total phosphorus content explained the highest variation in species richness (R^2 = 22.63 %) followed by pH (R^2 = 10.20 %), elevation (R^2 = 7.29 %), total dissolved solids (R^2 = 7.21 %), dissolved oxygen (R^2 = 6.49 %), temperature (R^2 = 4.88 %) and conductivity (R^2 = 3.34 %) (Table 3). The random forest model likewise identified total phosphorus as the variable possessing the greatest predictive power (Figure 6), aligning with the findings of the generalized linear model.

Discussion

Diversity of algal species in the Khumbu region. Totally 56 algal species from the water bodies (6 rivers, 3 lakes, 3 springs and 1 glacier) of the Khumbu

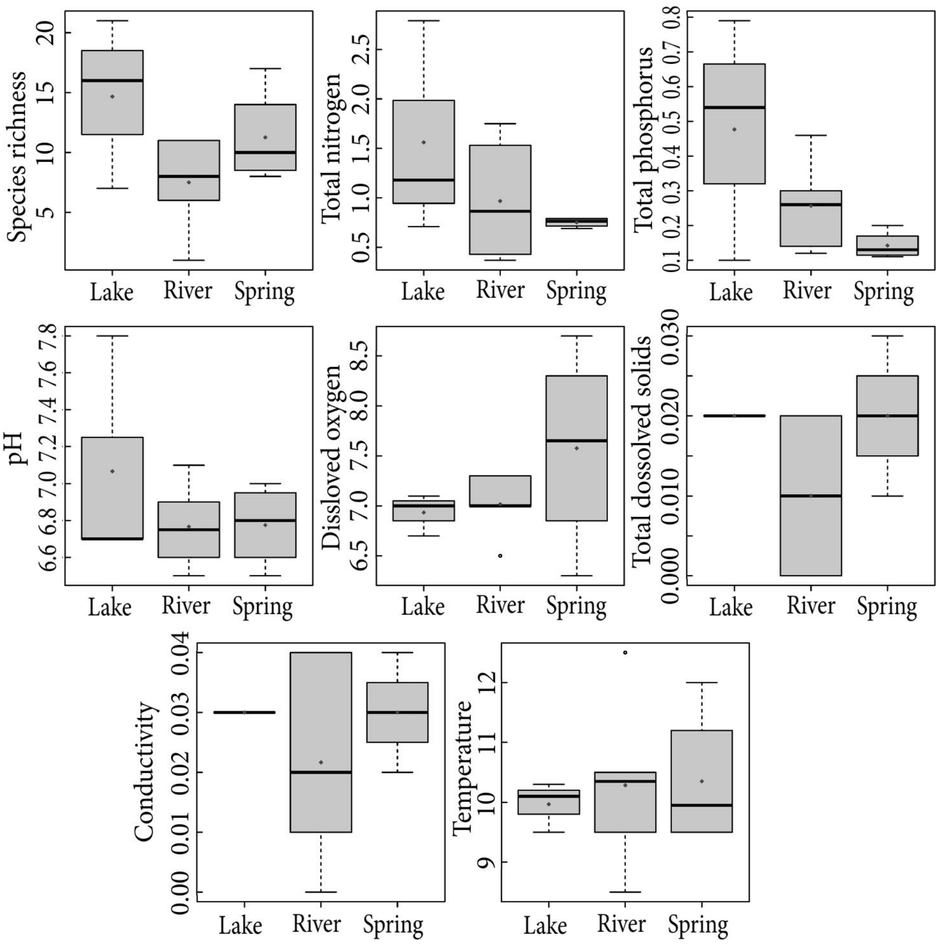

Fig 3. Boxplot showing differences in algal richness and physiochemical parameters of water across lake, river and spring of the Khumbu region. Mean values are represented by diamonds. The ANOVA and Tukey's HSD tests revealed no significant differences (p>0.05) in the mean values among three types of water ecosystems

region belonging to 6 classes (Bacillariophyceae, Zygnematophyceae, Cyanophyceae, Chlorophyceae, Trebouxiophyceae, and Euglenophyceae) were found during the period of investigations. The differences in the number of identified algae could be related to their growth and affected by competition between different algal groups, predation in the food chain, the level of pollution, and the flow characteristics of the water [39]. The significant differences in phytoplankton community structure with different nutrient states in water bodies directly reflect varying water quality status [16]. For example, the phytoplankton species belonging to Cyanophyceae and Chlorophyceae are good indicators of environmental change as they can detect the direct entry of domestic sewage, agricultural pollution, municipal waste, and effluents of organic waste from animals and human beings [22]. Similarly, the dominance of populations from the

