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The Effect of Human Activities on the Qualitative and Quantitative Composition of Epiphytic Algae on *Ceratophyllum demersum L.* In Al- Warar Canal, Ramadi, Iraq

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Abstract

The current study aims to investigate the qualitative and quantitative makeup of epiphytic algae on the aquatic host plant *Ceratophyllum demersum L.* Four locations along the Al-Warar Canal, Ramadi Governorate were surveyed during the seasons of Autumn 2022, Winter 2022, Spring 2023, and Summer 2023. The study also involved quantifying the physiochemical parameters, including Temperture of water, pH, Turbidity, Total Dissolved Solid (TDS), Electrical Conductivity (EC), Total Hardness (TH), Sulphate (SO_4^{-2}), Demand Oxygen (DO), Bio-chemical Oxygen Demand (BOD), Nitrate (NO_3^-), and Phosphate (PO_4^{3-}). The total number of epiphytic algae species was two hundred thirty-six communities of algae attached to the C. demersum L plant, belonging to 57 genera, which were identified during the study period. The phylum Cyanophyceae (8) genera, belonging to (34) species, were identified, while the green algae Chlorophyceae (20) genera, belonging to (65) species, were identified. The diatom algae Bacillariophyceae has been identified in (27) genera, dating back to (133) species. The order of the diatom algae, Bacillariophyceae-centralis, has been identified in (2) genera, dating back to (4) species. The order of the feathery diatom algae Bacillariophyceae-Pennales has been identified in (25) genera dating back to (125). As for the Euglenophyceae algae, (2) genus belonging to four species have been identified.

The study showed that the diatom algae Bacillariophyceae dominate the other algal groups, distinguishing Iraqi waters.

Introduction:

Epiphytic algae adhere to the surfaces of aquatic plants. They have a crucial role in freshwater ecosystems since they are responsible for primary production and serve as a nutritious food supply for aquatic animals. Additionally, they transfer energy from the sediment to the water column. And present a well-rounded argument on the aquatic organisms [1, 2]. In addition, algae play a role in the generation of oxygen through the process of photosynthesis[3]. Human activities in agriculture, industry, and urbanization generate significant quantities of pollutants that have a detrimental impact on aquatic organisms. Therefore, it is essential to utilize these organisms, particularly algae, as indicators to assess

water quality [4, 5]. *C. demersum* is universally recognized as an indigenous plant species. It is distinguished by its bifurcated shape and can be found either submerged or anchored to the ground by the rhizoidal shoots, which are the buried branching ends of the stem. The plant's leaves are sessile and have serrated margins. Each node has 5 to 12 leaves branching out from it. Several earlier research studies have been undertaken on epiphytic algae [6-8]. The objective of this study was to examine the population of epiphytic algae on the aquatic plant *C. demersum* and assess their diversity using indices in the water of Nazim Al-Warar in Anbar Governorate in western Iraq.

MATERIALS AND METHODS

2.1 Study area

Samples were collected from the water of the Al-Warar Canal in Anbar Governorate in western Iraq. Four sites were chosen for the study, starting from a site before the Al-Warar Dam all the way to the Humera area, from October 2022 until July 2023. Samples were collected from the study sites in depth. 30 cm from the surface of the river. These samples were placed in sterile, opaque glass bottles with a capacity of 2 liters and kept in special cork containers with pieces of ice added until reaching the laboratory to complete the chemical and physical analyses.

Table 1. Geographic coordinates of sampling sites in the Al-Warar Canal (Google Earth).

Sites	Latitudes (North)			Longitudes (East)		
	°	'	"	°	'	"
1 Euphrates River	**33**	**26**	**04**	**43**	**15**	**53**
2 Ramadi Women's Hospital	**33**	**26**	**25**	**43**	**16**	**47**
3 Al-Qasim Bridge	**33**	**25**	**09**	**43**	**17**	**15**
4 Hemerrah	**33**	**23**	**59**	**43**	**18**	**05**

