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Nutrients in sediment regulate benthic algal assemblages in the tropical Tri An reservoir of Vietnam

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Abstract:

Benthic algal assemblages are considered effective indicators of environmental change because they sensitively respond to a variety of environmental conditions. This study aims to investigate the relationship between the benthic algal community and nutrient variables in sediment using Spearman's correlation and linear regression analysis. A total of 27 genera of benthic algae were recorded during the study period where cyanobacteria and diatom were the two dominant groups. Several species belonging to *Achnanthidium, Luticola, Lyngbya, Navicula*, and *Phormidium* sensitively responded to nutrient levels and were used as bioindicators. Multiple stepwise linear regression revealed that ammonium (NH_4^+) and total nitrogen (TN) positively correlated with several indices of the benthic algae assemblage. This preliminary data set demonstrates the use of benthic algae as precise biological indicators for a measure of the ecological integrity of an aquatic ecosystem and contributes to the further study of benthic environments in tropical regions.

Keywords: **benthic diatom, bioindicators, linear regression analysis, nutrient factors, Spearman's correlation analysis.**

Classification number: **5.1**

Introduction

Sediments in lakes and reservoirs are home to a variety of benthic organisms including animals, plants, microorganisms, as well as a sink of pollutants [1]. Benthic algal communities in sediment play a vital role in the biogeochemical cycle of its primary elements and contribute to about 20-25% of global primary production [2, 3]. In addition, benthic algae are primary producers in freshwater ecosystems that consist of lentic (particularly lakes, reservoirs and wetlands) and lotic (including streams and rivers) waters [3]. Because of their taxonomic distinction, abundance, good preservation in sediment, and their rapid response to environmental changes, they have been widely used as environmental indicators in freshwater ecosystems [4].

Among environmental factors, phosphate and nitrate content have been identified as the two most important variables that can account for the distribution of benthic algal assemblages in lentic waters [5-7]. Nitrogen and phosphorus are important for the development of different groups of diatom and cyanobacteria [8]. Benthic diatoms have been used to assess the ecological status of standing waters [9, 10]. Some diatom species such as *Achnanthidium eutrophilu* and *Navicula cryptotenella* are bioindicators of ecological conditions from oligosaprobous to hypereutraphentic, while *A. minutissimum* was reported as tolerant to nutrient loadings [10]. Therefore, a definitive understanding of benthic algal assemblages is essential to assess water quality degradation and nutrient enrichment in inland reservoirs [2, 5, 11].

The Tri An reservoir (TAR), located in the Dinh Quan district, Dong Nai province, Vietnam, is about 70 km northeast of Ho Chi Minh city [12]. It receives water from the Dong Nai and La Nga rivers. The reservoir has had multiple purposes such as a hydroelectric power plant, flood

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*Corresponding author: Email: thanhluupham@gmail.com control, as a domestic and industrial water supply, fisheries, and for the irrigation of agricultural fields [13]. There are a large number of fish cages, as well as a sugar factory, at the inflow of the reservoir by the La Nga river that leads to nutrient enrichment [13, 14]. For this reason, cyanobacterial blooms and their toxins in the TAR have recently been reported [12, 13, 15] and the relationships between several environmental factors and the distribution of cyanobacteria have been investigated [13]. However, little is known about the composition of the benthic algal community and their distribution in the TAR or how they respond to changes in environmental variables. Therefore, the objectives of this study were to describe the benthic algal community's compositions and understand whether nutritional factors in sediment can influence their structure and distribution in the TAR.

Materials and methods

Field sampling and nutrient analyses

Sediment samples were collected monthly from 8 sampling stations from March to May (dry season) and from June to August (rainy season) in 2019 using a stainless-steel hand borer. The samples were stored in plastic boxes kept in an ice box and transferred to the laboratory for analysis. Nutrient concentrations in the sediment were analysed colorimetrically in triplicate with a spectrophotometer (Hach DR/2010) according to the methods described by APHA (2005): nitrate 4500NO_3^- (A); ammonium 4500NH_4^+ (B); total nitrogen Kjeldahl, 4500N (C); phosphate 4500PO_4^{3-} (D); and total phosphorous 4500P (E) [16]. The samples were collected in the TAR at three regions with distinct occupation characteristics: the lower Tri An dam site (DN), the reservoir sites (TA1-TA5), and the upstream sites of Dong Nai (DQ) and La Nga rivers (LN) (Fig. 1).

