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# Association between the alga *Spirogyra* (Zygnemataceae) and heavy metals in sediments of the Chi River Basin in Northeast Thailand

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**ABSTRACT**

**Introduction:** Sediments act as major sinks of heavy metals from human activities, while algae such as *Spirogyra* interact with these sediments through nutrient uptake and biogeochemical cycling. In the Chi River Basin, *Spirogyra* ("Tao") plays both ecological and socioeconomic roles, serving as food and a bioindicator of pollution.
**Objective:** To evaluate heavy-metal concentrations in sediments and *Spirogyra* and assesses their potential human health risks.
**Methods:** Water quality, *Spirogyra*, and sediment samples from the Chi River Basin were analyzed for heavy metals using ICP-OES, with human health risks assessed by *THQ* and *HI*, and sediment contamination evaluated through *Igeo*, *EF*, *CF*, *PLI* indices.
**Results:** Heavy metals (Cd, Cu, Fe, Mn, Zn) in sediments and *Spirogyra* from the Chi River Basin were assessed under average water conditions (pH 7.49, EC 497 $\mu S\ cm^{-2}$, NaCl 0.024 %, 24.9 ± 3.52 °C), showing concentrations in algae (Fe > Mn > Zn > Cu > Cd) with *THQ* and *HI* < 1 indicating low health risk, while sediments (Mn > Fe > Zn > Cu > Cd) exhibited *Igeo* < 0, *EF* < 2, *CF* < 1 except for Fe (*CF* = 8.31), *PLI* < 1, and significant correlations between Mn in algae with Cu and Mn in sediments, Cu with Fe in algae, and Cd in algae with Fe in sediments.
**Conclusions:** The findings indicate no significant heavy-metal pollution in sediments or *Spirogyra* from the Chi River Basin. However, due to elevated Fe contamination in sediments, periodic monitoring is recommended to safeguard ecological and food safety.

**Key words:** *Spirogyra*; sediment; heavy metals; Chi River Basin.

**RESUMEN**

**Asociación entre el alga *Spirogyra* (Zygnemataceae) y metales pesados en sedimentos de la cuenca del río Chi en el noreste de Tailandia**

**Introducción:** Los sedimentos actúan como sumideros principales de metales pesados provenientes de actividades humanas, mientras que algas como *Spirogyra* interactúan con estos sedimentos a través de la absorción de nutrientes y el ciclo biogeoquímico. En la cuenca del río Chi, *Spirogyra* ("Tao") cumple funciones tanto ecológicas como socioeconómicas, sirviendo como alimento y bioindicador de contaminación.

**Objetivo:** Evaluar las concentraciones de metales pesados en sedimentos y *Spirogyra*, y analiza sus posibles riesgos para la salud humana.
**Métodos:** Se analizaron la calidad del agua, *Spirogyra* y sedimentos de la cuenca del río Chi para detectar metales pesados mediante ICP-OES; los riesgos para la salud humana se evaluaron mediante *THQ* y *HI*, y la contaminación de los sedimentos se determinó con los índices *Igeo*, *EF*, *CF* y *PLI*.
**Resultados:** Los metales pesados (Cd, Cu, Fe, Mn, Zn) en sedimentos y *Spirogyra* de la cuenca del río Chi fueron evaluados bajo condiciones promedio del agua (pH 7.49, CE 497 µS cm-2, NaCl 0.024 %, 24.9 ± 3.52 °C), mostrando concentraciones en algas (Fe > Mn > Zn > Cu > Cd) con *THQ* y *HI* < 1 que indican bajo riesgo para la salud, mientras que los sedimentos (Mn > Fe > Zn > Cu > Cd) presentaron *Igeo* < 0, *EF* < 2, *CF* < 1 excepto para Fe (CF = 8.31), *PLI* < 1, y correlaciones significativas entre Mn en algas con Cu y Mn en sedimentos, Cu con Fe en algas, y Cd en algas con Fe en sedimentos.
**Conclusiones:** Los resultados indican que no existe una contaminación significativa por metales pesados en los sedimentos ni en *Spirogyra* de la cuenca del río Chi. Sin embargo, debido a la elevada contaminación por Fe en los sedimentos, se recomienda un monitoreo periódico para proteger la seguridad ecológica y alimentaria.

**Palabras clave:** *Spirogyra*; sedimento; metales pesados; Cuenca del río Chi

## INTRODUCTION

Sediments in water bodies contain the bulk of the heavy metals (HMs) produced by human activities (Guan et al., 2018; Liber et al., 2019; Sojka & Jaskuła, 2022). Fluvial processes (Foster & Charlesworth, 1996; Miller, 1997; Rahman et al., 2022; Schulte et al., 2024) and the decomposition of algae are involved in the biogeochemical cycles of HMs in the sediment (Ni et al., 2019; Wang et al., 2023; Xue et al., 2024). A variety of species, including algae, use silt as a source of nutrients (Park & Hwang, 2010; Xiang et al., 2023; Zhao et al., 2022), and thus may influence such cycles by removing HMs from the sediment (Machado et al., 2024; Sarma et al., 2024; Zada et al., 2021).

Algae are a diverse group of photosynthetic eukaryotes (Friedl et al., 2011; Kaštovsky et al., 2019) that have multiple uses, including as alternative energy sources (Hannon et al., 2010; Mohite et al., 2024; Neeti et al., 2023), in water treatment (Abdel-Raouf et al., 2012; Bhatt et al., 2022; Molinuevo-Salces et al., 2019), and in industry, especially in food production such as *Undaria pinnatifida*, *Wolffia globosa*, *Spirogyra* etc., (Matos et al., 2022; Naik et al., 2024). Algae are high in protein, vitamins, lipids, minerals, and essential fatty acids (de Oliveira & Bragotto, 2022; Kent et al., 2015; Wu et al., 2023).

The people inhabiting the Chi River Basin use algae of the genus *Spirogyra* (Phylum Chlorophyta, Family Spirogyraceae), known locally as *Tao*, as a commodity. *Spirogyra* can also be used as a bioindicator of pollution, including the HMs that can contaminate the water and sediments in reservoirs (Rajfur et al., 2010; Shing et al., 2018; Wijaya et al., 2025). The analysis of pertinent elements in ecosystems (Ewing & Weathers, 2021; Lobus & Kulikovskiy, 2023; Spohn et al., 2021) includes the interactions between HMs in sediments and algae. Understanding these relationships in ecosystems allows for better environmental management. In this study, we aimed to assess the hazards to human health posed by specific HMs in the sediments and *Spirogyra* of the Chi River basin in Thailand.

