

Influence of dissolved organic carbon on the limnochemical characteristics of a tropical endorheic wetland during a hydrological period: El Eneal Wetland, Colombia

Wilfer David Guzmán-López, Lina Claudia Giraldo, Fabio Vélez-Macías, & Néstor Aguirre

Escuela ambiental, Facultad de Ingeniería, Universidad de Antioquia, Medellín, Colombia. wilfer.guzman@udea.edu.co, lina.giraldo@udea.edu.co, fabio.velez@udea.edu.co, nestor.aguirre@udea.edu.co

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Abstract

The progressive darkening of inland waters globally has been attributed to increasing dissolved organic carbon (DOC) concentrations, yet this phenomenon remains understudied in tropical systems. This research investigated DOC's influence on the limnochemical characteristics of a tropical endorheic wetland through two sampling campaigns during different hydrological periods. DOC was measured alongside key limnological variables, with relationships analyzed through descriptive statistics and principal component analysis. Results demonstrated significant positive correlations between DOC and electrical conductivity ($r=0.78$, $p<0.001$) and turbidity ($r=0.74$, $p<0.001$), while inverse correlations emerged with bicarbonates ($r=-0.80$, $p<0.001$), ORP ($r=-0.61$, $p<0.001$), and chlorophyll-a ($r=-0.75$, $p<0.001$). However, these associations likely reflect synchronous responses to increasing water levels rather than direct DOC effects. Our findings suggest DOC potentially regulates phytoplankton biomass and optical properties in this tropical wetland system.

Keywords: DOC; tropical, wetland; limnochemistry; DOM; endorheic

Influencia del carbono orgánico disuelto en las características limnoquímicas de un humedal endorreico tropical durante un período hidrológico: Humedal El Eneal, Colombia

Resumen

El oscurecimiento progresivo de aguas continentales se ha atribuido al incremento de carbono orgánico disuelto (COD), fenómeno poco estudiado en sistemas tropicales. Esta investigación evaluó la influencia del COD en las características limnoquímicas de un humedal endorreico tropical mediante dos campañas de muestreo en diferentes periodos hidrológicos. Se midió el COD junto con parámetros limnológicos, analizando relaciones mediante estadística descriptiva y análisis de componentes principales. Los resultados mostraron correlaciones positivas significativas entre COD y conductividad eléctrica ($r=0,78$, $p<0,001$) y turbidez ($r=0,74$, $p<0,001$), y correlaciones inversas con bicarbonatos ($r=-0,80$, $p<0,001$), ORP ($r=-0,61$, $p<0,001$) y clorofila-a ($r=-0,75$, $p<0,001$). Estas asociaciones probablemente reflejan respuestas sincrónicas al aumento del nivel hídrico más que efectos directos del COD. Los hallazgos sugieren que el COD regula potencialmente la biomasa fitoplanctónica y las propiedades ópticas del sistema.

Palabras clave: DOC; humedal tropical; limnoquímica; DOM; endorreico

1. Introduction

In recent decades, a growing body of research has documented a progressive darkening of continental water bodies worldwide, a phenomenon known as "brownification" [1-3] This

change in coloration of aquatic ecosystems not only represents a visible alteration but also carries profound ecological and biogeochemical implications that affect the structure and functioning of these systems [4,5].

Research has established that this darkening is primarily

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linked to increasing concentrations of dissolved organic carbon (DOC) in water bodies [6]. DOC, composed of humic and fulvic substances derived from the decomposition of terrestrial plant material, has exhibited increasing trends in temperate and boreal regions, attributed to factors such as changes in land use, reduction of acid deposition, and notably, global climate change [7,8].

In contrast to the extensive research conducted at middle and high latitudes, there is a notable paucity of studies documenting analogous processes in tropical aquatic ecosystems, particularly in endorheic systems [9]. In Brazil, for instance, studies in urban wetlands have demonstrated that DOC significantly contributes to light attenuation, impacting primary productivity [10,11]. However, the existing literature has focused on lentic systems connected to hydrographic networks, thereby leaving a gap in the understanding of tropical endorheic wetlands. These wetlands are particularly vulnerable to the effects of DOC due to their hydrological isolation and dependence on rainfall inputs [9,12].

Tropical endorheic wetlands are recognized as ecosystems particularly vulnerable to the impacts of climate change and anthropogenic disturbances, distinguished by their hydrological isolation and dependence on precipitation-derived inputs [9,12]. These wetlands act as biodiversity reservoirs, providing vital ecosystem services such as water regulation, carbon sequestration, and flood mitigation [13]. However, the comprehension of their limnological processes, particularly those associated with carbon cycling, remains substantially underdeveloped [9].

The role of DOC in these ecosystems is of particular interest, as it has been shown to play a central role through complex regulatory mechanisms. These mechanisms include the absorption of photosynthetically active radiation (PAR), which reduces light penetration available for primary productivity, and the formation of complexes with essential nutrients, which limits their bioavailability for phytoplankton [14,15]. This dynamic alters the optical properties of water, increasing solar radiation absorption in the blue and ultraviolet spectral regions, with direct repercussions for photic zone depth and primary production [16-18].

In this context, this research addresses two key questions: (1) How does DOC influence the limnochemical characteristics of El Eneal wetland? and (2) What role do seasonal fluctuations in limnetic level play in these interactions? To answer these questions, in situ measurements of physical and chemical variables were combined with descriptive and multivariate analysis. The results will provide insight into the ecology of these unique systems and offer valuable information for their management and conservation over time.