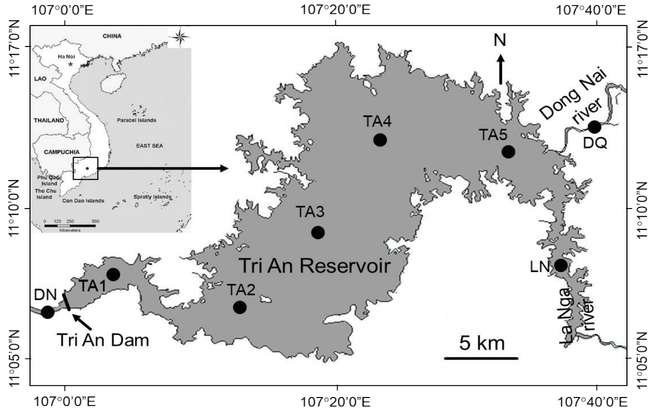

Fig. 1. Map of the Tri An reservoir and the 8 sampling locations.

Diatom sampling and identification

The benthic algae were collected on hard substrates in the field by using a toothbrush to scrape three stones over a surface area of 10 cm^2 [17]. Subsequently, the samples were preserved in plastic bottles and fixed in 4% neutralized formalin and then moved to the laboratory. The subsamples were concentrated to 10 ml and directly used for identification and to count the cyanobacteria. For diatom identification and counting, about 5-10 ml of a subsample was cleaned with concentrated nitric acid and washed with distilled water until it reached a circumneutral pH value. The morphology of the clear samples was observed with a light microscope (Olympus, Tokyo, Japan) at 400× magnification. The cell density was counted using a Sedgewick Rafter counting chamber. A minimum of 400 units were counted in each sample. The valves of the benthic algae were identified to the level of genera by using standard identification guides of several books [18-21].

Data analysis

Statistical calculations were performed using Statgraphic Centurion XV. One-way analysis of variance (ANOVA) was used to test the significance of the differences among the study sites and was based on the nutrient variables and diatom species structure metrics. The analysis was completed using Tukey's Honest Significant Difference (HSD). The correlation between the diatom species' structure metrics and environmental parameters was determined by Spearman's correlation analysis followed by linear regression analysis. All variables were log transformed (log+1) to normalize their distributions before analysis.

The diatom community's structural distribution, the Margalef's species richness index (d), Shannon-Weiner diversity index (H'), Pielou's evenness index (J'), and Simpson's diversity index (D'), which are commonly used in water quality bioassessments, were used to characterize each site [22].

Results and discussion

Nutritional concentration in sedimentary environment

The nutrient content of the sedimentary environment in TAR from the dates March to August in 2019 are illustrated in Fig. 2. During that time, the TN concentration in the rainy season was higher than in dry season and ranged from 0.04-0.19% and 0.04-0.29% in dry and rainy season, respectively. The TN concentration in the reservoir sites (TA1-TA5) were higher than that of the other sites ($p<0.05$). The N-NH_4^+ concentration was not much different in the dry and rainy season, which ranged from 3.6-29.6 mg/100 g and 1.4-22.7 mg/100 g in dry and rainy season, respectively. The N-NO_3^-

content varied from 1.4-5.4 mg/100 g and 0.1-3.2 mg/100 g in the dry and rainy season, respectively. The N-NO_3^- content in the dry season was higher than that of the rainy season, but it was not significantly different among reservoir sites ($p<0.05$). The TP value varied from 0.011-0.11% and from 0.01-0.07% in dry and rainy season, respectively. The TP content in the upstream sites (TA5, LN, DQ) were higher than that of the other sites during both seasons ($p<0.05$). The P-PO_4^{3-} content varied from 0.004-0.065% and 0.004-0.068% in the dry and rainy season, respectively. The concentration of P-PO_4^{3-} had almost the same trend as TP during both seasons.