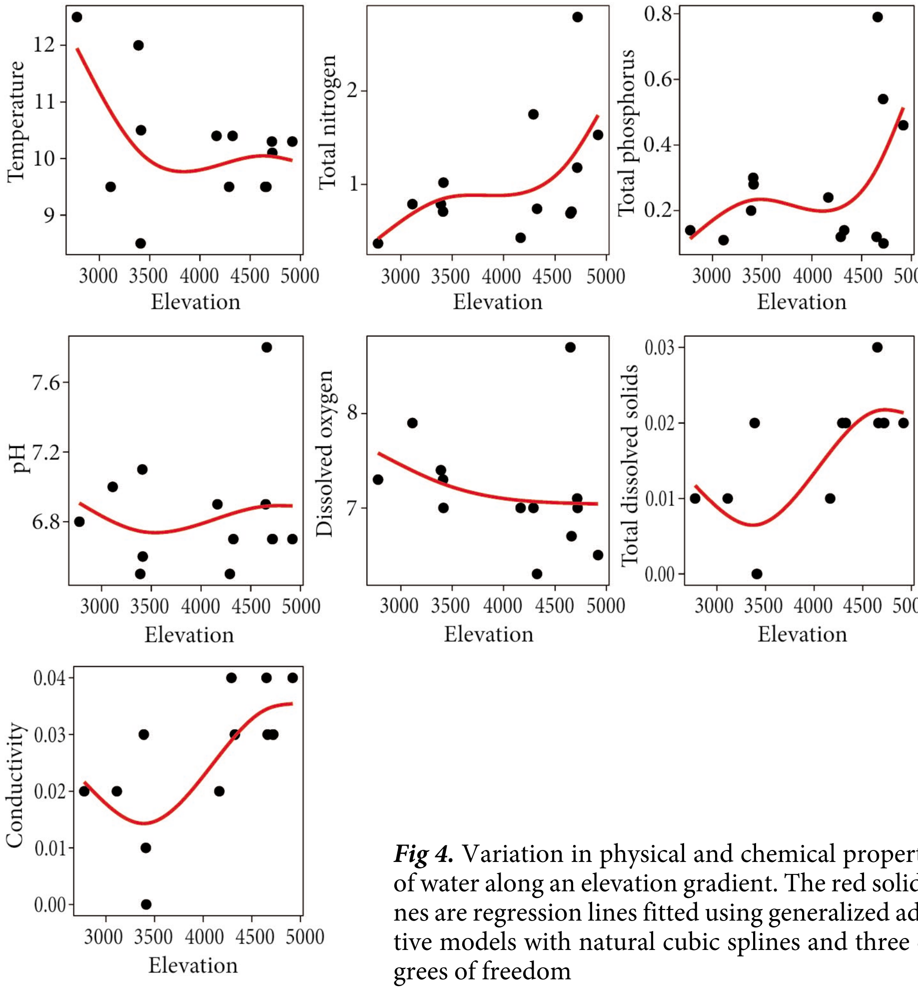

Fig 4. Variation in physical and chemical properties of water along an elevation gradient. The red solid lines are regression lines fitted using generalized additive models with natural cubic splines and three degrees of freedom

Bacillariophyceae class can indicate eutrophic conditions in reservoirs, as some species thrive in nutrient-rich environments and are sensitive to changes in water quality [36]. However, not all diatoms are associated with eutrophication, and their presence does not necessarily indicate a reservoir's trophic status [24]. For instance, the presence of *Fragilaria vaucheriae* is indicative of eutrophic conditions, whereas *Hannaea arcus* is associated with oligotrophic environments [44]. Eutrophication is a complex process influenced by a variety of factors, including nutrient levels, temperature, light, etc. As a result, while diatoms are important bioindicators, their role should be interpreted in light of the larger ecological and environmental context.

