



# Article Interaction Mechanism of Fe, Mg and Mn in Karst Soil-Mango System

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Abstract: Manganese (Mn), an essential trace element for plants in which it is involved in redox reactions as a cofactor for many enzymes, represents an important factor in environmental contamination. Excess Mn can lead to toxicity conditions in natural and agricultural sites. Manganese toxicity is one of the most severe growth limiting factors in acid soil, which accounts for 21% of the total arable lands in China. The more significant part of Mn-toxicity is its interactions with other mineral elements, in particular with phosphorus (P), calcium (Ca) and iron (Fe). The application of P or Ca can be beneficial in the detoxification of manganese, whereas Mn seems to interfere with Fe metabolism. Manganese toxicity varies with plant species, nutrients, and the soil environment. Mango is the main economic fruit in the karst area of the subtropical region of China. The karst soil in the mango orchard is characterized by high Fe, Mn and Mg. In order to explore the interaction among Fe, Mg, and Mn in karst soil and mango systems under high Mn conditions, a typical mango orchard in the karst depression landform in Baise in southern China was selected to study the effects of Fe and Mg on the toxic expression of Mn in mango plants and the interaction mechanism of Fe-Mn-Mg in mango plants. The results show that: (1) the mango growth status is closely correlated with  $Fe^{2+}$  (active iron) and Mg under the same soil Mn concentration; (2) The black spots on mango leaves were mainly caused by Fe and Mn. There is a lot of  $Fe^{3+}$  and  $Mn^{3+}$  in the black spots, which accounts for more than 90% of the total; (3) In addition, the studies also showed that the Fe and Mg inhibited the expression of Mn toxicity in mango. Conclusively, the interaction effect of Fe, Mn, and Mg is an important factor that affects mango growth, which can indicate the status of the soil and plants.

Keywords: karst area; Mn toxicity; soil-plant system; mango

## 1. Introduction

Manganese (Mn) is one of the essential trace elements in living organisms [1]. As an important nutrient element, Mn deeply controls a plant's physiological, metabolic, synthesis and enzymes activation [2–4]. Mn is also closely related to plant carbon and nitrogen uptake and utilization [5]. However, excessive Mn concentration in plants would affect the enzyme activity, and it always Mn toxicity. The high Mn poisoning reaction generally shows leaves' yellowness and blackness, roots lesions, and plant biomass reduction [6,7]. In acidic soil, the effects of Mn's toxicity are highlighted due to the inert Mn activated by acidic materials [8,9]. The strength of Mn toxicity lies on the conversion of Mn<sup>2+</sup> to Mn<sup>3+</sup> under photosynthetic oxidation. The cells and tissues around the Mn<sup>3+</sup> will then be over-oxidized [10].



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**Copyright:** © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). Iron (Fe) and magnesium (Mg) are two other important elements in the soil. Studies have shown that Fe and Mg can repair Mn toxicity [11,12]. Manganese and iron (Fe) showed antagonism. Fe<sup>2+</sup> can reduce  $Mn^{3+}$  to  $Mn^{2+}$  [13]. Magnesium can promote plant photosynthesis and product synthesis and then alleviates the Mn toxicity [14–16]. The  $Mn^{3+}$  reduced by Fe, Fe<sup>2+</sup>, and Fe, Fe<sup>2+</sup> would be oxidized to Fe<sup>3+</sup>, resulting in Fe deficiency and chlorosis in plant leaves [13]. Meanwhile, high Mn would inhibit the uptake of the Fe and Mg from soil to plants [17,18]. Mn poisoning has been demonstrated to occur when the Mn content is higher than 500 mg/kg in soil [19]. However, this toxic effect will be reduced when the high content of Fe and Mg co-exist with Mn in soil [20]. The mechanism of this phenomenon has not been clearly explained until now.

In the subtropical southwest of China, mangoes are widely cultivated. However, the mango trees differentiate in the karst and non-karst areas. Under the same content of Mn in the same climate environment, mango trees in karst areas are more susceptible to Mn toxicity when compared with non-karst areas. Therefore, we hypothesized that in the karst area, the contents of active Fe and Mg are low, and plants cannot absorb and utilize it enough to resist Mn toxicity compared to that in non-karst areas.