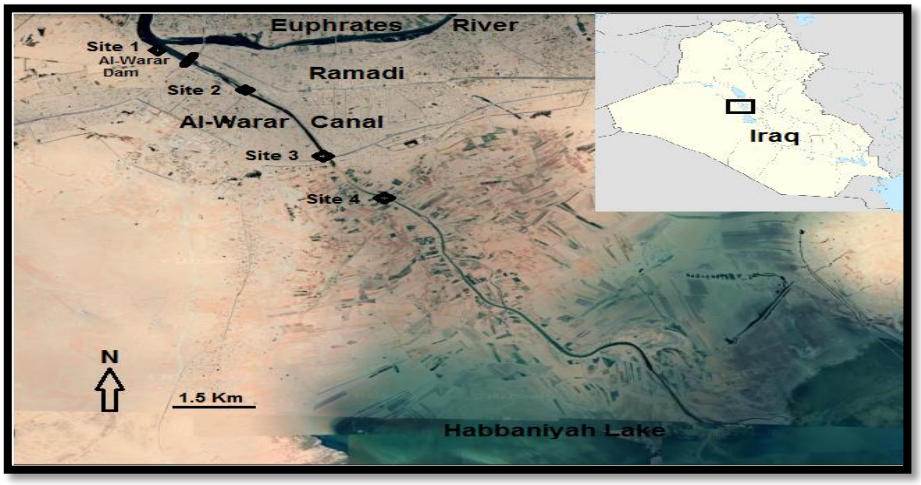


Fig. 1. Geographical representation of the locations where sampling was conducted in the Al-Warar Channel.

2.2 Physicochemical measurements

The water samples were collected from the designated sites (S1–S4) following the established sampling protocol (IS: 2498, 1966 – Part I). Periodically, data was documented to analyze variables like temperature, pH, dissolved oxygen, and turbidity in order to assess the type and magnitude of pollution. The American Public Health Association (APHA) [9]. has established standards and processes to assess several metrics, including Temperature of water, pH, Turbidity, Total Dissolved Solid (TDS), Electrical Conductivity (EC), Total Hardness (TH), Sulphate (SO_4^{-2}), Demand Oxygen (DO), Bio-chemical Oxygen Demand (BOD), Nitrate (NO_3^-), and Phosphate (PO_4^{3-}). The average result was calculated following three repetitions of each analysis to ensure precision.

2.3 Collection and identification of epiphytic algal samples

Plant samples were collected in plastic bags containing a tiny quantity of water from the river. The method of agitating and abrading was employed to separate the epiphytic algae from the surface of the plant. The plant samples were divided into 2-3cm long segments. These segments were agitated with 50-100ml of water from the surrounding environment. Subsequently, the plant's surface was gently scraped using a smooth brush. The samples were stored in a jar containing Lugol's solution for a qualitative study. In the quantitative study, samples were stored in 100ml cylinders for 10-15 days using Lugol's solution for preservation. The 20-30 ml sediments were preserved in containers labelled with the date and location, along with a few drops of Lugol's solution. Permanent slides were created to identify diatoms and examine them under a light microscope with magnifications of 40x to 100x. On the other hand, temporary slides were developed to identify non-diatom specimens with a magnification of 100x[10-12].

4. Statistical analysis

The data was analyzed using the statistical program Statistical Analysis System - SAS (2018) to investigate the impact of different sites on the researched attributes. The means were compared using the least significant difference (LSD) test to determine any significant changes.

Results and Discussion

3.1. Physical and Chemical Parameters

The current study was conducted to evaluate water quality in four different sites of the Al-Warar Canal in Al-Ramadi, western Iraq. Table 2 shows the results of some physical and chemical variables. During the summer, temperatures in Iraq reached high levels, while in winter, they dropped to their lowest values, as indicated by climate variations. The pH value of the study area was less than 8.5, which is the permissible limit. Turbidity values exceeded the permissible limit in all seasons of the year, which amounted to 5 NTU. The results obtained in the current study indicated higher values of EC and TDS in all seasons of the year, especially in the summer, with higher percentages compared to the year's seasons. The reason for this is the rise in temperatures, which leads to an increase in the dissolution of salts; the results agree with other studies conducted by Abdalhameed *et al.* [13]. Total hardness results showed that the canal has low hardness, especially in the Winter and Summer. At the same time, there were varying increases in hardness values for some study sites in the Autumn and

Spring seasons [14]. This result is consistent with this previous study [15]. Sulfates were higher than the permissible limits for all seasons of the year.

The dissolved oxygen (DO) levels exhibited an increase throughout the Winter and a decline during the Summer since the rising temperatures led to a reduction in dissolved oxygen [16]. The rise in BOD during the Springer was attributed to the activity of organisms and the accumulation of organic materials[16]. The results of the current study indicated a decrease in nitrate values for all seasons of the year in the Al-Warar Canal, which is less than the permissible limit. While there was an apparent increase in phosphate levels to exceed the permissible limit of 0.1mg/L[17] , this is because the canal is affected by sewage water, which contains many phosphate compounds such as detergents [12] .

