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Diversity and Structure of the phytoplankton of Lake Ahémé, a part of the Ramsar 1017 (Bénin)

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Abstract
This study aims to assess the phytoplankton community of Lake Ahémé, subject to anthropogenic pressures. For this purpose, eight sites were sampled from September 2014 to October 2016. A total of 274 taxa were recorded and were mainly dominated by Bacillariophyceae (54%), followed by Chlorophyceae (12.77%), Cyanophyceae (10.22%), Euglenophyceae (9.49%), Conjugatophyceae (7.66%), Trebouxiophyceae (2.18%), Dinophyceae (1.82%), Xanthophyceae (0.73%), Ulvophyceae (0.73%), Chrysophyceae (0.36%). Overall, diversity was high during the long wet season and low during the short dry season, with imbalanced distribution of phytoplankton taxa over the study period. Biodiversity indices varied throughout seasons : between 4 and 16 for Margalef index, between 1.2 and 3 bits for Shannon index, between 0.46 and 0.9 for Simpson index, between 0.22 and 0.72 for Berger-Parker index and between 0.04 and 0.15 for evenness index. The particular abundance of diatoms observed is due to strong mineralization of the lake at the time of the study. These observations suggest that phytoplankton diversity and structure can be used in conjunction with physical and chemical indicators to assess water quality.

1. Introduction
Wetlands are important habitats for biological resources and provide benefits for domestic activities such as consumption and recreation, agriculture, pastoralism, and economic activities [1]. Due to the current population explosion, aquatic ecosystems are increasingly threatened by human activities (agriculture, fisheries, industrial activities, tourism, etc.), [2]. These problems affect aquatic communities and thus induce changes in the environment.

Due to its hydrographic network, Benin has significant water resources, including Lake Ahémé, one of the largest surface water resources in Benin. It is a tropical coastal ecosystem characterized by important lagoon-marine exchanges. It is classified as a site of international importance and is part of the Ramsar 1017 list [3]. According to the Beninese Environment Agency (ABE), Lake Ahémé is the most intensively exploited water resource due to the large number of residents who practice several activities in the area (industries, agriculture, fishing, etc.). Lake Ahémé has extraordinary assets such as its richness in fisheries resources and ecosystem services (ecotourism, trade) [4]. Unfortunately, this ecosystem faces problems of degradation, such as eutrophication, which disrupts the environment, limiting the services it can provide and leading to the loss of biodiversity [5] [6] [7].

Studies carried out by [5] [8] [9] on the physico-chemical quality of Lake Ahémé have shown the impact of anthropogenic activities (eutrophication) on this water body. However, no study so far has measured the impact of these activities on the phytoplankton communities responsible for primary production in aquatic ecosystems. Biodiversity is an integrated component of the environment that can be used to understand changes in the environment [10]. Phytoplankton, an autotrophic microorganism, is the key element that contributes to the structuring of food webs in water bodies [11]. It is used as a biological indicator of aquatic ecosystems and its density is likely to vary according to the environmental conditions in the middle. Thus, special attention must be given to the phytoplankton communities of Lake Ahémé to understand the mechanisms that drive their functioning so that they can be better used as indicators of water quality in this ecosystem.
The aim is to study the composition and spatio-temporal variations of phytoplankton diversity in Lake Ahémé. More specifically, the limnological variables that affect phytoplankton diversity on the one hand, and the biodiversity indices on the other, were presented.

2. Material and Methods

2.1. Study area

Lake Ahémé in south-west Benin is an integral part of the Mono-Ahémé-Couffo fluvial-lagunar complex and is included in the Rasmar 1017 Convention, making it a site of community importance. Its surface area varies between 78 km^2 in low-water periods and 100 km^2 in high-water periods [8]. Mangroves can be found there, extending along the western rivers (Mono and Sazué) and the edges of the coastal lagoons (Djègbadji, Togbin, and Djondji). Lake Ahémé communicates with the Atlantic Ocean through the outlet called "Bouche du Roy" and is influenced by the brackish and marine waters of the coastal lagoon which it communicates through the Aho channel. It is also influenced by the floodwaters of the Mono River, which can flow up to the Aho Canal, and by the freshwater of the Couffo River in the north [8] [12] [4]. The main activity of the lake is traditional fishing, which employs more than 68% of the riparian population [13]. Some use it for market gardening and washing and others sometimes use it in households. It constitutes an important ecosystem because of the wide variety of plant and animal species present. This nature reserve is an asset for the promotion of biodiversity through challenges related to environmental issues. The average water temperature of the lake is 27.2°C.

2.2. Sampling of environmental variables

Samples were collected during field campaigns between September 2014 and September 2016 covering four seasons: the long dry season (LDS) from mid-November to mid-March, the long wet season (LWS) from mid-March to mid-July, the short dry season (SDS) from mid-July to mid-September and the short wet season (SWS) from mid-September to mid-November. Samples were collected at approximately two-month intervals by adjusting the precise timing of sampling depending on the beginning of each of these seasons [14]. Sampling was repeated in the same localities during the same seasons over the years. Two sampling areas were selected and is composed by Eight sampling sites (Figure 1) : the first is called "river site" near the mouth of the Couffo River and Toho Lake (Site 5, Site 6, Site 7, Site 8) and the second is called "marine site" near the coastal area connected to the Atlantic Ocean (Site 1, Site 2, Site 3, Site 4). The so-called marine sites are characterized by activities such as agriculture and livestock farming; household and industrial waste (liquid waste from the Possotomè thermal water station) is dumped into the river. The so-called river sites are characterized by the effluents of household and agricultural waste and some are outlets for the sale of petroleum products.

Measurements of water temperature (T), pH, and dissolved oxygen (DO), total dissolved solids (TDS), electrical conductivity, and salinity were made *in situ* using a multi-parameter (HANNA HI 9829) and transparency was measured using the Secchi disk method. Turbidity was measured using a turbidimeter Eutech (TN-100). Water samples collected in 1.5 L bottles were transported to the laboratory in a cold room. Nutrients such as orthophosphate ($PO4^{3-}$), nitrite ($N\text{-}NO_2$), nitrate ($N\text{-}NO_3$), ammonium ($N\text{-}NH_4^+$) were measured by spectrophotometry (spectrophotometer HACH Dr 6000) according to the conventional method described by [15].

2.3. Phytoplankton sampling

The phytoplankton sampling was carried out using plankton nets with mesh size 20 µ. Samples were taken at the water surface by dragging the net 10m horizontally against the current (at low speed).