## MATERIAL AND METHODS

**Water conditions and study sites:** The basic data characterizing the aquatic environments at the study sites are provided in Table 1. The pH, electrical conductivity, percentage of sodium chloride (NaCl) in the water, and the water temperature were measured, and the sample locations were recorded as Universal Transverse Mercator (UTM) coordinates. Fig. 1 shows the area of the river basin. The types of


**Fig. 1.** Sample Locations in the Chi River Basin.

**Table 1**
Water characteristics and types of water bodies at the study sites.

| Location | Water Characteristics | | | | Coordinates (UTM) | | | Water Body |
|---|---|---|---|---|---|---|---|---|
| | pH | EC (µS cm$^{-2}$) | % NaCl | Temperature | Northing | Easting | Zone | |
| 1 | 7.97 | 495 | 0.05 | 27.3 °C | 809847 | 1727556 | 47P | Lake |
| 2 | 7.67 | 194 | 0 | 23.7 °C | 818007 | 1836476 | 47Q | Canal |
| 3 | 7.4 | 525 | 0.02 | 29.5 °C | 267554 | 1805080 | 48Q | Paddy Field |
| 4 | 7.78 | 1.083 | 0.05 | 19.1 °C | 290743 | 1809612 | 48Q | Lake |
| 5 | 6.66 | 188 | 0 | 25.2 °C | 422056 | 1714464 | 48P | Canal |
| Average | 7.49 | 497 | 0.024 | 24.9 °C | -- | -- | -- | -- |
| SD | 0.456 | 326 | 0.022 | 3.52 | -- | -- | -- | -- |

Note: EC = electrical conductivity; % NaCl = percentage of sodium chloride (NaCl) in water soluble; SD = standard deviation.

water bodies the *Spirogyra* samples were collected from are listed in Table 1. However, the environmental conditions, including pH, electrical conductivity (EC), and salinity, influence the accumulation of toxins by altering their chemical properties and bioavailability to living organisms. They also influence the health and functionality of living organisms regarding toxin buildup.

***Spirogyra* collection:** The *Spirogyra* samples were scooped from the water by hand, after which they were cleaned with pure water, packed in plastic zipper bags, and kept at -4 °C before being sent to the laboratory. In the laboratory, the alga were air-dried for five h, then dried at 60 °C in a hot oven for 120 h. The water content of the *Spirogyra* was calculated to have been around 91.9 ± 0.730 %. The dried samples were ground and sifted through a Nº. 20 sieve, then collected in polyethylene tubes before being maintained in the refrigerator at a temperature of 4 °C.

**Algal extraction and analysis of heavy metals:** The algae were prepared for extraction by mixing 0.1 mol $l^{-1}$ of nitric acid ($HNO_3$) and 0.1 mol $l^{-1}$ of sodium hydroxide (NaOH) in a 50 ml volume with 1 g of sample in 100 ml conical flasks. The flasks were agitated at 200 rpm for 6 h (Mehta & Gaur, 2005; Sheng et al., 2004). The solution was then filtered. The HM analysis was performed on the residue using inductively coupled plasma atomic emission spectroscopy (ICP-OEM) (PlasmaQuant 9100 series, Germany). Quality assurance and quality control procedures were performed on all 20 samples, as well as the duplicates and blanks. The samples were compared against multi-element standard solutions from AccuStandard (USA).

**Algae and health risk indicators:** From the algal samples, the target hazard quotient (*THQ*) and hazard index (*HI*) (or sum of the *THQs*) (Guerra et al., 2012; Javed & Usamni, 2016; Thummajitsakul et al., 2018) were calculated to assess the risk to human health. A *THQ* of < 1 indicates little chance of the exposed person experiencing any adverse effects. The *THQ* was calculated as follows:

$$THQ = \frac{(E_F \times E_D \times F_{IR} \times C) \times 10^{-3}}{(RFD \times WAB \times TA)} \quad (1)$$

where $E_F$ is the exposure frequency from the consumption of HMs in vegetables (365 days $year^{-1}$), with the assumption that the *Spirogyra* is being eaten in the same way as fresh vegetables; $E_D$ is the exposure duration during the average lifetime of a Thai citizen (i.e., 75.2 years for adult males and 81.3 years for adult females) (Thailand Board of Investment, 2025); $F_{IR}$ is the food ingestion rate of a Thai citizen, equal to 0.268 kg $person^{-1}$ $day^{-1}$ for adult males and 0.283 kg $person^{-1}$ $day^{-1}$ for adult females (Thummajitsakul et al., 2018); *C* is the quantity of HMs in the algae (mg $kg^{-1}$); *RFD* is the oral references dose, which is 0.001 mg $kg^{-1}$ $day^{-1}$ for cadmium (Cd), 0.04 mg $kg^{-1}$ $day^{-1}$ for copper (Cu), 0.7 mg $kg^{-1}$ $day^{-1}$ for iron (Fe), 0.14 mg $kg^{-1}$ $day^{-1}$ for manganese (Mn), and 0.3 mg $kg^{-1}$ $day^{-1}$ for zinc (Zn) (Thummajitsakul et al., 2018; U.S. Environmental Protection Agency, 1988); *WAB* is the average body weight of a Thai citizen (68.83 kg for adult males and 57.4 kg for adult females) (Thummajitsakul et al., 2018); and $T_A$ is the average exposure time for HMs ($E_D \times 365$ days $year^{-1}$).

The *HI* was used to estimate the overall potential health risk from more than one HM, based on the following:

$$HI = THQ_{Cd} + THQ_{Cu} + THQ_{Fe} + THQ_{Mn} + THQ_{Zn} \quad (2)$$

where $HI < 1$ indicates no significant health risk.

**Sediment collection and extraction:** Approximately 5 kg of sediment was collected at each site, placed in plastic bags, and kept in a box for transport from the field to the laboratory, where the sediment was dried at 105 °C in a hot oven for 120 h, after which it was ground using a mortar and pestle. After being passed through a Nº. 4 (10 mm) sieve, the sediment was extracted for use in ICP-OES analysis. For this, 2 g of each prepared soil sample was mixed with concentrated hydrofluoric acid (HF), concentrated perchloric acid ($HClO_4$), and concentrated $HNO_3$ at a ratio of 1 : 1 : 1 for a total volume of 20 ml. Extraction was performed at 500 °C in a SpeedDigester K-425 BUCHI instrument (Switzerland). Each residue was rinsed with 1 % $HNO_3$ and passed through filter paper. The supernatant was transferred to a 50 ml volumetric flask, and 1 % $HNO_3$ was added for continued ICP-OES analysis (PlasmaQuant 9100 series, Germany). The quality assurance and quality control procedures ensured that all 20 samples, as well as duplicates and blanks, were collected, processed, and examined to standard. The samples were compared against ICP-OEM multi-element standard solutions (AccuStandard, USA).