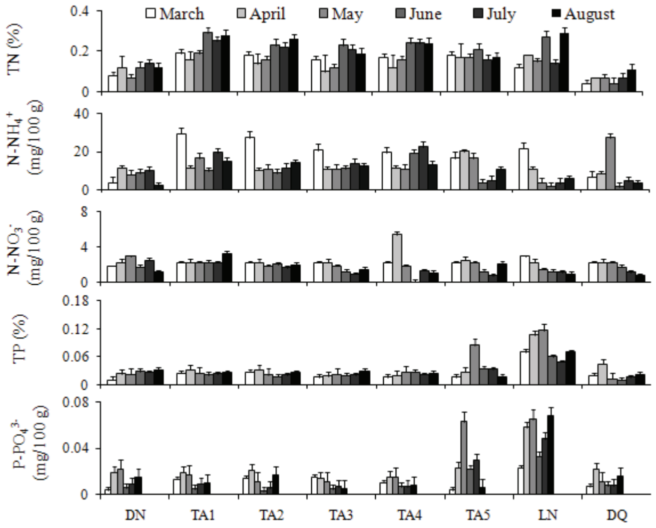

Fig. 2. The content of nutrients in the sedimentary environment.

Sediment in reservoirs, where nitrogen and phosphorus compounds are stored, can operate as a source of nutrient enrichment that could lead to eutrophication [23]. During the survey period, the main reservoir area (reservoir bed) had a higher nutrient content than the rest, particularly in TN and NH_4^+ concentrations. This may affect the sedimentation process and create a high potential for nutrient accumulation. In addition, the high nutrient content of TP and P-PO_4^{3-} at upstream sites such as TA5 and LN, especially in both seasons, may be due to the excessive food and excretion that is dumped directly into the environment from nearby cage fish farming. Many studies have already addressed the negative impact of fish cage culture on benthic organism communities of rivers and lakes. The sediment of organic matter released from the farming process could discharge between 67 to 89% of its nitrogen and phosphorus into the surrounding environment. Over time, the excess accumulation of phosphorus and nitrogen compounds can easily cause benthic algal communities, as well as phytoplankton, to grow in abundance and biomass that can lead to the eutrophication of cyanobacteria and diatom adapted to high concentrations [24, 25].

Benthic algal community composition and abundance

The benthic algal community structure from March to August in 2019 is illustrated in Fig. 3. A total of 27 genera of benthic algae were identified during the study period of which 13 genera belonged to cyanobacteria and 14 genera belonged to diatom. The benthic algal communities at the reservoir sites were more diverse than at the lower and upper sites. *Navicula, Nitzschia, Luticola, Thalassionema, Phormidium, Planktothrix, Plectonema,* and *Lyngbya* were the most common genera. While most of the genera of cyanobacteria were observed to be of the filamentous form, the diatoms were of the genera with order centrales and pennales.

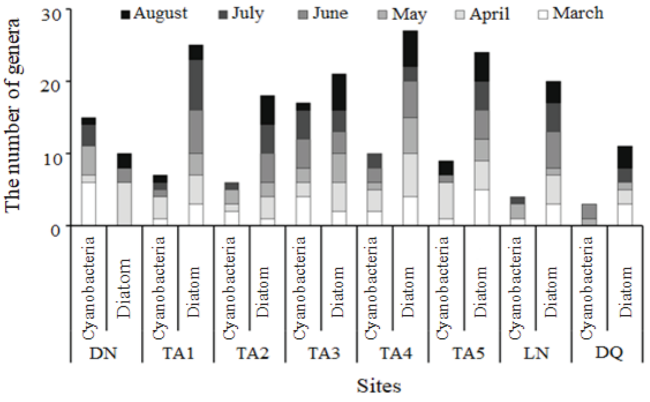

Fig. 3. The number of genera at each survey site.