To understand the relationship between the conditions of Mn toxicity with Fe and Mg contents, this study choose one typical karst that has been planting mango plants for more than ten years, and the same site with the same experience in mango planting in non-karst areas as the control in Baise, which is located in the southwest of China. We studied the Mn toxicity under different Mn content both in the soil and in mango leaves. In addition, other soil properties, particularly the Fe and Mg content, were also determined to reveal the influencing factors of Mn toxicity.

#### 2. Research Methodology and Data Sources

## 2.1. Study Area

Both karst and non-karst areas are located at Tianyang District, Baise City, China (Figure 1a). The karst landscape here is karst peak-cluster depression and the non-karst area is the thick Quaternary fluvial impact plain. The annual average temperature is 18–22 °C and the annual average precipitation is 1100–1200 mm. The annual sunshine duration is 1600–1900 h. This is the main mango producing area in China and in all of Southeast Asia.

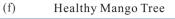


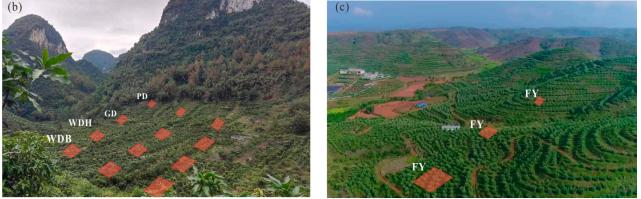
(d) Nutritional toxicity Mango Tree



Sub-healthy Mango Tree







Sampling sites in karst areas

500

500

(a)

0

1000 Kilometers

Baise, Guangxi

Sampling sites in non-karst areas

Figure 1. (a). The geographical location of the study area; (b). The sampling sites in karst areas; (c). The sampling sites in non-karst areas; (d). Nutritional toxicity mango tree; (e). Sub-healthy mango tree; (f). Healthy mango tree.

## 2.2. Sampling

The plant leaves samples in karst areas were collected from three landforms: the slope, depression, and transition zones (Figure 1b). The leaves' mango samples in karst areas show yellow or black spots (Figure 1d). The leaf samples in the non-karst areas were collected from healthy mango trees (Figure 1c). The soil samples including topsoil and the rhizosphere soil were collected from the same site at the same time (Table 1). The rhizosphere soil's physical and chemical properties are shown in Table S1.

Sampling Site	Locations	Landform	Health Degree	Sample Types	Stage
FY	Non karst area	Plain	Healthy leaves	Soil and leaves	sprout, young leaves and old leaves
PD WDH WDB		Sloping bottomland bottomland	Unhealthy leaves (include sub-healthy leaves and nutrional toxicity leaves)	Soil samples and plant samples were collected according to different landforms. The plant samples were divided	sprout, young leaves and old leaves
GD	Karst area	transitional zone	- · ·	into four types: unhealthy leaves, sub-healthy leaves, nutritional toxicity leaves, and black spots on nutritional toxicity leaves	

Table 1. The sample information.

Note: Fy, non-karst area, PD, slope landform in karst area; WDH, depression landform in karst area with relatively good growth; WDB, depression landform in karst area with relatively poor growth; GD, transition zone in karst area.

The health degree of the leaves is divided into healthy (Figure 1f), sub-healthy (Figure 1e) and nutritional toxicity (Figure 1d) according to the number of black spots on mango leaves under the advice of experts in the field of plant nutrition. The sprout leaves, young leaves and old leaves were distinguished and collected separately. The black spots on nutritional toxicity leaves were gathered to analyze their composition.

Sample plots of  $10 \times 10$  (Figure 1c) were randomly set in different slope positions of both the karst and non-karst area according to Ding [21]. Both of the 0–20 cm and rhizosphere soils were taken from the four corners and the center in each plot. We sampled the mango leaves from the same place. Soil samples in each plot were mixed together and screened to 2 mm for the next laboratory analysis. The leaf samples were dried at 50 °C for 24 h for laboratory analysis.