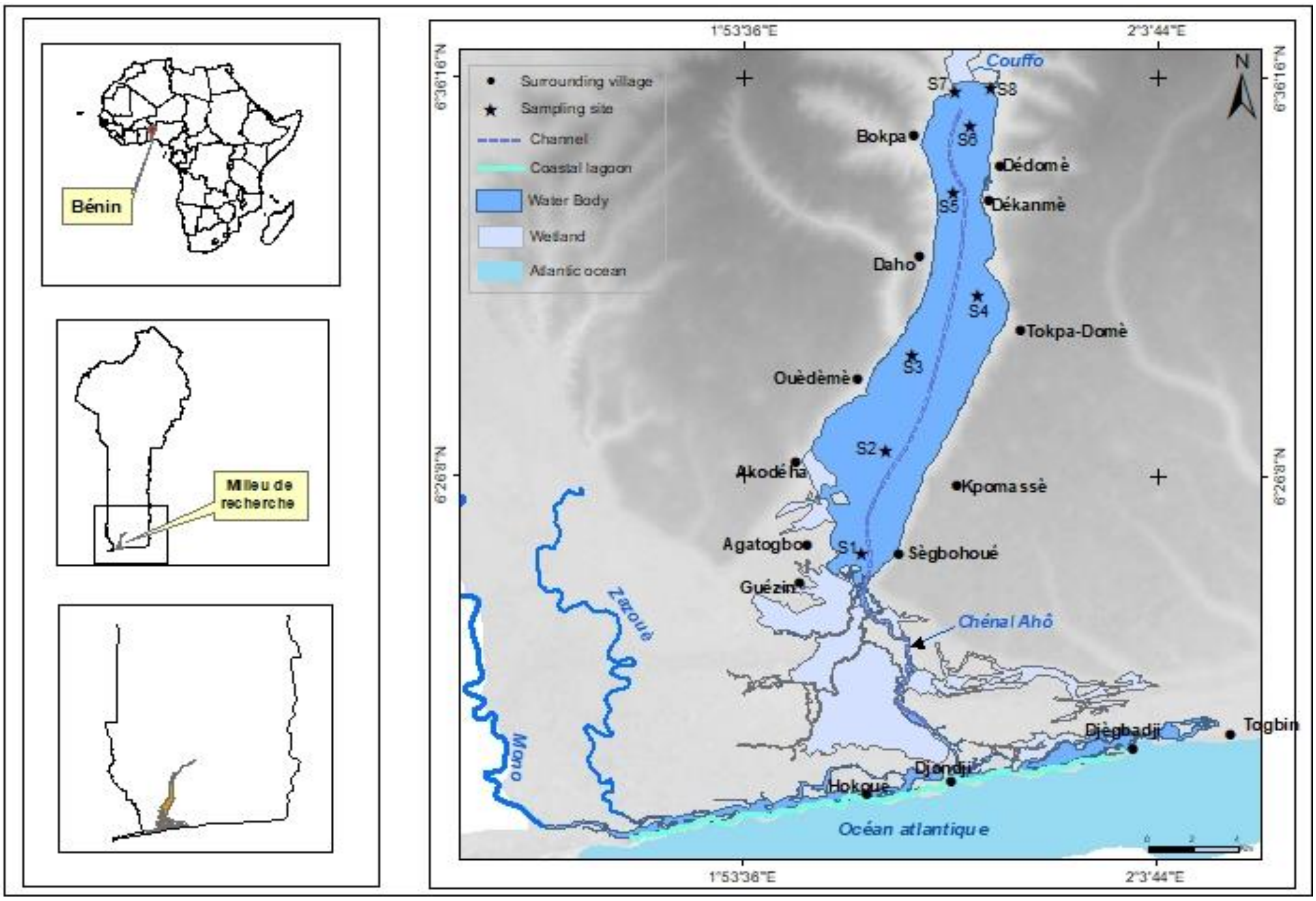


Figure 1: Location of the Lake Ahémé with the sampling sites.

These samples were then collected in labeled 250 ml plastic containers and were immediately fixed with an alkaline solution of Lugol and preserved with 5% formaldehyde. The sample is left for 48 hours to allow the phytoplankton to settle to the bottom of the bottle. The supernatant is then removed by pipetting off [16]. 0.1 ml of the concentrated sample were taken and placed in a Bürker cell for microscopic observation using an optical microscope (Olympus CX30 at a magnification of 400x) [14]. Identifications were made at the lowest taxonomic level possible as, according to the identification keys of [17] [18] [19] [20] [21].

2.4. Data analysis

Diversity indices are used in ecology to assess environmental quality and the effect of disturbances on biological communities. The data obtained were averaged and Excel software was used to set out the curves of relative abundances and environmental variables. The PAST software (Version 3.14) was used to perform the diversity indices.

To assess the structure of the phytoplankton community, five diversity indices were evaluated. The species richness (Margalef index), species diversity (Shannon-Wiener (H')), dominance (Simpson (1-D) and Berger and Parker), evenness (Pielou index (J)) of each station were evaluated using:

Margalef index: $D = \frac{S-1}{\ln(N)}$; S= species number in the sample and N the total number of sampled species.

Shannon-Wiener index: $H' = -\sum(p_i \times \log_2 p_i)$; pi = number of individuals of the taxon/total number of individuals of the sample and log2 = 2 base logarithms

Simpson index: $D = \frac{S-1}{\ln(N)}$; ni = number of species in the sample and N the total number of individuals.

Equitability index: $E = H' / \log_2 S$; H′ is the Shannon-Wiener diversity index and S the total number of taxa.

Berger-Parker index: $d = N_{max} / N_T$; the maximal proportion of a species in the sample and NT = total number of sampled species.

3. Results and discussion

The physico-chemical parameters varied spatially and temporally over the study period. The highest depth value (1.91 m) was obtained during the long-wet season (LWS) and the lowest depth value (1.05 m) was obtained during the short wet season (SWS) at sites S4 and S1 respectively with mean values of 2.70 m and 1.17 m each (Figure 2).

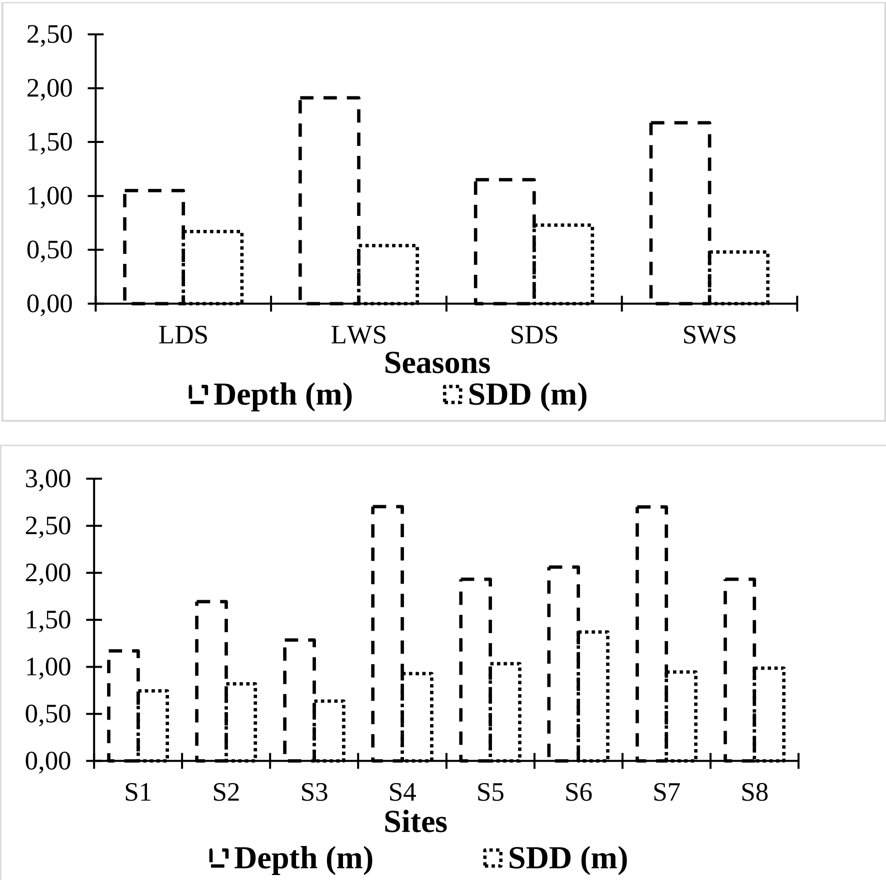
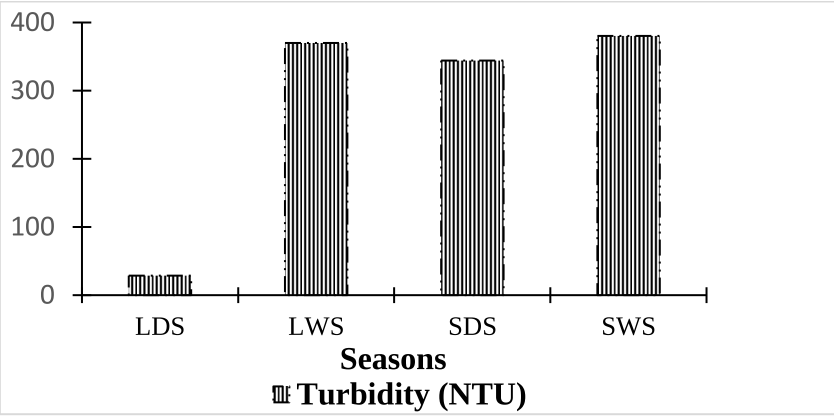


Figure 2: Seasonal and spatial variations of depth and SDD in Lake Ahémé. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8.

