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Biomass Quality Variations over Different Harvesting Regimes and Dynamics of Heavy Metal Change in *Miscanthus lutarioriparius* around Dongting Lake

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Abstract: *Miscanthus lutarioriparius* has a growing area of 100,000 ha and an annual biomass production of 1 Mt around Dongting Lake. However, due to serious soil pollution, there is a concern that the *M. lutarioriparius* biomass could have high heavy metal (HM) concentrations. This necessitates investigation of biomass quality to find the appropriate end use. Thus, this study aims to investigate the dynamics of HM elements in the *M. lutarioriparius* biomass and their impact on biomass quality across different growing areas and harvest times. We analyzed the HM concentrations in soil and biomass from 11 sites under different harvesting times (April, August and December). Results showed that Cd in soil samples was 9.43-fold higher than the national standards. The heavily polluted soil caused a high HM concentration in the biomass and the accumulation increased with the delayed harvest. The fresh young shoots in April met the food limitation for Cd and Cr, whereas Pb concentration was slightly higher than the threshold limit. The mature biomass from the southern part had higher Mn, Cd and Pb, but lower Cu, Zn and Cr concentrations than that from the eastern part. These results can provide guidance for guaranteeing the consistent quality of the *M. lutarioriparius* biomass for bio-based industry.

Keywords: biomass quality; eco-industrial crop; heavy metal; *Miscanthus*; phytoremediation; wetland plant

1. Introduction

Dongting Lake, the second largest freshwater lake in China, plays a vital role in maintaining the ecosystem functioning of the Yangtze River Basin. This is mainly achieved through supporting the wild *Miscanthus lutarioriparius* community with an area of approximately 100,000 ha [1]. *M. lutarioriparius* is a perennial herbaceous plant characterized by tall stems, high biomass yield and extensive belowground systems [2]. The developed rhizome network can reduce soil erosion and simultaneously adsorb and accumulate contaminants from soil or water [3,4]. In addition, its high biomass can sequester a great amount of atmospheric CO_2 with a potential of approximately 100 t CO_2 ha^{-1} annually [5]. The tall stems provide an ideal shield for wild animals, indicating a biodiversity-increasing potential. Moreover, the *M. lutarioriparius* community also supports the biomass-based industry in the Dongting Lake region through production of 1 Mt cellulose-rich biomass.

Since the 1950s, this biomass feedstock has been used to produce pulp [6,7]. However, due to the serious water pollution concerns associated with the paper mills, almost all the paper-making plants have been closed by the Hunan provincial government since 2019. At present, there are still no appropriate alternate pathways identified for the commercial utilization of the *Miscanthus* biomass. With the closure of paper mills and lack of industrial demand, management practices and harvesting of the *M. lutarioriparius* biomass has been almost abandoned. Thus, there is an increasing concern that the unharvested plants will cause a series of ecological problems such as water eutrophication caused by the leaching of nutrients and an increasing risk of wildfire causing negative impacts on biodiversity. The aforementioned issues indicate that harvesting and commercial utilization of the biomass are the key management measures to preserve the *M. lutarioriparius* community and the Dongting Lake wetland ecosystem [4].

Compared to the most prevalent herbaceous plant species, the biomass of *M. lutarioriparius* is characterized with high contents of lignin (132 g kg^{-1}) and cellulous (620 g kg^{-1}) and a low content of ash (84 g kg^{-1}) [8]. These biomass components can be utilized to produce a wide range of biobased products such as biofuel [9], light-weight concrete [10], biochar and mushroom growth substrate [1], etc. Of all these utilization possibilities, biochar production and use as growth substrate for mushroom and bio-fertilizer are mainly recommended because of the availability of well-established methods and matured technology. In the short term, these utilization pathways can immediately accommodate huge quantities of available unharvested *Miscanthus* biomass and address the aforementioned imminent ecological challenge. Furthermore, the young shoots of *M. lutarioriparius* harvested in early spring can be used to produce pickles. The production of pickled shoots has reached approximately 20,000 t with production values of about CNY0.5 billion [2], which can also contribute toward overcoming the ecological problem and maintaining the ecosystem functioning.

The main water sources of the Dongting Lake are the Xiangjiang River, Yuanshui River, Zishui River and Lishui River [11], which flow through the main mining area of Hunan. Consequently, the water and soil around Dongting Lake are contaminated by heavy metal (HM) elements such as Cd, Cu, Mn, Zn, Cr and Pb [3,11]. The miscanthus species, especially *Miscanthus floridulus*, is known for their high HM adsorption and accumulation capabilities and is considered as a promising candidate plant for phytoremediation [12]. From this, it can be deducted that the *M. lutarioriparius* biomass from Dongting Lake is contaminated by the HM elements. Currently, *M. lutarioriparius*-based bioproducts (e.g., biochar-based fertilizer, pickles and mushroom growth substrate) are mainly used in agriculture and food production. Therefore, there is a growing concern about the safety of these bioproducts, especially their impact on food safety. This is also the main barrier in the development and expansion of bio-based industries.

M. lutarioriparius is a perennial herbaceous plant with three main growth phases, including seedling, mature and senescence. There are differences in the ability to bioaccumulate HM elements by the plant organs [3]. However, it is still unclear how they vary and what determines the uptake of HM elements in *M. lutarioriparius* over different growing phases. In addition, *M. lutarioriparius* is a plant with a nutrients re-translocation ability at the end of the growth [13]. It is still not known if HM elements in the leaves and stems also return back into the underground organs or not. The aim of this study is to investigate the dynamics of HM elements in the *M. lutarioriparius* biomass and their impact on biomass quality across different growing areas and harvest times. Based on the outcomes of this study, recommendations will be formulated to ensure the efficient and safe use of the *M. lutarioriparius* biomass around the Dongting Lake area.

2. Materials and Methods

2.1. Study Area and Sampling Strategy

The total water-flooded area in the Dongting Lake region (28°38′–29°45′ N, 111°40′–113°10′ E) is approximately 20,000 km^2 during the wet season (from April to September), whereas during the dry season (from November to next March) it shrinks to 16,400 km^2. This region is characterized by a subtropical monsoon climate with a mean annual temperature of 17 °C and precipitation of 1200–1400 mm. The study area was comprised of 11 sampling sites evenly distributed across the whole Dongting Lake region as shown in Figure 1. There are six sampling sites locating in the southern Dongting Lake (S1–S6) and five in the eastern Dongting Lake (S7–S11). Each sampling site was comprised of evenly distributed areas of more than 1 ha of *M. lutarioriparius*.

