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Iron Availability in Tropical Soils and Iron Uptake by Plants

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ABSTRACT: Given the increase in crop yields and the expansion of agriculture in low fertility soils, deficiency of micronutrients, such as iron, in plants grown in tropical soils has been observed. The aim of this study was to evaluate Fe availability and Fe uptake by corn (*Zea mays* L.) plants in 13 different soils, at two depths. Iron was extracted by Mehlich-1, Mehlich-3, and $CaCl_2$ (Fe-CC) and was fractionated in forms related to low (Fe_o) and high (Fe_d) crystallinity pedogenic oxyhydroxides, and organic matter (Fe_p) using ammonium oxalate, dithionite-citrate, and sodium pyrophosphate, respectively. In order to relate Fe availability to soil properties and plant growth, an experiment was carried out in a semi-hydroponic system in which part of the roots developed in a nutrient solution (without Fe) and part in the soil (the only source of Fe). Forty-five days after seeding, we quantified shoot dry matter and leaf Fe concentration and content. Fe_d levels were high, from 5 to 132 g kg^{-1}, and Fe_o and Fe-CC levels were low, indicating the predominance of Fe as crystalline oxyhydroxides and a low content of Fe readily available to plants. The extraction solutions showed significant correlations with various soil properties, many common to both, indicating that they act similarly. The correlation between the Mehlich-1 and Mehlich-3 extraction solutions was highly significant. However, these two extraction methods were inefficient in predicting Fe availability to plants. There was a positive correlation between dry matter and Fe levels in plant shoots, even within the ranges considered adequate in the soil and in the plant. Dry matter production and leaf Fe concentration and content were positively correlated with Fe_p concentration, indicating that the Fe fraction related to soil organic matter most contributes to Fe availability to plants.

Keywords: Mehlich, corn (*Zea mays*), micronutrient, nutrient solution, *Latossolo*.

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INTRODUCTION

Humid tropical climate soils generally exhibit a reduced number of mineral species and advanced pedogenetic development. *Latossolos* (Oxisols), the soil class of greatest geographical distribution in Brazil, are most prominent in this climate, in which minerals such as kaolinite and Fe and Al oxyhydroxides (hematite, goethite, and gibbsite, in addition to quartz) predominate. As a consequence of the variation in their charges and degree of crystallinity, these oxides exercise considerable influence on the physical and chemical properties of the soils (Kämpf et al., 2000).

In spite of the high total concentrations of Fe in the soil, this element, which is originally in the structure of primary minerals, undergoes oxidation and precipitates as compounds of low solubility, limiting its availability to plants (Kämpf and Curi, 2000). The state of oxidation of Fe and its bond to soil components, as well as the degree of crystallinity of the mineral, also affect its availability (Abreu et al., 2007).

So that the availability of any nutrient in the soil is correctly evaluated, selection of an adequate extractant is fundamental. The methods traditionally used to extract Fe from the soil involve dilute acids and chelating agents; Mehlich-1 and DTPA are most used. The Mehlich-1 extraction solution, composed of strong dilute acids, acts on labile and non-labile forms of metals, and is relevant in the evaluation of their availabilities (Abreu et al., 2007). The Mehlich-3 multielemental extraction method has also been the object of considerable study; the purpose of this extraction method is to simplify analytical procedures and reduce laboratory costs in routine analysis, due to its greater versatility (Schoninger et al., 2012). Nevertheless, results with diverse extractants are not yet consistent, and the correlation between concentrations in the soil and in the plant vary from low and not significant to high and significant for various crops (Rodrigues et al., 2001; Simonete and Kiehl, 2002; Abreu et al., 2004; Moreira et al., 2009).

Fractionation techniques also play an important role in understanding the physical and chemical behavior of soils (Inda Junior and Kämpf, 2003) and in predicting the availability of elements in the soil-plant system (Nascimento et al., 2005). Thus, Fe concentrations in the soil have basically been evaluated under three forms: constituents of high and low crystallinity pedogenetic oxyhydroxides, and forms related to soil organic matter (Kämpf et al., 2000).

In well-drained and highly-weathered soils, such as *Latossolos* in general, the Fe_o/Fe_d ratio tends to be quite low, showing a predominance of Fe oxides of greater crystallinity (Melo et al., 2001). Although the influence of mineralogy, texture, and organic matter on the Fe dynamics in the soil is known, it is still necessary to relate knowledge of soil mineralogy and chemistry to plant fertility and mineral nutrition.

Little is mentioned in the literature regarding Fe deficiency in plants grown in tropical soils, although there are reports that have shown typical symptoms of this deficiency in the field even in acid soils and soils with high total Fe concentrations (Nunes et al., 2004; Schulthais, 2013). In fact, through the increase in nutrient export from increasing crop yield, and expansion of agricultural activity into soils of low natural fertility and with more sandy texture, more cases of deficiency of Fe and of other micronutrients can be expected (Abreu et al., 2004).

We hypothesize that, despite of the high amount of Fe in our highly weathered soils, it is found in chemical species that are very stable in the environment and not readily available to plants. Therefore, the aim of this study was to evaluate availability of Fe and its uptake by plants in tropical soils in which this element is, in a general sense, abundant.

MATERIALS AND METHODS

Soils were selected with a preference for clayey soils under natural vegetation or in regeneration. This is because sandy soils have quartz as the main mineral and exhibit low natural Fe concentrations (Abreu et al., 2007). In contrast, Fe is abundant in clayey soils and is present in various minerals, above all in oxyhydroxides (Ferreira et al., 1999). The preference for non-cultivated areas is explained by the fact that the effects of soil fertility management practices, such as liming and P fertilization, have been widely studied, and Fe deficiency is much more understood in these cases (Nunes et al., 2004).