## 2.3. Laboratory Analysis

Soil organic carbon (SOC) was measured using the K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>-H<sub>2</sub>SO<sub>4</sub> oxidation method; total nitrogen (TN) concentration was measured with the Semi-Micro Kjeldahl method; total P was determined using HClO<sub>4</sub>-H<sub>2</sub>SO<sub>4</sub> digestion followed by a Mo–Sb colorimetric assay, and total potassium (TK) concentration was measured with the HF-HClO4 flame photometric method [22]. Available P (AvP) was determined by the NaHCO<sub>3</sub>-extraction method [23]. Soil calcium (Ca), magnesium (Mg), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) were extracted by  $HNO_3$ -HF-HClO<sub>4</sub> and analyzed by inductively coupled plasma atomic emission spectroscopy (ICP-AES, Icap6300, Thermo Fisher Scientific, Waltham, MA, USA). The available elements were determined by ICP-AES after extraction of diethylene triamine Penta acetic acid-calcium chloride-triethanolamine buffer solution. Soil pH was determined at a 1:2.5 (*w:v*) soil: water ratio by a DMP-2 mV/pH detector (Quark Ltd., Nanjing, China). The S contents were determined using the high frequency combustion infrared absorption method. The contents of P, K, Ca, Mg, Fe, Mn, Cu, Zn, and B were digested using nitric acid and hydrogen peroxide and determined by inductively coupled plasma mass spectrometry (ICP-MS, IcapQc, Thermo Fisher Scientific, USA). The leaf active iron (Fe<sup>2+</sup>) was extracted using 1 mol/L of HCl at a ratio of 1:10 (continuous oscillation for 5 h) in the Kunming Natural Resources Comprehensive Survey Center of China Geological Survey and analyzed and tested using ICP-AES according to Zou [24], Takkar and Kaur [25] and Pierson and Clark [26]. Three replicates were performed for each soil sample.

#### 2.4. Statistical Analysis

The statistical analysis was undertaken by EXCEL2016 (Microsoft, Washington, DC, USA), SPSS24 (IBM SPSS Corp., Chicago, IL, USA), and Canoco5 (Microcomputer Power, New York, NY, USA). A Spearman correlation analysis was used to study the correlation between the soil and plant nutrient elements. A redundancy analysis was used to analyze the explanatory degree of Fe, Mn, and Mg and Fe/Mn, Fe/Mg, and Mg/Mn in the soil and plant leaves. The Diagnosis and Recommendation Integrated System (DRIS) nutritional diagnosis of the plant leaves was carried out using Python. Multiple comparisons were conducted by the Duncan method when the variance was homogeneous or by the T2 Tamhane test when the variance was not homogeneous.

#### 3. Results

#### 3.1. The Background Nutrient Elements in the Soil

The topsoil and plants analysis results showed that there was a significant difference in the topsoil element contents between the different sampling sites in karst areas and non-karst areas (P < 0.05), but there was no significant difference in the karst areas (Table 2). The effective Fe content of soil in karst areas is less than 50% of that in non-karst areas, and most of the available P was lower than the detection threshold of 0.25 mg/kg. In addition, the contents of all the nutrient elements in the soils of the different landforms in the karst areas were much higher than those in the non-karst areas. The available Mn content was 20–30 times higher than that in the non-karst areas, and the total Mn content in the soil (1560 mg/kg) was nearly two times higher than the national average Mn content 850 mg/kg [27]. The exchangeable Mg content was also several times higher than that in the non-karst areas.