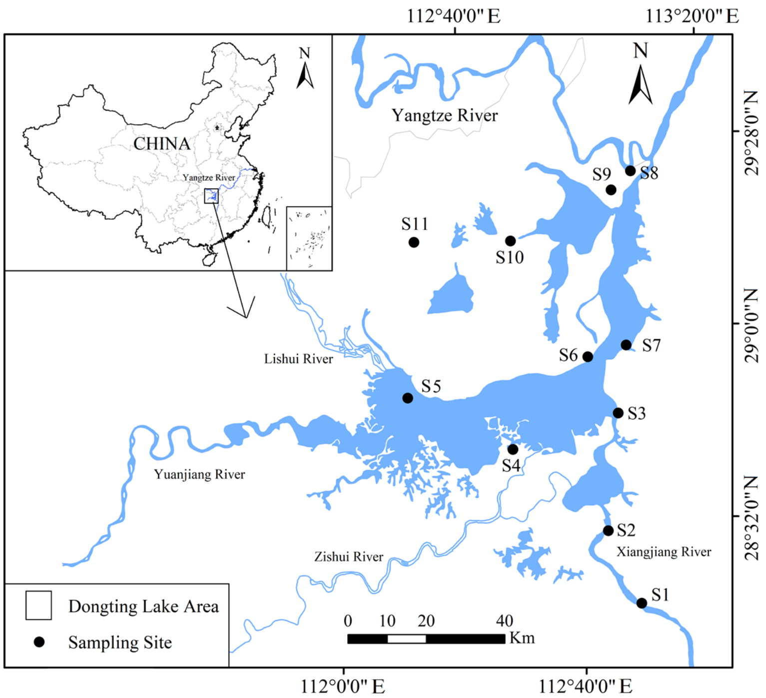

Figure 1. Location of the 11 in situ sampling sites within the Dongting Lake area.

Plant and soil sampling were conducted in 2018 at three different harvest times (3–8 April, 20–25 August and 15–20 December). Prior to the sampling, three 1 × 1 m quadrats were randomly selected at each site. The distance between each sampling quadrat was set to be more than 100 m. For each sampling, all the aboveground plants within the quadrat were harvested and weighed; then stems and leaves were separated; the belowground rhizomes and roots within the depth of 0–30 cm were collected, washed and weighed. About 500 g of fresh stems, leaves and underground organs (rhizomes and roots together) were collected and then taken to the laboratory for biomass yield determination and quality analysis. Afterward, all the plant samples were oven-dried at 80 °C until constant weight [8] and then milled to a powder (100 mesh) for the HM concentration determination. For soil sampling, five points were randomly selected within each sampling quadrat by following the "S"-shape principle. Five soil cores (0–30 cm) within the quadrat were mixed thoroughly to prepare a composite sample (approximately 500 g) and taken to the laboratory. The soil samples were air dried and then ground to pass through a 100-mesh sieve for the soil chemical analysis and HM concentration determination.

2.2. Chemical Analysis of Soils and Plants

Soil pH and electrical conductivity were determined using a portable pH meter (Bante-221, BANTE, Shanghai, China) and a conductivity meter (Bante-950-UK, BANTE, Shanghai, China), respectively. The soil suspension was prepared at soil: distilled water (*w*/*v*) of 1:2.5. Soil organic matter (SOM) was determined by the $K_2Cr_2O_7$–H_2SO_4 oxidation method. Soil total nitrogen (TN) was determined using the Kjedahl method. Both total phosphorous (TP) and available phosphorous (AP) of the soil were determined using the molybdenum antimony colorimeter method after samples were digested in $HClO_4$–H_2SO_4 (1:10, v/v) and extracted in $NaHCO_3$ (0.5 mol L^{-1}), respectively. Soil total potassium (TK) and available potassium (AK) were determined using the flame photometry method after soil samples were digested in $HClO_4$–H_2SO_4 (1:10, v/v) and extracted in NH_4OAc (1.0 mol L^{-1}). Plant and soil samples were digested by $HClO_4/HNO_3$ (1:4, v/v) and HCl/HNO_3 (1:3, v/v), respectively. The concentrations of Cu, Mn and Zn were determined by a flame atomic absorption spectrometer (TAS-A3, PERSEE, Beijing, China) with air-acetylene, while concentrations of Cd, Cr and Pb were determined by an inductively coupled plasma spectrometer (ICAP-7200, Thermo Fisher Scientific, Waltham, MA, USA). The procedures of soil and plant chemical analysis followed in this study are described in detail by Bao [14].

2.3. Data Analysis

The bioaccumulation factor (BF) is used to evaluate capability to absorb a specific HM element from soils by a plant [15,16], while the translocation factor (TF) is a measure of the internal mobility of a given HM element across plant organs [17]. They are calculated by the following equations:

$$BF = C_{underground}/C_{soil} \quad (1)$$

$$TF_{stem/underground} = C_{stem}/C_{underground} \quad (2)$$

$$TF_{leaf/underground} = C_{leaf}/C_{underground} \quad (3)$$

$$TF_{leaf/stem} = C_{leaf}/C_{stem} \quad (4)$$

where C_{soil}, C_{stem}, C_{leaf} and $C_{underground}$ are the concentrations (mg kg^{-1} DW) of a given HM element in soils, stems, leaves and underground organs (rhizomes and roots), respectively.

For each HM element, its total amount of element bioaccumulation (TAB) by each plant part (stem, leaf, rhizome and root) was calculated based on the following Equation (5):

$$TAB_{HM} = \sum^{organ=3} BW_{organ} \times C_{organ} \quad (5)$$

where BW_{organ} is the dry weight of the stems, leaves and underground organs (rhizomes and roots), which was calculated based on fresh weight and their corresponding water content; C_{organ} is the HM concentrations in the corresponding organs of stems, leaves and underground organs.

The statistical analysis was based on log-transformed data. A one-way ANOVA followed by pairwise *t*-tests was used to assess the effects of sampling location on soil chemical properties and HM concentrations. The significances of variance in terms of HM concentration in biomass among different sampling months and plant organs were determined using a one-way ANOVA followed by Duncan's post hoc test. The significances of variation in terms of soil chemical properties, HM concentrations in soils and biomass among different sampling months, sites and plant organs were evaluated using a two-way ANOVA. Pearson's coefficient analysis was used to reveal relationships between soil chemical properties and HM absorption-translocation capability and changes of HM concentrations in plant organs over growing seasons. Statistical analysis was performed using the statistical software IBM SPSS Version 22.0 (IBM, Armonk, NY, USA).