**Sediment indicators:** The quality of the sediment was assessed by several indicators: the geo-accumulation index ($I_{geo}$), the enrichment factor (*EF*), and the contamination factor (*CF*). The $I_{geo}$ was originally formulated by Muller (1980) and is a quantitative measure of pollution in aquatic sediments (Kroeksakul et al., 2023; Nobi et al., 2010), developed through an understanding of the lithogenic effect, and calculated using the following formula:

$$I_{geo} = log2\ [C_n / 1.5B_n] \quad (3)$$

where $C_n$ is the measured concentration of an element in the sediment; and $B_n$ is the background value of the element. The interpreted values of $I_{geo}$ are: < 0 = not polluted; 0-1 = not polluted to moderately polluted; 1-2 = moderately polluted; 2-3 = moderately to strongly polluted; 3-4 = strongly polluted; 4-5 = strongly to extremely polluted, and > 5 = extremely polluted.

The *EF* was calculated in the following way:

$$EF = (\mathrm{C} / \mathrm{RE})\ \text{sample} / (\mathrm{C} / \mathrm{RE})\ \text{background} \quad (4)$$

where (C/RE) sample (background) is the concentration (C) of an element compared to a reference element (RE) in the sample (background). Aluminum (Al) was used as the reference element because it is a major component of clay. The interpreted values of the *EF* are: < 2 = deficiency in mineral enrichment; 2-5 = moderate enrichment; 5-20 = significant enrichment; 20-40 = very high enrichment; and > 40 = extremely high enrichment.

The *CF* was approximated as the observed concentration of an element in the sample (*Ci*) to the background level of the same element (*Cb*), as follows:

$$CF = Ci / Cb \quad (5)$$

The interpreted values of the *CF* are: < 1 = low pollution level; 1-3 = moderate pollution level; 3-6 = considerable pollution level; and > 6 = very high pollution level. The background element values for the $I_{geo}$, *EF*, and *CF* calculations were taken from reference data for Al, Cu, Fe, and Zn in Potipat et al. (2015). The Cd background references were obtained from the Land Development Department (2012), and the Mn background references were from Czarnowska and Gworek (1990). The *CFs* were used to calculate the pollution load index (PLI) that was used to measure the general contamination level, as follows:

$$PLI = \ \text{x}\ CF_2\ \text{x}\ CF_3 \ldots .CF_n \quad (6)$$

where *n* is the number of observed elements. The interpreted values of the *PLI* are: 0 = unpolluted; < 1 = baseline level for no pollution; and > 1 = polluted.

**Statistical analysis**: The data were analyzed using one-way analysis of variance, and the differences between datasets were compared using a least significant difference (LSD) test with p < 0.05. The correlations were assessed using Pearson's correlation coefficient (r). All analyses were conducted using Statistical Package for the Social Sciences (SPSS) v.22 software.

## RESULTS

**Heavy metal content in *Spirogyra*:** The HM contents determined in the *Spirogyra* samples are listed in Table 2. The Cd and Cu contents in the algae from Site 5 were significantly greater than those from Sites 1, 2, and 3 (p < 0.05), and the Fe content in the algae from Site 5 was significantly greater than at Sites 1, 2, and 3 (p < 0.05). The Mn content in the algae from Site 5 was less than at Sites 1 and 2, and the Zn content in algae. The HM contents in the *Spirogyra* samples were mostly Fe (about 80 %) (Fig. 2).

**Human health risk assessment:** The people of the Chi River Basin use *Spirogyra* to make a salad the locals call "Laab Tao", and this may become useful as a food resource in the future. The *THQs* occurred in the order Mn > Fe > Cu > Zn > Cd for males and Cu > Fe > Mn > Zn > Cd for females (Table 3). The THQ values were < 1, and the *HI* value for the adult males averaged $5.490 \pm 1.512 \times 10^{-4}$ higher than the adult female average of $122 \pm 63.9 \times 10^{-4}$. The HI values for the adult males and females were both < 1, indicating no significant risk to human health.

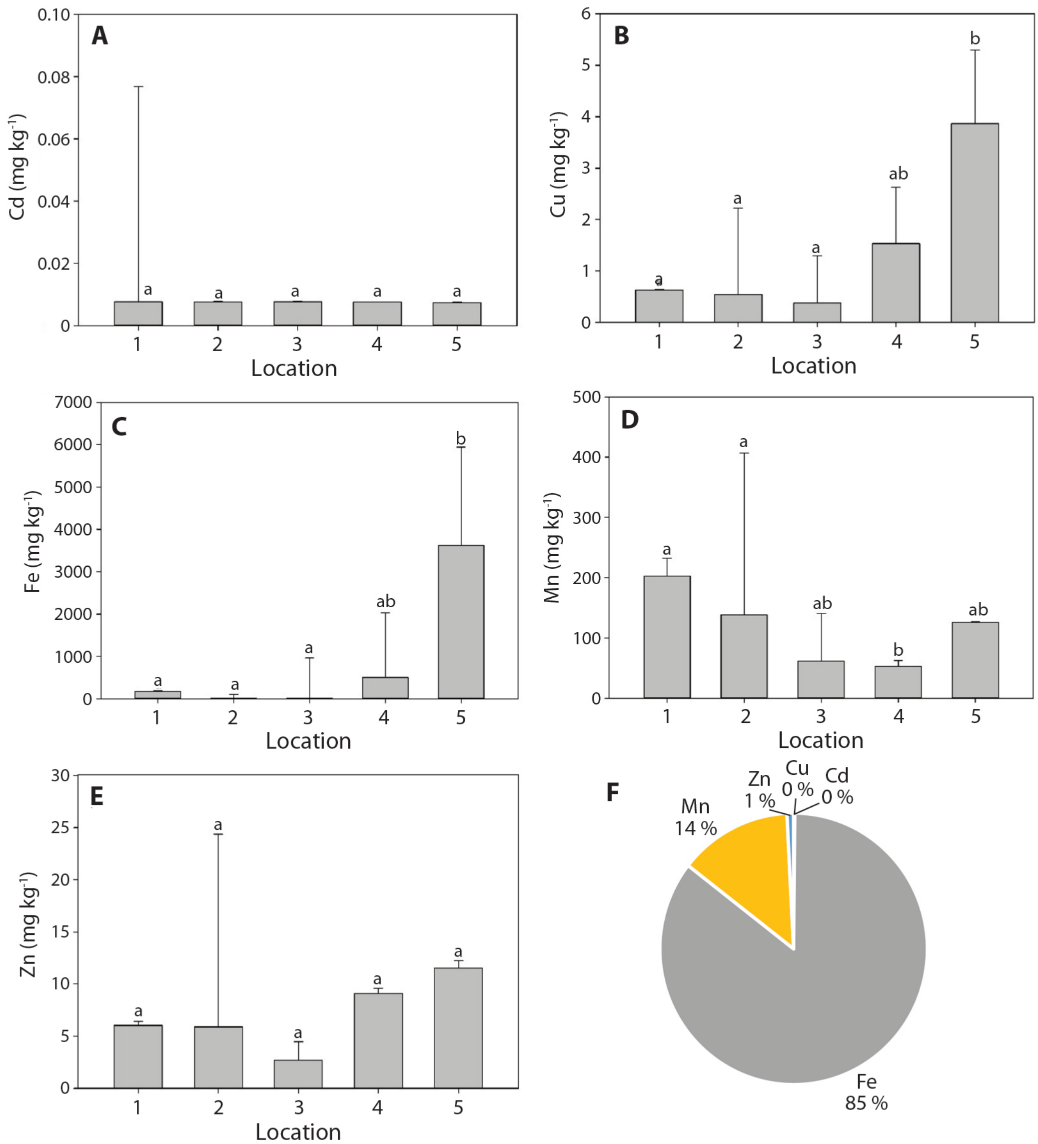


**Fig. 2.** The HM content in *Spirogyra* by site. **A.** Cd, **B.** Cu, **C.** Fe, **D.** Mn, **E.** Zn, **F.** Percentage of five HMs in *Spirogyra*.