Physiochemical properties of water along an elevation gradient. As indicated by our data, the temperature is primarily altitude-dependent as we observed that the temperature of surface water decreased linearly with increasing elevation up to 4000 m after which the temperature reaches an asymptote and decreases again after 4700 m. This is plausible because the air temperature is strongly related to altitude and decreases as altitude increases in mountain are-

Table 2

Total number of species recorded from various algal classes at different locations and altitude

SN	Locality	Alt. (m)	Trebouxiophyceae	Bacillariophyceae	Cyanophyceae	Zygnematophyceae	Chlorophyceae	Euglenophyceae	Total species
1	Lake Gokyo III	4720	1	4	—	2	—	—	7
2	Lake Gokyo II	4716	1	8	3	4	—	—	16
3	Lake Gokyo I	4661	—	7	5	7	2	—	21
4	Ngozumpa glacier	4650	—	2	1	4	1	—	8
5	Luza river	4325		3	—	6	—	—	9
6	Thame spring	3692	—	6	1	1	—	—	8
7	Bhotekoshi River	3410	—	8	2	1	—	—	11
8	Namche spring	3417	—	3	1	4	2	1	11
9	Dudhkoshi River	2777	—	2	1	3	—	—	6
10	Lobuche River	4919	—	3	2	—	2	—	7
11	Lower Lobuche River	4219	1	4	3	2	1	—	11
12	Imja River	5007	—	—	1	—	—	—	1
13	Pheriche spring	4279	—	7	—	8	2	—	17

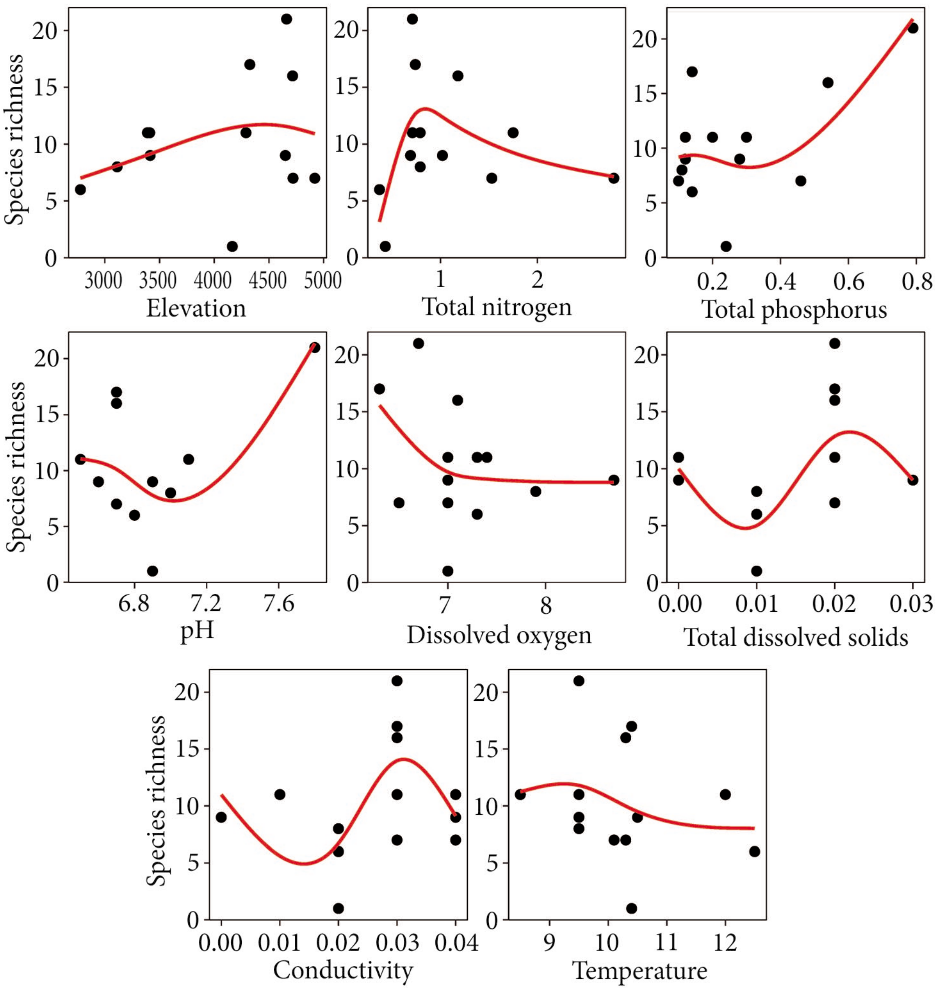

Fig 5. Relationship between algal species richness and different physical and chemical properties of water. The red solid lines are regression lines fitted using generalized additive models with natural cubic splines and three degrees of freedom