3. Results

3.1. Variation in Soil Chemical Properties and HM Concentrations across 11 Sampling Sites within the Dongting Lake Region

Results presented in Table 1 showed the variation in soil chemical properties and HM concentrations among the 11 sampling sites. The data supported that there was less variation in soil chemical properties than HM concentrations. The average variation coefficient of the eight soil chemical properties was 38.93% (10.68–69.82%), while it was 43.41% (22.18–95.17%) of the HM concentrations. Of the eight soil chemical properties, different sites had significant differences in terms of TN (p = 0.001), TP (p = 0.047), AP (p = 0.026), AK (p = 0.023) and SOM (p = 0.016). There was a general trend that the sites in the southern Dongting were characterized to have higher TN (0.10 vs. 0.06%), TP (0.07 vs. 0.06%), AP (26.67 vs. 16.99 mg kg^{-1}), AK (172.55 vs. 112.17 mg kg^{-1}) and SOM (2.72 vs. 2.11%) than that in the eastern Dongting. This trend was stable across different sampling months as indicated by the nonsignificant ($p > 0.05$) effect of 'Month' and the interaction of 'Month × Site'. Also, the southern Dongting sites generally had more serious HM pollution problems than the eastern Dongting sites. For example, the average Mn concentration of the southern Dongting sites (368.38 mg kg^{-1}) was 1.63-fold higher (p = 0.001) than that of the eastern Dongting sites (226.65 mg kg^{-1}). The significant differences between sampling sites in terms of Zn, Cd, Cr and Pb concentrations were recorded as indicated by their p values of <0.001. The variation coefficient of soil Cd concentration was the highest (95.17%), followed by soil Mn (37.89%), Cu (36.58%), Pb (34.47%), Cr (23.56%) and Zn (22.18%). Soil from Dongting Lake had 9.43-fold higher concentration of Cd than the national standards of China (GB15618-2018) for HM contamination. Although soil Cu concentration was still high in the southern Dongting sites, these differences were not significant (p = 0.346). The above results analysis suggest that the southern Dongting area is characterized by more fertile and seriously polluted soil than the eastern Dongting area.

Table 1. Soil chemical properties and heavy metal concentrations in the study area.

	Parameter [a]	Location [b]		Statistic		Sources of Variance in ANOVA (p-Value)		
		Southern Dongting	Eastern Dongting	Mean	Variation Coefficient (%)	Month	Site	Month × Site
Chemical properties	pH	7.34	7.62	7.47	10.68	0.106	0.350	0.131
	EC (μs cm^{-1})	425.83	367.89	399.49	36.63	0.961	0.487	0.994
	TN (%)	0.10A	0.06B	0.08	37.70	0.984	0.001	0.997
	TP (%)	0.07a	0.06b	0.06	28.45	0.668	0.047	0.378
	TK (%)	1.12	1.05	1.09	34.24	0.397	0.435	0.826
	AP (mg kg^{-1})	26.67a	16.99b	22.27	69.82	0.993	0.026	0.680
	AK (mg kg^{-1})	172.55a	112.17b	145.11	61.03	0.341	0.023	0.340
	SOM (%)	2.72a	2.11b	2.44	32.85	0.339	0.016	0.094
Heavy metal concentration (mg kg^{-1})	Cu	32.14	29.94	31.14	36.58	0.995	0.346	0.995
	Mn	368.38A	226.65B	303.96	37.89	0.998	0.001	0.997
	Zn	190.77A	141.44B	168.35	22.18	0.882	<0.001	0.867
	Cd	4.06A	1.37B	2.83	95.17	0.715	<0.001	0.903
	Cr	78.00A	57.89B	68.86	23.56	0.997	<0.001	0.999
	Pb	78.87A	51.21B	66.30	45.10	0.245	<0.001	0.730

[a] pH—soil pH; EC—soil electrical conductivity; TN—soil total nitrogen content; TP—soil total phosphorous content; TK—soil total potassium content; AP—soil available phosphorous content; AK—soil available potassium content; SOM—soil organic matter content. [b] Values are means of three sampling times (April, August and December) in 2018. Different lowercase and uppercase letters within the same row indicate significant differences at $p < 0.05$ and at $p < 0.01$ for the same indicators, respectively.

3.2. Seasonal Changes in HM Concentrations in Different Plant Organs

The pattern of the HM concentrations in the plant organs over the growing season was complex and varied for each HM element (Figure 2). Except for Zn and Pb, there was a general trend that HM concentrations in the underground organs decreased toward the end of the growing season. This was particularly true for Mn (Figure 2d) with significant changes in concentrations in the underground organs. However, a general opposite trend of dynamics through the growing seasons was observed for the HM concentrations in leaves and stems. This was especially true for Mn (Figure 2d) and Zn (Figure 2f) with a significant decrease in stems and a significant increase in leaves over the growing season. For example, the Mn concentrations in stems in April (49.66 mg kg^{-1}) were 33.7% and 49.7%, higher than that in August (32.91 mg kg^{-1}) and December (25.00 mg kg^{-1}), respectively. The Mn concentrations in leaves increased from 34.24 mg kg^{-1} in April to 39.74 mg kg^{-1} in August and then finally reached to 75.97 mg kg^{-1} in December. At the end of growth

(December), there was a significant increase in the HM concentrations in leaves, especially for Cd (Figure 2b), Mn (Figure 2d), Pb (Figure 2e) and Zn (Figure 2f).