**Heavy metals in the sediments:** In the sediment samples, Cd averaged 0.005 ± 0.000 mg $kg^{-1}$ dry weight, and the amount from Site 2 (0.004 ± 0.000 mg $kg^{-1}$) was significantly less ($p < 0.05$) than at Sites 1, 3, 4, and 5. The Cu content from Site 4 averaged 0.713 ± 0.026 mg $kg^{-1}$, which was significantly less ($p < 0.05$) than at Sites 1 and 2. At all sites, the Fe and Mn contents were both significant ($p < 0.05$). The Zn content at Site 3 averaged 36.0 ± 16.0 mg $kg^{-1}$ and was significantly higher ($p < 0.05$) than at the other sites. The HM contents in the sediments are presented in Table 4 and illustrated in Fig. 3. The Mn content was about 59 % of the total HMs in the sediments, and Fe was the most abundant HM in the algae (Fig. 3F).

**Table 2**
Heavy metal contents in *Spirogyra* in the Chi River Basin.

| Location | Heavy Metal Dry Weight in *Spirogyra* (mg kg$^{-1}$) | | | | |
|---|---|---|---|---|---|
| | Cd | Cu | Fe | Mn | Zn |
| Site 1 | 0.030(± 0.039)$^{a}$ | 0.626(± 0.015)$^{a}$ | 175(± 6.91)$^{a}$ | 207(± 21.1)$^{a}$ | 6.09(± 0.263)$^{a}$ |
| Site 2 | 0.007(± 0.000)$^{a}$ | 1.09(± 0.981)$^{a}$ | 37.2(± 49.4)$^{a}$ | 222(± 160)$^{a}$ | 11.6(± 11.0)$^{a}$ |
| Site 3 | 0.007(± 0.000)$^{a}$ | 0.670(± 0.538)$^{a}$ | 326(± 546)$^{a}$ | 86.9(± 43.3)$^{ab}$ | 2.63(± 1.58)$^{a}$ |
| Site 4 | 0.007(± 0.000)$^{a}$ | 1.65(± 0.842)$^{ab}$ | 900(± 6.91)$^{ab}$ | 55.7(± 5.52)b | 8.97(± 0.539)$^{a}$ |
| Site 5 | 0.007(± 0.000)$^{a}$ | 3.13(± 1.87)$^{b}$ | 2.920(± 2.616)$^{b}$ | 125(± 0.463)$^{ab}$ | 11.4(± 0.759)$^{a}$ |
| Average | 0.012(± 0.017) | 1.43(± 1.30) | 872(± 1541) | 139(± 92.9) | 8.13(± 5.50) |

Note: $^{a, b}$ Difference is significant at $p < 0.05$ (LSD); Cd = cadmium; Cu = copper; Fe = iron; Mn = manganese; Zn = zinc.

**Table 3**
*THQs* and *HIs* for *Spirogyra* as a measure of heavy metals contamination in male and female adults (Aged over 15 years).

| Location | *THQ* (mean ± SD) | | | | | *HI* |
|---|---|---|---|---|---|---|
| | Cd (x $10^{-4}$)* | Cu (x $10^{-4}$)* | Fe (x $10^{-4}$)* | Mn (x $10^{-4}$)* | Zn (x $10^{-4}$)* | (mean ± SD) (x $10^{-4}$)* |
| **Male** | | | | | | |
| Site1 | 1.20(± 1.27) | 0.61(± 0.01) | 9.79(± 0.31) | 8.095(± 673) | 0.79(± 0.03) | 8.108(± 672) |
| Site2 | 0.29(± 0.01) | 1.06(± 0.78) | 2.07(± 2.25) | 8.644(± 5.095) | 1.51(± 1.17) | 8.649(± 5,099) |
| Site3 | 0.30(± 0.00) | 0.65(± 0.43) | 18.1(± 24.8) | 3.385(± 1.473) | 0.34(± 0.17) | 3.405(± 1,499) |
| Site4 | 0.29(± 0.00) | 1.61(± 0.67) | 50.0(± 44.5) | 2.169(± 175) | 1.16(± 0.06) | 2.222(± 167) |
| Site5 | 0.28(± 0.01) | 3.05(± 1.49) | 162(± 118) | 4.900(± 14.7) | 1.48(± 0.08) | 5.067(± 123) |
| Average | 0.47(± 0.26) | 1.40(± 0.68) | 48.5(± 38.1) | 5.438(± 1,486) | 1.06(± 0.30) | 5.490(± 1,512) |
| **Female** | | | | | | |
| Site1 | 1.51(± 1.61) | 12.39(± 0.40) | 73.2(± 6.09) | 7.51(± 0.26) | 1.00(± 0.04) | 95.6(± 4.48) |
| Site2 | 0.37(± 0.01) | 2.62(± 2.84) | 78.1(± 46.0) | 14.3(± 11.1) | 1.91(± 1.48) | 97.3(± 61.5) |
| Site3 | 0.38(± 0.00) | 23.0(± 31.4) | 30.6(± 13.3) | 3.24(± 1.60) | 0.43(± 0.21) | 57.7(± 46.2) |
| Site4 | 0.37(± 0.00) | 63.4(± 56.4) | 19.6(± 1.59) | 11.0(± 0.54) | 1.47(± 0.07) | 95.9(± 56.1) |
| Site5 | 0.36(± 0.01) | 205(± 150) | 44.3(± 0.13) | 14.0(± 0.76) | 1.87(± 0.10) | 266(± 151) |
| Average | 0.60(± 0.33) | 61.4(± 48.3) | 49.1(± 13.4) | 10.0(± 2.86) | 1.34(± 0.38) | 122(± 63.9) |

Note: *x $10^{-4}$ means the value in the table must be divided by 10 000 to obtain the actual value; Cd = cadmium; Cu = copper; Fe = iron; Mn = manganese; Zn = zinc.