as [34]. This is a common global trend and has been reported by a number of other studies [17]. The decrease in dissolved oxygen along with the rise in elevation might be due to the decrease in atmospheric pressure as we move upward. It has been found [1] that the concentration of total dissolved solids decreased with altitude which is in contrast to our observation. This disparity may be due to differences in location and uneven human pressures between the two study areas. Another research [53] also discovered that the concentration of dissolved solids decreased with increasing altitude due to an unequal distribution of rock type along altitude. A negative correlation was established between altitude and other physical parameters from the Sagarmatha National Park, a positive correlation — between altitude and pH, and a near-neutral cor-

Table 3

Explanatory power (R^2, %) of elevation and other water-related parameters for the algal diversity in the lake and river cluster of the Sagarmatha National Park evaluated by negative binomial generalized linear model

Variables	R-square (%)	Coefficient	p-value
Elevation	7.29	0.0002	0.28
Total nitrogen (NO_3^-)	0.34	-0.0541	0.81
Total phosphorus (PO_4^{3-})	**22.62**	1.1202	0.04
pH	10.20	0.4486	0.24
Dissolved oxygen	6.49	-0.2281	0.34
Total dissolved solids	7.21	17.2277	0.29
Conductivity	3.34	8.762	0.47
Temperature	4.88	-0.1188	0.38

Note. Statistically significant (p<0.05) variable is shown in bold font face.

relation — between altitude and conductivity [33]. In contrast, our data showed that as altitude increased, so did total dissolved solids and conductivity. This could be due to the presence of more anions and cations, as well as the higher concentration of degradable matter dissolved in the water. This might also explain why algal richness increased as we moved upward.

Correlation among physical and chemical properties of the water. Dissolved oxygen showed a positive correlation with total dissolved solids and a negative correlation with phosphorus, nitrogen, pH, temperature, and conductivity. Consistent with [21], we found a negative correlation of pH with the dissolved oxygen. The pH was negatively correlated with the phosphorus content and this relationship was also reported in [7]. The pH affects the concentration of orthophosphate in the aquaponics nutrient solution. The increase in pH lowers the overall concentration of orthophosphate as high pH influences the formation of insoluble calcium phosphate that precipitates from the solution. The temperature has an opposing effect on the concentration of dissolved oxygen in a water body. The higher the temperature, the lower the concentration of dissolved oxygen [48] and our findings clearly support this relationship. Total dissolved solids and conductivity were very strongly correlated with each other (r = 0.93) and such significant association has also been reported in [28]. Conductivity is a useful indicator of total dissolved solids. Water's electrical conductivity is provided by dissolved ions of various metallic and organic salts. The rate of dissociation of salt and organic matter increases with temperature [18]. However, it has also been argued that the relationship between conductivity and total dissolved solids is not directly linear as it depends on the strength and activity of the dissolved ions [15].

Algal diversity along an elevation gradient. In general, the algal richness increased with rising elevation. This could be due to multiple factors. However,