The abundance ranged from 1000-9600 (cells/cm^2) and from 800-15600 (cells/cm^2) in dry and rainy season, respectively (Fig. 4). Generally, the cell density was higher in the rainy season than in the dry season and the cell density at the reservoir sites were higher than those in the downstream and upstream sites during both seasons. The abundance in August was significantly higher than the other months (10000-15000 cells/cm^2) owing to the dramatically increasing cell density of several diatoms, namely *Luticola, Navicula,* and *Nitzschia.*

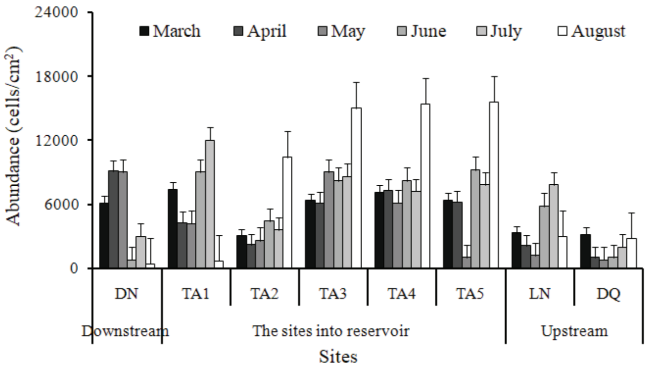

Fig. 4. The abundance of benthic algal communities in Tri An reservoir.

For the cyanobacterial communities in the TAR, the species of *Phormidium* spp. and *Plectonema* spp. were the two most abundant at 18 and 17%, respectively (Fig. 5). In addition, the other species belonging to the *Planktolyngbya*, *Planktothrix*, *Lyngbya*, and *Oscillatoria* genera also commonly found while the species of *Anabaena, Pseudanabaena,* and *Spirulina* were not common. As for the diatom, the species of *Navicula* spp. and *Nitzschia* spp. were dominant and accounted for the largest percentage of the total density, 20 and 13%, respectively, while the other species belonging to the genera of *Synedra*, *Gyrosigma*, *Achnanthidium*, and *Amphora* were rarely recorded (Fig. 5).

Because this study was based on the distribution of benthic microalgae assemblages in the surface sediments of the TAR, the distribution of these dominant species could reflect nutrient levels in the environment. Particularly, diatoms are used as main food sources for many consumers in the food chain because they are rich in protein and fatty acids [26]. At some sites with high nutrient concentrations such as TA1-TA5, some genera such as *Navicula*, *Nitzschia*, *Thalassionema*, *Phormidium*, and *Lyngbya* had a significantly higher percentage of total density (over 70%) while the other stations with low nutrient concentrations, such as the upstream and downstream (DN, DQ, and LN), many species of *Plectonema*, *Achnanthidium*, and *Gomphone* existed. Our results are consistent with previous observations that the species belonging to the *Gomphoneis* and *Achnanthidium* genera are related to low TP content in the sediment whereas the species belonging to the *Luticola* and *Navicula* genera indicate a high TP and nitrogen content [27, 28].

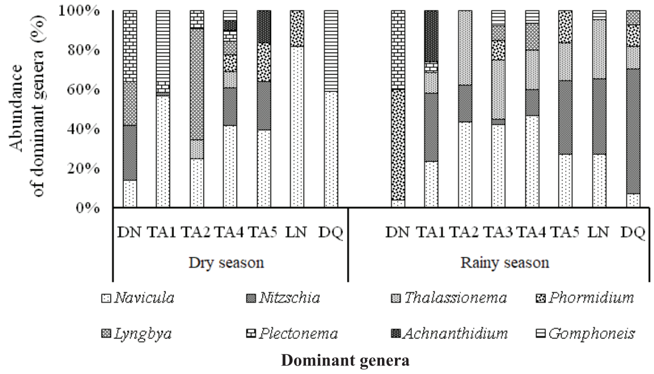

Fig. 5. The distribution of the dominant genera.

Benthic algal assemblage species diversity metrics

Diversity is a composite parameter that integrates the number of species and the distribution of biomass among these species [29]. The temporal and spatial variations of benthic algal metrics are shown in Fig. 6. From this figure, Margalef's diversity (d) ranged from 0.14-0.92 and 0.13-0.75 in the dry and rainy season, respectively (see Fig. 6A). The evenness (J') was also high as all of values surpassed 0.7 with a minimum value of 0.72 (Fig. 6B). The Shannon-Weiner diversity index (H') ranged from 0.88-3.04 and from 0.88-2.79 in the dry and rainy season, respectively (Fig. 6C). Overall, the H' index was relatively low, which indicated a low biodiversity of benthic algae communities and ongoing pollution in the environment while the classification systems of SFT (1997) [30] revealed that the quality of water was indeed polluted. Simpson's diversity index (D') ranged from 0.38-0.87 and from 0.42-0.84 in dry and rainy season, respectively (Fig. 6D). The high value of the dominance index (D) reflected the dominance of the two diatom *Luticola* sp. and *Navicula* sp. at all reservoir sites.