Table 2. Means and standard method for drinking water of physical and chemical parameters of samples

Parameters	Standard WHO	Rang			
		Autumn	Winter	Springer	Summer
Temperature (C^o)	25	21	12	19	33
pH	6.5-8.5	6.6-7.7	7.2-7.6	7.3-7.5	7.6-7.8
Turbidity(NTU)	5	5.4-15	5.5-10.5	6-7.5	6-12
Total Dissolved Solids(mg/L)	500	784-1168	975-1331	989-1409	1062-1518
Electrical Conductivity (µ.s/cm)	1000	1189-1765	1453-1996	1641-2241	1960-2705
Total Hardness (mg $CaCO_3$/L)	500	458-664	129-155	448-643	127-145
Sulphate (mg/L)	200	378-615	388-597	406-499	416-686
Dissolved Oxygen (mg / L)	4-6	4-7	4-8	3.3-4.6	5.8-7.9
Biochemical Oxygen Demand (mg/L)	5	7-12	8.5-15.5	11-16.5	9-14
Nitrate (mg/L)	50	1.6-3.8	2.80-10.70	2.5-3.4	0.66-0.81
Phosphate (mg/L)	0.1	0.16-0.51	0.05-0.14	0.18-1.71	0.14-1.27

3-2 Qualitative study

Two hundred thirty-six communities of algae attached to the *Ceratophyllum demersum* L plant, belonging to 57 genera, were identified during the study period. The Class Cyanophyceae (8) genera, belonging to (34) species, were identified, while the green algae Chlorophyceae (20) genera, belonging to (65) species, were identified. The diatom algae Bacillariophyceae has been identified in (27) genera, dating back to (133) species. The order of the diatom algae, Bacillariophyceae-centralis, has been identified in (2) genera, dating back to (4) species. The order of the feathery diatom algae Bacillariophyceae-Pennales has been

identified in (25) genera dating back to (125). As for the Euglenophyceae algae, (2) genus belonging to four species have been identified.

The study showed that the diatom algae Bacillariophyceae dominate the other algal groups, distinguishing Iraqi waters**.**

Table 3: Number of genera and their species identified in the sampling sites during the period of the study.

Sites		*Cyanophyceae*	*Chlorophyceae*	Bacillariophyceae		*Euglenophyceae*	Total
				Centrales	*Pennales*		
Site 1	Genera	6	13	2	19	2	42
	%	14.3	30.9	4.8	45.2	4.8	100
	Species	15	29	5	57	2	108
	%	13.9	26.9	4.6	52.8	1.9	100
Site 2	Genera	5	11	2	17	1	36
	%	13.9	30.6	5.6	47.2	2.8	100
	Species	15	21	3	51	3	93
	%	16.1	22.6	3.2	54.8	3.2	100
Site 3	Genera	6	10	2	19	2	39
	%	15.4	25.6	5.1	48.7	5.1	100
	Species	12	19	5	56	2	94
	%	12.8	20.2	5.3	59.6	2.1	100
Site 4	Genera	7	15	2	15	1	40
	%	17.5	37.5	5.0	37.5	2.5	100
	Species	16	33	6	53	2	110
	%	14.5	30.0	5.5	48.2	1.8	100

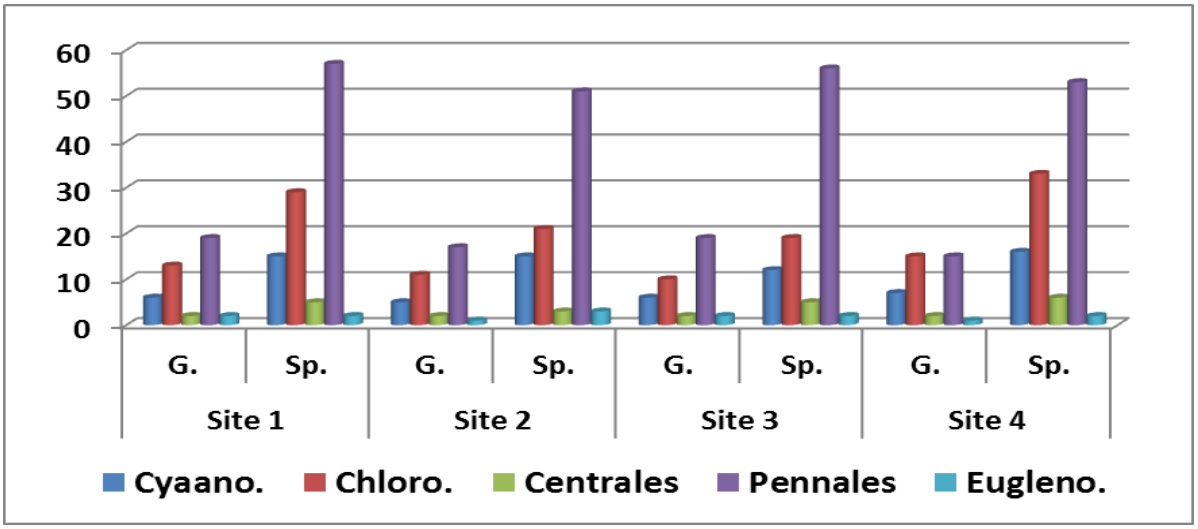


Figure 2: *Numbers of identified genera and species of algae attached to C. demersum plants at sampling sites for the waters.*