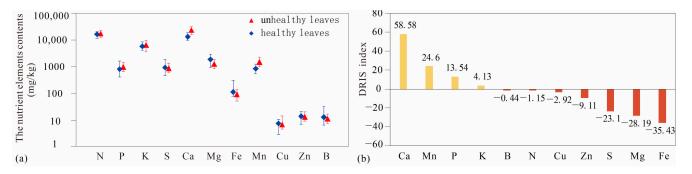
Parameter	Non-Karst Area	Karst Area				
	Fy	PD	WDH	WDB	GD	
SOM (gC/kg)	$20.5\pm3.0~\mathrm{a}$	$35.86\pm4.65~\mathrm{b}$	$25.27\pm0.61~\mathrm{b}$	$28.7\pm2.66~\mathrm{b}$	$30.2\pm9.64b$	
TN (gN/kg)	$1.11\pm0.07~\mathrm{a}$	$2.5\pm0.21b$	$2.12\pm0.05b$	$2.2\pm0.17~\mathrm{b}$	$2.32\pm0.33b$	
C/N	$10.69\pm1.12~\mathrm{a}$	$8.32\pm0.38b$	$6.9\pm0.15\mathrm{b}$	$7.57\pm0.37~\mathrm{b}$	$7.44\pm1.26\mathrm{b}$	
pН	$4.79\pm0.07~\mathrm{a}$	$5.91\pm0.43$ a	$5.75\pm0.26$ a	$6.34\pm0.61~\mathrm{a}$	$6.22\pm1.14$ a	
CEC (cmol/kg)	$4.09\pm1.88~\mathrm{a}$	$15.21\pm1.66~b$	$12.32\pm1.56~\text{b}$	$14.08\pm2.18~\text{b}$	$13.23\pm4.28~b$	
N (mg/kg)	$40.83\pm18.46~\mathrm{a}$	$150\pm10.82~\mathrm{b}$	$114.33\pm2.08~\mathrm{b}$	$117\pm5.57~\mathrm{b}$	$127\pm14.73\mathrm{b}$	
P (mg/kg)	$3.85\pm2.97~\mathrm{a}$	$0.25\pm0.25$ a	$0.7\pm0.45~\mathrm{a}$	$0.99\pm0.78~\mathrm{a}$	$1.1\pm1.1$ a	
K (mg/kg)	$37.77 \pm 22.73$ a	$175\pm41.58~\mathrm{b}$	$91\pm10.11~\mathrm{ab}$	$97.67 \pm 16.44~\mathrm{ab}$	$123\pm23.43b$	
S (mg/kg)	$12.97\pm3.13~\mathrm{a}$	$26.3\pm12.88~\mathrm{a}$	$9.1\pm1.29~\mathrm{a}$	$8.78\pm5.6~\mathrm{a}$	$12.49\pm3.80~\mathrm{a}$	
Ca <sub>ex</sub> (cmol/kg)	$0.73\pm0.15$ a	$9.37\pm3.3b$	$6.88\pm1.30~\text{b}$	$10.96\pm3.94b$	$11.34\pm9.84b$	
Mg <sub>ex</sub> (cmol/kg)	$0.17\pm0.01~\mathrm{a}$	$0.69\pm0.17\mathrm{b}$	$0.5\pm0.03~\mathrm{ab}$	$0.6\pm0.08~{ m b}$	$0.5\pm0.18b$	
Fe (mg/kg)	$108.4\pm84.83~\mathrm{a}$	$35.73\pm9.47~b$	$50.1\pm4.45~\mathrm{b}$	$44.03\pm19.67b$	$38.3\pm21.11\mathrm{b}$	
Mn (mg/kg)	$3.58\pm1.8$ a	$65.77\pm19.42\mathrm{b}$	$134\pm14.8b$	$127.3\pm39.92\mathrm{b}$	$104.87\pm64\mathrm{b}$	
Cu (mg/kg)	$0.91\pm0.39~\mathrm{a}$	$1.03\pm0.07~\mathrm{a}$	$1.8\pm0.03~\mathrm{a}$	$1.82\pm0.47$ a	$1.55\pm0.62~\mathrm{a}$	
Zn (mg/kg)	$0.9\pm0.61~\mathrm{a}$	$0.7\pm0.11$ a	$1.26\pm0.09~\mathrm{a}$	$1.3\pm0.21~\mathrm{a}$	$0.99\pm0.35~\mathrm{a}$	
B (mg/kg)	$0.08\pm0.04~\mathrm{a}$	$0.2\pm0.05~\mathrm{a}$	$0.21\pm0.07~\mathrm{a}$	$0.2\pm0.02~\mathrm{a}$	$0.16\pm0.04~\mathrm{a}$	

**Table 2.** The topsoil physical and chemical properties of the different landforms in the karst areas and non-karst areas.