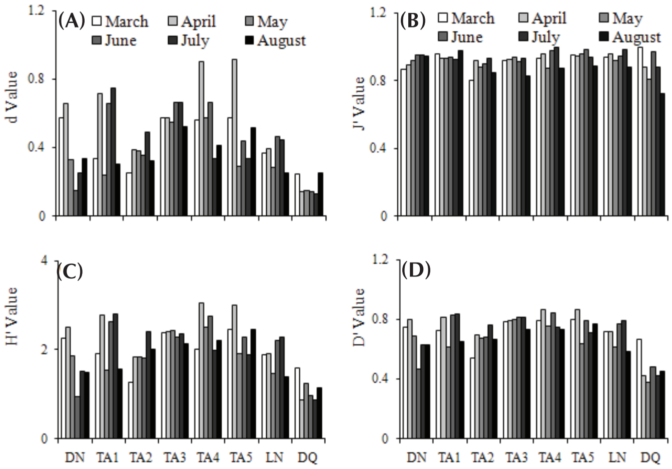

Fig. 6. Benthic algal metrics (d, J', H', and D') in Tri An reservoir.

Relationship between benthic algae distribution and nutrients in sedimentary environment

The correlation between the benthic algae distribution and the nutrient-related variables in sediment are depicted in Fig. 7. The results of Spearman's correlation analysis showed that NH_4^+ and TN correlated with benthic algal metrics in which NH_4^+ was associated with the H' index ($p=0.0352$) while TN was related to a few indices such as H', D, and d ($p=0.0241$, $p=0.0187$, and $p=0.0379$, respectively). The results from the linear regression analysis indicated that there was a strong correlation between the TN value and the H', D, and d indices with $R^2=26.8702\%$ and $p=0.0002$, $R^2=25.3578\%$ and $p=0.0003$, and $R^2=31.8274\%$ and $p<0.001$, respectively. There was little correlation found between the H' index and NH_4^+ concentration with $R^2=15.3685\%$ and $p=0.0093$.