Transparency is higher (0.73 m) during the short dry season (SDS) and lower during the short wet season (SWS) at sites S6 and S3 respectively with mean values of 1.37 m and 0.64 m each (Figure 2). The low transparency values obtained in the wet season may be due to a high content of suspended solids from runoff to the lake during rainfall. The higher transparency, as obtained in the dry season, will allow deeper light penetration, which could be favorable to the development of some phytoplankton species. Turbidity (Figure 3) is high (380.53 NTU) in the short wet season (SWS) and low (28.65 NTU) in the long dry season (LDS). At the site level, the highest turbidity values were recorded at S5 (326.09 NTU) and the lowest at S7 (24.14 NTU). Turbidity was higher during the short wet season, which confirms the effect of precipitation on the physical aspect of the aquatic ecosystem of Lake Ahémé. These results are in agreement with those of [22], which state that transparency values vary inversely with those of turbidity.



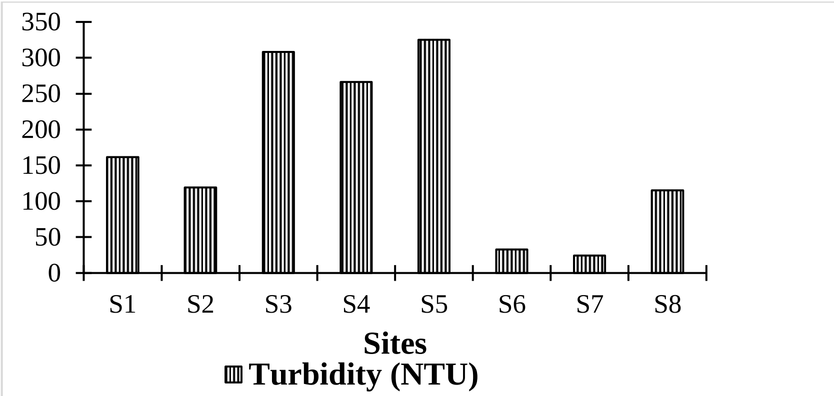


Figure 3: Seasonal and spatial variations of Turbidity in Lake Ahémé. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

The temperature oscillated between 27.36°C and 29.83°C during the short dry season (SDS) and short wet season (SWS) at sites S4 and S3 respectively, with mean values of 29.56°C and 30.47°C at each site (Figure 4). These variations in temperature are probably due to the specific characteristics of the aquatic environment of Lake Ahémé (ranging between 26°C-33°C) [13]. The values recorded in this study for temperature are similar to those obtained by [23], who stated that temperatures between 24 and 35°C are favorable for good growth of fish species commonly found in Benin. The pH was higher (7.47) in the long dry season (LDS) and lower (6.85) in the short wet season (SWS) precisely at sites S1 (8.26) and S8 (7.37) (Figure 5). These pH values are consistent with those obtained by [24] on the same lake. The pH basicity could be related to the salinity of these sites which is probably the result of marine water intrusion. However, this result is within the acceptable range of 6.5 - 8.5 recommended by [25] and is favorable for aquaculture.

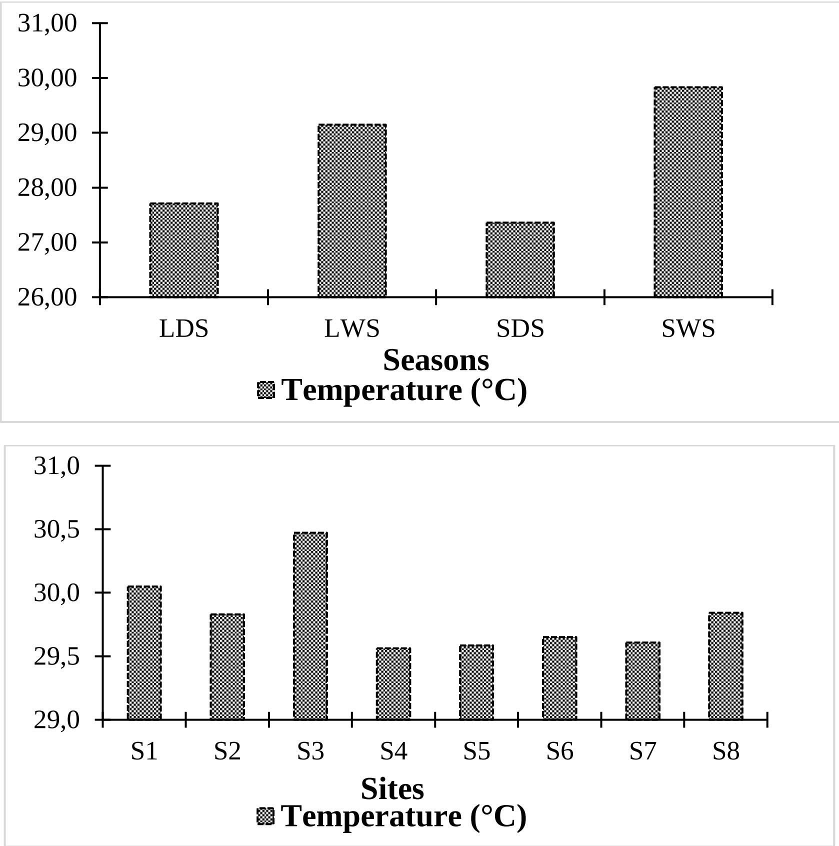


Figure 4: Seasonal and spatial variations of Temperature in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