Samples of 13 soils from Minas Gerais were collected at depths of 0.00-0.20 and 0.20-0.40 m, including regions where there is evidence of Fe deficiency in plants, such as in Patos de Minas (Table 1).

After obtaining air dried fine earth (ADFE), particle size analysis and determination of moisture equivalent (ME) were carried out according to Claessen (1997). Determination was made of pH in water (1:2.5) and of organic C by the modified Walkley-Black method (Nelson and Sommers, 1982). Ca^{2+}, Mg^{2+}, and Al^{3+} were extracted with 1 mol L^{-1} KCl and K, P, Zn, Cu, and Mn by the Mehlich-1 extractant (Claessen, 1997). The P-rem was determined according to Alvarez V et al. (2000) and H+Al with a 0.5 mol L^{-1} calcium acetate solution in acetic acid at pH 7.0 (Table 2).

For the extractions of Fe by Mehlich-1 (0.0125 mol L^{-1} H_2SO_4 and 0.05 mol L^{-1} HCl), Mehlich-3 (HOAc, NH_4NO_3, NH_4F, HNO_3, and EDTA at 0.2, 0.25, 0.015, 0.013, and 0.001 mol L^{-1}, respectively), and $CaCl_2$ (0.01 mol L^{-1}), the soil/solution ratio of 1:10 was used, with shaking for 5 min and rest for 16 h.

In extraction of Fe from high crystallinity pedogenetic oxyhydroxides in the clay fraction (Fe_d), dithionite-citrate (0.2 mol L^{-1} $Na_3C_6H_5O_7$ and 500 mg of $Na_2S_2O_4$) was used in a water bath at 70 °C for 4 h. For the low crystallinity pedogenetic oxyhydroxides (Fe_o), extraction in the clay fraction was carried out with ammonium oxalate (0.2 mol L^{-1} $C_2H_8N_2O_4$, pH 3.0) in the absence of light for 4 h (McKeague and Day, 1966). To extract the Fe bound to organic matter of the ADFE (Fe_p), sodium pyrophosphate in alkaline medium (0.1 mol L^{-1} $Na_4P_2O_7$, pH 10.0) was used for 2 h (Wang, 1978). After the samples were centrifuged, Fe was determined by atomic absorption spectrophotometry.

An experiment with corn (*Zea mays* L.) plants in a greenhouse was carried out to relate Fe availability in the soil with uptake by plants. To ensure supply of Fe exclusively from the soil, a system analogous to that of paired pots was developed in which pots containing soil were matched up to a center channel containing a nutrient solution without Fe (Figure 1a). Thus, all plants received all nutrients, except for Fe, in the same amounts. The 26 treatments (13 soils at two depths) were arranged in four blocks, with two plants per pot in each replication.

Table 1. Classification, textural class, and location of the soils sampled

Soil	Classification	Textural class	Location
1	*Latossolo Vermelho*	Very clayey	Varjão de Minas
2	*Latossolo Vermelho-Amarelo*	Very clayey	Coromandel
3	*Latossolo Vermelho-Amarelo*	Very clayey	Coromandel
4	*Latossolo Vermelho*	Very clayey	Serra do Salitre
5	*Latossolo Vermelho*	Clay	Lagoa Grande
6	*Neossolo Quartzarênico*	Loamy sand	Bela Vista
7	*Latossolo Vermelho*	Very clayey	Presidente Olegário
8	*Latossolo Vermelho-Amarelo*	Very clayey	Presidente Olegário
9	*Latossolo Vermelho-Amarelo*(1)	Clay	Coromandel
10	*Latossolo Vermelho*(1)	Clay	Patos de Minas
11	*Latossolo Vermelho*(1)	Clay	Patos de Minas
12	*Latossolo Vermelho*(1)	Very clayey	Sete Lagoas
13	*Latossolo Vermelho*(1)	Very clayey	Lavras

(1) Soils with high magnetic attraction.

Table 2. Mean, minimum, and maximum values and coefficient of variation (CV) of the results of chemical analysis of the 26 soil samples

Property	Mean	Minimum	Maximum	CV
				%
pH(H_2O)	5.12	4.01	6.28	11.7
P (mg dm^{-3})	8.5	0.0	48.8	160.6
P-rem (mg L^{-1})	13.6	3.2	40.3	70.2
K (mg dm^{-3})	80	8	494	135.0
Ca^{2+} ($cmol_c$ dm^{-3})	1.44	0.04	8.56	167.4
Mg^{2+} ($cmol_c$ dm^{-3})	0.60	0.02	3.29	152.9
Al^{3+} ($cmol_c$ dm^{-3})	0.64	0.00	1.70	82.2
H+Al ($cmol_c$ dm^{-3})	7.1	3.3	15.3	43.0
SB ($cmol_c$ dm^{-3})	2.25	0.09	12.32	155.3
t ($cmol_c$ dm^{-3})	2.89	0.22	12.32	111.8
T ($cmol_c$ dm^{-3})	9.36	3.37	17.66	43.5
V (%)	18.5	1.1	78.9	120.5
m (%)	45.9	0.0	90.2	70.8
OM (dag kg^{-1})	4.50	1.14	10.77	53.4

pH in water (1:2.5) (v:v); P and K: Mehlich-1; P-rem: remaining phosphorus (Alvarez V et al., 2000); Ca^{2+}, Mg^{2+}, Al^{3+}: 1 mol L^{-1} KCl; H+Al: 0.5 mol L^{-1} calcium acetate solution in acetic acid at pH 7.0 (Claessen, 1997); OM: organic matter, modified Walkley-Black method (Nelson and Sommers, 1982). SB: exchangeable bases; t: cation exchange capacity at soil pH; T: cation exchange capacity at pH 7; V: base saturation; m: aluminum saturation.