2. Materials and methods

2.1 Study area

The El Eneal wetland is located in the municipality of San Onofre, Sucre Department, Colombia, between the coordinates W75° W76° and N9° N10° (Fig. 1). This water body, which is part of the Sanguare Civil Society Nature

Reserve (598 ha), is the result of the isolation of an ancient coastal lagoon, a process that began approximately fifty years ago. This transformation has led to the emergence of a limnetic aquatic environment, which in turn has favored the colonization of freshwater species and the subsequent disappearance of marine species [19,20].

The wetland, which covers an area of 36 hectares and has a perimeter of 4.6 km, is situated between 0 and 3 meters above sea level. The topography of the area is characterized by Pleistocene soils from the Magdalena formation and alluvial sediments [19,20]. Seismic studies have identified diapiric submarine structures that have been colonized by reefs in the area [21].

The climate of the region is characterized by an annual precipitation of 1000 mm, a mean temperature of 27°C, and a bimodal rainfall regime, with dry periods from December to April and from July to September, and rainy periods from April to May and from October to November [22,23]. The region is also subject to the influence of trade winds, which contribute an average wind speed of 3.3 meters per second.

2.2 Sampling stations and periods

Two sampling campaigns were conducted in February and December of 2024. These periods correspond to the high water (February) and low water (December) seasons of the system. Within these campaigns, 10 sampling points were selected using the Random Point tool of ArcGIS Pro software to ensure independence between samples and avoid bias in the collected data. It is noteworthy that the same sites were sampled during both campaigns (Fig. 1).

2.3 Field measurements

Samples were collected at a depth of 10 centimeters at all ten sampling sites. At each site, a comprehensive assessment of the physical and chemical variables was conducted. In situ measurements were performed using WTW 2FD47D and Hach HQ40D multiparametric equipment, both of which had been previously calibrated. Additionally, a HACH 2100Q turbidimeter was utilized for turbidity measurement and a 20-centimeter Secchi disk was employed to estimate water

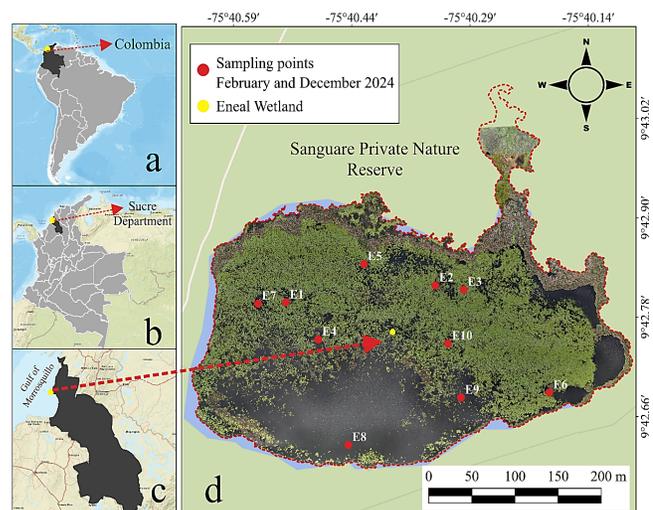


Figure 1. Study Area
Source: Own elaboration

Figure 2. *in situ* measurement of variables.

Source: Own elaboration

transparency. Samples intended for laboratory analysis were stored in sterilized bottles and preserved in thermal coolers at temperatures between 0 and 4°C until processing at the University of Antioquia laboratories (Fig 2).

Amber glass bottles with a capacity of 150 milliliters were utilized for the storage of DOC and TOC samples. For bicarbonates (HCO_3^-), carbonates (CO_3^{2-}), and sulfates (SO_4^{2-}), 250-ml HDPE bottles were utilized. The containers used for nutrients (NO_3^- , NO_2^- , NH_4^+ and PO_4^{3-}) were also 250 ml HDPE bottles. Finally, samples of chlorophyll-a were collected in 1000 ml amber plastic bottles.

Table 1 details the analyzed variables, differentiating those measured *in situ* from those determined in the laboratory, along with the equipment used and the methodologies applied.

Table 1.
Variables analyzed in this study.

Variable	Unit	Equipment	Methodology
Environmental Variables			
Ambient temperature	°C	Thermometer / meteorological station	IDEAM data
Precipitation	mm/month	Meteorological station	IDEAM data
Physical Variables			
Water temperature	°C	Multiparameter HQ40d	<i>In situ</i>
Turbidity	NTU	Turbidimeter Hach 2100Q	<i>In situ</i>
Transparency	m	Secchi disk	<i>In situ</i>
Water level	m	Limnimeter	<i>In situ</i>
Chemical Variables			
pH	pH units	Multiparameter WTW	<i>In situ</i>
Conductivity	µS/cm	Multiparameter WTW	<i>In situ</i>
Dissolved oxygen	mgO ₂ /L	Multiparameter WTW	<i>In situ</i>
Redox potential	mV	Multiparameter HQ40d	<i>In situ</i>
Chlorophyll-a	µg/L	Spectrophotometry	Laboratory
Ammonium (NH ₄ ⁺)	mgNH ₄ ⁺ /L	Photometer	<i>In situ</i>
Nitrite (NO ₂ ⁻)	mgNO ₂ /L	Photometer	<i>In situ</i>
Nitrate (NO ₃ ⁻)	mgNO ₃ /L	Photometer	<i>In situ</i>
Phosphate (PO ₄ ³⁻)	mgPO ₄ ³⁻ /L	Photometer	<i>In situ</i>
Sulfate (SO ₄ ²⁻)	mgSO ₄ /L	Ion chromatography (SM 4110-B)	Laboratory
Bicarbonate (HCO ₃ ⁻)	mgCaCO ₃ /L	Titrimetric (SM 2320-B)	Laboratory
Carbonate (CO ₃ ²⁻)	mgCaCO ₃ /L	Titrimetric (SM 2320-B)	Laboratory
Dissolved organic carbon (DOC)	mgDOC/L	SM 5310 B, Ed. 23, 2017	Laboratory
Total organic carbon (TOC)	mgTOC/L	ASTM D7573-18 ac1	Laboratory