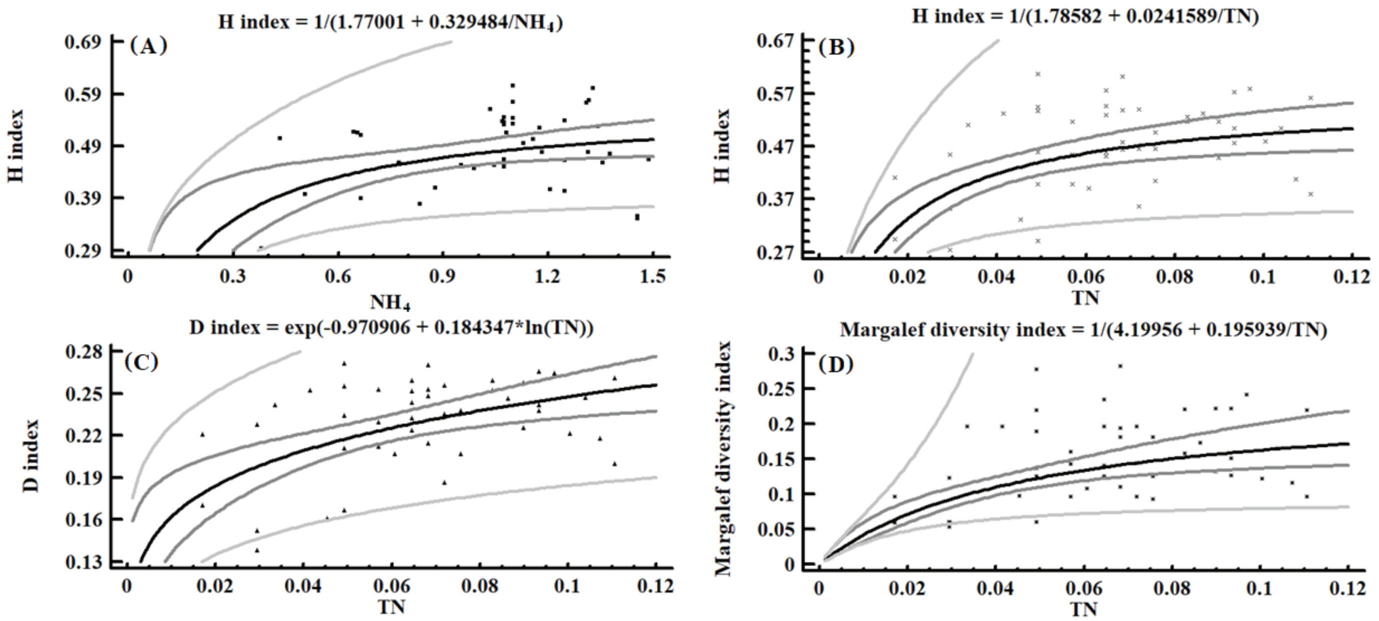

Fig. 7. Model of linear regression analysis: (A) The correlation between NH_4^+ concentration and H′ index; (B), (C), and (D) are the correlations between TN concentration and the H′, D, and d indices, respectively.

Nutrients are a key factor influencing diatom activity and growth rate as well as any variations in nutrients can change the community structure of the diatoms [31]. In this study, NH_4^+ and TN are considered associated with the diversity of benthic algae assemblage. That means, if NH_4^+ and TN values grow by accretion, the diversity indices will increase and vice versa. Previous studies have demonstrated that the benthic microalgae community are strongly influenced by nutrient concentrations [26]. According to Saros, et al. (2011) [32], increases in the two diatom taxa *Asterionella formosa* and *Fragilaria crotonensis* were indicators of N enrichment in an N-limited reservoir and these two species responded positively to moderate N enrichment. The results of Hillebrand, et al. (2000) [29] showed that the nutrient and spring grazing experiment influenced the biomass and species composition of benthic microalgae in which species richness was significantly decreased by grazer presence (ANOVA, $p=0.013$) and increased with nitrogen supply. It is well known that sediments are an important source of nutrients to freshwater ecosystems. For example, nutrient cycling at the sediment-water interface is affected by both dissolved oxygen and by nutrient concentration gradients that result from algal decay [33]. The release of nutrients from the bottom sediment can have a significant impact on water quality and can result in continued eutrophication by excessive input of phosphorus and nitrogen that has become one of the most common damages to surface waters [34]. Thus, our results indicate that the nutrients in sediment may restructure the distribution of several diatom and cyanobacteria in the TAR.

Conclusions

The benthic algae assemblage in the TAR was investigated in this study. The presence of species such as *Navicula, Nitzschia,* and *Phormidium* represented eutrophic environments in reservoir sites whereas *Plectonema* and *Achnanthidium* were indicators of nutrient-poor environments in the upstream and downstream sites. The TN and NH_4^+ concentration were the two main environmental variables that regulated the benthic algae assemblage in the TAR. The present work has provided a first framework on the distribution and diversity of benthic algae in response to the nutrients in this area. Further studies focusing on the impact of benthic algae in response to other components, as well as using them as bioindicators in aquatic ecosystems, should be implemented.

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COMPETING INTERESTS

The authors declare that there is no conflict of interest regarding the publication of this article.

REFERENCES

[1] M. Wang, W. Cheng, T. Wan, T. Meng, Z.J. Hao, L.D. Shi (2018), “Effects of water ecological environment on benthic algae in a water source reservoir”, *IOP Conference Series: Earth and Environmental Science*, **191(1)**, DOI: 10.1088/1755-1315/191/1/012056.

[2] J. Yang, H. Jiang, W. Liu, B. Wang (2018), “Benthic algal

community structures and their response to geographic distance and environmental variables in the Qinghai-Tibetan lakes with different salinity", *Frontiers in Microbiology*, **9**, DOI: 10.3389/fmicb.2018.00578.