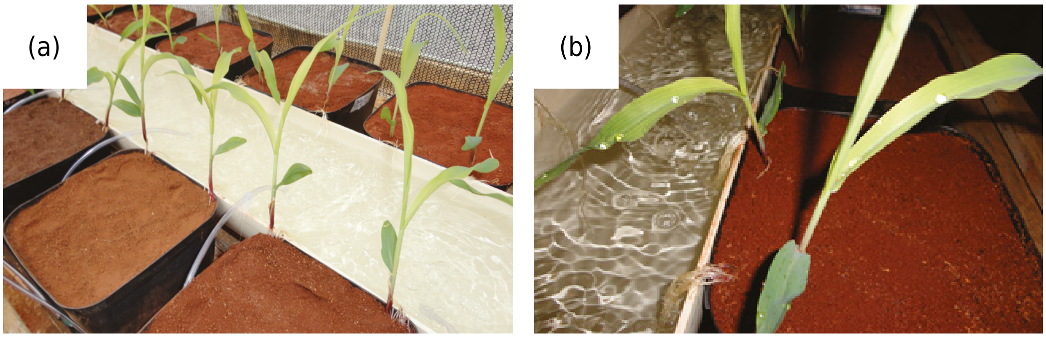

Figure 1. Semi-hydroponic system of pots paired to the center channel (a), with the plant roots growing in both containers (b).

The seeds germinated in paper toweling and, at three days, the tip of the main root of each seedling was cut to favor secondary root growth. They were kept for seven days in a container with Hoagland and Arnon (1950) nutrient solution at ¼ of its full strength. At that time, the plants were selected and transferred to a semi-hydroponic system, and their roots were divided between the two containers (Figure 1b). The pots received 3 dm^3 of soil and the center channel had 40 L of nutrient solution at ½ ionic strength. After seven days, the concentration of the solution was increased to full strength.

The pH of the prepared nutrient solution was approximately 5.7. This pH was maintained between 5.7 and 6.5 throughout the experiment. As the plant took up the nutrients of this solution, pH increased. When the pH of this solution reached 6.5, it was corrected to 5.7 with dilute HCl solution. Due to continuous uptake of nutrients by plants, pH continued to rise. When the pH of 6.5 was reached once more, the nutrient solution was replaced. This was therefore done in an alternating fashion throughout the experiment, either correcting the pH or replacing the solution. These corrections/replacements were more frequent as the plants had greater development since nutrient uptake was greater. The pH of the solution was corrected a total of four times, and the solution was replaced five times. Shading of 50 % was maintained to avoid drying out the roots that were exposed. Irrigation of the pots containing soil was carried out twice a day, nearly up to field capacity.

Plant shoots were harvested at 45 days and the vegetative material was dried in a laboratory oven at 65 °C until constant weight and ground in a Willey mill with a 20 mesh screen (0.841 mm). The Fe concentrations in the plant were determined after nitric perchloric digestion and quantified by atomic absorption spectrophotometry.

Data were subjected to descriptive statistical analysis and correlation by simple linear regression. Correlations significant up to 10 % (F value of significance less than 0.1) were combined by multiple linear regression among the Fe concentrations extracted, and the variables were related to the plant and to the soil.

RESULTS AND DISCUSSION

Soil Fe-pyrophosphate (Fe_p) ranged from 0.98 to 16.36 g kg^{-1}, with a mean value of 5.54 (Table 3). These values are consistent with those found in *Latossolos* by Vendrame et al. (2011), except for the concentrations found in soils 1 and 8. Considering that sodium pyrophosphate acts on the Fe bound to soil organic fractions, positive correlation between the Fe_p values and the organic matter (OM) content of the soils is to be expected (Figure 2). This can explain the high Fe_p concentrations in soils 1 and 8, which exhibited high OM contents. The positive correlation between Fe_p and the OM content of the soil was also reflected in the Fe_p/Fe_d ratio (Figure 3), showing that in soils with greater OM contents, like soil 3, a considerable proportion of Fe was found bound to the organic fraction. Zanelli et al. (2007) and Inda Junior et al. (2013) explained that OM, moisture, and microbial activity can change the pedoenvironmental conditions and the dynamics of mineral formation, favoring dissolution and remobilization of Fe oxides. Thus, in soils with high OM content, the Fe tends to remain in less crystalline forms and is associated with organic compounds, since there is less crystallization of the oxides (Schwertmann and Kämpf, 1983).

The concentrations of low (Fe_o) and high (Fe_d) crystallinity pedogenetic oxyhydroxides of Fe were consistent with those obtained in *Latossolos* by various authors (Melo et al., 2001; Inda Junior and Kämpf, 2003; Vendrame et al., 2011). The high concentrations of Fe_o in soils 9, 10, and 11 were accompanied by greater concentrations of Fe_d, such that a large fraction of Fe was found in crystalline mineral forms. In fact, in the great majority of soils, except for 1, 3, and 8, the Fe_p/Fe_d and Fe_o/Fe_d ratios were quite low (Table 3). The mean value for the Fe_p/Fe_d ratio, excluding soils 1, 3, and 8, was 0.07, and for the Fe_o/Fe_d ratio, a mean of 0.08, indicating that the Fe of the soils was associated mainly with crystalline oxyhydroxides. The crystalline forms of Fe oxyhydroxides tended to predominate in clayey soils, and the Fe_o/Fe_d ratio decreased with an increase in the clay content (Figure 4).