Source: Own elaboration

2.4 Laboratory analysis

The determination of DOC and TOC was conducted at the Pollution Diagnostic Laboratory (GDECON) of the University of Antioquia in accordance with standardized protocols outlined in the Standard Methods for the Examination of Water and Wastewater [24]. The determination of bicarbonates (HCO_3^-), carbonates (CO_3^{2-}), and sulfates (SO_4^{2-}) was conducted at the Environmental Studies Laboratory (LEA) of the aforementioned university. These determinations were made using titrimetric techniques and ion chromatography. Finally, the concentration of chlorophyll-a was analyzed at the Sanitary Hydrobiology Laboratory of the University of Antioquia, following the spectrophotometric methodology proposed by Aguirre 2013 [25].

The determination of nutrients (NO_3^- , NO_2^- , NH_4^+ and PO_4^{3-}) was carried out at the Sanitary Hydrobiology Laboratory of the University of Antioquia. For this, samples were initially filtered through 0.45 µm Whatman GF/F glass fiber filters and analyzed using the Visicolor colorimetric kit, with readings performed on a PF-12 portable photometer.

2.5 Statistical analysis

Statistical analysis was conducted using R 4.4.3 software [26]. Initially, variables with observations below the quantification limit or, alternatively, those with more than 20% observations below the quantification limit, were excluded from the analysis. Subsequently, a descriptive analysis was performed on the remaining variables, calculating measures of central tendency (mean, median) and dispersion (standard deviation, interquartile range). Additionally, box plots were constructed to visualize and compare the variation of these variables during the evaluated periods.

For the multivariate analysis, a correlation matrix was initially constructed using Spearman's coefficient, given that many variables did not meet the normality assumption. This approach facilitated the identification of significant associations between DOC and limnological variables. Subsequently, variables with correlation coefficients greater than 0.6 or less than -0.6, and with a coefficient of variation exceeding 20%, were selected. Finally, with the variables that met both criteria, a principal component analysis (PCA) was performed with the objective of reducing the dimensionality of the data and analyzing their underlying structure.

3 Results

3.1 Water level

The water level of the wetland, measured monthly between February and December 2024, was linked to the bimodal rainfall regime of the region. The minimum level was recorded in April (1.40 m), while the maximum level was reached in December (3.10 m). Additionally, it was observed that the wetland's response to the increase in water level was delayed with respect to the maximum peak of precipitation recorded in October, as evidenced in Figure 3a.

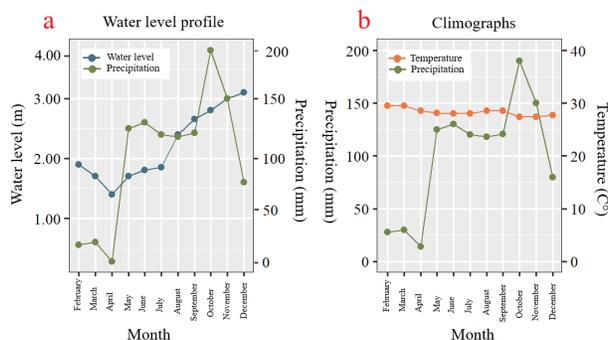


Figure 3. Water level profile and climograph for the year 2024. Source: Own elaboration

On the other hand, the climograph showed characteristic ambient temperature behavior for the region, where temperatures remained relatively high and constant throughout the year. Values fluctuated within a narrow range between approximately 28°C and 32°C, without presenting significant seasonal variations. It was observed that the warmest months corresponded to the January-February period, with a slight decrease during the intermediate months and a subtle increase towards September-October (Fig. 3b).

Table 2.

Summary statistics of the analyzed variables.

Variable	DOC	COT	HCO ₃ ⁻	pH	E.C	ORP	Turb	DO	PO ₄ ³⁻	Temp	Chl-a
February (n=8)											
Mean	12.77	14.43	101.48	7.22	850.75	195.36	6.91	3.85	0.26	31.54	0.54
Median	12.78	14.46	101.42	7.17	852	201.15	6.69	4.03	0.2	32	0.6
SD	0.29	0.52	0.67	0.15	4.40	52.22	0.69	1.55	0.11	1.37	0.29
Max.	13.31	15.39	102.42	7.42	855	266.8	7.79	5.52	0.5	32.9	1.04
Min.	12.3	13.6	100.7	7.02	843	128.3	6.18	1.56	0.2	29.3	0.15
December (n=10)											
Mean	11.81	12.72	116.29	6.87	443.1	481.65	3.98	3.02	0.54	32.59	1.48
Median	11.81	12.86	116.42	6.91	438	546.5	3.81	3.1	0.5	32.6	1.22
SD	0.26	0.37	1.46	0.13	10.76	222.47	0.6	1.45	0.16	0.86	0.46
Max.	12.19	13.09	118.39	7.02	468	748	4.79	4.87	0.8	34.5	2.38
Min.	11.45	12.15	113.84	6.53	437	160.6	3.27	1.07	0.3	31.4	1.19

Abbreviation: SD = Standard Deviation; E.C = Electrical Conductivity (µS/cm); Turb. = Turbidity; DO = Dissolved Oxygen; Temp. = Temperature; Chl-a = Chlorophyll-a; PO₄³⁻ = Phosphates.