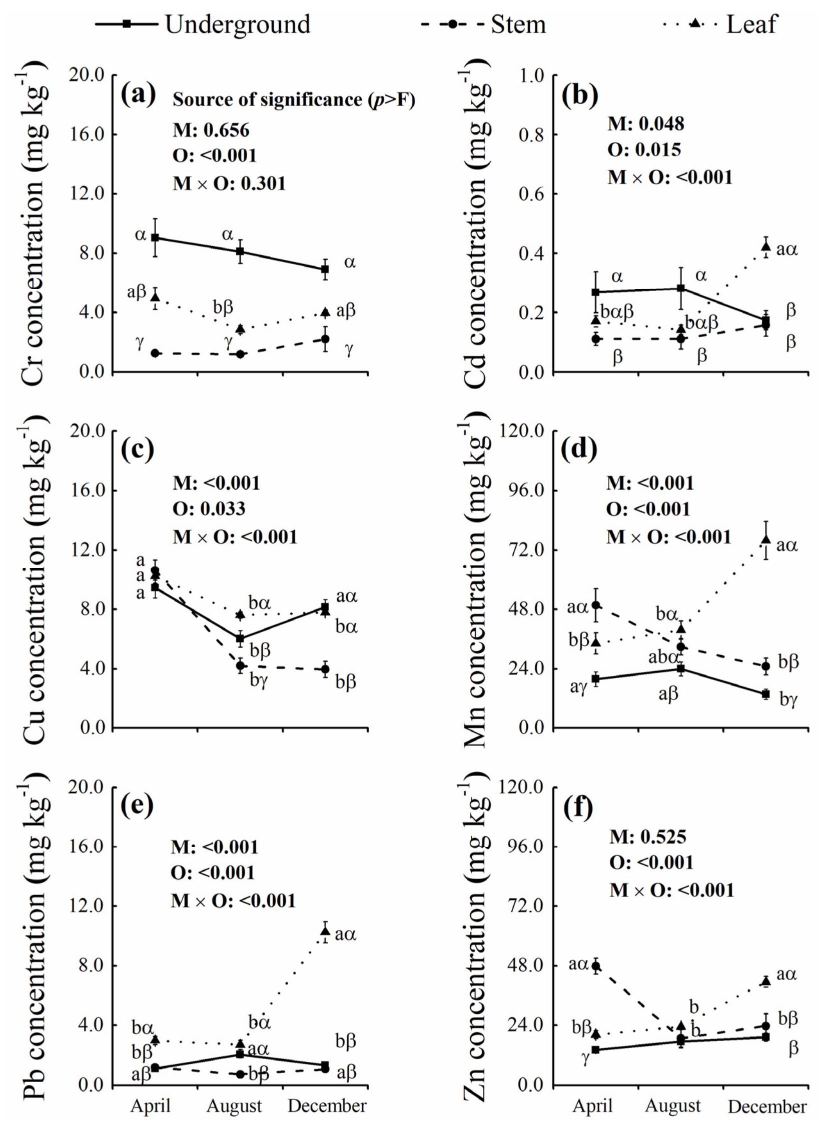

Figure 2. Seasonal changes of heavy metal concentrations in the underground organs, stems and leaves. M and O and represent sampling months and plant organs. Bars indicate standard errors of means. Different English letters indicate significant differences among the three sampling months at $p < 0.05$. Different Greek letters indicate significant differences among the underground organs, stems and leaves at $p < 0.05$.

April is the harvest time of aboveground young shoots for pickle production and December is the harvest time for mature biomass (Table 2). Based on dry weight, young shoots from only two sites (S5 = 0.05 mg kg^{-1}, S9 = 0.04 mg kg^{-1}) met the food safety standard (<0.05 mg kg^{-1}) of China (GB 2762–2017) for Cd concentration (Table 2). Furthermore, the young shoots in all sites had two- to fourfold higher Cr concentrations (0.84–1.78 mg kg^{-1}) and nine- to nineteenfold higher Pb concentrations (0.86–1.85 mg kg^{-1}) than the standard limitation (0.5 mg kg^{-1} for Cr, 0.1 mg kg^{-1} for Pb) (Table 2). The young shoots harvested during April are generally used to produce pickles or eaten freshly. Therefore, it is more practical to assess the quality of the April-harvested biomass based on the fresh weight instead of dry weight. Based on the fresh weight (converted by 85% water content), the young shoots from all the sampling sites met the food safety standards for Cd and Cr, but not for Pb. The fresh young shoots had varied Pb concentrations (0.13–0.28 mg kg^{-1}), which was still one- to threefold higher than the standard limits. For the mature biomass harvested in December, the concentrations of different HM elements were shown in a descending order as Mn (36.32 mg kg^{-1}), Zn (27.89 mg kg^{-1}), Cu (4.92 mg kg^{-1}), Pb (3.18 mg kg^{-1}), Cr (2.74 mg kg^{-1}) and Cd (0.22 mg kg^{-1}) (Table 2). In comparison with eastern Dongting Lake, the mature biomass in southern Dongting Lake was generally characterized to have higher Mn, Cd and Pb concentrations (Table 2). For example, the average Mn concentration in the harvested biomass was 40.73 mg kg^{-1} in southern Dongting Lake, which was 21.7% higher than that of eastern Dongting Lake (31.91 mg kg^{-1}). In addition, the produced biomass in southern Dongting Lake had a higher Cd concentration than that of eastern Dongting Lake by 9.1% (0.22 vs. 0.2 mg kg^{-1}) (Table 2). The average Pb concentration (3.21 mg kg^{-1}) in the harvested biomass from southern Dongting Lake was slightly higher (1.6%) than that in eastern Dongting Lake (3.16 mg kg^{-1}) (Table 2). A different trend was found in terms of Cu, Zn and Cr concentrations. In particular, biomass in eastern Dongting Lake had higher Cu, Zn and Cr concentrations than that in southern Dongting Lake by 28.1% (5.73 vs. 4.12 mg kg^{-1}), 6.9% (28.89 vs. 26.89 mg kg^{-1}) and 35.2% (3.32 vs. 2.15 mg kg^{-1}), respectively (Table 2).

Table 2. Heavy metal concentration of the April-harvested stems and December-harvested aboveground biomass (stems and leaves together) across the 11 sampling sites.

Sampling Sites	April (mg kg^{-1})						December (mg kg^{-1})					
	Cu	Mn	Zn	Cd	Cr	Pb	Cu	Mn	Zn	Cd	Cr	Pb
S1	7.64	28.36	37.09	0.13	1.20	1.13	4.02	38.15	33.58	0.26	1.45	3.34
S2	7.82	92.72	45.80	0.12	0.84	0.95	3.55	56.29	31.87	0.17	1.53	3.43
S3	10.96	76.75	62.50	0.31	1.46	1.02	3.80	42.13	26.72	0.36	4.77	2.93
S4	11.13	42.29	46.74	0.07	0.91	0.91	4.62	13.13	15.33	0.12	1.71	2.48
S5	10.84	29.28	49.88	0.05	1.14	0.86	3.87	43.66	19.66	0.12	1.52	2.49
S6	10.95	42.70	50.36	0.09	1.49	1.20	4.85	51.02	34.17	0.33	1.94	4.58
S7	n/a	n/a	n/a	n/a	n/a	n/a	4.59	35.33	17.35	n/a	n/a	n/a
S8	12.25	50.12	54.58	0.09	1.29	1.85	5.15	46.10	33.10	0.12	1.73	2.89
S9	7.69	60.44	26.37	0.04	1.25	1.38	5.05	22.07	14.99	0.14	7.85	2.81
S10	13.06	22.85	44.61	0.13	1.78	1.65	5.06	31.15	20.95	0.19	1.95	3.55
S11	14.13	51.08	60.86	0.08	1.17	1.03	8.78	24.92	58.08	0.38	1.74	3.38
Southern Dongting	9.89	52.02	48.73	0.13	1.17	1.01	4.12	40.73	26.89	0.22	2.15	3.21
Eastern Dongting	11.78	46.12	46.61	0.09	1.37	1.48	5.73	31.91	28.89	0.20	3.32	3.16
Overall mean	10.65	49.66	47.88	0.11	1.25	1.20	4.92	36.32	27.89	0.21	2.74	3.18