**Table 4**
Heavy metal contamination in the sediment by study site.

| Location | Heavy metal dry weight in sediment (mg kg$^{-1}$) | | | | |
|---|---|---|---|---|---|
| | Cd | Cu | Fe | Mn | Zn |
| Site1 | 0.005(± 0.000)$^{a}$ | 2.07(± 0.228)$^{a}$ | 725(± 23.6)$^{a}$ | 857(± 30.5)$^{a}$ | 5.64(± 0.114)$^{a}$ |
| Site2 | 0.004(± 0.000)b | 2.72(± 1.07)$^{a}$ | 260(± 22.5)$^{b}$ | 920(± 27.0)$^{b}$ | 6.51(± 1.03)$^{a}$ |
| Site3 | 0.005(± 0.000)$^{ac}$ | 1.41(± 0.029)$^{ab}$ | 47.2(± 7.94)$^{c}$ | 37.7(± 1.23)$^{c}$ | 36.0(± 16.0)$^{b}$ |
| Site4 | 0.005(± 0.000)$^{ac}$ | 0.713(± 0.026)$^{b}$ | 211(± 10.1)$^{d}$ | 208(± 13.8)$^{d}$ | 3.21(± 0.356)$^{a}$ |
| Site5 | 0.005(± 0.000)$^{c}$ | 1.52(± 0.052)$^{ab}$ | 164(± 26.8)$^{e}$ | 90.6(± 10.9)$^{e}$ | 5.37(± 0.103)$^{a}$ |
| Average | 0.005(± 0.000) | 1.68(± 0.811) | 281(± 241) | 422(± 398) | 11.3(± 14.2) |

Note: $^{a, b, c, d, e}$ Difference is significant at $p < 0.05$ (LSD); Cd = cadmium; Cu = copper; Fe = iron; Mn = manganese; Zn = zinc.

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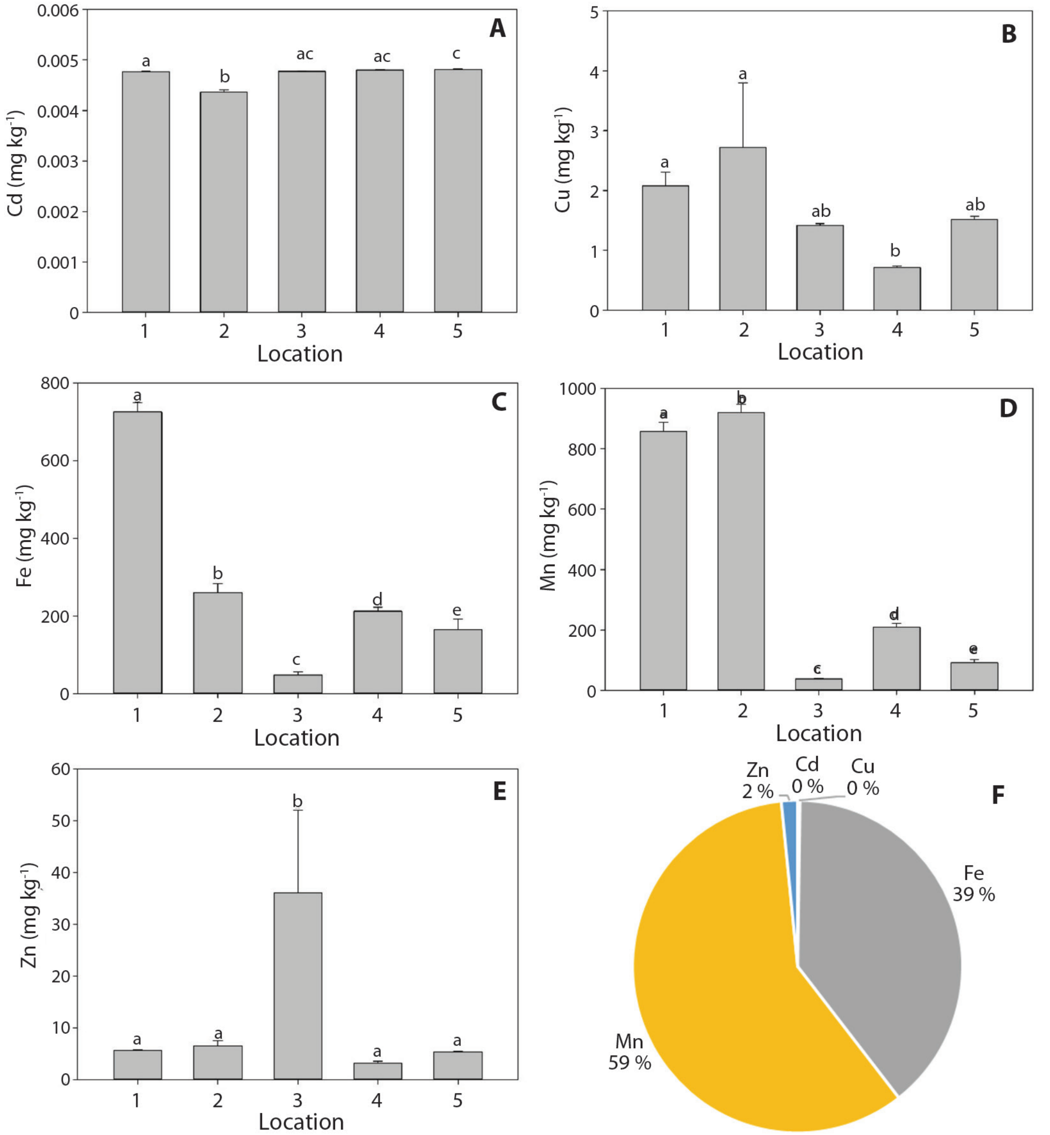


**Fig. 3.** Heavy metal content in the sediment by Site. **A.** Cd, **B.** Cu, **C.** Fe, **D.** Mn, **E.** Zn, **F.** Percentages of five HMs in *Spirogyra*.