Note: Fy, non-karst area, PD, slope landform in karst area; WDH, depression landform in karst area with relatively good growth; WDB, depression landform in karst area with relatively poor growth; GD, transition zone in karst area; Ca<sub>ex</sub>, exchangeable Ca; Mg<sub>ex</sub>, exchangeable Mg; different letters in the same line indicate significant differences in nutrient elements in different landforms at the level of P < 0.05.

#### 3.2. The Nutrient Status of Leaves with Excessive Mn

The laboratory analysis showed that the contents of Ca, Mg, Fe, and Mn in the mango leaves in the karst areas and non-karst areas showed significant differences (Figure 2a). Among them, the Ca content in the unhealthy leaves is 24.18 g/kg, which was much higher than that in the healthy leaves (13.03 g/kg) due to the difference in the geological background. The Mn content in the unhealthy leaves  $(1333.7 \pm 625 \text{ mg/kg})$  was also much higher than that in the healthy leaves  $(816.7 \pm 155.35 \text{ mg/kg})$ . The Mn content in the leaves has exceed the critical value of Mn toxicity in plant leaves (500 mg/kg) that was defined by An and Fang [19]. The Fe and Mg were also significantly higher than those in the unhealthy leaves.



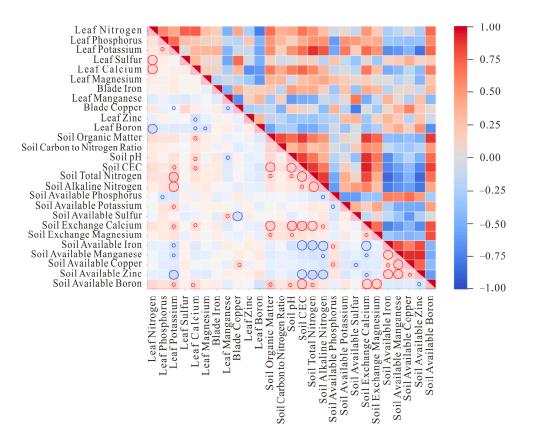
**Figure 2.** (a) The nutrient elements contents of the unhealthy leaves in the karst areas and healthy leaves in the non-karst areas; (b) The Diagnosis and Recommendation Integrated System (DRIS) nutritional diagnosis: yellow represents excess and red represents deficiency; the bar chart shows the abundance and deficiency of the nutrient elements in the unhealthy leaves compared to the healthy leaves.

Healthy leaves in the non-karst areas were used as the background value. Therefore, the DRIS nutritional diagnosis according to Gott et al. [28] and Huang et al. [29] was carried out on the karst unhealthy mango leaves. The results showed that the unhealthy leaves were severely deficient in Fe and Mg, and severely in excess in Mn when compared with the healthy leaves (Figure 2b).

### 3.3. The Fe, Mn, and Mg and the Nutrient Status of the Mango Leaves

A Spearman correlation analysis showed that there was no significant correlation between most of the elements in the leaves, and the correlation between the Mg, Fe, and Mn elements in plant leaves and most of the soil indexes was also not significant (Figure 3). Although the exchangeable Ca had a significant negative correlation with the available Fe, Mn, and Zn in the soil, it had no direct correlation with the nutrient elements in the leaves. Similarly, the significantly high Ca content in the leaves was only negatively correlated with Zn and B but had no correlation with the other elements such as Mg, Fe, and Mn.

The redundancy analysis (RDA) that used Fe, Mn, Mg and Fe/Mn, Fe/Mg, Mg/Mn as the environmental factors had high explanatory rates (Figure 4). The explanatory rate for the nutrient elements in the leaves was 91.5% (Figure 4a), and the explanatory rate for the soil physical and chemical indexes was 99.9% (Figure 4d). Fe, Mn, and Mg and their combined effects (Fe/Mn, Fe/Mg, and Mg/Mn) had significant differences in the explanatory rates). While each of the environmental factors had low explanatory rates for the plant nutrient elements (Figure 4h,i), the explanatory rate of the combined factors reached 99.8% (Figure 4g).