Source: Own elaboration

3.2 Physical and chemical variables

Table 2 presents a summary of the descriptive statistics for variables that did not show values below the quantification limit during the months of February and December 2024. It is worth noting that, for February, only eight observations were analyzed, as two of the DOC data points were identified as outliers. The decision was made to eliminate all observations from this sampling point. The reason for these atypical values is that, when navigating through the wetland, sediment solids were resuspended, which affected the sample taken. In contrast, during the December sampling, environmental conditions were calmer and no sediment resuspension occurred, resulting in no outliers for that month.

DOC showed a decreasing trend between sampling periods. In February, a mean of 12.77 mg/L was recorded (range: 12.30-13.31 mg/L, SD=0.29), while in December it decreased to 11.81 mg/L (range: 11.45-12.19 mg/L, SD=0.26). TOC followed a similar pattern, reducing from 14.43 mg/L in February (SD=0.52) to 12.72 mg/L in December (SD=0.37), with less variability in the latter period (Fig. 4a and 4b).

Throughout the evaluated period, average concentrations were 12.24 mg/L for DOC and 13.48 mg/L for TOC, with low standard deviations (DOC: 0.56; TOC: 0.97). For DOC, 25% of the data were below 11.80 mg/L and 75% below 12.73 mg/L, while for TOC these percentiles were 12.80 mg/L and 14.36 mg/L, respectively (Fig. 4a and 4b).

On the other hand, bicarbonates (HCO₃⁻) showed behavior

opposite to that of organic and total carbon species, registering an increase in December. The lowest concentrations were observed in February, with a mean of 101.48 mg CaCO₃/L and an SD of 0.67, while in December they increased to reach a mean of 116.29 mg CaCO₃/L and a standard deviation of 1.46 (Fig. 4c). The mean bicarbonate concentration in the system was 109.71 mg CaCO₃/L with an overall SD of 7.66.

Regarding carbonates (CO₃²⁻), these remained at very low levels during both sampling periods, recording values below the detection limit (< 1.00 mg CaCO₃/L).

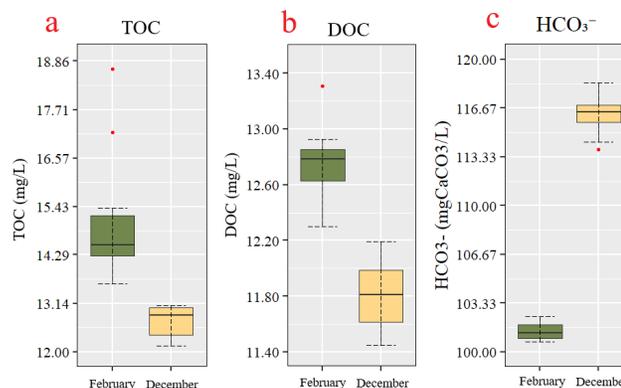


Figure 4. Carbon species. Source: Own elaboration

Electrical conductivity and oxidation-reduction potential (ORP) showed contrasting changes between both sampling periods. In February, the average electrical conductivity was 850.75 $\mu\text{S}/\text{cm}$, with an SD of 4.40. Meanwhile, in December, conductivity was reduced by almost half, recording a mean of 443.10 $\mu\text{S}/\text{cm}$, accompanied by an increase in data dispersion (SD = 10.76). On the other hand, ORP showed inverse behavior: in February, values were significantly lower, with a mean of 195.36 mV and relatively low variability (SD = 52.22). In December, ORP increased considerably, reaching an average of 481.65 mV, with a range oscillating between 160.60 mV and 748.00 mV, reflecting greater redox variability in December (SD = 222.47) (Fig 5a and 5b).

Water temperature and dissolved oxygen levels presented slight variations between both sampling periods. In February, water temperature ranged between 29.30°C and 32.90°C with a mean of 31.54°C and an SD of 1.37. This compared with December, when the mean rose to 32.59°C and the SD decreased to 0.86. Dissolved oxygen, meanwhile, exhibited opposite behavior: in February, the average value was 3.85 mg/L, with an SD of 1.55, while in December it decreased to 3.02 mg/L, with a slight reduction in variability (SD = 1.45). Overall, the system has average water temperatures of 32.12°C and dissolved oxygen levels of 3.39 mg/L. (Fig. 5c and 5d)

The turbidity of the system showed a decrease between February and December, consistent with transparency levels and total suspended solids (TSS), the latter measured only in December. In February, average turbidity was 6.91 NTU, with a standard deviation of 0.694. During December, these values decreased, reaching a mean of 3.98 NTU (SD = 0.60). In this same period, water transparency varied between 0.60 m and 0.75 m, while TSS remained between <5 mg/L and 15 mg/L. (Fig. 5e)

The pH showed a slight trend toward more alkaline values in February and more acidic in December, remaining near neutrality in both periods. In February, values ranged between 7.02 pH units and 7.42 pH units, with a mean of 7.22 pH units and a standard deviation of 0.15. In December, however, values were more acidic, with an average of 6.87 and an SD of 0.13 (Fig. 5f).