Various studies carried out in weathered tropical soils, such as *Latossolos* (Kämpf et al., 1988; Melo et al., 2001; Oliveira et al., 2006; Pereira et al., 2010), showed quite low relationships between the amorphous and crystalline forms of Fe oxyhydroxides, arriving at values as low as 0.01. Inda Junior et al. (2014) explained that in aerobic pedoenvironments, the Fe oxides exhibited high stability and were able to persist in the soil for long periods. Values lower than 0.05 have often been cited as consistent with soils of a *Latossolo* nature (Inda Junior and Kämpf, 2003).

The concentrations of Fe extracted in $CaCl_2$ were the lowest and varied little among the soils (coefficient of variation - CV near 20 %), with values from 2.29 to 5.82 mg dm^{-3} (Table 3). This small concentration extracted in low ionic strength saline solution was indicative that only a small concentration of the element was readily available to the plant. In well-drained soils, such as *Latossolos* in general, the soluble form of the element occurs in very small proportions, except in very low pH conditions, near 3.2 to 3.7 (Lindsay, 1979), which rarely occurs.

Table 3. Concentration of Fe extracted by pyrophosphate (Fe_p), ammonium oxalate (Fe_o), dithionite-citrate (Fe_d), calcium chloride (Fe-CC), Mehlich-1, and Mehlich-3 and their relationships in samples of 13 soils at two depths (0.00-0.20 and 0.20-0.40 m)

$\text{Soil}_{\text{layer}}$	**Fe_p**	**Fe_o**	**Fe_d**	**Fe_p / Fe_d**	**Fe_o / Fe_d**	**Fe-CC**	**Mehlich-1**	**Mehlich-3**	**Fe-CC / Mehlich**	**Mehlich-1 / Mehlich-3**
m	g kg^{-1}					mg dm^{-3}				
$1_{0.00\text{-}0.20}$	14.93	2.85	49.96	0.30	0.06	4.32	105.2	116.6	0.04	0.90
$1_{0.20\text{-}0.40}$	12.01	2.70	47.17	0.25	0.06	3.06	75.2	59.0	0.05	1.27
$2_{0.00\text{-}0.20}$	3.76	1.78	39.66	0.09	0.04	3.41	44.6	42.1	0.08	1.06
$2_{0.20\text{-}0.40}$	6.30	2.08	37.89	0.17	0.06	3.21	44.9	65.3	0.06	0.69
$3_{0.00\text{-}0.20}$	3.44	1.81	6.42	0.54	0.28	2.55	57.3	64.8	0.04	0.88
$3_{0.20\text{-}0.40}$	3.20	1.77	5.47	0.59	0.32	3.17	52.6	57.2	0.06	0.92
$4_{0.00\text{-}0.20}$	2.54	2.23	40.21	0.06	0.06	3.29	62.9	57.3	0.05	1.10
$4_{0.20\text{-}0.40}$	2.50	1.81	39.21	0.06	0.05	3.13	37.8	41.5	0.08	0.91
$5_{0.00\text{-}0.20}$	4.65	4.05	67.69	0.07	0.06	2.29	196.4	61.8	0.02	3.18
$5_{0.20\text{-}0.40}$	4.18	3.23	61.54	0.07	0.05	3.32	150.2	52.1	0.03	2.88
$6_{0.00\text{-}0.20}$	1.16	3.18	19.27	0.06	0.16	5.82	106.5	117.9	0.05	0.90
$6_{0.20\text{-}0.40}$	1.07	2.85	26.52	0.04	0.11	4.56	110.4	115.3	0.04	0.96
$7_{0.00\text{-}0.20}$	3.09	2.93	58.26	0.05	0.05	3.72	48.8	65.1	0.07	0.75
$7_{0.20\text{-}0.40}$	3.70	2.95	72.80	0.05	0.04	4.19	42.0	51.5	0.09	0.81
$8_{0.00\text{-}0.20}$	14.47	4.90	40.13	0.36	0.12	3.15	106.5	105.5	0.03	1.01
$8_{0.20\text{-}0.40}$	16.36	4.67	39.91	0.41	0.12	3.58	99.8	79.1	0.04	1.26
$9_{0.00\text{-}0.20}$	3.91	13.04	74.44	0.05	0.18	3.99	39.8	64.5	0.08	0.62
$9_{0.20\text{-}0.40}$	3.48	14.32	77.54	0.04	0.18	3.59	38.4	67.6	0.07	0.57
$10_{0.00\text{-}0.20}$	5.53	7.57	103.45	0.05	0.07	3.34	18.2	58.6	0.09	0.31
$10_{0.20\text{-}0.40}$	5.51	9.53	124.82	0.04	0.08	4.14	37.2	52.7	0.09	0.71
$11_{0.00\text{-}0.20}$	2.80	8.45	85.97	0.03	0.10	3.35	14.4	50.9	0.10	0.28
$11_{0.20\text{-}0.40}$	4.22	10.96	131.59	0.03	0.08	4.37	44.8	60.6	0.08	0.74
$12_{0.00\text{-}0.20}$	7.18	3.39	51.63	0.14	0.07	3.27	39.0	67.5	0.06	0.58
$12_{0.20\text{-}0.40}$	7.31	2.94	51.18	0.14	0.06	3.65	24.0	36.4	0.12	0.66
$13_{0.00\text{-}0.20}$	0.98	3.27	92.86	0.01	0.04	4.65	28.5	32.0	0.15	0.89
$13_{0.20\text{-}0.40}$	5.66	4.36	89.44	0.06	0.05	4.39	48.8	51.7	0.09	0.94
Mean	5.54	4.75	59.04	0.15	0.10	3.67	64.39	65.17	0.07	0.99
CV (%)	76.9	75.6	54.8	110.6	75.5	20.1	118.9	36.2	45.1	65.2