**Figure 3.** A Spearman correlation between the soil index and leaf nutrient elements in the karst areas. The red and blue circles represent positive and negative correlations, respectively, and the large circles and small circles represent a significance level of 0.01 and 0.05, respectively.

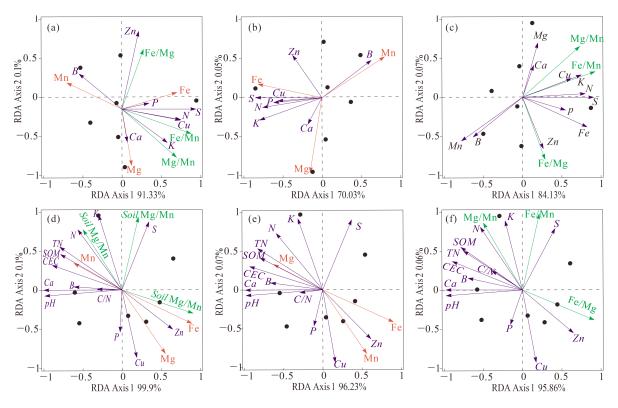
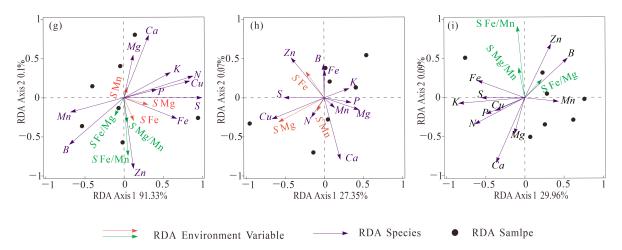


Figure 4. Cont.

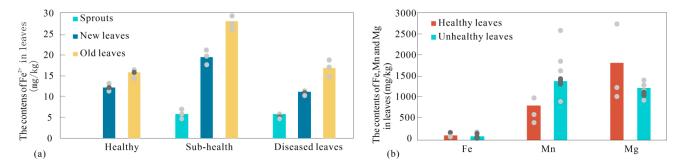


**Figure 4.** The explanation rate of Fe, Mn, and Mg in the plants and soil. Fe/Mn, Fe/Mg, and Mg/Mn are environmental factors that affecting the plant nutrients and soil's physical and chemical properties, respectively. (**a**–**c**): A redundancy analysis (RDA) for plant nutrition. (**d**–**f**) A redundancy analysis for the soil; (**g**–**i**). A redundancy analysis for the soil-plant indices.

The RDA results showed that Fe, Mn, and Mg and their combined effects had significant effects on plant nutrition and the soil's physical and chemical properties. These results could explain the nutrient status of the plants and soil adequately.

## 3.4. The Relationship between the Fe, Mn, and Mg in Mango Leaves

The detection and analysis results of the healthy leaves, sub-healthy leaves, and nutritional toxicity leaves showed that the Fe<sup>2+</sup> content in the healthy leaves was lower than that in the sub-healthy leaves at the stage of leaf growth. However, the Fe<sup>2+</sup> content in the sub-healthy leaves was significantly higher than that in the nutritional toxicity leaves at all leaf growth stages (Figure 5a). Additionally, the total Fe (113.3 mg/kg) and total Mg (1823.3 mg/kg) contents in the healthy leaves were significantly higher than those in the unhealthy leaves (sub-health and nutritional toxicity leaves mixed). The total Fe and total Mg in unhealthy leaves were 86.7 mg/kg and 1230 mg/kg, respectively (Figure 5b). The total Mn content (816.7 mg/kg) in the healthy leaves was significantly lower than that in the unhealthy leaves (1333.6 mg/kg). Through analyzing the contents of Fe<sup>2+</sup>, Fe<sup>3+</sup>, and Mn in the black spots of the nutritional toxicity leaves, the results showed that the total Fe content was 95.86 mg/kg, the Fe<sup>2+</sup> content was 6.79 mg/kg, and the total Mn content was 92.9%.