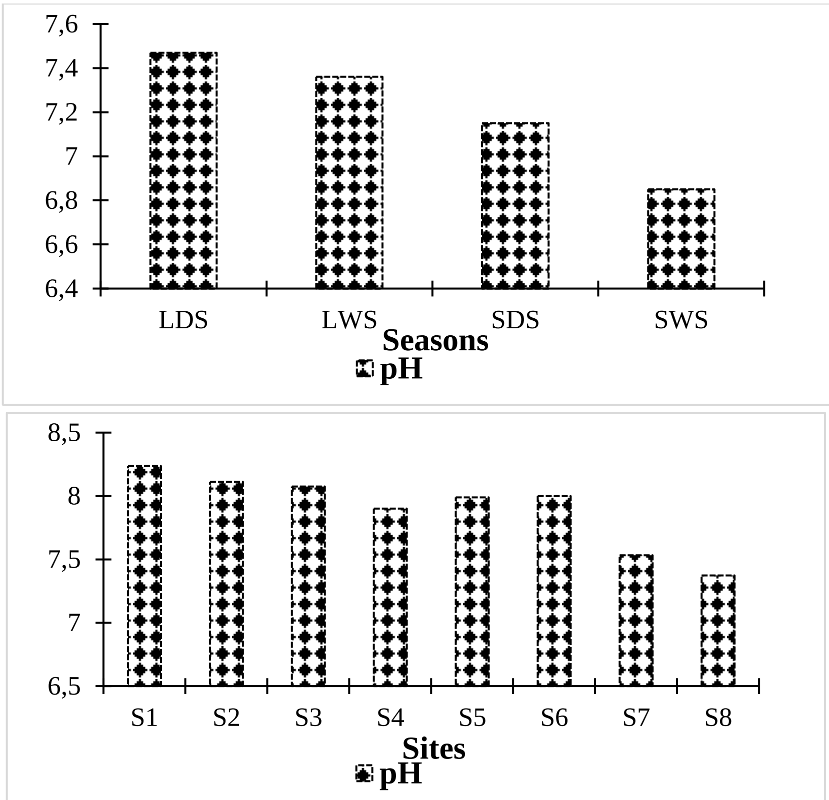
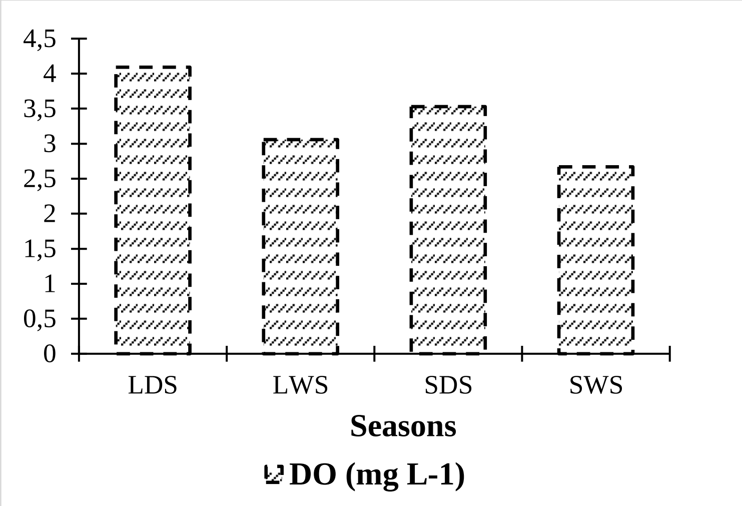


Figure 5: Seasonal and spatial variations of pH in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

As for dissolved oxygen, it varies between 2.67 mg L^{-1} and 4.09 mg L^{-1} respectively during the short wet season (SWS) and the long dry season (LDS), particularly at sites S5 (2.23 mg L^{-1}) and S1 (4.88 mg L^{-1}) (Figure 6). These values are slightly higher than those recorded by [23], which vary between 1.0 mg L-1 and 4.1 mg L^{-1}. Although, the low dissolved oxygen values during the short wet season (SWS), inversely proportional to turbidity, could be related to the oxidation process of organic matter.
TDS, conductivity and salinity followed the same trend of variation (Figure 7, Figure 8 and Figure 9, respectively). Salinity, conductivity and TDS revealed their highest values in the long dry season (LDS), respectively 18.53 PSU, 29.43 g L^{-1} and 15.00 g L^{-1} and their lowest values in the short wet season (SWS), respectively 0.19 PSU, 0.46 g L^{-1} and 0.46 g L^{-1}.



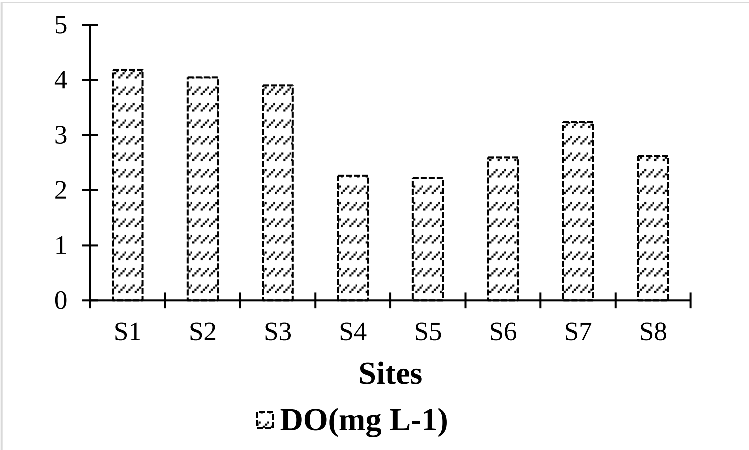
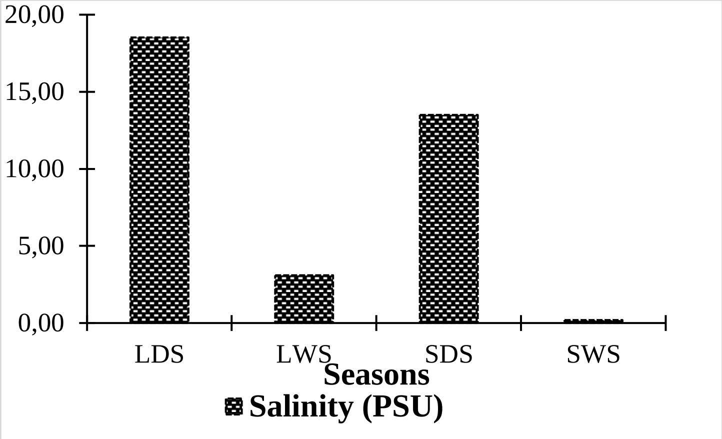


Figure 6: Seasonal and spatial variations of Dissolved Oxygen in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

Similarly, salinity, conductivity and TDS reached their high values (7.14 PSU, 12.82 g L^{-1} and 8.41 g L^{-1}, respectively) at site S1 which is close to the sea and their low values (3.75 PSU, 7.00 g L^{-1} and 4.30 g L^{-1}, respectively) at site S8 which is much more fluvial. These seasonal values show a sharp drop in salinity, conductivity and TDS during the wet season, which is probably related to the heavy precipitation during this period, which induces water dilution in the lake. However, the values of these parameters are higher than those obtained by [23] on the same lake, who obtained lower salinity and conductivity concentrations.



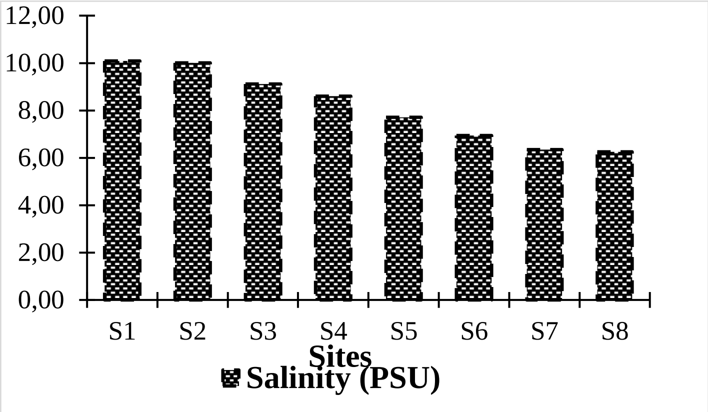
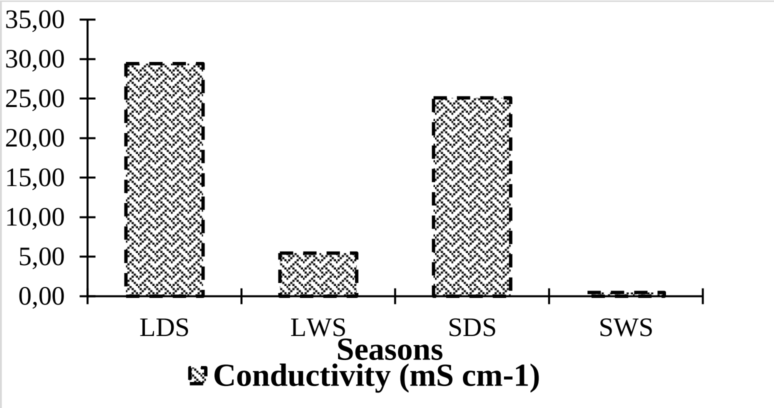