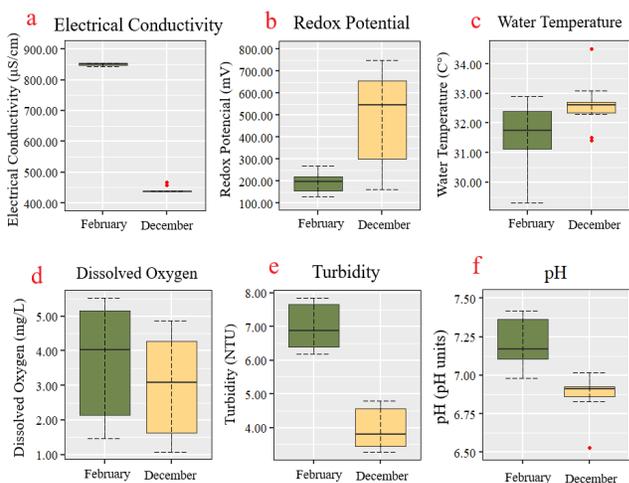


Figure 5. Physical and chemical variables. Source: Own elaboration

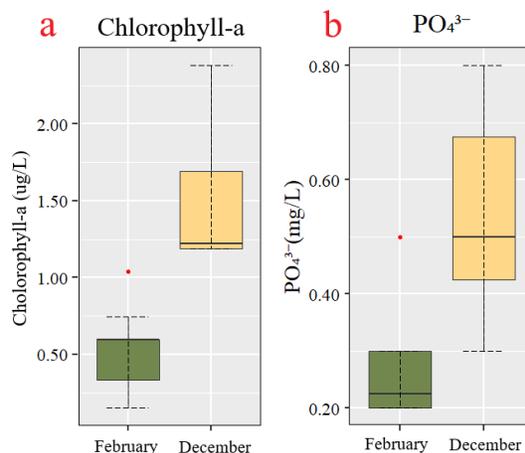


Figure 6. Chlorophyll and phosphates. Source: Own elaboration

The concentration of chlorophyll-a, an indicator of primary biomass, increased from February to December. In February, 25% of the data was below 0.33 $\mu\text{g}/\text{L}$ and 75% below 0.59 $\mu\text{g}/\text{L}$, with a mean of 0.54 $\mu\text{g}/\text{L}$. By December, the average rose to 1.48 $\mu\text{g}/\text{L}$ (SD = 0.46) (Fig. 6a).

Phosphorus (PO_4^{3-}) oscillated between 0.20 and 0.80 mg/L, with an average of 0.40 mg/L. In February, the mean was 0.26 mg/L (SD = 0.11), with relatively homogeneous variation among sampling stations. In December, the concentration nearly doubled (0.54 mg/L, SD = 0.16), with maximum values at E4 and E5 (0.7-0.8 mg/L respectively) and minimums at E1, E2, E7, and E9 (0.2 mg/L) (Fig. 6b).

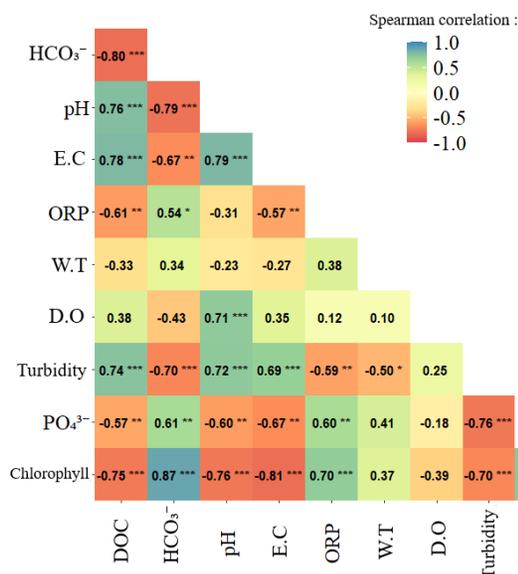
Regarding nitrogenous species, these exhibited different patterns of seasonal variation. For example, in February, nitrate (NO_3^-) concentrations ranged between 1.1 and 1.9 mg/L with a mean of 1.36 mg/L and SD of 0.21. Meanwhile, in December, all measurements remained below the detection limit (<1.0 mg/L). On the other hand, nitrites (NO_2^-) presented a mean of 0.012 mg/L and SD = 0.004 in February, and records for December were below the detection limit (<0.02 mg/L).

Ammonium (NH_4^+), for its part, showed a decrease between both periods. In February, average values were 0.79 mg/L (SD = 0.16), with some stations recording concentrations below the detection limit (< 0.40 mg/L). In December, all stations recorded levels below 0.5 mg/L.

3.3 Correlation matrix

After completing the descriptive analysis, a correlation analysis was conducted with all variables, except those that presented values below the quantification limit (QL) or, failing that, more than 30% of the data had records below the QL. Of these selected variables, more than 30% did not follow a normal distribution, so the Spearman coefficient was chosen to evaluate the degree of association between these variables.