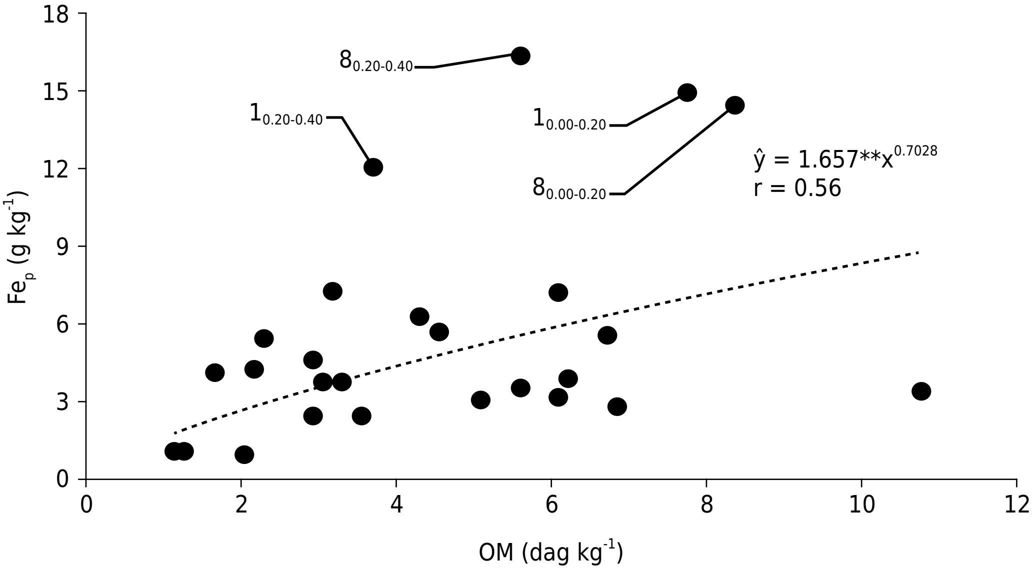

Figure 2. Ratio between concentration of Fe-pyrophosphate (Fe_p) and organic matter (OM) of the soils. **: significant at 1 % by the F test.

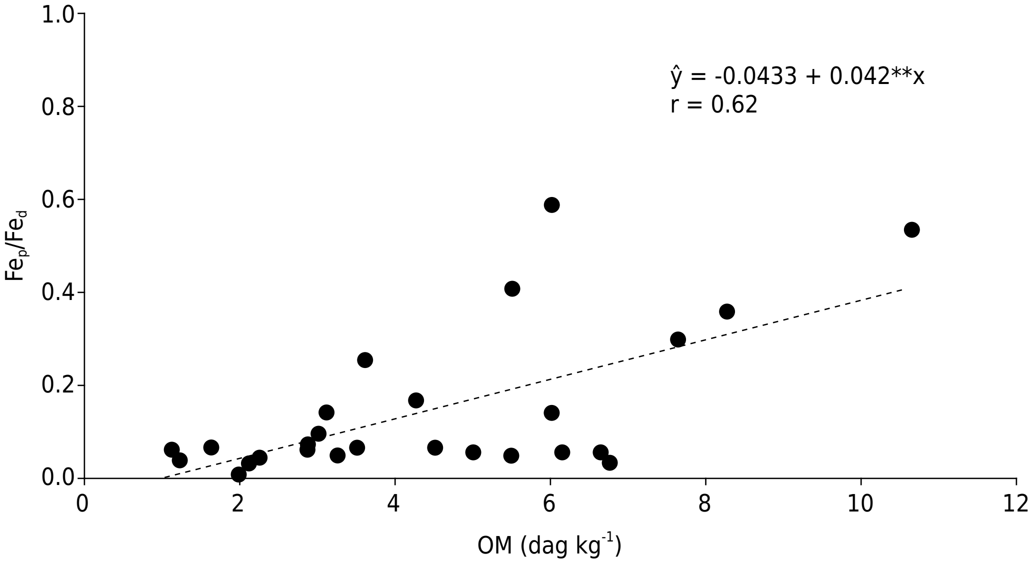

Figure 3. Ratio between the Fe-pyrophosphate (Fe$_p$) and Fe-dithionite-citrate (Fe$_d$) fractions and organic matter content of the soils. **: significant at 1 % by the F test.

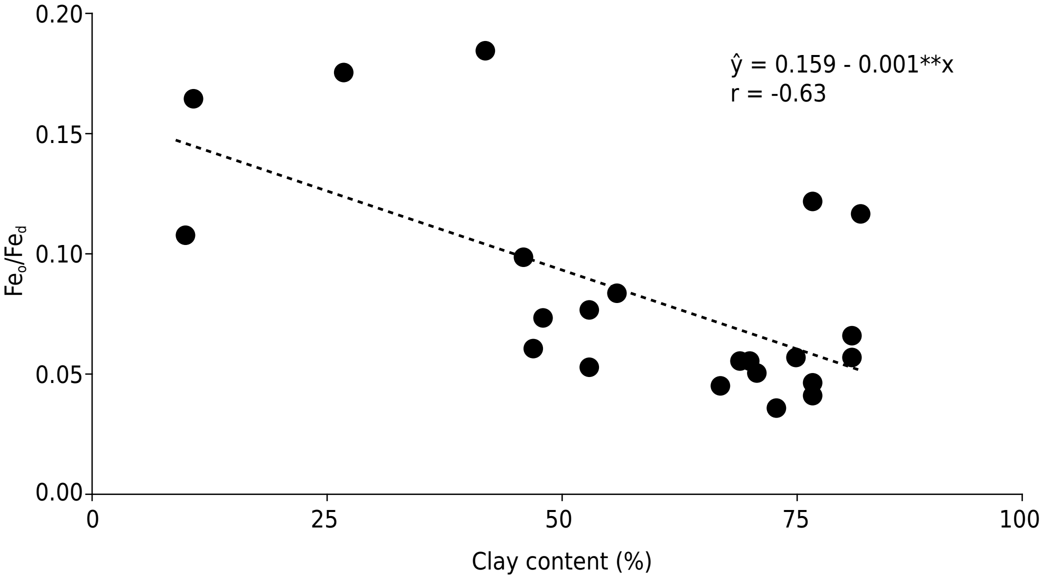

Figure 4. Ratio between low and high crystallinity Fe oxyhydroxides (Fe$_o$/Fe$_d$) and clay content of the soils. **: significant at 1 % by the F test.