Figure 7: Seasonal and spatial variations of Salinity in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8



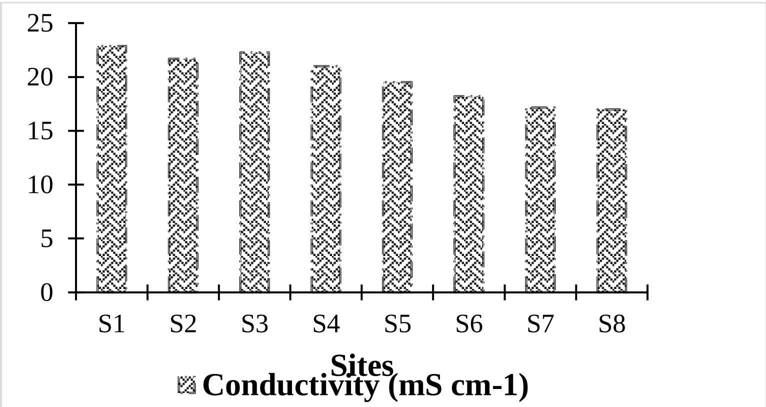
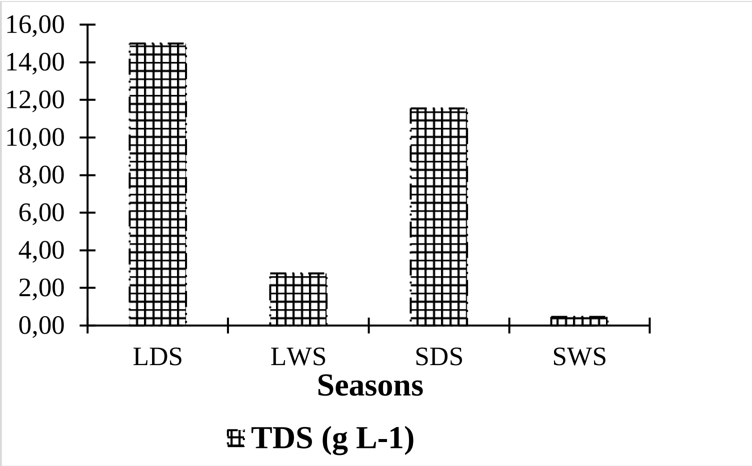


Figure 8: Seasonal and spatial variations of Conductivity in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8



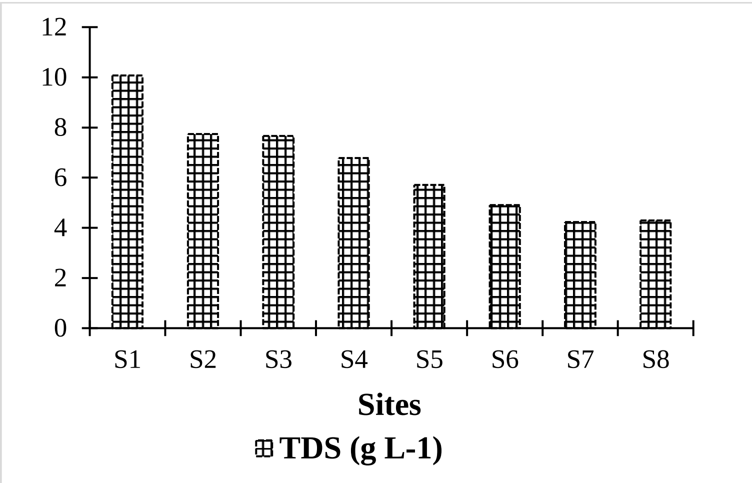
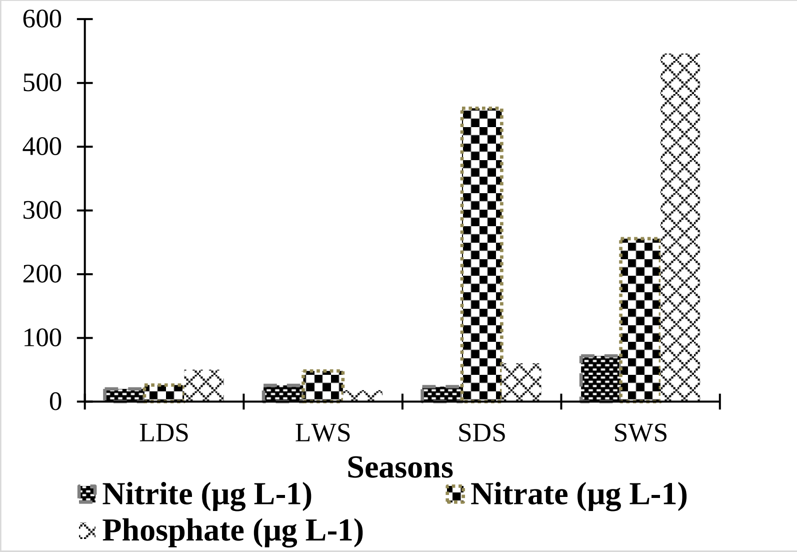


Figure 9: Seasonal and spatial variations of TDS in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

Nitrites and nitrates showed their lowest values in the long dry season (LDS) at 19.74 µg L^{-1} and 25.94 µg L^{-1} respectively, while phosphates had the lowest values (18.18 µg L^{-1}) in the long wet season (LWS) (Figure 10). Furthermore, the highest values for nitrites and phosphates (71.50 µg L^{-1} and 546.23 µg L^{-1} respectively) are obtained during the short wet season (SWS), while the highest values for nitrates (459.92 µg L^{-1}) are obtained during the short dry season (SDS) (Figure 10). However, the nitrate, phosphate and nitrite values obtained in this study are much lower than those obtained by [23] on the same ecosystem, who obtained nitrate, nitrite and phosphate concentrations greater than 25 mg L^{-1}. In contrast, the values obtained for nitrates and phosphates are close to those observed by [22] in Ivory Coast also during the wet season. The increase in nitrite concentration during the wet season is the result of the leaching of agricultural land from the upstream part where agricultural activities are more intense and thus enriched the lake with nutrients. The high values of phosphates are believed to come from domestic water discharges which are generally the main source of water pollution by phosphates. Domestic wastewater composed of detergents (used for dishwashing) is also a source of phosphates as well as urine [26]. Moreover, the high nitrate values, although obtained during the short dry season, could be due to domestic discharges of riparian populations into the lake, which enriches the environment.