Fig. 7 shows the results obtained from evaluating the degree of relationship between the different variables and



Note: The significance of the correlation is indicated by an asterisk (*) next to each coefficient: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Figure 7. Spearman correlation matrix.

Source: Own elaboration

their statistical significance. First, a strong negative association is observed between DOC and bicarbonate concentrations (HCO_3^-), with a correlation coefficient of -0.80 ($p < 0.001$), indicating that as bicarbonate levels increase, dissolved organic carbon concentrations tend to decrease.

pH showed a positive and significant correlation with DOC ($r = 0.76$, $p < 0.001$), suggesting that higher pH values are associated with higher DOC concentrations. Similarly, electrical conductivity showed a strong positive correlation with DOC ($r = 0.78$, $p < 0.001$), suggesting that waters with higher dissolved solute content present higher levels of dissolved organic carbon.

Turbidity also showed a strong positive correlation with DOC ($r = 0.74$, $p < 0.001$), implying that higher levels of suspended particles in the water are closely related to an increase in dissolved organic carbon concentrations.

On the other hand, oxidation-reduction potential (ORP) presented a moderate negative correlation with DOC ($r = -0.61$, $p < 0.01$), implying that more reducing conditions, characterized by low ORP values, are associated with an increase in dissolved organic carbon concentrations. Likewise, phosphates (PO_4^{3-}) showed a moderate negative correlation with DOC ($r = -0.57$, $p < 0.05$). This result suggests that an increase in phosphate concentrations is associated with a reduction in dissolved organic carbon levels. Finally, chlorophyll-a presented a strong negative correlation with DOC ($r = -0.57$, $p < 0.05$), indicating that higher chlorophyll concentrations, linked to photosynthetic activity and algal biomass, are related to lower dissolved organic carbon concentrations.

Regarding water temperature ($r = -0.33$) and dissolved oxygen ($r = 0.38$), these did not show significant correlations with DOC, indicating that a clear relationship cannot be established between both variables and DOC in this analysis.

Table 3.

Spearman correlation coefficients and coefficients of variation (CV) associated with COD.

Variable	Spearman	CV (%)
HCO ₃ ⁻	-0.8	7.08
pH	0.76	3.19
E.C.	0.78	32.3
ORP	-0.61	63.9
Water Temp	-0.33	3.64
DO	0.38	45.7
Turbidity	0.74	30.4
PO ₄ ³⁻	-0.57	47.1
Chlorophyll-a	-0.75	60.1

Source: Own elaboration

3.4 Principal component analysis

Table 3 presents the Spearman coefficient and coefficient of variation for each of the evaluated variables. These results show which variables meet the established criteria. In particular, the variables that satisfy these criteria are electrical conductivity (EC), oxidation-reduction potential (ORP), turbidity, and chlorophyll-a.

The PCA was performed with the selected variables. This identified a dimensional structure in the variation of the limnochemical variables of El Eneal wetland. The eigenvalues and cumulative variance showed that the first two principal components explained 88.55% of the total variability of the data. The first principal component (PC1) captured 77.59% of the variability, while the second component (PC2) added an additional 10.96%.

PC1 was primarily defined by electrical conductivity (EC), which showed a contribution of 23.34%, followed by turbidity, with a contribution of 21.49%, and chlorophyll, with 20.45%. DOC also contributed slightly to this component, with 17.21%, while ORP showed a similar contribution, with 17.52%. This component captures most of the variability in the data and represents a combination of all variables, with greater weight on EC, turbidity, and chlorophyll-a.

On the other hand, PC2 was marked by a high contribution from ORP, which explained 40.98% of the variance in this component, followed by DOC, with a contribution of 31.42%. Chlorophyll-a also contributed, albeit to a lesser extent, with 15.86%. This component highlights the relationship between ORP and DOC, suggesting a significant association between these two variables.

Additionally, \cos^2 values (quality of representation of variables in the components) indicate that electrical conductivity (EC) and turbidity are well represented in PC1, with values of 0.95 and 0.91, respectively. DOC also showed moderate representation in PC1, with a \cos^2 value of 0.67, and a positive correlation of 0.82 with this component. On the other hand, in PC2, DOC showed a correlation of 0.42, indicating a weaker but still significant relationship with this component. ORP, for its part, showed high representation in PC2, with a \cos^2 value of 0.68 and a correlation of 0.47, highlighting its importance in the structure of the second component.

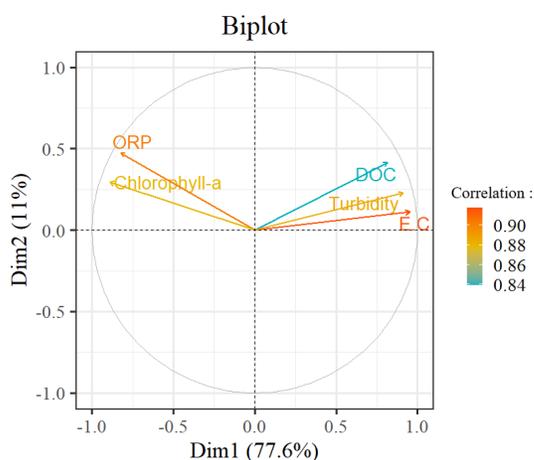


Figure 8. Biplot of Principal Component Analysis.
Source: Own elaboration

The graphical representation of the biplot (Figure 8) allowed visualization of the correlations between the limnochemical variables studied. DOC showed a strong positive correlation with electrical conductivity (EC), evidenced by the proximity and similar orientation of their vectors in the biplot. Additionally, a moderate positive correlation was observed with turbidity, in the angle of their vectors. On the other hand, DOC presented a moderate negative correlation with redox potential (ORP) and chlorophyll, indicated by the opposite position of their vectors in the graph.