**Figure 5.** Relationship between leaf growth and contents of Fe, Mn and Mg elements. (**a**) The active iron (Fe<sup>2+</sup>) in the leaves at different leaf growth stages; (**b**) The total Fe, Mn, and Mg contents in the healthy and unhealthy leaves.

## 4. Discussion

## 4.1. The Relationship between Mn Toxicity and Fe, Mg Activity

In the study of trace elements, the relationship between manganese and iron is the closest, and the antagonism between them was shown [12]. Previous studies have proved that excessive Mn can cause the loss of intracellular Fe [18]. Similarly, the application of iron fertilizer can also significantly reduce the absorption and transport of manganese by plants [30]. In this study, the DRIS nutritional diagnosis showed that the Mn content in the leaves of the unhealthy plants in the karst areas was higher than that in the non-karst area, while the Fe and Mg contents were lower. Accordingly, soils in the karst areas showed a high available Mn content and low available Fe content compared to the soil in the non-karst areas. Research showed that both deficit or excess manganese nutrition could induce disorders in the uptake of Mn and Fe nutrients, which may influence plant yielding [31]. Our nutritional toxicity mango trees have significantly higher levels of manganese and significantly lower levels of iron and magnesium than healthy mango trees. Although healthy mango trees also have higher levels of manganese, the higher levels of iron and magnesium mitigate the manganese poisoning and the mango trees still grow healthily [32,33].

In karst soil, Mn and Ca would inhibit the absorption of Fe and My in plants [34]. For this reason, the nutrient elements in karst soils may cause the imbalance of nutrient elements in karst plants. The redundancy analysis in this study proved that the comprehensive effects of Fe-Mn-Mg explained the nutritional status of plants perfectly. Previous studies found that mango's healthy growth status is greatly related to Mn and Fe abundance but Mg deficiency in plants. In these conditions, the mango leaves always appeared with wrinkled, yellow, black spots, and the trees were short, had low biomass, and were of low yield [13,35,36]. Our studies also showed that it was difficult to control the mango nutritional status with one single element. The Fe-Mn-Mg and its combined effects can explain this.

Lambers et al. [37] believed that the Mn content in plant leaves was significantly negatively correlated with the available P content in the soil. The P acquisition strategy of plants in p-restricted acidic soil can promote the activation of P and Mn [38]. The soil P in the karst area was significantly restricted. The available P, Mn, Fe, and Zn in the rhizosphere soil was significantly activated relative to the surface soil. These results are highly consistent with the previous study of Lambers et al. [39]. The available P in the rhizosphere soil was more than 100 times higher than that of the topsoil (Figure 6). In addition, the Mn was also greatly activated in this soil environment. Therefore, the P acquisition may be one of the main reasons for the high Mn content in the mango leaves.

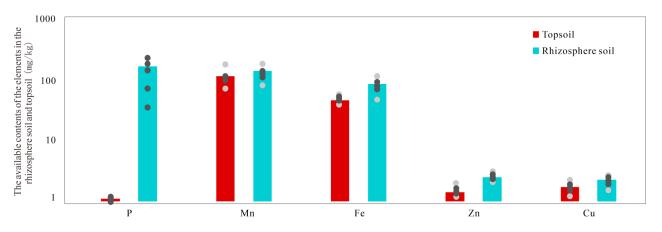


Figure 6. The available contents of the elements in the rhizosphere soil and topsoil.