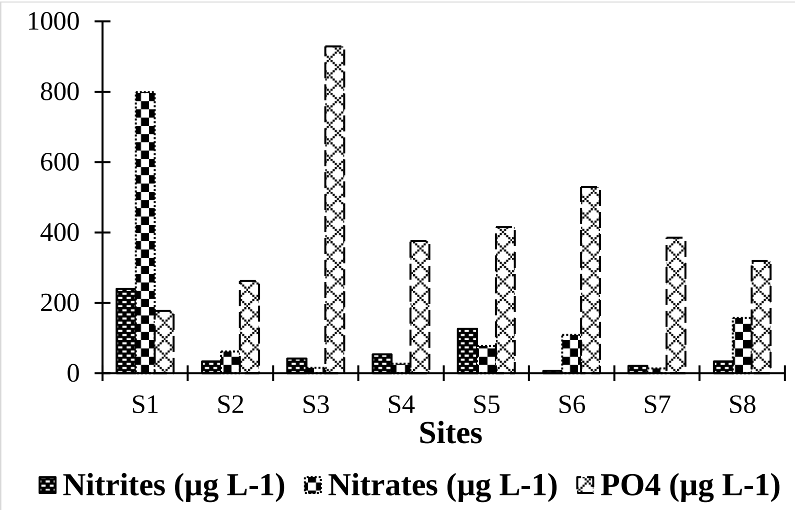


Figure 10: Seasonal and spatial variations of Nitrite, nitrate and phosphate in Lake Ahémé. The upper graph shows the mean values for all stations over the four seasons and the lower graph shows the mean values for each station. LDS: Long Dry Season; LWS: Long Wet Season; SDS: Short Dry Season; SWS: Short Wet Season. S1 to S8= Site 1 to Site 8

The phytoplankton identified comprises a total of 274 species belonging to the families: Bacillariophyceae (148), Chlorophyceae (35), Cyanophyceae (28), Euglenophyceae (26) Conjugatophyceae (21), Trebouxiophyceae (6), Dinophyceae (5), Xanthophyceae (2), Chrysophyceae (1) and Ulvophyceae (2). Bacillariophyceae are the most dominant group, accounting for (54%) of the total species inventoried (Figure 7). It was followed by Chlorophyceae (12.77%), Cyanophyceae (10.218%), Euglenophyceae (9.49%), Conjugatophyceae (7.66%), Trebouxiophyceae (2.18%), Dinophyceae (1.82%), Xanthophyceae (0.73%), Ulvophyceae (0.73%) and Chrysophyceae (0.36%), (Figure 11). These groups are typical for West African coastal ecosystems, including Togo, Nigeria and Ivory Coast [27] [28] [29] [30] [31] [22] [32] [33]. These taxa are common and generally observed in ecosystems in Benin [26] [34] [35]. In this study, Bacillariophyceae was more abundant during SDS in all the sites and less abundant during LDS especially in sites S2 and S6 (Figures 11 and 12) while Chlorophyceae were more abundant during SWS mainly in sites S6 and S8 and less dominant during SDS mainly in sites S3 and S4. These different observations show that Bacillariophyceae are abundant with higher values of nitrite and phosphate while Chlorophyceae are more favourable to large variations in nitrate. However, Cyanophyceae was found more abundant during LDS and less abundant during

SWS. This group seems to be influenced by large variations in temperature, salinity, conductivity and TDS which are in very high concentrations during this period. This apprehension is corroborated by the results of [36] who found that cyanophyceae are abundant at high temperatures and their density is low at low temperatures. In addition, other classes such as Dinophyceae, Conjugatophyceae, Ulvophyceae, Xanthophyceae, Trebouxiophyceae and Chrysophyceae also identified in this study were observed in much smaller proportions during the seasons.

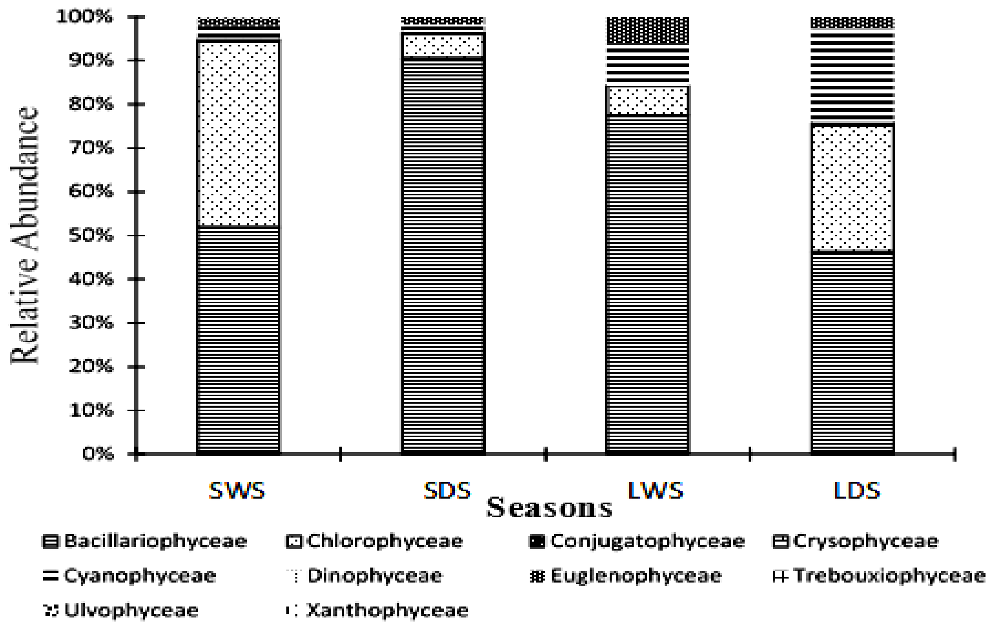


Figure 11: Relative abundance of phytoplankton classes during seasons in Lake Ahémé (Bénin). LWS : Long Wet Season; SWS : Short Wet Season; LDS : Long Dry Season; SDS : Short Dry Season

Biodiversity indices are used for biomonitoring of aquatic ecosystems [37]. The Shannon diversity index (H) fluctuated both seasonally and at the site level. Shannon diversity was highest during the long wet season (LWS) especially at site S7 and lowest during the short dry season at site S3 (Figures 13 and 14). Low values of Shannon Index less than 1 bit observed at S3 were likely related to the degradation of habitat structure. Similarly, Evenness Index values are well below 1 in all seasons and across all sites, indicating that individuals belonging to the recorded classes are not evenly distributed across stations. The Simpson's diversity index also indicates low diversity during the short dry season, especially at site S3. According to [38], low values of the diversity index indicate probable eutrophication. The same indices were calculated at the same sites and during the same periods for diatoms, which represent 53.5% of the total phytoplankton in Lake Ahémé [14]. These indices show the same trend as the biodiversity indices obtained for phytoplankton as a whole. However, the values of these indices are higher for phytoplankton than for diatoms, which could be related to the specific composition of each of these compartments (274 species for phytoplankton and 148 species for diatoms).

However, [11] has already reported that in Lake Ahémé, climatic phenomena (e.g., fluctuations in precipitation) strongly influence species diversity. This difference could also be explained by the ever-increasing population growth, generating strong anthropogenic pressures (activities of the Possotomè industry, garbage discharges, etc.) on the various water resources. These activities result in the loss of species habitats and the decrease in species diversity [13] [4] [7] [39].