Regarding the component equations, the first principal component (PC1) is dominated by positive contributions from electrical conductivity (EC), turbidity, and DOC, with loadings of 0.48, 0.46, and 0.41, respectively. On the other hand, chlorophyll-*a* and ORP show significant negative contributions, with loadings of -0.47 and -0.38, respectively, indicating an inverse relationship with this component (eq 1).

$$PC_1 = 0.41DOC - 0.48EC - 0.41ORP - 0.46Turbidity - 0.40Chl-a \quad (1)$$

The second principal component (PC2) is mainly influenced by ORP, which has a loading of 0.64, highlighting its central role in this component. DOC and turbidity also contribute, albeit to a lesser extent, with loadings of 0.56 and 0.30, respectively. Chlorophyll-*a* and electrical conductivity (EC) have contributions of 0.39 and 0.15, suggesting that this component is more related to redox processes in the system (eq 2).

$$PC_2 = 0.56DOC - 0.15EC - 0.64ORP - 0.30Turbidity - 0.3Chl-a \quad (2)$$

4 Discussion

4.1 Water level

The results obtained from the principal component analysis indicate that the dynamics of DOC in El Eneal wetland are primarily influenced by the variables that

dominate the first component (PC1), which explains 77.59% of the data variability. Despite DOC having a strong contribution to the variance of PC2, according to Kaiser's criterion, PC1 is the most relevant because it presents an eigenvalue greater than 1, while PC2 has a lower eigenvalue, suggesting that the first component should be the primary focus of analysis [27].

Within PC1, electrical conductivity emerged as the most influential variable, with a contribution of 23.34%. Its decreasing trend as water level increased suggests a dilution process in this endorheic system, a phenomenon previously documented by Ríos et al. (2008; 2009) [19,20], who noted how hydrological increase dilutes dissolved solutes, altering multiple physicochemical parameters.

This dilution process not only affects conductivity but also explains the synchronous behavior of DOC, evidenced by its strong correlation ($r = 0.78$, $p < 0.001$) and significant contribution (17.10%) to the variance of PC1. It is essential to highlight that this correlation does not indicate a direct causal relationship between electrical conductivity and DOC, but rather that both variables are responding independently, yet similarly, to the same factor: the increase in water volume that causes the simultaneous dilution of both parameters [9,12].

The strong positive correlation between turbidity and DOC ($r = 0.74$, $p < 0.001$) reinforces this interpretation, as when DOC is diluted during periods of higher water levels, its concentration decreases and, consequently, so does the water color. This occurs because DOC, particularly in the form of humic and fulvic substances, imparts dark hues to the water. During December, higher water levels notably improved transparency, as demonstrated by the low values of suspended solids (< 5 mg/L - 10 mg/L) and the consequent reduction in both turbidity and transparency.

4.2 DOC Mineralization

In addition to the dilution effect, our results point toward a possible increase in DOC mineralization during periods of higher water levels. This hypothesis is based on the significant inverse correlation between DOC and ORP ($r = -0.61$, $p < 0.001$). The increase in water level in December, along with the greater presence of vegetation, appears to have generated more oxidative conditions that favored the microbial decomposition of DOC and the consequent release of CO_2 [28,29].

This interpretation is reinforced by the inverse correlation between DOC and bicarbonates ($r = -0.80$, $p < 0.001$). Although the latter were not included in the PCA due to their low coefficient of variation ($< 20\%$), their increase during periods of higher water levels suggests that the CO_2 released during DOC decomposition is transformed into bicarbonates. This transformation process aligns with previous research on organic matter mineralization in similar aquatic systems [30,31].

However, this hypothesis should be approached with caution, as the formation of bicarbonates could also be influenced by other factors, such as the near-neutral pH (6.7-7.2) that characterizes the El Eneal wetland. These conditions allow the CO_2 released from DOC degradation to rapidly

transform into bicarbonates (HCO_3^-), which would explain the absence of carbonates (CO_3^{2-}) and the predominance of the bicarbonate form in the system [30,31].

4.3 Biological processes

The biological processes in the system appear to be closely conditioned by the concentration of DOC. In the principal component analysis, chlorophyll-a showed a high contribution to the first component (20.45%) and a significant negative correlation with DOC ($r = -0.75$, $p < 0.001$), suggesting that a decrease in DOC concentration associated with dilution processes favors phytoplankton growth. This effect is reinforced by the inverse relationship between chlorophyll-a and turbidity, as the lower presence of suspended particles allows greater light penetration and, consequently, improved photosynthetic activity, as also noted by Ríos et al. [19,20].

Furthermore, Jackson and Hecky (1980) [33] highlighted that DOC can form complexes with essential trace elements such as iron, reducing their bioavailability and limiting phytoplankton metabolism. Similarly, previous studies [3,16] have reported that DOC can bind to key nutrients like phosphorus and nitrogen, decreasing their availability to photosynthetic organisms.