Note: n/a represents the data unavailable because of the missing samplings. Southern Dongting includes sampling sites S1–S6 and eastern Dongting includes sampling sites S7–S11.

3.3. Adsorption and Translocation Characteristics of HM Elements in Different Organs

The average TF values of all HM elements were higher than that of BF during the whole growth season (Figure 3). Among the three sampling dates, the $BF_{underground/soil}$ value of Cu was higher than that of the other HM elements (Figure 3a). It is particularly true for the $BF_{underground/soil}$ value (0.32) of Cu in April, suggesting a strong Cu absorption ability of the underground organs during this time period. The $BF_{underground/soil}$ value of Pb (0.016–0.037) was always the lowest among all the HM elements. For all the HM elements except Cu, no variations were observed between different sampling dates in terms of the $BF_{underground/soil}$ value. More HM elements absorbed by the underground organs were transformed to leaves than stems. In April, a higher $TF_{leaf/underground}$ value (Figure 3c) was observed for the most HM elements than the $TF_{stem/underground}$ value (Figure 3b). For example, the $TF_{leaf/underground}$ values for Pb, Cu, Cd and Cr in April reached to 5.46, 1.16, 1.04 and 0.76, respectively, whereas values for $TF_{stem/underground}$ were 1.82, 1.15, 0.56 and 0.17, respectively. Toward the end of the growing season, more HM elements were transformed to leaves than stems. The average $TF_{leaf/underground}$ value for all the HM elements was 2.36, whereas for the $TF_{stem/underground}$ it was only 1.13. At the end of the growing season (December), the highest $TF_{leaf/stem}$ value (10.16) for Pb was recorded.

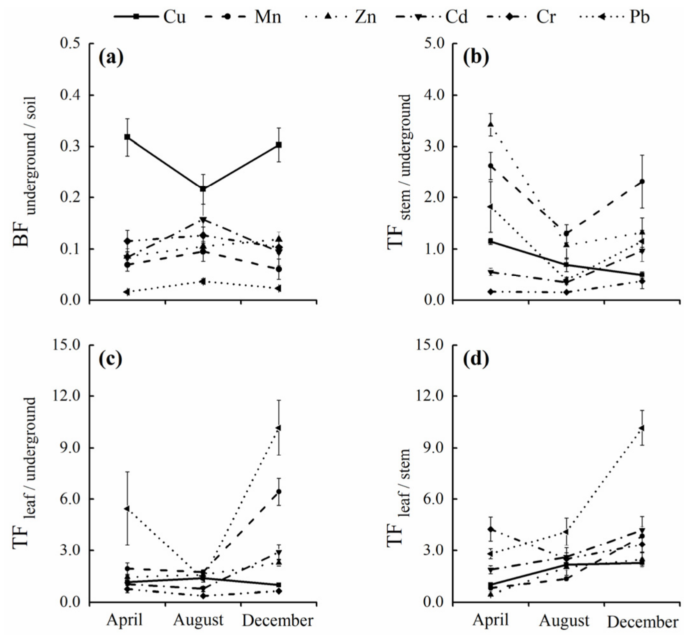

Figure 3. The bioaccumulation factor (BF) and translocation factor (TF) in the different parts (underground organs, stems and leaves) of *Miscanthus lutarioriparius* for Cu, Mn, Zn, Cd, Cr and Pb across different sampling time. Bars indicate standard errors of means.

3.4. Factors Contributing to the Variation in HM Absorption and Translocation Capabilities of M. lutarioriparius

Pearson's analysis results presented the contributions of soil chemical properties and HM absorption-translocation capability on the changes in HM concentrations in different plant organs over different harvest times (Figure 4). For the effects of the initial soil HM concentrations, the Cd concentrations in the underground organs were positively correlated with the soil Zn (r = 0.62, $p < 0.01$), Cd (r = 0.72, $p < 0.001$) and Cr (r = 0.50, $p < 0.05$) concentrations. The Mn and Pb concentrations in the underground organs positively (r = 0.56–0.60, $p < 0.05$) correlated to soil Cd concentration. The Mn, Zn, Cr and Pb concentrations in the underground organs were also found to be positively (r = 0.52–0.88, $p < 0.05$) associated with their bioaccumulation factors. However, to assess the contribution of the translocation factor, the Mn, Cd, Cr and Pb concentrations in the underground organs were negatively (r = 0.65–0.73, $p < 0.01$) correlated with the $TF_{leaf/underground}$ of the corresponding HM elements. The Cu, Mn, Zn, Cr and Pb concentrations in stems were positively (r = 0.53–0.91, $p < 0.05$) associated with the $TF_{stem/underground}$ of the corresponding HM elements. The concentrations of the HM elements except Pb in stems showed negative (r = 0.70–0.81, $p < 0.001$) relationships with the $TF_{leaf/stem}$ of the corresponding HM elements. The Mn, Zn, Cr and Pb concentrations in leaves were positively correlated with the translocation capability of the corresponding HM elements from the underground organs (r = 0.60–0.78, $p < 0.01$) and stems (r = 0.53–0.87, $p < 0.05$) to leaves. Furthermore, the Zn and Cd concentrations in leaves were positively correlated with the translocation capability of Mn (r = 0.62–0.71, $p < 0.01$) and Pb (r = 0.73–0.79, $p < 0.001$) from stems to leaves.