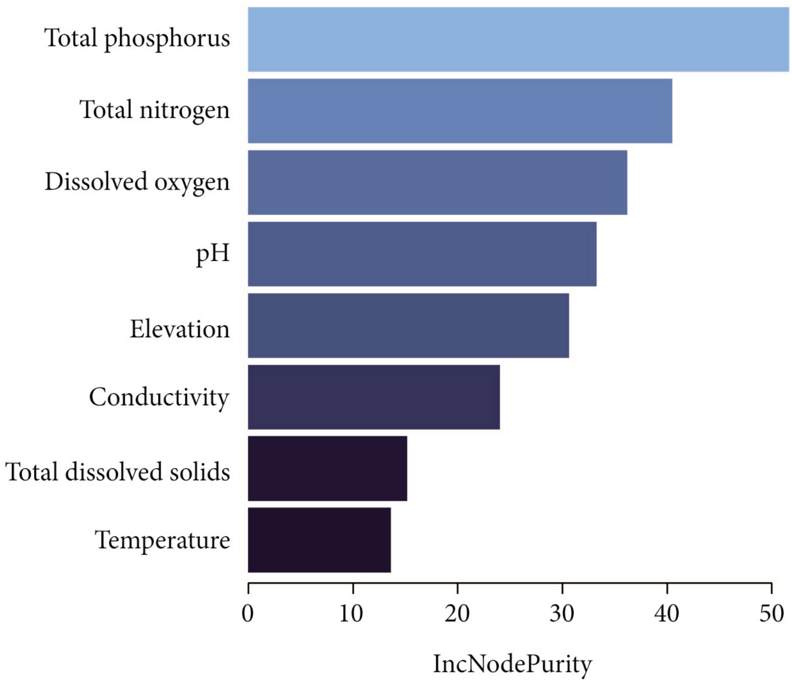

Fig 6. Variable importance plot showing increase in node purity (MeanDecreaseGini) of the random forest model. Variables are arranged in order from highest to lowest predictive capacity

the increase in total nitrogen and phosphorus content along an elevation gradient (see Figure 3) offers a more plausible explanation because both nitrogen and phosphorus have been shown to cause an overgrowth of algae in a relatively short period of time, also known as algal blooming. Our finding is in contrast to several previous studies, which have shown a decrease in algal diversity with increasing elevation. For example, the algal species diversity was relatively high at the lowest and the middle elevation [42]. Similarly, in another study [52], it has been shown that the overall phytoplankton abundance in the Lhasa River decreased with increasing elevation. It has been also reported [50] that the algal biomass in the glacier region decreased as the altitude increased. Studies have suggested that the aquatic vegetation is impacted by altitude only occasionally and usually on a wide geographical or altitudinal scale [26]. It has been found [23] that altitude was the strongest predictor of species richness among all the environmental factors examined. Aquatic ecosystems at high altitudes have extreme environments in which physical stressors associated with ice and snow (winter cover, scouring, and avalanche) and severe climate are limiting factors for the distribution of aquatic plants. However, contrasting finding of algal distribution observed in our study reveals that the algal communities have well fostered in high-altitude lakes and rivers in the Khumbu region, suggesting that the pollution level in high-altitude water bodies is higher than the anticipated scenario. This could be the result of increased deposition of phosphorus and nitrogen in the lakes and rivers likely due to excessive human activities in the Khumbu region.

Does physiochemical properties of the water influence algal diversity? Our findings show that pH, temperature, dissolved oxygen and phosphorus content are the key factors affecting phytoplankton abundance, which is consistent with studies showing that the growth and reproduction of phytoplankton are affected by various environmental factors [45, 51]. The elevation gradient is one of the main gradients, and it reflects the change in all environmental factors with increasing altitude [8]. The physical and chemical properties of water are the two main aspects that directly affect the growth and reproduction of phytoplankton [17]. Similarly, water temperature is one of the most important physical factors influencing the phytoplankton community because it directly controls lake ecosystems while also indirectly influencing physical and chemical parameters of the water [14]. Although solar radiation is higher in higher elevations than in lower elevations, low temperatures become the primary constraint for vegetation growth in higher elevations [47]. Different pH values of water lead to the growth of different algal species. Phosphorus is the easiest and most readily available nutrient for algae at pH 6—7. The pH of water ranged between 6 and 7 in 11 out of 13 sampling sites, indicating that the pH of water in our study sites is suitable for algal growth [10]. High levels of phosphorus in water bodies accelerate the growth of algae and other aquatic plants. Therefore, phosphate levels also indicate the eutrophic condition of a fresh water body and determine its pollution level [49]. Phosphorus content is an important water parameter because it contributes significantly to water quality degradation [19]. It is also known as the key nutrient for algal growth, as well as the cause of eutrophication and toxic algal blooms in lakes and rivers [4, 11].