**Sediment quality analysis:** The $I_{geo}$ values for Cu and Fe in the sediments from all sites were < 0, indicating no significant pollution. The Mn from Sites 1 and 2 had values > 0, indicating moderately polluted, with Sites 3, 4, and 5 being < 0. The Zn at all sites had $I_{geo}$ < 0. The ranking of the mean $I_{geo}$ values was in the order Mn > Zn > Cu > Fe > Cd. The statistics for the comparisons between sites and $I_{geo}$ values are listed in Table 5 and shown in Fig. 4. The *EF* values were < 1, indicating no enrichment or a minimal effect from human activities. The mean *EF* values were in the order Mn > Zn > FE > Cu > Cd. The statistics for the sites and EF values are presented in Table 5 and Fig. 4. The *CF* values for Cd, Cu, Mn, and Zn were < 1,

**Table 5**
Heavy metal contamination of *Spirogyra* reservoir habitats in the Chi River basin.

| Indicator | HM | | | | |
|---|---|---|---|---|---|
| | Cd | Cu | Fe | Mn | Zn |
| **$I_{geo}$** | | | | | |
| Site1 | -9.06(± 0.002)[a] | -4.62(± 0.164)[ad] | -6.48(± 0.047)[a] | 0.827(± 0.051)[a] | -5.40(± 0.029)[a] |
| Site2 | -9.19(± 0.014)[b] | -4.29(± 0.526)[a] | -7.97(± 0.127)[b] | 0.929(± 0.042)[a] | -5.20(± 0.239)[a] |
| Site3 | -9.06(± 0.003)[ac] | -5.16(± 0.029)[b] | -10.4(± 0.242)[c] | -3.67(± 0.046)[b] | -2.83(± 0.721)[b] |
| Site4 | -9.05(± 0.002)[ac] | -6.15(± 0.053)[c] | -8.26(± 0.068)[b] | -1.21(± 0.095)[c] | -6.21(± 0.163)[c] |
| Site5 | -9.04(± 0.003)[c] | -5.06(± 0.050)[d] | -8.64(± 0.239)[d] | -2.42(± 0.169)[d] | -5.47(± 0.027)[a] |
| Average | -9.08(± 0.056) | -5.05(± 0.687) | -8.36(± 1.32) | -1.11(± 1.86) | -5.02(± 1.22) |
| **EF** | | | | | |
| Site1 | 0.002(± 0.000)[a] | 0.061(± 0.006)[ab] | 0.016(± 0.000)[a] | 0.266(± 0.009)[a] | 0.035(± 0.000)[a] |
| Site2 | 0.002(± 0.000)[b] | 0.080(± 0.031)[a] | 0.005(± 0.000)[b] | 0.285(± 0.008)[b] | 0.040(± 0.006)[a] |
| Site3 | 0.002(± 0.000)[ac] | 0.041(± 0.000)[bc] | 0.001(± 0.000)[c] | 0.011(± 0.000)[c] | 0.226(± 0.100)[b] |
| Site4 | 0.002(± 0.000)[ac] | 0.021(± 0.000)[c] | 0.004(± 0.000)[d] | 0.064(± 0.004)[d] | 0.020(± 0.002)[a] |
| Site5 | 0.002(± 0.000)[c] | 0.044(± 0.001)[bc] | 0.003(± 0.000)[e] | 0.028(± 0.003)[e] | 0.033(± 0.000)[a] |
| Average | 0.002(± 0.000) | 0.049(± 0.023) | 0.06(± 0.005) | 0.131(± 0.123) | 0.071(± 0.089) |
| **CF** | | | | | |
| Site1 | 0.000(± 0.000)[a] | 1.22(± 0.134)[ac] | 21.3(± 0.697)[a] | 0.266(± 0.009)[a] | 0.035(± 0.000)[a] |
| Site2 | 0.000(± 0.000)[b] | 1.60(± 0.629)[a] | 7.67(± 0.664)[b] | 0.285(± 0.008)[b] | 0.040(± 0.006)[a] |
| Site3 | 0.000(± 0.000)[a] | 0.834(± 0.017)[bc] | 1.39(± 0.234)[c] | 0.011(± 0.000)[c] | 0.226(± 0.100)[b] |
| Site4 | 0.000(± 0.000)[ac] | 0.419(± 0.015)[b] | 6.24(± 0.298)[d] | 0.064(± 0.004)[d] | 0.020(± 0.002)[a] |
| Site5 | 0.000(± 0.000)[c] | 0.894(± 0.030)[c] | 4.85(± 0.791)[e] | 0.028(± 0.003)[e] | 0.033(± 0.000)[a] |
| Average | 0.000(± 0.000) | 0.994(± 0.477) | 8.31(± 7.12) | 0.131(± 0.123) | 0.071(± 0.089) |

Note: [a, b, c, d, e] Difference significant at $p < 0.05$ (LSD); *Igeo* = Geo-Accumulation index; *EF* = Enrichment Factor; *CF* = Contamination Factor; Cd = cadmium; Cu = copper; Fe = iron; Mn = manganese; Zn = zinc.

indicating low pollution levels, although the *CF* value of Fe (8.31 ± 7.12) indicates a very high pollution level. The *PLI* values at each site were < 1, indicating no significant pollution in the *Spirogyra* habitat in the Chi River basin (Table 6).

**Relationship between heavy metal contents in *Spirogyra* and sediments:** The relationship between the HMs in the Spirogyra and sediment samples was evaluated using Pearson's r. There were positive correlations between Cd in the algae and Fe in the sediment ($r = 0.530$, $p < 0.05$), between Mn in the algae and Cu in the sediment ($r = 0.920$, $p < 0.01$), Mn in the algae and Mn in the sediment ($r = 0.669$, $p < 0.01$), and Zn in the algae and Mn in the sediment ($r = 0.920$, $p < 0.01$). There was a negative correlation between Mn in the algae and Cd in the sediment ($r = -0.530$, $p < 0.05$). The correlations among the other HMs are presented in Table 7.

**Factor analysis of heavy metal contents in *Spirogyra* and sediments:** The HMs were used as parameters in a principal component analysis (PCA). Prior to the analysis, the HM contents of the *Spirogyra* and sediment samples were tested using the Kaiser-Meyer-Olkin (KMO) and Bartlett tests. The KMO measure of sampling adequacy was 0.419, and there was a significant difference between the eigenvalues ($p < 0.001$) (Table 8). The PCA identified three principal components (PCs) with eigenvalues > 1.5 that collectively explained 80.3 % of the total variance in the dataset (Table 9 and