The first site for the Cyanophyceae class recorded (15) species (13.9%) belonging to 6 genera (14.3%), while the green algae class Chlorophyceae recorded (29) species (26.9%)

belonging to (13) genera (30.9%). The class Bacillarophyceae-Centralis recorded (5) species (4.6%) belonging to (2) genera (4.8%), and the feathered diatom order Bacillarophyceae-Pennales recorded (57) genera (52.8%) belonging to (19 genera). At a rate of (45.2%), while the class Euglenophyceae recorded (2) species at a rate of (1.9%) belonging to (2) genera at a rate of (4.8%) Table 3 and Figure 2. The increase in species of algae in this site is Due to the high biological diversity. Also, it indicates the presence of a moderate percentage of pollution due to the dilution process, as this site is considered a source of feeding the water of the Warar Canal from the Euphrates River [18]. The highest number of species of feathery diatom algae has been recorded, and this confirms that the site contains a percentage of Large amounts of silicates [19].

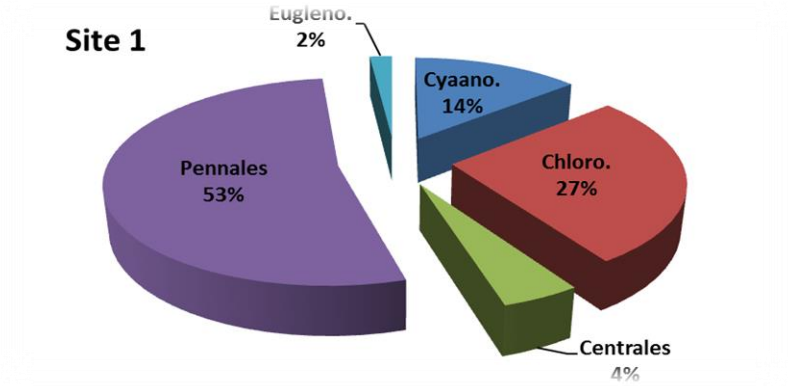


Figure 3: *Percentages of the number of identifiable species of algae attached to the Ceratophyllum demersum L plant at site (1) in the water*

The numbers of algae in Site 2 were found to be lower than in Site 1, where 36 species were identified, and 15 species were recorded in the class Cynophyceae (16.1%), belonging to 5 genera (13.9%), and the class Chlorophyceae was recorded. 21 species (22.6%) belonging to (11 genera (30.6%). As for the central Diatomaceous algae order Bacillrophyceae-centralis, (3) species (3.2%) were recorded belonging to (2) genera (5.6%). The feather algae order Bacillarophyceae-Pennales recorded (51) species (54.8%) belonging to (17) genera (47.2%), and the Euglenophyceae class recorded (3) species (3.2%) belonging to (1) genus (2.8%). Table 2 and Figure 4 This indicates good biodiversity because the site is affected by the presence of wastewater points [20]

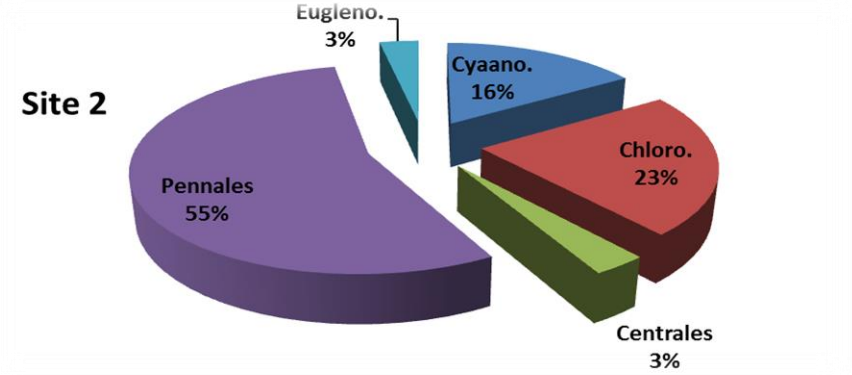


Figure 4: *Percentages of the number of identifiable species of algae attached to the Ceratophyllum demersum L plant at site (2) in the water .*

In the third site, there were (94) species belonging to (39) genera. The blue-green algae Cyanophyceae (12) species were recorded (12.8%) belonging to (6) genera (15.4%).