[3] J. Wang, Q. Liu, X. Zhao, A.G. Borthwick, Y. Liu, Q. Chen, J. Ni (2019), "Molecular biogeography of planktonic and benthic diatoms in the Yangtze River", *Microbiome*, **7(153)**, pp.1-15.

[4] S.B. Faustino, L. Fontana, E.C.R. Bartozek, C.E.D.M. Bicudo, D.D.C. Bicudo (2016), "Composition and distribution of diatom assemblages from core and surface sediments of a water supply reservoir in Southeastern Brazil", *Biota Neotropica*, **16(2)**, DOI: 10.1590/1676-0611-BN-2015-0129.

[5] A.I. Negro, C.D. Hoyos (2005), "Relationships between diatoms and the environment in Spanish reservoirs", *Limnetica*, **24(1-2)**, pp.133-144.

[6] S. McGowan, H.V. Gunn, E.J. Whiteford, N.J. Anderson, V.J. Jones, A.C. Law (2018), "Functional attributes of epilithic diatoms for palaeoenvironmental interpretations in South-West Greenland lakes", *Journal of Paleolimnology*, **60(2)**, pp.273-298.

[7] R.V.H. Hansika, S.K. Yatigammana (2019), "Distribution of diatom assemblages in the surface sediments in Sri Lankan reservoirs located in the main climatic regions and potential of using them as environmental predictors", *Tropical Ecology*, **60(3)**, pp.415-425.

[8] B. Sabour, M. Loudiki, V. Vasconcelos (2009), "Growth responses of *Microcystis ichthyoblabe* Kützing and *Anabaena aphanizomenoides* Forti (cyanobacteria) under different nitrogen and phosphorus conditions", *Chemistry and Ecology*, **25(5)**, pp.337-344.

[9] M.H. Novais, S. Blanco, C. Delgado, M. Morais, L. Hoffmann, L. Ector (2012), "Ecological assessment of Portuguese reservoirs based on littoral epilithic diatoms", *Hydrobiologia*, **695(1)**, pp.265-279.

[10] D. Fidlerová, D. Hlúbiková (2016), "Relationships between benthic diatom assemblages structure and selected environmental parameters in Slovak water reservoirs (Slovakia, Europe)", *Knowledge and Management of Aquatic Ecosystems*, **417**, DOI: 10.1051/kmae/2016014.

[11] Y. Pan, et al. (2016), "Variation of benthic algal assemblages among habitats in subalpine Karstic lakes and implications for bioassessment", *Hydrobiologia*, **777(1)**, pp.183-196.

[12] H.Q. Nguyen, N.T. Ha, T.L. Pham (2020), "Inland harmful cyanobacterial bloom prediction in the eutrophic Tri An reservoir using satellite band ratio and machine learning approaches", *Environmental Science and Pollution Research*, **27**, pp.9135-9151.

[13] T.S. Dao, J. Nimptsch, C. Wiegand (2016), "Dynamics of cyanobacteria and cyanobacterial toxins and their correlation with environmental parameters in Tri An reservoir, Vietnam", *Journal of Water and Health*, **14(4)**, pp.699-712.

[14] V.H. Nguyen, V.T. Dinh, D.R. Nguyen, D.L. Le (2013), "Study on the impact of the Tri An reservoir on its downstream groundwater level regime", *Journal of Sciences of the Earth*, **34(4)**, pp.465-476.

[15] T-L. Pham, T.H.Y. Tran, N.S. Hoang, X.Q. Ngo, T.T. Tran (2020), "Co-occurrence of microcystin-and geosmin-producing cyanobacteria in the Tri An reservoir, a drinking water supply in Vietnam", *Fundamental and Applied Limnology*, **193(4)**, DOI: 10.1127/fal/2020/1296.

[16] A.D. Eaton, L.S. Clesceri, E.W. Rice, A.E. Greenberg, M.A.H. Franson (2005), *Standard Methods for the Examination of Water and Wastewater (21st edition)*, American Public Health Association, 1325pp.

[17] J.D. Wehr, G.S. Robert, J.K. Patrick (2003), *Freshwater Algae of North America: Ecology and classification*, Elsevier, 935pp.