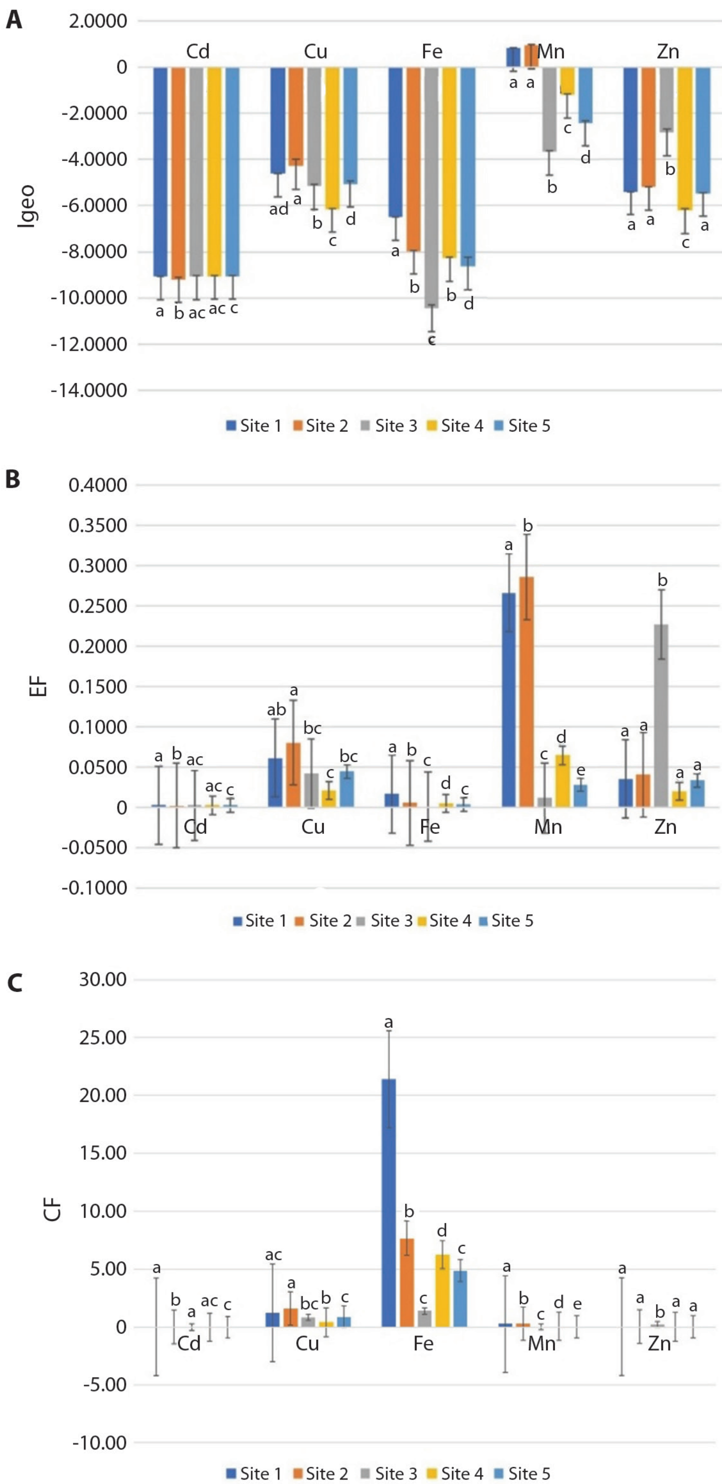


**Fig. 4.** Sediment quality in the reservoir habitat of *Spirogyra* in the Chi River basin. **A.** $I_{geo}$, **B.** EF, **C.** CF.

**Table 6**
*PLI* values for the sediments from *Spirogyra* reservoir habitats in the Chi River basin.

| Location | *PLI* | Meaning |
|---|---|---|
| Site1 | 0.0001169(± 0.00000609)[a] | No pollution |
| Site2 | 0.0000855(± 0.00002124)[b] | No pollution |
| Site3 | 0.0000127(± 0.00000156)[c] | No pollution |
| Site4 | 0.0000139(± 0.00000118)[c] | No pollution |
| Site5 | 0.0000152(± 0.00000196)[c] | No pollution |
| Average | 0.0000448(± 0.00000641) | No pollution |

Note: [a, b, c, d, e] Difference is significant at $p < 0.05$ (LSD).

**Table 7**
Correlation between heavy metals contents in the sediment (Soi) and *Spirogyra* (Al) in the Chi River basin.

| | Cd-Al | Cu-Al | Fe-Al | Mn-Al | Zn-Al | Cd- Soi | Cu- Soi | Fe- Soi | Mn- Soi | Zn- Soi |
|---|---|---|---|---|---|---|---|---|---|---|
| Cd-Al | 1 | –– | –– | –– | –– | –– | –– | –– | –– | –– |
| Cu-Al | –– | 1 | –– | –– | –– | –– | –– | –– | –– | –– |
| Fe-Al | –– | .935** | 1 | –– | –– | –– | –– | –– | –– | –– |
| Mn-Al | –– | –– | –– | 1 | –– | –– | –– | –– | –– | –– |
| Zn-Al | –– | .568* | –– | .645** | 1 | –– | –– | –– | –– | –– |
| Cd- Soi | –– | –– | –– | –.530* | –– | 1 | –– | –– | –– | –– |
| Cu- Soi | –– | –– | –– | .920** | .520* | –.719** | 1 | –– | –– | –– |
| Fe- Soi | .530* | –– | –– | –– | –– | –– | –– | 1 | –– | –– |
| Mn- Soi | –– | –– | –– | .669** | –– | –.687** | .706** | .728** | 1 | –– |
| Zn- Soi | –– | –– | –– | –– | –– | –– | –– | –– | –– | 1 |

Note: * two-tailed $p < 0.05$; ** two-tailed $p < 0.01$; Cd-Al = cadmium in algae, Cu-Al = copper in algae, Fe-Al = iron in algae, Mn-Al = manganese in algae, Zn-Al = zinc in algae, Cd-Soi = cadmium in sediment, Cu-Soi = copper in sediment, Fe-Soi = iron in sediment, Mn-Soi = manganese in sediment, Zn-Soi = zinc in sediment.

**Table 8**
Results of the KMO and Bartlett tests on the heavy metal contents of *Spirogyra* and the Sediments in the Chi River basin.

| | | |
|---|---|---|
| KMO Measure of Sampling Adequacy | | 0.419 |
| Bartlett's Test of Sphericity Approximate Chi Square | | 177.592 |
| | Degrees of Freedom | 45 |
| | Significance | .000 |

Fig. 5A), with each of the three components explaining > 10 % of the variance. The most important contributors to PC1 were Mn in the algae (0.878), Cu in the sediment (0.927), and Mn in the sediment (0.714). This group is indicated by the red ellipse in Fig. 5B. For PC2, the factor loading of Cu of the algae was 0.976 and Fe in the algae was 0.882. This group appears in the green ellipse in Fig. 5B. For PC3, the factor loading of Cd in the algae was 0.734 and Fe in the sediment was 0.907. This group is shown in the yellow ellipse in Fig. 5B.

## DISCUSSION

This study found that the measured heavy metal levels in Spirogyra were below Thailand's food safety standards, which stipulate that cadmium concentrations in dried seaweed must not surpass 2 mg $kg^{-1}$, and the test results confirmed compliance with these standards.