The green algae Chlorophyceae (19) species were recorded (20.2%) belonging to (10) genera, with a percentage of (25.6%), and the class Bacillarophyceae was classified into two orders. The first order Centrales was recorded (5) species, with a percentage of (5.3%), belonging to (2) genera, with a percentage of (5.1%). The second order, the feathery diatom order Bacillarophyceae - Pennales has recorded (56) species (59.6%) belonging to (19) genera (48.7%). The phylum of diatom algae, Bacillarophyceae, was divided into two orders: the first order Centrales recorded (5) species, with a percentage of (5.3%) belonging to (2) genera, with a percentage of (5.1%), and the second order, the feathery diatom order Bacillarophyceae-Pennalis recorded (56) species, with a percentage of (59.6%). It belongs to (19) genera (48.7%), while the Euglenophyceae algae has recorded (2) species (2.1%) belonging to (2) genera (5.1%). The species identified in the third site are considered less than the species diagnosed in the site. First this gives the impression that the third site was affected by the presence of sand washing quarries close to the study sites, in addition to the presence of a sewage point, which led to a slowdown in the water recycling process in this site, and thus a negative impact on the biological diversity of algae attached to the *C. demersum* L plant, as in Table 3 and Figure 5

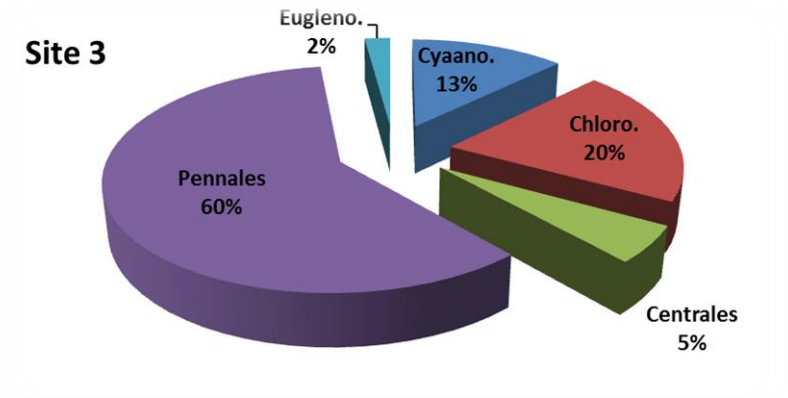


Figure 5: *Percentages of the number of identifiable species of algae attached to the Ceratophyllum demersum L plant at site (3) in the water of the Al-Warar Canal*

The S4 recorded (110) species belonging to (40) genera. The class Cyanophyceae recorded (16) species (14.5%) belonging to (7) genera (17.5%), while the class Chlorophycea*e* (33) was identified. A species with a percentage of (30.0%) belonging to (15) genera with a percentage of (37.5%). As for the order of Centralis - Bacillarophyceae, (6) species with a percentage of (5.5%) were identified, belonging to (2) genera with a percentage of (5.0%). As for the order Diatomaceous feathers Pennales has been identified (53) species (48.2%) belonging to (15) genera (37.5%). As for the Euglenophyceae algae, (2) species (1.8%) belonging to (1) genus (2.5%) were identified in Table 2 and Figure 6. The largest number of species was recorded compared to the rest of the study sites because they are agricultural lands rich in chemical fertilizers. With nitrogen and phosphorus, as it is agricultural land, which encouraged the bloom and growth of algae during the study year, and this is known as the phenomenon of eutrophication [21].

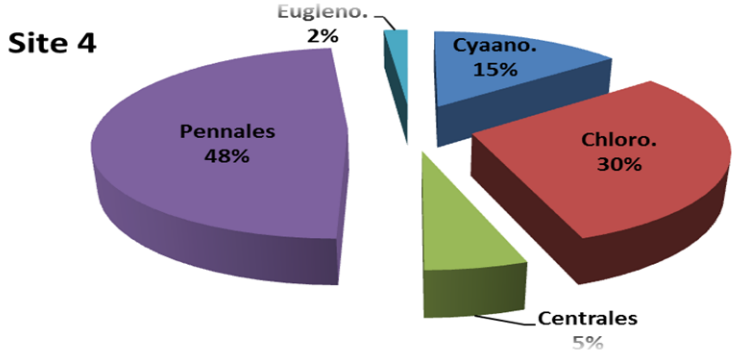


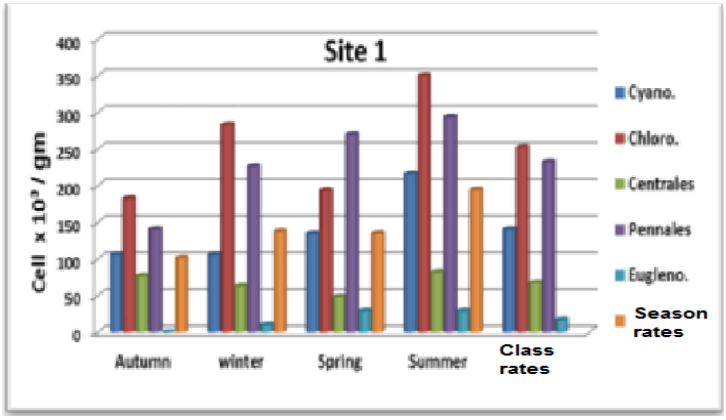
Figure 6: *Percentages of the number of identifiable species of algae attached to the Ceratophyllum demersum L plant at site (4) in the water*