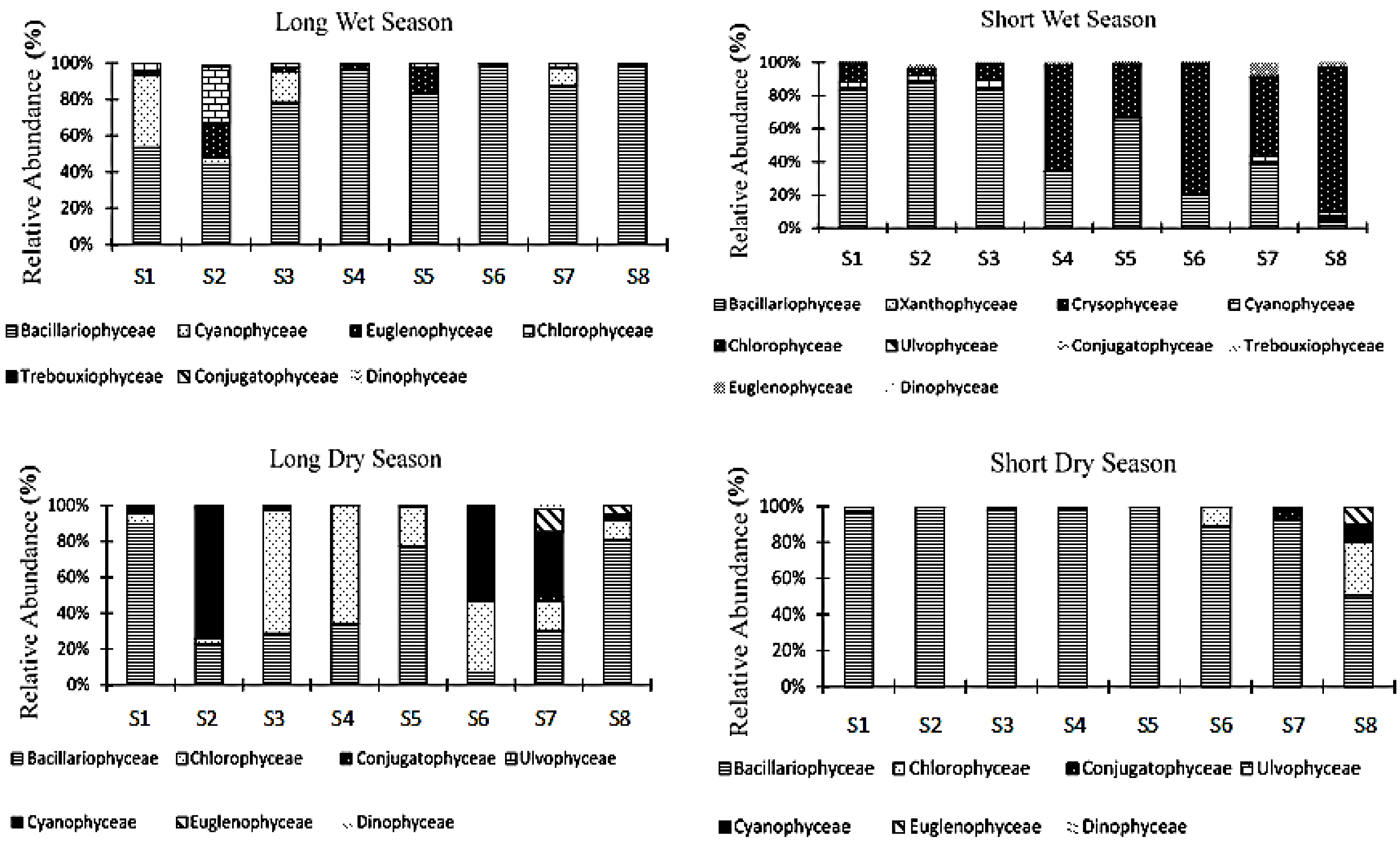


Figure 12: Relative abundance of phytoplankton classes during seasons in Lake Ahémé (Bénin). LWS : Long Wet Season; SWS : Short Wet Season; LDS : Long Dry Season; SDS : Short Dry Season. S1= site 1, S2= site 2, S3= site 3, S4= site 4, S5= site 5, S6= site 6, S7= site 7, S8= site 8.

In general, *Entomoneis paludosa, Iconella capronii, Iconella robusta, Melosira* sp, *Navicula* sp, *Nitzschia bacata, Nitzschia palea, Nitzschia* sp, *Pinnunavis elegantoides, Coscinodiscus wailesii, Coscinodiscus* sp, *Pleurosigma* sp., *Terpsinoe musica, Diploneis didyma, Tryblionella scalaris, Lyngbya* sp., *Synechococcus* sp. are the taxa that were permanently present in all seasons. As regards Bacillariophyceae, the species *Amphora ovalis, Aulacoseira* sp, *Bacillaria paxilifera, Cerataulina bicornis, Cocconeis placentula, Coscinodiscus wailesii, Craticula cuspidata, Cyclotella radiosa, Entomoneis paludosa, Gyrosigma fasciola, Iconella capronii, Iconella robusta, Nitzschia palea, Nitzschia sigma, Nitzschia closterium, Pinnunavis elegantoides, Terpsinoe musica*, and *Tryblionella scalaris* were the most dominant genera during the long wet season, while during the short dry season, *Bacillaria* sp. , *Campylodiscus* sp., *Cerataulina bicornis, Coscinodiscus* sp., *Cyclotella* sp., *Entomoneis paludosa, Navicula* sp., *Pinnunavis elegantoides, Ulnaria* sp. are the most dominant.