Another indication of the low Fe concentration in the soil readily available to plants is the ratio between the contents extracted with the $CaCl_2$ solution and the mean value of the concentrations extracted by Mehlich-1 and Mehlich-3 (Table 3). The values of the Fe-CC/Fe-Mehlich ratio ranged from 0.02 to 0.15, with a mean value of 0.07, indicating that the content of immediately available Fe is very small, as observed by Oliveira et al. (2006). These authors carried out sequential extraction studies to characterize forms of Fe and concluded that there was low availability of the element to the plants, given the low contents in the exchangeable fraction and predominance of the element bound to crystalline oxyhydroxides.

The concentrations of Fe extracted by Mehlich-1 (M-1) ranged from 14.4 to 196 mg dm^{-3}, consistent with the concentrations reported in *Latossolos* by other authors (Oliveira et al., 2006; Silva et al., 2009; Soares et al., 2013). Thus, it is expected that the vast majority of the soils can supply Fe to the plants in the short and medium term, with little chance of deficiency, according to the interpretation of Alvarez V et al. (1999). The concentrations of Fe extracted by Mehlich-3 (M-3) ranged from 32.0 to 118 mg dm^{-3}, consistent with the concentrations found in *Latossolos* by Oliveira et al. (2006) and Silva et al. (2009).

The ratio of the concentrations extracted by the extractants (M-1/M-3) ranged from 0.28 to 3.18 (Table 3). Nevertheless, for most (20 out of 26) of the samples, this ratio was between 0.70 and 1.30, indicating that these extractants exhibited similar extraction capacity. The mean value of the Fe concentrations extracted, around 65 mg dm^{-3} for both, reflected in the mean ratio between the extractants, was very near 1.0 (Table 3); this also indicated that for many soils, the methods were equivalent. In contrast, some authors, such as Abreu et al. (2004), found higher Fe concentrations in extraction with M-3. Considering that M-3 has an acidic and chelating nature, it could be expected that it would extract greater Fe contents from the soil. However, this was not observed in many cases. A possible explanation would be the greater acidity of M-1 (pH around 1.2, whereas M-3 is 2.5), which increases its extraction capacity.

The similarity between the M-1 and M-3 methods was also clear from the high correlation and the regression slope close to one (Figure 5). Positive correlations between the two extractants were also observed by Abreu et al. (2004) and Silva et al. (2009), who obtained correlation coefficients of 0.97 and 0.89, respectively.

Both M-1 and M-3 were affected in a similar way by soil properties (Table 4) and exhibited eight significant correlations in common (pH, Al^{3+}, Fe_d, Fe_p, P-rem, ME, H+Al, and Fe_p/Fe_d). In addition, M-1 exhibited significant correlation with 16 soil properties, whereas M-3 exhibited 12, showing that the former extractant was influenced by a greater number of variables, explaining the greater amplitude of values observed for M-1. In general, the properties that affected the concentrations of Fe extracted by M-1 and M-3 can be placed in three groups: (a) chemical (pH, cation concentrations and their relationships); (b) physical (content of clay, P-rem, and ME); and (c) availability and forms of Fe (OM, Fe_p, Fe_d, Fe-CC, and their relationships).

In the group of properties related to soil chemistry, there was a contrary relationship between the concentrations of Fe extracted and the pH of the soils (Equations 2 and 23). For M-1, this property explained 48 % of the variations in the Fe concentrations ($R^2 = 0.48$) and was that which best correlated with the extractant. As for M-3, only 20 % of the variation ($R^2 = 0.20$) was explained by the property. In a similar way, the concentrations

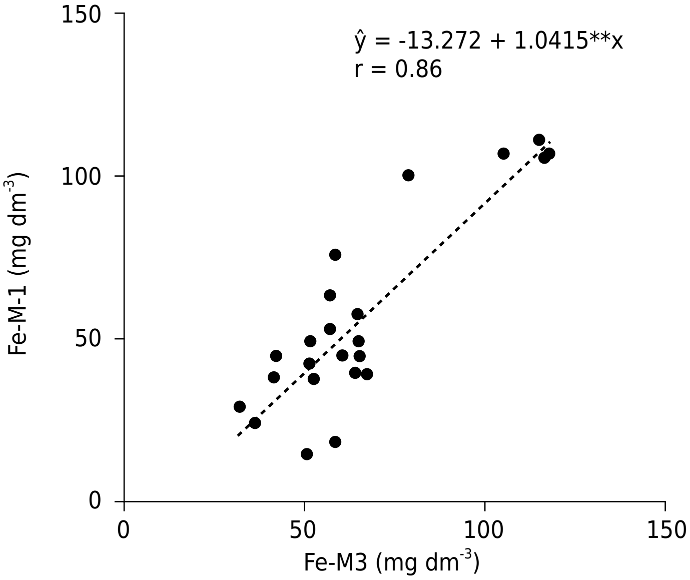

Figure 5. Correlation between the concentrations of Fe obtained by Mehlich-1 (M-1) and Mehlich-3 (M-3). **: significant at 1 % by the F test.