According to the Pollution Control Department's criteria (2022), the concentration of heavy metals in sediments does not surpass the critical contamination thresholds for freshwater sources, which specify that Cd must be below

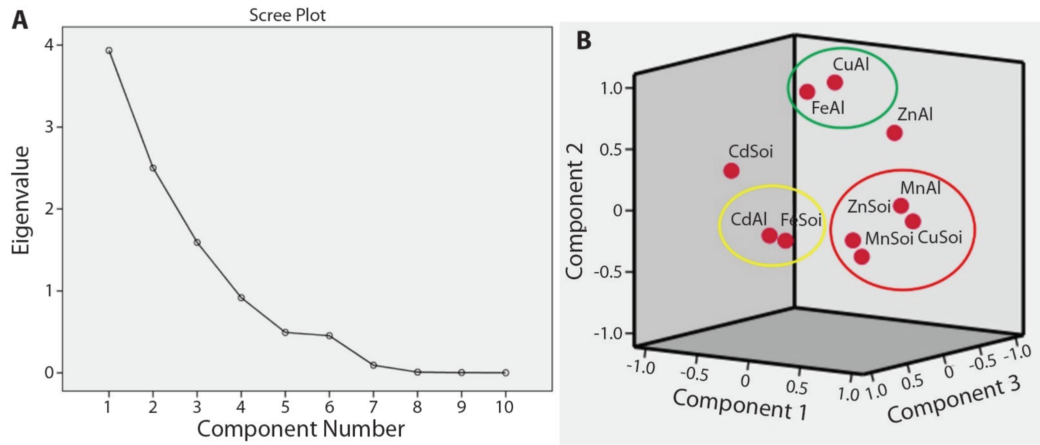


**Fig. 5.** Results of the PCA of the heavy metal contents of *Spirogyra* and the sediments in the Chi River basin. **A.** Eigenvalues of the PCA components, **B.** PC Loadings.

**Table 9**
Results of the PCA of the heavy metal contents of *Spirogyra* and the sediments in the Chi River basin.

| PC | Component | | |
|---|---|---|---|
| | 1 | 2 | 3 |
| % Variance | 39.4 | 25 | 15.9 |
| Cumulative % | 39.4 | 64.4 | 80.3 |
| Eigenvalue | 3.93 | 2.5 | 1.59 |
| Cd-Alg | −0.061 | −0.152 | 0.734 |
| Cu-Alg | −0.029 | 0.976 | −0.126 |
| Fe-Alg | −0.265 | 0.882 | −0.079 |
| Mn-Alg | 0.878 | 0.116 | 0.253 |
| Zn-Alg | 0.630 | 0.648 | −0.010 |
| Cd-Sed | −0.853 | 0.209 | 0.132 |
| Cu-Sed | 0.927 | −0.020 | 0.158 |
| Fe-Sed | 0.215 | −0.141 | 0.907 |
| Mn-Sed | 0.714 | −0.271 | 0.562 |
| Zn-Sed | −0.169 | −0.391 | −0.576 |

Note: PCs = Components; Cd-Al = cadmium in algae; Cu-Al = copper in algae; Fe-Al = iron in algae; Mn-Al = manganese in algae; Zn-Al = zinc in algae; Cd-Soi = cadmium in sediment; Cu-Soi = copper in sediment; Fe-Soi = iron in sediment; Mn-Soi = manganese in sediment; Zn-Soi = zinc in sediment.

1 mg kg$^{-1}$, Cu must be below 31 mg kg$^{-1}$, and Zn must be below 120 mg kg$^{-1}$ (calculated on a dry weight basis), Fe and Mn are excluded from the standards due to the significant variability in their baseline values, which rely on the sediment source and mineral composition, rendering a constant control value unattainable.

However, in the natural aquatic ecosystems, the concentration of heavy metals in sediments is influenced by both geological factors (parent material) and anthropogenic activities associated with the adjacent land use, including agriculture, industry, and transportation (Akhtar et al., 2021; Zha et al., 2024). Consequently, even though Fe concentrations may be elevated, they typically do not reach levels that are considered to be severely polluted when combined with the *Igeo* and *EF*. The accumulation of minerals from source materials that have been deposited in sediments for an extended period or from lithogenic materials within the reservoir may be the cause of high Fe values (Abdullah et al., 2020; Fadlillah et al., 2023).

*Spirogyra* as a possible future food, the "Future food" is a concept representing food security and sustainability, referring to the food's safety and nutritional value (Çakmakçı et al., 2024; Galanakis, 2024). The concept also covers food processing and production. The dry-weight nutrient content of *Spirogyra* includes 12-24 % protein by dry weight, about 43-62 % carbohydrates, and about 15-21 % fats (Saragih et al., 2019; Sitthiwong, 2019; Yongkhamcha & Buddhakala, 2023). Regarding the dietary requirements of minerals, such as Cu, Fe, Mn, and Zn, *Spirogyra* had a Cu content of 1.43 ± 1.30 mg $kg^{-1}$, with an Fe content averaging 872 ± 1.541 mg $kg^{-1}$, Mn averaging 139 ± 92.9 mg $kg^{-1}$, and Zn averaging 18.13 ± 5.50 mg $kg^{-1}$. Our results indicate that the mineral content in the *Spirogyra* is sufficient to meet recommended daily consumption levels (U. S. Department of Health and Human Services, 2025). However, there have been concerns about the quantity of Cd in the algae associated with the Fe content in the sediment. We found that Fe in the sediment averaged 281 ± 241 mg $kg^{-1}$ dry weight, and the CF was relatively high (8.31 ± 7.12), indicating a very high pollution level. Thus, it will be necessary to monitor the Cd contents of the algae for consumer safety. However, the efficacy of each algal species as bioindicators is contingent upon the environmental and climatic conditions (Anabtawi et al.,2024; Omar, 2010).

The HM contents in *Spirogyra* samples from the Chi River basin occurred in the order Fe > Mn > Zn > Cu > Cd (85.4 : 13.6 : 0.8 : 0.1 : 0, respectively). The *THQ* and *HI* values for adult males and females indicated no significant risks to human health from consuming the algae. The HM contents in the sediments occurred in the order Mn > Fe > Zn > Cu > Cd (58.9 : 39.2 : 1.6 : 0.2:0, respectively). The $I_{geo}$ values at all sites were < 0. A deficiency in HM enrichment was indicated by *EF* values < 2, and the CF values for Cd, Cu, Mn, and Zn were < 1, indicating low pollution levels. The *CF* value for Fe (mean = 8.31) indicated a high pollution level, however, especially at Site 1 (21.3), possibly due to human activity around the reservoir. A *PLI* value of < 1 indicated no pollution in the aquatic habitats of *Spirogyra* in the Chi River basin. An analysis of the relationships between the HMs and the algae and the sediments identified three associations-Mn in the algae and Cu and Mn in the sediment (PC1), Cu and Fe in the algae (PC2), and Cd in the algae and Fe in the sediment (PC3). *Spirogyra* in the Chi River Basin is suitable for future food development, but the Fe content in the sediment should be monitored because it was positively correlated with Cd in the algae.

**Ethical statement:** The authors declare that they all agree with this publication and made significant contributions; that there is no conflict of interest of any kind; and that we followed all pertinent ethical and legal procedures and requirements. All financial sources are fully and clearly stated in the acknowledgments section. A signed document has been filed in the journal archives.

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