3-3 Quantitative study :

The quantitative study results of algae attached to the *C. demersum* L plant showed clear seasonal and locational variations during the study year. The highest average total number was recorded in the spring, amounting to 868.4 cell x 10^3 g^{-1}. The highest total number was recorded in the fourth site in the summer, which amounted to 1323.7 cell x10^3 g^{-1} , the main nutrients supplied starting from the site where the water enters the Al-Warar Canal from the Euphrates River, as well as the increase in their numbers during the summer due to the long duration of light intensity during the day [22].

Seasonal variations of the cell density of the rows of algae attached to the *C. demersum* L plant at the first site before the Warar Canal showed that the dominance of green algae Chlorophyceae amounted to (350.4 cell×10^3 g^{-1}) in the summer, while the blue-green algae Cyanophyceae recorded (216 cell × 10^3 g^{-1}) , while the central diatoms recorded (81.6 x 10^3 cell g^{-1}) , and the feathery diatoms reached (293 x 10^3 cell g^{-1}) , while the Euglenophyceae class reached (28.8 x 10^3 cell g^{-1}) during the spring and summer, and the dominance of green algae in the first location gives an indication that the waters of the Euphrates River are Fresh water and low salt concentration [23]

Figure (4) showed seasonal variations in the percentage density of algae cells attached to the *C. demersum* L plant, where the highest percentage was recorded in the summer, reaching 33%, then in the winter, the percentage reached 24%. At the same time, in the spring, it reached 22%. In the fall season, it reached (21%). The high percentage of algae cell population density during the summer is due to high temperatures and the availability of basic nutrients that help algae grow and flourish[24]



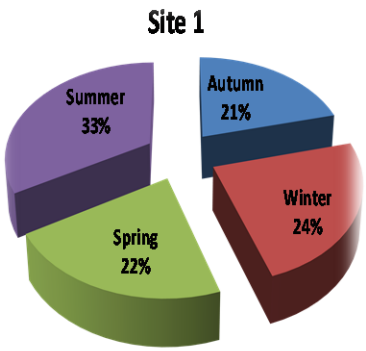


Figure 7: *Seasonal variations in the density of the number of cells of rows of algae attached to the Ceratophyllum demersum L plant at site (1).*

The second site showed that the blue-green algae species Cyanophyceae recorded the highest density in the spring, reaching (360 x 10^3 cell g^{-1}) , followed by the green algae species in the winter (345.6 x 10^3 cell g^{-1}) , and then the diatom algae. The feathery algae reached (294.1 x 10^3 cell g^{-1}) , then the Euglenophyceae species recorded in the summer (38.4 x 10^3 cell g^{-1}), then the central Diatomaceous algae order recorded in the winter (25.5 x 10^3 cell g^{-1}), and the dominance of blue-green algae in this site is due to the presence of sanitation points. Various human activities cause high organic pollution in the water, and these algae resist various environmental variations. Species of algae indicative of water quality (*Oscillatoria princeps*) were recorded at a rate of (4.73%), species of diatom algae (*Caloneis placentula* (0.90%), *Nitzschia palea* (1.30%), and a species from the phylum *Euglena acus* (1.18%) *E. elastica. E. minus* (1.57%) , as the presence of these species indicates that the water contains organic materials [20]. Figure 8 seasonal variations in the percentage density of algae cells showed algae cells attached to the champion plant, where The highest percentage was recorded in the spring (40%), then in the winter (34%). In contrast, in the summer it was recorded (18%) and in the fall the percentage was (8%). The high value of the percentages during the winter and spring is due to rainfall soil erosion. and increase in organic nutrients as a result of waste from human activities [25].