[18] U. Rumrich, H. Lange-Bertalot, M. Rumrich (2000), *Diatoms of the Andes: From Venezuela to Patagonia/Tierra del Fuego*, Gustav Fischer, 671pp.

[19] D. Metzeltin, H. Lange-Bertalot (2002), *Diatoms from the "Island Continent" Madagascar*, A.R.G. Gantner Verlag K.G. Koenigstein, 286pp.

[20] D. Metzeltin, H. Lange-Bertalot (2007), *Tropical Diatoms of the South America II*, A.R.G. Gantner Verlag K.G. Koenigstein, 877pp.

[21] J.C. Taylor, J. Prygiel, A. Vosloo, A. Pieter, L. van Rensburg (2007), "Can diatom-based pollution indices be used for biomonitoring in South Africa? a case study of the crocodile west and Marico water management area", *Hydrobiologia*, **592(1)**, pp.455-464.

[22] A. Agrawal, K. Gopal (2013), "Biomonitoring of water and wastewater", *Springer Science Business Media*, DOI: 10.1007/978-81-322-0864-8.

[23] R.G. Wetzel (2001), *Limnology: Lake and River Ecosystems*, Gulf Professional Publishing, 1006pp.

[24] L. Nordvarg, T. Johansson (2002), "The effects of fish farm effluents on the water quality in the Åland archipelago, Baltic Sea", *Aquacultural Engineering*, **25(4)**, pp.253-279.

[25] L. Mwebaza-Ndawula, V. Kiggundu, G. Magezi, J. Naluwayiro, W. Gandhi-Pabire, H. Ocaya (2013), "Effects of cage fish culture on water quality and selected biological communities in northern Lake Victoria, Uganda", *Uganda Journal of Agricultural Sciences*, **14(2)**, pp.61-75.

[26] L.D.N. Silvano Filho, E.J. de França, M.M. de Júnior, A.D.N. Moura (2019), "Interactions between benthic microalgae, nutrients and benthic macroinvertebrates in reservoirs from the semi-arid Neotropical region", *Fundamental and Applied Limnology/Archiv für Hydrobiologie*, **192(3)**, pp.237-254.

[27] M.G. Potapova, D.F. Charles, K.C. Ponader, D.M. Winter (2004), "Quantifying species indicator values for trophic diatom indices: a comparison of approaches", *Hydrobiologia*, **517(1-3)**, pp.25-41.

[28] S.A. Wood, R.G. Young (2012), *Benthic Cyanobacteria and Toxin Production in the Manawatu-Wanganui Region*, Prepared for Horizons Regional Council, Cawthron Report No.1959, 36pp.

[29] H. Hillebrand, B. Worm, H.K. Lotze (2000), "Marine microbenthic community structure regulated by nitrogen loading and grazing pressure", *Marine Ecology Progress Series*, **204**, pp.27-38.

[30] SFT (1997), "Classification of environmental quality in fiords and coastal waters (in Norwegian)", SFT report **97(03)**, *Norwegian Pollution Control Authority, Oslo*.

[31] R. Shen, H. Ren, P. Yu, Q. You, W. Pang, Q. Wang (2018), "Benthic diatoms of the Ying river (Huaihe river basin, China) and their application in water trophic status assessment", *Water*, **10(8)**, DOI: 10.3390/w10081013.

[32] J.E. Saros, D.W. Clow, T. Blett, A.P. Wolfe (2011), "Critical nitrogen deposition loads in high-elevation lakes of the western US inferred from paleolimnological records", *Water, Air & Soil Pollution*, **216(1-4)**, pp.193-202.

[33] M. Zhu, G. Zhu, L. Zhao, X. Yao, Y. Zhang, G. Gao, B. Qin (2013), "Influence of algal bloom degradation on nutrient release at the sediment-water interface in lake Taihu, China", *Environmental Science and Pollution Research*, **20(3)**, pp.1803-1811.

[34] W.H. Nowlin, J.L. Evarts, M.J. Vanni (2005), "Release rates and potential fates of nitrogen and phosphorus from sediments in a eutrophic reservoir", *Freshwater Biology*, **50(2)**, pp.301-322.