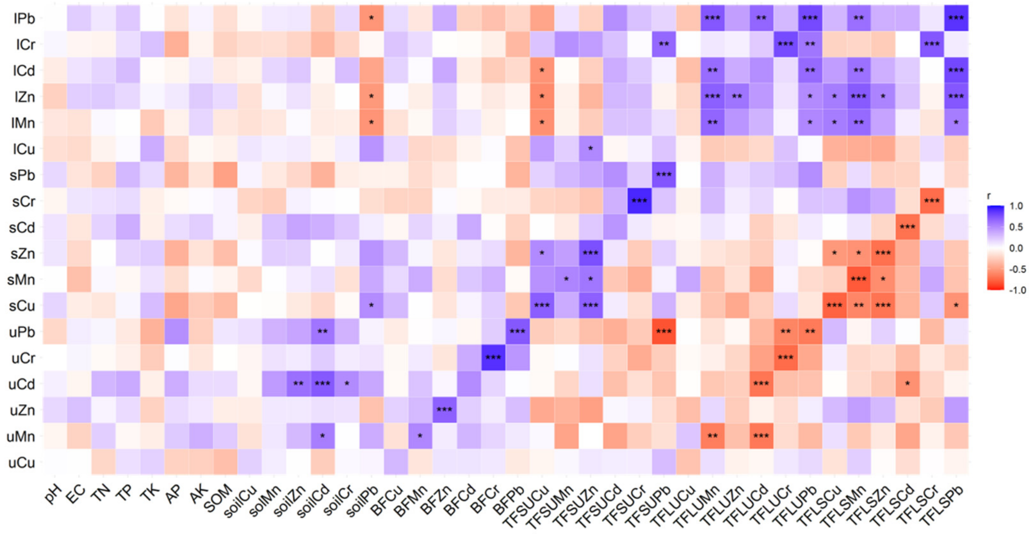

Figure 4. Pearson's coefficient analysis of heavy metal concentrations in different plant organs, soil chemical properties and heavy metal absorption-translocation capability. The first letters of the vertical axis indicate: 'l' represent plant leaves, 's' indicates stems and 'u' for underground organs followed by the chemical symbol of each heavy metal. In the horizontal axis, pH—soil pH; EC—soil electrical conductivity; TN—soil total nitrogen; TP—soil total phosphorous; TK—soil total potassium; AP—soil available phosphorous; AK—soil available potassium; SOM—soil organic matter. Moreover, the indicators of "BF + heavy metal chemical symbol" represent the bioaccumulation factor of the corresponding heavy metal. TFSUCu, TFSUMn, TFSUZn, TFSUCd, TFSUCr and TFSUPb represent translocation factor for Cu, Mn, Zn, Cd, Cr and Pb from underground organs to stems, respectively. It is same for TFLUCu, TFLUMn, TFLUZn, TFLUCd, TFLUCr, TFLUPb, TFLSCu, TFLSMn, TFLSZn, TFLSCd, TFLSCr and TFLSPb, where LU and LS represent translocation from the underground organs to leaves, and stems to leaves, respectively. Asterisks indicate significance: * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

4. Discussion

4.1. Factors Affecting the Seasonal Dynamics of HM Concentrations in M. lutarioriparius Biomass

Miscanthus, a rhizomatous perennial herbal species, is characterized by the translocation of nutrients back to rhizomes before harvest to be used in next growth season [2]. Cu, Mn and Zn, as micro-nutrients, participate in plant growth and metabolism [17]. This can explain that why more Cu, Mn and Zn were transferred from the underground organs to stems and leaves at the seedling stage (Figure 2). For a compartmentalization tolerance strategy [16], nonessential phytotoxic elements Cd and Cr were mainly stored in the underground organs (rhizomes and roots) (Figure 2) and enabled *M. lutarioriparius* to tolerate a high level of HM contamination. During the growth season, substantial amounts of essential nutrients are consistently taken up by the plant for photosynthesis, growth and reproduction [13], which lead to a co-transfer of HM elements to aboveground organs [18]. At the end of the growth season, unlike the macro-nutrients of N, P and K, the HM elements (especially for Zn, Mn, Cd and Pb) in the aboveground were not re-transferred to the underground but accumulated in leaves of *M. lutarioriparius* (Figure 2). These HM elements in leaves may be stored in the cell wall structure and retained by secondary metabolites [19].

The outcomes of correlation analysis between soil HM concentration and plant HM tolerance ability indicate that there is no consistent trend. A few studies have reported that soil HM concentration is positively correlated with the HM uptake amount by miscanthus plants [20,21]. The difference in genetic make-up could be the leading factor in terms of defining the ability to absorb, accumulate and tolerate. In the present study, the Cd concentrations in the underground organs were positively correlated with the soil Cd concentration (Figure 4). Previous studies indicated that HM elements might be co-taken with other essential elements [18,22]. In contrast with this, our results showed that HM uptake by *M. lutarioriparius* had no significant correlations with soil nutrients, but significantly correlated to other soil HM elements (Figure 4). For example, the Mn and Pb concentrations in the underground organs positively correlated to the soil Cd concentration and the Cd concentrations in the underground organs positively correlated with the soil Zn and Cr concentrations. Overall, the uptake of a specific HM element by the underground organs of *M. lutarioriparius* can be promoted by other HM elements. More HM elements are transferred and stored in the leaves during the whole growth season.

4.2. Recommendations for the Utilization of M. lutarioriparius Biomass Based on the Seasonal Dynamics of HM Concentrations

In our study, the sampling sites suffer different extents of HM contamination. Most sampling sites have high soil HM concentrations by exceeding the sediment background values, which is consistent with most previous studies [23–25]. High translocation potential from the underground to the aboveground organs would pose serious food safety risk and also negatively affect other industrial applications. At the seedling phase (in April), there were relatively high stem–underground translocation potentials for Cd, Cr and Pb (Figure 3), which would increase their concentrations in plant shoots (Figure 2 and Table 2) and consequently influence pickle quality. For pickled shoot manufacturing, the Pb concentrations in fresh young stems across the sampling Dongting Lake areas slightly exceeded the limitation set by the food safety standard of China (GB 2762–2017). The young shoots are generally comprised of sheath and unelongated stems. For pickle production, only the unelongated stems are used. It is evident from the literature that a high HM concentration is observed in sheaths [26]. This indicates that the young shoots with slightly excessive Pb can still be used for pickle processing. The processed sheath and substandard young shoots can be used as superior raw materials for silage production, according to the China Hygienical Standard for Feed GB13078-2017 (1, 30 and 5 mg kg^{-1} set limitations for Cd, Pb and Cr, respectively). The mature *M. lutarioriparius* biomass produced in Dongting Lake can be used for biogas production because of its high cellulose content [7] and the biogas residues with HM elements can be further processed safely as biofertilizers [15]. According to the current standards for fertilizers NY/T 3618-2020, permissible values for Cd, Pb and Cr are 3, 50 and 150 mg kg^{-1}, respectively. The *M. lutarioriparius* biomass produced within the Dongting Lake area is safe to be exploited to produce biofertilizers [6,27]. However, one must be careful not to exploit such contaminated biomass as substrate for mushroom cultivation because edible fungi have a high ability to absorb HM elements, which can consequently pose serious health risks [28]. As no HM elements in the aboveground organs were transferred back to the underground organs of *M. lutarioriparius* at the end of growth (Figure 2), this suggests that a delayed harvest will not improve the biomass quality in terms of HM contents. In addition, the delayed harvest will reduce the biomass yield because of the foliage falling [13]. For these reasons, the optimal harvest time for *M. lutarioriparius* in Dongting Lake is in August because of the high biomass yield and relatively low HM contents.