We acknowledge that, in addition to the factors that were analyzed in the current study, other natural factors such as flash floods, avalanches, landslides, and soil disintegration or soil erosion and man-made factors such as the construction of highways and expressways, eradication of substratum, cutting down of forests, forest fires, farming practices, substantial sedimentation, and open utilization of river water could potentially determine the abundance of phytoplankton in water bodies [29, 37]. As comparisons of algal richness and water properties revealed no significant differences among three types of ecosystems, our judgement of combining them together for statistical analyses is fully justified. Although planktonic algae are usually much more diverse and abundant in lakes compared to running water such as rivers, springs or glacier, we did not find such differences in our study region. Therefore, our findings are not biased by the selection of different ecosystems along an elevation gradient.

Conclusion

Totally 56 species of algae from the water bodies (6 rivers, 3 lakes, 3 springs and 1 glacier) of the Khumbu region belonging to 6 classes (Bacillariophyceae, Zygnematophyceae, Cyanophyceae, Chlorophyceae, Trebouxiophyceae and Euglenophyceae) were found during the period of investigations. The dominance of algal populations from the class Bacillariophyceae (37.5 % of the total number of species) across all sampling sites suggests the potential for eutrophic conditions in water bodies in the Khumbu region. However, while certain diatom species are known as indicators of nutrient-rich environments, their overall dominance does not conclusively determine eutrophication because some species are associated with oligotrophic conditions. A comprehensive evaluation of species composition, along with supporting environmental data such as nutrient levels, oxygen concentrations, and water clarity, is essential to accurately assess the trophic status of these water bodies. An interesting elevation-dependent increase in phosphorus and nitrogen content, as well as total dissolved solids, observed in our study further indicates high pollution levels in high-altitude water bodies. Our study also revealed a significant association between phosphorus content in the water and algal species richness, which is a clear indication that the increased levels of phosphorus in water bodies accelerate algal bloom. Except for the Imja River, which occurs at an elevation of 5007 m, all the water bodies in the Khumbu region below 5000 m have abundant algal species, indicating increased pollution levels. This is a worrying scenario because the Khumbu region is a popular tourist destination and is one of the most visited places in Nepal. Consuming such contaminated water (mostly indirectly because villages and settlements in the Khumbu region obtain their water directly from open water sources) may thus not only pose a serious health risk to tourists, but may also have a long-term impact on the local people and economy. Retrospectively, increased human activity as a result of a thriving tourism industry in the Khumbu region has significantly contributed to increased pollution levels in water bodies. With climate change and increased tourism already increasing the vulnerability of the Khumbu region, our findings highlight the need of constantly monitoring the environment therein.

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DIVERSITY OF PLANKTON ALGAE ALONG AN ELEVATION GRADIENT IN THE FRESHWATER ECOSYSTEMS OF THE KHUMBU REGION, NEPAL

Understanding the diversity and composition of algal communities across different elevation gradients and factors affecting these patterns can provide valuable insights into the ecological processes that shape these ecosystems. In this study, we aimed to investigate the diversity and composition of algae in the Khumbu region of Nepal and assess their relationship with environmental variables along an elevation gradient. We collected algal samples from 13 lakes and rivers ranging in elevation from 2700 m to 5000 m and analyzed the relative influence of various environmental factors on algal richness, including altitude, temperature, nitrogen and phosphorus content, dissolved oxygen concentration, total dissolved solids content, and conductivity. A total of 56 species from six classes were recorded from various sampling sites, with Bacillariophyceae being the most abundant (20 species). Species richness exhibited both increasing and decreasing trends in relation to various physiochemical properties of the water. The region exhibited a general trend of rising algal richness with increasing elevation. This phenomenon can be attributed to the elevation-dependent rise in total dissolved solids, along with higher nitrogen and phosphorus levels, caused by intensified human activity. Of all the variables, phosphorus content had the strongest effect on algal richness, indicating that phosphorus is the most significant predictor of water quality. The increase in phosphorus content and subsequent algal richness in these high-altitude fresh water lakes and rivers points to the deterioration in water quality in the Himalayan reservoirs. With climate change and rising tourism already increasing the vulnerability of the Khumbu region, our findings underline the importance of regularly monitoring the ecosystem therein.

Keywords: *algal diversity, diatoms, the Khumbu region, physiochemical properties of the water, the Sagarmatha National Park, Nepal.*