of base cations (Ca, Mg, and K), as well as the derived properties (SB and V), exhibited a contrary relationship to the concentration of extracted Fe, since they followed the increase in the pH of the soils (Equations 6, 7, 8, 14, and 16). Furthermore, the properties that had an inverse relationship to pH (Al^{3+}, Al saturation - m, and potential acidity - H+Al) exhibited a direct relationship to the amount of Fe extracted by the extractants (Equations 3, 4, 13, 19, and 24). The high correlations between M-1 and m and M-1 and Al^{3+} are noteworthy, and each variable individually explains around 40 % of the variation in the concentration of Fe extracted ($R^2 = 0.42$ and 0.39, for M-1 correlated with m and correlated with Al^{3+}, respectively). The high correlation between M-3 and H+Al is also noteworthy, since this variable explained 65 % of the variation in the concentration of the Fe extracted.

The increase in clay content and, consequently, in the value of moisture equivalent (ME) of the soils, led to a reduction in the concentration of Fe extracted by the extractants (Equations 11, 25, and 26), which is contrary to what occurred with P-rem, which exhibited a positive correlation with the extractants (Equations 10 and 18).

Table 4. Correlation between Fe concentrations obtained by Mehlich-1 (M-1) and Mehlich-3 (M-3) and the individualized properties of the soil

No.	Equation	r
	Mehlich-1	
1	$M\text{-}1 = -13.272 + 1.041^{**}\ M\text{-}3$	0.87
2	$M\text{-}1 = 223.607 - 32.971^{**}\ pH$	-0.69
3	$M\text{-}1 = 29.662 + 0.574^{**}\ m$	0.65
4	$M\text{-}1 = 34.440 + 35.285^{**}\ Al^{3+}$	0.62
5	$M\text{-}1 = 81.945 - 0.455^{**}\ Fe_d$	-0.52
6	$M\text{-}1 = 64.343 - 5.846^{*}\ Ca^{2+}$	-0.49
7	$M\text{-}1 = 64.775 - 4.002^{*}\ SB$	-0.49
8	$M\text{-}1 = 64.126 - 14.34^{*}\ Mg^{2+}$	-0.47
9	$M\text{-}1 = 38.104 + 3.058^{*}\ Fe_p$	0.46
10	$M\text{-}1 = 37.552 + 1.316^{*}\ P\text{-}rem$	0.45
11	$M\text{-}1 = 101.473 - 0.178^{*}\ ME$	-0.45
12	$M\text{-}1 = 66.569 - 3.808^{*}\ t$	-0.44
13	$M\text{-}1 = 28.061 + 3.766^{\circ}\ H+Al$	0.40
14	$M\text{-}1 = 63.679 - 0.562^{\circ}\ V$	-0.39
15	$M\text{-}1 = 44.9109 + 68.368^{\circ}\ Fe_p/Fe_d$	0.39
16	$M\text{-}1 = 62.755 - 0.093^{\circ}\ K$	-0.36
	Mehlich-3	
17	$M\text{-}3 = 26.253 + 0.716^{**}\ M\text{-}1$	0.87
18	$M\text{-}3 = 37.934 + 1.671^{**}\ P\text{-}rem$	0.82
19	$M\text{-}3 = 23.184 + 5.092^{**}\ H+Al$	0.81
20	$M\text{-}3 = 41.618 + 3.263^{**}\ Fe_p$	0.73
21	$M\text{-}3 = 36.805 + 5.045^{**}\ OM$	0.61
22	$M\text{-}3 = 52.937 + 51.715^{*}\ Fe_p/Fe_d$	0.46
23	$M\text{-}3 = 155.659 - 17.663^{*}\ pH$	-0.45
24	$M\text{-}3 = 53.179 + 17.782^{*}\ Al^{3+}$	0.41
25	$M\text{-}3 = 93.957 - 0.470^{*}\ Clay$	-0.41
26	$M\text{-}3 = 97.483 - 0.127^{*}\ ME$	-0.40
27	$M\text{-}3 = 81.009 - 0.268^{\circ}\ Fe_d$	-0.37
28	$M\text{-}3 = 23.345 + 11.386^{\circ}\ Fe\text{-}CC$	0.36

°, *, and **, significant at 10, 5, and 1 %, respectively, by the F test.

The most available forms of Fe, such as Fe-CC and Fe_p, correlated positively with the contents extracted by M-1 and M-3 (Equations 9, 15, 20, 22, and 28), corroborating the data found by Oliveira et al. (2006). These authors found high coefficients of determination between the two extractants and forms of Fe bound to organic matter and amorphous oxyhydroxides of Fe. Soumaré et al. (2003) also concluded that the concentration of available Fe widely depends on the C content of the soils. The concentration of high crystallinity oxyhydroxides (Fe_d) had a negative correlation with the concentrations of Fe shown by the extractants (Equations 5 and 27), indicating that these forms of Fe significantly limited Fe availability to the plants. Thus, the more weathered and clayey the soils are, the greater the proportion of Fe associated with the crystalline oxyhydroxides and, therefore, the lower the plant availability of the element.