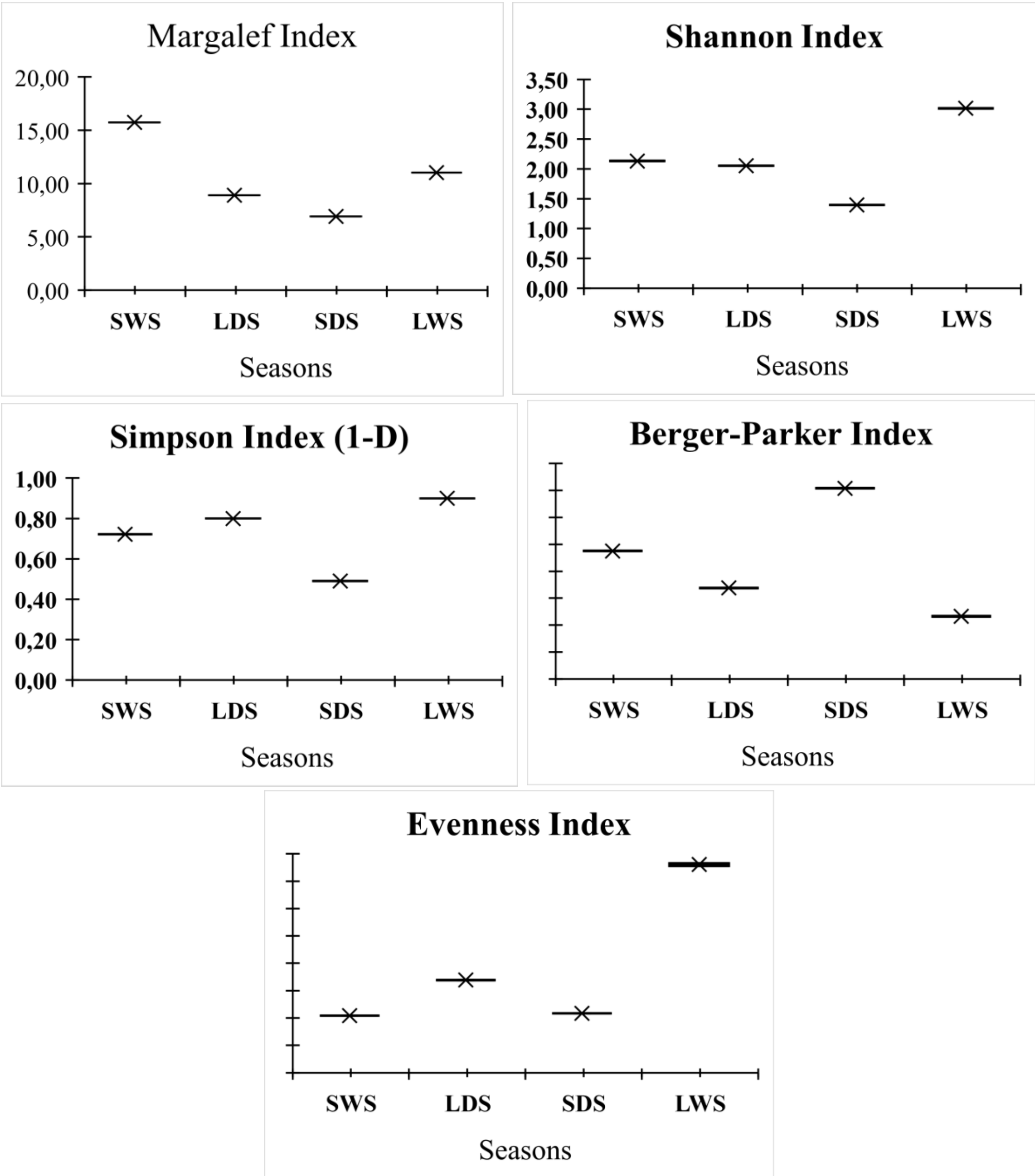


Figure 13: Seasonal variation of diversity indices. a: Margalef; b: Berger_Parker; c: Simpson (1-D); d: Shannon index; e: Evenness. SWS= short wet season, LDS= long dry season, SDS= short dry season, LWS= long wet season. Median value is shown in each box, vertical bars correspond to the minimum and maximum values.

The most dominant taxa of Chlorophyceae in the long wet season are *Pandorina morum*, *Eudorina elegans*, while in the short dry season *Stigeoclonium subsecundum* dominates. Cyanophyceae, *Chroococcus* sp., *Anabaena* sp., *Lyngbya majuscula*, *Microcystis* sp. and *Synechococcus* sp. were the most dominant during the long wet season while in the short dry season *Synechococcus* sp., *Anabaena* sp. dominate. As for Euglenophyceae, the most dominant during the long wet season were *Euglena allorgei*, *Euglena* sp., *Trachelomonas oblonga*, *Phacus contortus* and *Phacus gigas* while in the short dry season, *Trachelomonas superba* and *Phacus* sp. were the most dominant. The other taxa of groups such as Dinophyceae, Conjugatophyceae, Trebouxiophyceae, Ulvophyceae, Xanthophyceae and Chrysophyceae were recorded in small proportions during the different periods.

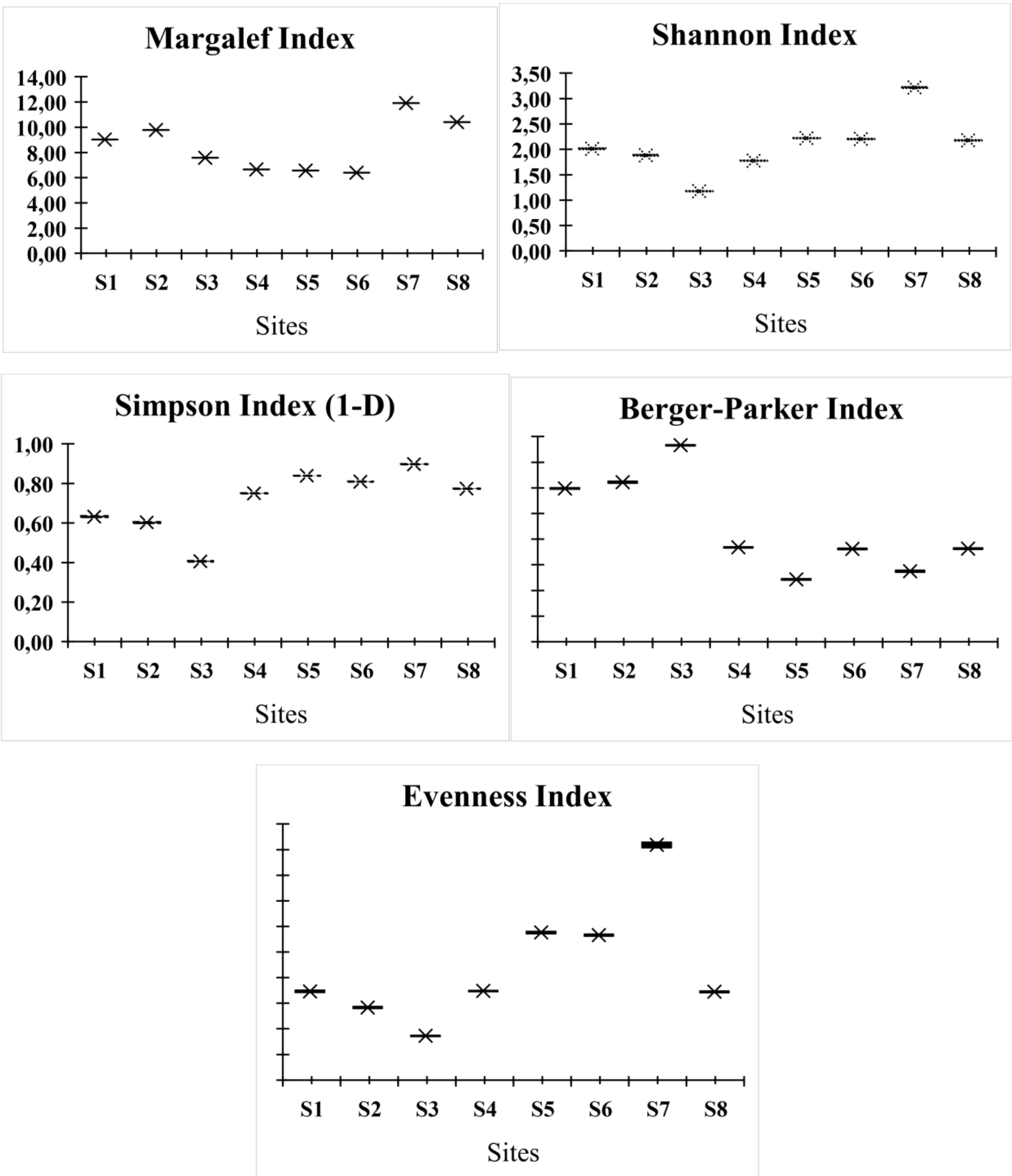


Figure 14: Spatial variation of diversity indices. a: Margalef; b: Berger_Parker; c: Simpson (1-D); d: Shannon index; e: Evenness. S1= site 1, S2= site 2, S3= site 3, S4= site 4, S5= site 5, S6= site 6, S7= site 7, S8= site 8. Median value is shown in each box, vertical bars correspond to the minimum and maximum values.

Moreover, the diversity values observed in this study are lower than those observed in Lake Hlan in Benin (ranged between 4.8bit $cell^{-1}$ and 5.1bit $cell^{-1}$) [35] and higher than those of the Ivory Coast (0.001 bit $cell^{-1}$ and 2.35 bit $cell^{-1}$) [40]. This could be explained by the difference in the flow of each water body [28] [26] [33] and also linked to the sampling period. To some extent, the observed trends could result from the inability of some taxa to develop at certain times of the year when hydrological and limnological characteristics are unfavorable [41] [39].

Conclusion

This study showed that the algal community in Lake Ahémé is unevenly distributed. Bacillariophyceae are the most important group of the phytoplankton found in Lake Ahémé during this study. The high Shannon-Wiener Index and low Evenness Index confirm that the distribution of phytoplankton communities in Lake Ahémé is inequitable. In this study, some particular species appeared at specific times, which means that environmental conditions play an important role in the composition and structure of the phytoplankton community. This suggests an assessment of the direct or indirect causes and effects of types of pollution or stress on the ecosystem.

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