4.3. Potential Ecological Risks Posed by the Unharvested M. lutarioriparius Biomass around the Dongting Lake

Although *M. lutarioriparius* is not a hyperaccumulator, a great amount of HM elements is removed annually by the aerial shoots with the harvested biomass. It is estimated that 2.02–2.64 $\times 10^4$ kg Cu, 1.54–1.77 $\times 10^5$ kg Mn, 1.02–1.19 $\times 10^5$ kg Zn, 0.56–0.84 $\times 10^3$ kg Cd, 0.79–1.05 $\times 10^4$ kg Cr and 0.62–1.24 $\times 10^4$ kg Pb can be absorbed by the plants annually and removed from the soil (Table S1). These potentials are achieved by harvesting the *M. lutarioriparius* biomass and using it to produce bio-products (mainly making paper during last decades). However, since 2019, all the paper mills around Dongting Lake were closed because of the concerns that they are obstacles to improving the water quality of Dongting Lake. With the shutdown of the paper mills and a lack of cost-effective and environmentally friendly techniques, *M. lutarioriparius* biomass utilization is impeded. In addition, during recent years, a lack of management practices and harvesting of the *M. lutarioriparius* biomass have posed serious ecological threats, such as water eutrophication risks [29]. The unharvested biomass would submerge into the water and become a source of soil and water contamination from decomposition and the leaching of biomass constituents. In addition, carbon sequestered by the *M. lutarioriparius* biomass would be released into the atmosphere in the form of greenhouse gases (CO_2 and CH_4) because of microbial decomposition under aerobic and anaerobic conditions, consequently reducing the carbon sequestration potential of the Dongting Lake wetland ecosystem [30,31]. Dead plant biomass releases large amounts of dissolved organic resources into waters and affects microbial communities' diversity [32]. From this it can be concluded that the unharvested *M. lutarioriparius* biomass will induce a series of ecological issues in the Dongting Lake wetland. Thus, cost-effective and environmentally friendly techniques are needed to expand the applications of the miscanthus biomass.

5. Conclusions

Across the Dongting Lake area, the soil is facing a severe Cd contamination problem. The heavily polluted soil finally results in a high HM concentration of the *M. lutarioriparius* biomass. The HM concentrations in the aboveground organs, except Mn and Zn in stems, do not reduce but keep increasing with the ongoing growing season. This is particularly true for the HM concentrations in leaves. The young shoots harvested in April can meet the food safety standard limitation of Cd and Cr, but not Pb. For the mature biomass harvested in December, the concentrations of different kinds of HM elements show in a descending order as Mn, Zn, Cu, Pb, Cr and Cd. For differences between southern and eastern Dongting Lake, the mature biomass from the southern part generally has higher Mn, Cd and Pb, but lower Cu, Zn and Cr concentrations than that from the eastern part.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/agronomy12051188/s1, Table S1: Average amounts of heavy metal bioaccumulated by *M. lutarioriparius* at different harvest time.

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References

1. Xu, Y.; Wu, S.; Huang, F.; Huang, H.; Yi, Z.; Xue, S. Biomodification of feedstock for quality-improved biochar: A green method to enhance the Cd sorption capacity of *Miscanthus lutarioriparius*-derived biochar. *J. Clean. Prod.* **2022**, *350*, 131241. [CrossRef]
2. Xue, S.; Kalinina, O.; Lewandowski, I. Present and future options for *Miscanthus* propagation and establishment. *Renew. Sustain. Energy Rev.* **2015**, *49*, 1233–1246. [CrossRef]
3. Yao, X.; Niu, Y.; Li, Y.; Zou, D.; Ding, X.; Bian, H. Heavy metal bioaccumulation by *Miscanthus sacchariflorus* and its potential for removing metals from the Dongting Lake wetlands, China. *Environ. Sci. Pollut. Res.* **2018**, *25*, 20003–20011. [CrossRef]
4. Xue, S.; Yi, Z.; Huang, H.; Yang, S. Development of the Dongting Lake grown *Miscanthus lutarioriparius* derived bio-industries: Problems and solutions. *Environ. Ecol.* **2022**, *4*, 33–38.
5. Xue, S.; Lewandowski, I.; Wang, X.; Yi, Z. Assessment of the production potentials of *Miscanthus* on marginal land in China. *Renew. Sustain. Energy. Rev.* **2016**, *54*, 932–943. [CrossRef]