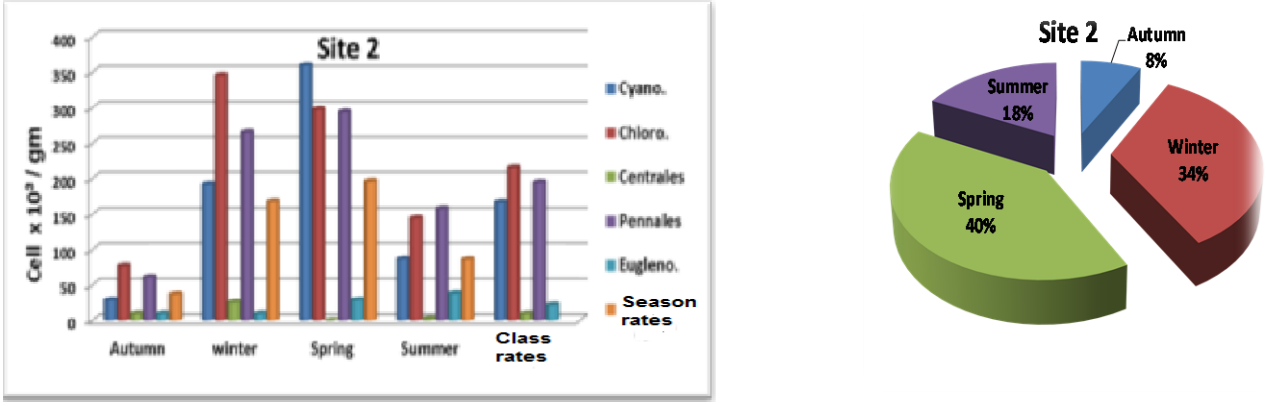


Figure 8: *Seasonal variations in the density of the number of cells of rows of algae attached to the Ceratophyllum demersum L plant at site (2)*

Site 3 showed the highest total number of algae adhered to the *C. demersum* plant was recorded for the blue-green algae Cyanophyceae in the spring season (316.3 x 10^3 cell g^{-1}), followed by the green algae Chlorophycaea (218.7 x 10^3 cell g^{-1}), and then the feathery diatom algae (209.3 x 10^3 cell g^{-1}). The centrales order of diatoms was recorded as winter (32.4 %) Summer (35%), while in winter (16%) were recorded. Many species that indicate the quality of water quality were recorded. Many species belonging to the genus *Oscillatoria* were recorded, such as *O.princeps* (3.93%), *O.formosa* (2.95%), and *O.splendida* (4.42%), and two species belonging to the Euglenophyceae algae class were identified: *Euglena acus* (1.47%) and *E. elastic* (0.98%), which are considered among the species that indicate water pollution with organic materials [17].

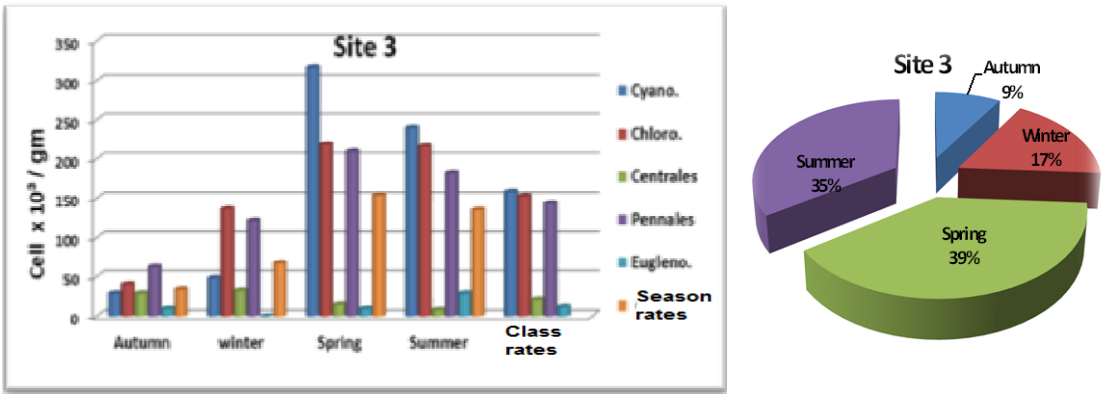


Figure 9*: Seasonal variations in the density of the number of cells of rows of algae attached to the Ceratophyllum demersum L plant at site (3)*

The highest total number of algae adhered to the *C. demersum* plant for the green algae row in the summer was (768 x 10^3 cell g^{-1}), while the blue-green algae recorded (412.6 x 10^3 cell g^{-1}), while the numbers of feathery diatom algae reached (315.7 x 10^3 cell g^{-1}) in the spring. The centrales order of diatoms was recorded in the winter (28.8 x 10^3 cell g^{-1}), while the *Euglen*ophyceae algae class were recorded in the spring (28.8 x 10^3 cell g^{-1}). Figure 9 show the seasonal variations of the percentage density of algae cell numbers. In the fourth location, the highest percentage was recorded in the summer, amounting to (43%), and the lowest percentage in the fall, amounting to (4%). In the spring, it was (35%), and in the winter, it was (16%). and it was recorded that there are many species whose presence indicates water pollution. The predominant two species in all study sites of the genus *Oscillatoria* are *O.formosa* (2.47%) and *O.splendida* (5.88%). Species of the diatom *Cocconeis placentula* (1.59%) and the species *Syndra ulna* (3.26%) were recorded. Two species of *Euglena* sp. , *E.acus* (1.23%) and *E. minuta* (0.61%) were recorded. The presence of these species indicates the abundance of organic materials in the water