## 4.2. Expression of the Mn Toxin in the Plants

Studies showed that plant photosynthesis could oxidize  $Mn^{2+}$  to  $Mn^{3+}$  [10] due to the reversible redox reaction of the  $Mn^{3+} + Fe^{2+} \rightleftharpoons Fe^{3+} + Mn^{2+}$  [13]. The higher the available Mn in the soil, the more the Mn absorption in the plants. However, when the Fe<sup>2+</sup> content in plants is not enough to reduce  $Mn^{3+}$  to  $Mn^{2+}$ , more  $Mn^{3+}$  and  $Fe^{3+}$  will be accumulated in the plants [10]. As Fe<sup>3+</sup> and  $Mn^{3+}$  has no physiological effect but a side-effect function on plants, this will result in poorer plant growth. This hypothesis is consistent with the finding in this study that the unhealthy leaves contained more  $Mn^{3+}$  and  $Fe^{3+}$  than the healthy leaves. Mg is considered to be an important element for photosynthesis, promoting and photosynthate movement [14,35]. When the Mg is not sufficient in plants, the inertial  $Mn^{3+}$  and  $Fe^{3+}$  will accumulate. Their strong oxidation makes the surrounding cells and tissues over-oxidized [10]. In this study, a large amount of  $Fe^{3+}$  and  $Mn^{3+}$  were found in the black spots on the mango leaves (Figure 7). Some studies have also indicated that the Mn in the black spots is  $Mn^{4+}$ , i.e., the MnO<sub>2</sub> deposit [13]. However, we did not find this phenomenon here, and the higher valence of Mn is probably related to the higher intensity of oxidation.

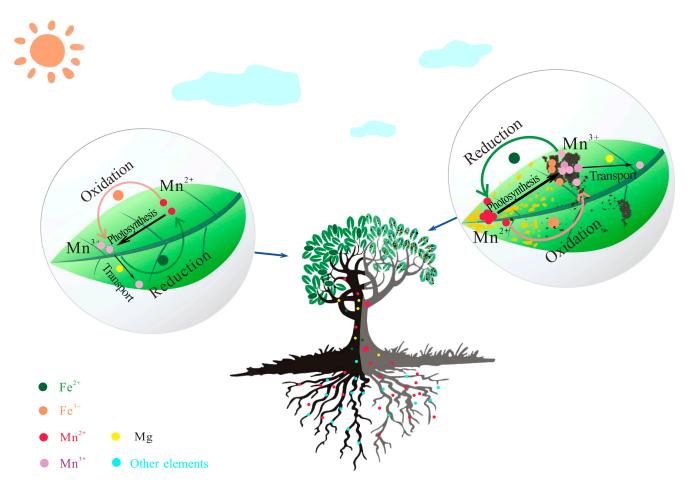


Figure 7. The Mn toxicity expression mechanism under the interaction of Fe, Mn, and Mg.

On the contrary, when the contents of Fe and Mg are at a sufficient level, the Mn toxicity will be reduced and even not shown on the leaves. This study proved this result, that is, mangoes with high Mn content are still healthy when the content of Fe and Mg are at a relatively high levels. Thus, the expression of the Mn toxicity is strictly restricted by Fe and Mg in both soil and plants.

## 5. Conclusions

The measurement of element contents in soil and mango leaves combined with the  $Fe^{2+}$  contents in mango leaves revealed that the leaves of the mango plants in karst areas have a high Mn content but insufficient contents of Fe and Mg. The lack of available Fe content in the soil and the inhibition of Fe activity by high concentrations of Ca and Mn are the main reasons for Fe deficiency in the plant leaves, and the physiological activity of Fe is also inhibited by the high Mn content in the leaves. The results of this study confirm our previous hypothesis that both Fe and Mg are important factors restricting the expression of Mn toxicity symptoms in mango plants. Specifically, the reduction effect of  $Fe^{2+}$  on  $Mn^{3+}$  is an important mechanism for Fe to alleviate Mn toxicity. Meanwhile, this study also found that the balance between Fe, Mn and Mg elements is an important factor affecting plant growth, and the imbalance of Fe/Mn and Mg/Mn ratios will affect the growth of plants. These findings can contribute to improve degraded soil restoration in areas of rocky karst desertification.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/land12010256/s1, Table S1: Total and available content of nutrient elements in rhizosphere soil.

**Author Contributions:** Investigation, C.H.; Methodology, M.L.; Project administration, L.Z.; Validation, F.Z.; Visualization, Z.Z.; Writing—original draft, C.X.; Writing—review & editing, H.Y. All authors have read and agreed to the published version of the manuscript.

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