6. Liao, X.; Long, Q.; Wang, H.; Yi, Z.; Xue, S. Interaction effects of *Miscanthus lutarioriparius*-derived biochar and cadmium passivators on rice cadmium content and yield. *J. Agro-Environ. Sci.* **2018**, *37*, 1818–1826.
7. Xue, S.; Guo, M.; Iqbal, Y.; Liao, J.; Yang, S.; Xiao, L.; Yi, Z. Mapping current distribution and genetic diversity of the native *Miscanthus lutarioriparius* across China. *Renew. Sustain. Energy. Rev.* **2020**, *134*, 110386. [CrossRef]
8. Zheng, C.; Iqbal, Y.; Labonte, N.; Sun, G.; Feng, H.; Yi, Z.; Xiao, L. Performance of switchgrass and *Miscanthus* genotypes on marginal land in the Yellow River Delta. *Ind. Crops Prod.* **2019**, *141*, 111773. [CrossRef]
9. Pidlisnyuk, V.; Stefanovska, T.; Lewis, E.E.; Erickson, L.E.; Davis, L.C. *Miscanthus* as a productive biofuel crop for phytoremediation. *Crit. Rev. Plant Sci.* **2014**, *33*, 1–19. [CrossRef]
10. Alexopoulou, E. *Perennial Grasses for Bioenergy and Bioproducts: Production, Uses, Sustainability and Markets for Giant Reed, Miscanthus, Switchgrass, Reed Canary Grass and Bamboo*; Academic Press: Cambridge, MA, USA, 2018.
11. Liu, J.; Liang, J.; Yuan, X.; Zeng, G.; Yuan, Y.; Wu, H.; Huang, X.; Liu, J.; Hua, S.; Li, F.; et al. An integrated model for assessing heavy metal exposure risk to migratory birds in wetland ecosystem: A case study in Dongting Lake Wetland, China. *Chemosphere* **2015**, *135*, 14–19. [CrossRef]
12. Wang, C.; Kong, Y.; Hu, R.; Zhou, G. *Miscanthus*: A fast-growing crop for environmental remediation and biofuel production. *GCB Bioenergy* **2021**, *13*, 58–69. [CrossRef]
13. Yang, S. Research on Genetic Diversity and Cultivation Techniques for High Biomass of *Miscanthus lutarioriparius*. Doctoral Dissertation, Hunan Agricultural University: Changsha, China, 2019.
14. Bao, S.D. *Soil Agrochemical Analysis*, 3rd ed.; China Agricultural Press: Beijing, China, 2000.
15. Ali, H.; Khan, E.; Sajad, M.A. Phytoremediation of heavy metals—Concepts and applications. *Chemosphere* **2013**, *91*, 869–881. [CrossRef] [PubMed]
16. Bonanno, G.; Vymazal, J.; Cirelli, G.L. Translocation, accumulation and bioindication of trace elements in wetland plants. *Sci. Total Environ.* **2018**, *631–632*, 252–261. [CrossRef] [PubMed]
17. Bonanno, G.; Cirelli, G.L. Comparative analysis of element concentrations and translocation in three wetland congener plants: *Typha domingensis*, *Typha latifolia* and *Typha angustifolia*. *Ecotoxicol. Environ. Saf.* **2017**, *143*, 92–101. [CrossRef] [PubMed]
18. Ghori, Z.; Iftikhar, H.; Bhatti, M.F.; Nasarum, M.; Sharma, I.; Kazi, A.G.; Ahmad, P. Chapter 15—Phytoextraction: The Use of Plants to Remove Heavy Metals from Soil. In *Plant Metal Interaction*; Ahmad, P., Ed.; Elsevier: Amsterdam, The Netherlands, 2016; pp. 385–409.
19. Krzesłowska, M. The cell wall in plant cell response to trace metals: Polysaccharide remodeling and its role in defense strategy. *Acta Physiol. Plant.* **2011**, *33*, 35–51. [CrossRef]
20. Adamczyk-Szabela, D.; Markiewicz, J.; Wolf, W.M. Heavy metal uptake by herbs. IV. influence of soil pH on the content of heavy metals in *Valeriana officinalis* L. *Water Air Soil Pollut.* **2015**, *226*, 106. [CrossRef]
21. Antonkiewicz, J.; Kołodziej, B.; Bielińska, E.J. The use of reed canary grass and giant miscanthus in the phytoremediation of municipal sewage sludge. *Environ. Sci. Pollut. Res.* **2016**, *23*, 9505–9517. [CrossRef]
22. van der Ent, A.; Baker, A.J.M.; Reeves, R.D.; Pollard, A.J.; Schat, H. Hyperaccumulators of metal and metalloid trace elements: Facts and fiction. *Plant Soil* **2013**, *362*, 319–334. [CrossRef]
23. Li, F.; Huang, J.; Zeng, G.; Yuan, X.; Li, X.; Liang, J.; Wang, X.; Tang, X.; Bai, B. Spatial risk assessment and sources identification of heavy metals in surface sediments from the Dongting Lake, Middle China. *J. Geochem. Explor.* **2013**, *132*, 75–83. [CrossRef]
24. Liang, J.; Liu, J.; Yuan, X.; Zeng, G.; Lai, X.; Li, X.; Wu, H.; Yuan, Y.; Li, F. Spatial and temporal variation of heavy metal risk and source in sediments of Dongting Lake wetland, mid-south China. *J. Environ. Sci. Health A* **2015**, *50*, 100–108. [CrossRef]
25. Peng, D.; Liu, Z.; Su, X.; Xiao, Y.; Wang, Y.; Middleton, B.A.; Lei, T. Spatial distribution of heavy metals in the West Dongting Lake floodplain, China. *Environ. Sci.-Proc. Impacts* **2020**, *22*, 1256–1265. [CrossRef] [PubMed]
26. Xiao, J. Study on Flavonoids and Analyse on Ingredients of *Triarrhena lutarioriparia* L. Liu in the Dongting Lake. Master's Thesis, Hunan Agricultural University, Changsha, China, 2016.
27. Liao, X.; Zhou, X.; Cai, D.; Wang, H.; Yi, Z.; Xue, S. Effects of application of *Miscanthus lutarioriparius*-derived biochar based-soil conditioner on photosynthetic characteristics and yield of rice (*Oryza sativa* L.). *J. Agric. Sci. Technol.* **2019**, *21*, 132–139.
28. Ab Rhaman, S.M.; Naher, L.; Siddiquee, S. Mushroom quality related with various substrates' bioaccumulation and translocation of heavy metals. *J. Fungi* **2022**, *8*, 42. [CrossRef] [PubMed]
29. Geng, M.; Wang, K.; Yang, N.; Li, F.; Zou, Y.; Chen, X.; Deng, Z.; Xie, Y. Evaluation and variation trends analysis of water quality in response to water regime changes in a typical river-connected lake (Dongting Lake), China. *Environ. Pollut.* **2021**, *268*, 115761. [CrossRef]
30. Kayranli, B.; Scholz, M.; Mustafa, A.; Hedmark, Å. Carbon storage and fluxes within freshwater wetlands: A critical review. *Wetlands* **2010**, *30*, 111–124. [CrossRef]
31. Carmichael, M.J.; Helton, A.M.; White, J.C.; Smith, W.K. Standing dead trees are a conduit for the atmospheric flux of CH_4 and CO_2 from wetlands. *Wetlands* **2018**, *38*, 133–143. [CrossRef]
32. Geng, M.; Zhang, W.; Hu, T.; Wang, R.; Cheng, X.; Wang, J. Eutrophication causes microbial community homogenization via modulating generalist species. *Water Res.* **2022**, *210*, 118003. [CrossRef]