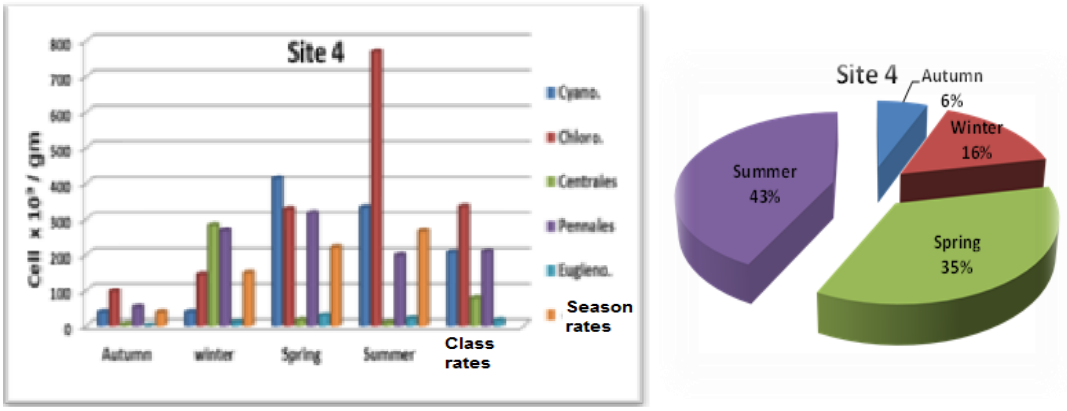


Figure 10: *Seasonal variations in the density of the number of cells of rows of algae attached to the Ceratophyllum demersum L plant at site (4).*

Conclusion

The study findings indicate that the presence of epiphytic algae is influenced by the type of human and industrial activities occurring in the surrounding areas and by seasonal changes in temperature, transparency, and nutrient levels. The class Bacillariophyceae was the most prevalent.

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تأثير الأنشطة البشرية في التركيب النوعي والكمي للطحالب الملتصقة على نبات الشمبلان. في قناة الورار، الرمادي، العراق

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الخلاصة:

هدفت الدراسة دراسة التركيب النوعي والكمي للطحالب الملتصقة على العائل المائي للنبات *Ceratophyllum demersum L.*تم مسح أربعة مواقع على طول قناة الورار في محافظة الرمادي خلال مواسم خريف 2022، شتاء 2022، ربيع 2023، وصيف 2023. وتضمنت الدراسة أيضًا قياس العوامل الفيزيائية والكيميائية، بما في ذلك درجة حرارة الماء، ودرجة الحموضة، والعكارة، وإجمالي المواد الصلبة الذائبة(TDS) ، والموصلية الكهربائية(EC) ، والصلابة الكلية(TH) ، والكبريتات(SO_4^{-2}) ، والطلب على الأكسجين(DO) .، الطلب على الأكسجين الحيوي والكيميائي(BOD) ، والنترات(NO_3^-) ، والفوسفات (PO_4^{3-}) بلغ إجمالي عدد أنواع الطحالب الملتصقة مائتان وستة وثلاثون مجتمعاً من الطحالب المرتبطة بنبات*C. demersum L* ، تنتمي إلى 57 جنساً، خلال فترة الدراسة. تم التعرف على شعبة Cyanophyceae (8)أجناس تنتمي إلى (34) نوعا، في حين تم التعرف على شعبة الطحالب الخضراء Chlorophyceae (20) جنسا تنتمي إلى (65) نوعا. تم التعرف على طحالب الدياتوم Bacillariophyceae في (27) جنساً يعود تاريخها إلى (133) نوعاً. تم تحديد رتبة الطحالب الدياتومية Bacillariophyceae-centralis في (2) أجناس يعود تاريخها إلى (4) أنواع. تم تحديد رتبة الطحالب المشطورة الريشي-Bacillariophyceae Pennalesفي (25) جنسًا يعود تاريخها إلى (125). أمّا بالنسبة لطحالب Euglenophyceaeفقد تم التعرف على جنس (2) ينتمي إلى أربعة أنواع.
وأظهرت الدراسة أن طحالب الدياتوم Bacillariophyceae تهيمن على مجموعات الطحالب الأخرى التي تميز المياه العراقية.