Additionally, various authors [34-36] have indicated that, at high concentrations, DOC acts as an absorbent of photosynthetically active radiation (PAR), significantly reducing light penetration in the water column. This light limitation directly affects photosynthesis and, consequently, phytoplankton development.

Collectively, these mechanisms could explain the low productivity recorded in the system (25–27 mgC/h/m² in February and December respectively) by Macías et al. 2024 [37], a condition that, as documented by Ríos et al. [19,20], has been persistent in the wetland, which is classified as oligoproductive. For example, high DOC concentrations darken the water, reducing light penetration and limiting photosynthetic activity, which in turn constrains phytoplankton growth. Therefore, DOC dynamics emerges as a regulating factor of key ecological processes, although further experimental studies are needed to validate these hypotheses.

4.4 Nutrients

On the other hand, nutrient dynamics did not show significant relationships with DOC, except for phosphorus, which presented a significant inverse relationship ($r = -0.75$, $p < 0.001$). This relationship has been interpreted by various authors as a consequence of DOC's ability to bind to phosphorus compounds, forming stable associations that limit its availability in the aquatic environment. This interaction is particularly relevant in systems where DOC concentrations are high, as this compound acts as a "sink" for nutrients, reducing the bioavailable fraction for biological processes. In this case, the decrease in DOC in December could have facilitated the release of phosphorus into the water column, contributing to the increase in its concentration during this period [3,16].

Although nitrogen species did not show a significant correlation with DOC, this could be influenced by limitations in the detection of these variables. Many of the measurements were below the quantification limit, making it difficult to identify possible relationships. This aspect highlights the need to employ analytical methods with lower quantification limits for a more precise assessment of the interaction between DOC and nitrogen forms. In previous studies where DOC has been the response variable, associations with nitrogen species have been found, suggesting that this relationship could exist but may not have been detected in this case due to methodological restrictions.

4.5 Unrelated variables

No significant relationships were found between DOC and dissolved oxygen ($r = 0.38$) or water temperature ($r = -0.33$), variables that were also not prominent in the PCA. While these results suggest an absence of direct influence, scientific literature indicates that DOC can affect water temperature through infrared radiation absorption. Rüdiger et al. [38], Bonelli et al. [39], and Sahay et al. [40] have documented how chromophoric dissolved organic matter (CDOM) absorbs light, contributing to water warming and modulating thermal transfer. The lack of correlation observed may be attributed to methodological limitations of this preliminary study, including a reduced number of observations (only two periods) and potential measurement inaccuracies due to temperature effects on multiparametric equipment. Implementation of the Winkler method for dissolved oxygen determination could provide more accurate measurements and facilitate a more robust assessment of its relationship with DOC.

5 Conclusions and recommendations

This research determined that DOC influences the limnological characteristics of El Eneal wetland through dilution processes and optical alterations of water. During high water periods, the increase in water level dilutes DOC, electrical conductivity, and turbidity, generating synchronous behavior among these variables. This suggests that their correlations depend primarily on water volume rather than direct interactions. Additionally, an inverse correlation between DOC and bicarbonates was identified, suggesting a mineralization process during the water level increase phase. Similarly, an inverse relationship with phosphorus was observed, indicating possible adsorption mechanisms, likely associated with DOC's capacity to form organic complexes with this nutrient. On the other hand, no significant relationship was found between DOC and nitrogenous compounds. The negative correlation between DOC and chlorophyll-a suggests that DOC reduces light availability for phytoplankton due to absorption and scattering of solar radiation, directly affecting its productivity. This effect is supported by the decrease in primary productivity of this system. As a recommendation, it is proposed to expand the study with a greater number of samplings and include additional variables, such as trace metals, specific optical parameters (e.g., absorbance and

fluorescence of organic matter), and dissolved organic matter fractions (e.g., humic substances), in order to deepen understanding of DOC's role in the dynamics of this ecosystem.

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- W.D. Guzman-López**, received the BSc. Eng. in Environmental Engineering in 2020, from the Universidad de Antioquia, Colombia. Currently, he is a Master's student in Environmental Engineering and a part-time lecturer at the Faculty of Engineering, Universidad de Antioquia. His research focuses on water quality, wetland ecosystems, and dissolved organic carbon (DOC) dynamics. ORCID: 0009-0007-4494-1231
- L.C. Giraldo**, received the BSc. and MSc. in Environmental Engineering, and the PhD in Engineering, all of them from the Universidad de Antioquia, Colombia. She is a full-time faculty member at the Faculty of Engineering, Universidad de Antioquia. Her research interests include water treatment, aquatic ecology, and environmental management. ORCID: 0000-0002-1801-6612
- F.V. Macías**, received the BSc. and MSc. in Environmental Engineering from the Universidad de Antioquia, Colombia, and the PhD in Geography from Heinrich-Heine-Universität Düsseldorf, Düsseldorf, Germany. He is a retired faculty member of the Faculty of Engineering, Universidad de Antioquia. His research experience includes water quality, environmental modeling, and ecological restoration. ORCID: 0000-0001-6348-6405
- N. Aguirre**, received the BSc. and MSc. in Biology from the Universidad de Antioquia, Colombia. PhD in Natural Resources, Universität Giessen, Germany. He is a full-time faculty member at the Faculty of Engineering, Universidad de Antioquia. His research focuses on aquatic ecosystems, water quality, and environmental engineering. ORCID: 0000-0002-0847-7335