The diverse soil properties that individually exhibited significant correlations with the extractants (Table 4) were combined to develop equations through multiple regression (Table 5). Fitting these equations allowed the influence of the Fe forms to be verified in a combined manner on the extractants without influence from other soil properties (Equations 39, 51, and 52). From the values of $R^2 = 0.74$ for equation 39, 0.61 for equation 51, and 0.57 for equation 52, it can be perceived that the forms of Fe exhibited greater correlation and better explained the variations in the concentrations of Fe extracted by M-1 than by M-3. It was noteworthy that the Fe_o property, related to Fe in low crystallinity oxyhydroxides, did not show significant correlation with either of the two extractants;

Table 5. Multiple linear regression between the Fe concentrations obtained by Mehlich-1 (M-1) and Mehlich-3 (M-3) and soil properties

No.	Equation	R²
29	$\text{M-1} = 19.822 + 0.337^{**}\ \text{m} + 5.928^{**}\ \text{OM} + 1.132^{\circ}\ \text{P-rem} + 2.792^{**}\ \text{Fe}_\text{p} + 4.519^{**}\ \text{Fe}_\text{o} - 5.850^{**}\ \text{SB} - 0.198^{\circ}\ \text{ME}$	0.93
30	$\text{M-1} = 105.837 + 5.678^{**}\ \text{OM} + 3.483^{**}\ \text{Fe}_\text{p} + 5.099^{**}\ \text{Fe}_\text{o} - 4.508^{**}\ \text{SB} - 0.361^{**}\ \text{ME} - 0.287^{*}\ \text{Fe}_\text{d}$	0.91
31	$\text{M-1} = 33.855 + 0.828^{**}\ \text{M-3} + 0.256^{**}\ \text{m} - 8.726^{\circ}\ \text{pH}$	0.91
32	$\text{M-1} = 75.661 + 0.301^{*}\ \text{m} + 7.530^{**}\ \text{OM} + 2.710^{**}\ \text{Fe}_\text{p} + 4.851^{**}\ \text{Fe}_\text{o} - 3.963^{**}\ \text{SB} - 0.379^{**}\ \text{ME}$	0.90
33	$\text{M-1} = -18.758 + 0.880^{**}\ \text{M-3} + 0.361^{**}\ \text{m}$	0.89
34	$\text{M-1} = -1.792 + 0.978^{**}\ \text{M-3} - 3.075^{**}\ \text{SB}$	0.89
35	$\text{M-1} = 98.137 + 4.123^{**}\ \text{Fe}_\text{o} + 8.263^{**}\ \text{OM} + 3.251^{**}\ \text{Fe}_\text{p} - 0.413^{**}\ \text{ME} - 5.263^{**}\ \text{SB}$	0.86
36	$\text{M-1} = 67.505 + 4.908^{**}\ \text{OM} + 3.137^{**}\ \text{Fe}_\text{p} + 3.000^{*}\ \text{Fe}_\text{o} + 0.470^{**}\ \text{m} - 0.340^{**}\ \text{ME}$	0.84
37	$\text{M-1} = 64.790 + 1.045^{\circ}\ \text{P-rem} + 4.565^{**}\ \text{Fe}_\text{p} - 4.526^{**}\ \text{SB} - 0.620^{*}\ \text{Clay}$	0.82
38	$\text{M-1} = -5.180 + 1.766^{**}\ \text{P-rem} + 3.592^{**}\ \text{Fe}_\text{p} + 0.367^{**}\ \text{m}$	0.78
39	$\text{M-1} = -23.714 + 3.638^{**}\ \text{Fe}_\text{p} + 24.237^{**}\ \text{Fe-CC} - 0.551^{**}\ \text{Fe}_\text{d}$	0.74
40	$\text{M-1} = 95.999 - 3.141^{\circ}\ \text{SB} - 0.128^{\circ}\ \text{ME}$	0.34
41	$\text{M-3} = 69.817 + 7.966^{**}\ \text{OM} + 2.329^{**}\ \text{Fe}_\text{p} + 4.379^{**}\ \text{Fe}_\text{o} + 7.929^{*}\ \text{Fe-CC} - 2.032^{*}\ \text{SB} - 0.341^{**}\ \text{ME} - 0.199^{*}\ \text{Fe}_\text{d}$	0.91
42	$\text{M-3} = 67.679 + 6.280^{**}\ \text{OM} + 2.778^{**}\ \text{Fe}_\text{p} + 3.156^{**}\ \text{Fe}_\text{o} + 9.193^{**}\ \text{Fe-CC} - 0.314{**}\ \text{ME} – 0.253{*}\ \text{Fe}_\text{d}$	0.88
43	$\text{M-3} = 60.231 + 12.096^{\circ}\ \text{Al}^{3+} + 0.523{**}\ \text{M-1} - 0.489{**}\ \text{Clay}$	0.86
44	$\text{M-3} = 54.977 + 3.712^{**}\ \text{H+Al} + 3.435^{*}\ \text{OM} + 1.512^{*}\ \text{Fe}_\text{p} + 2.240^{**}\ \text{Fe}_\text{o}$	0.86
45	$\text{M-3} = 69.777 + 6.822^{**}\ \text{OM} - 0.392^{*}\ \text{Clay} - 0.237^{**}\ \text{ME} + 3.027^{**}\ \text{Fe}_\text{p} + 8.795^{*}\ \text{Fe-CC}$	0.84
46	$\text{M-3} = 49.740 + 0.674^{**}\ \text{M-1} - 0.340^{**}\ \text{Clay}$	0.83
47	$\text{M-3} = 47.572 + 4.556^{**}\ \text{H+Al} + 1.547^{*}\ \text{Fe}_\text{p} - 0.451^{**}\ \text{Clay}$	0.80
48	$\text{M-3} = 55.527 + 3.187^{**}\ \text{Fe}_\text{p} + 12.173^{**}\ \text{Fe-CC} + 2.271^{\circ}\ \text{OM} - 0.725^{**}\ \text{Clay} - 0.313^{**}\ \text{Fe}_\text{d}$	0.79
49	$\text{M-3} = 44.097 + 6.057^{**}\ \text{H+Al} - 0.428^{*}\ \text{Clay}$	0.74
50	$\text{M-3} = 28.623 + 3.469^{**}\ \text{OM} + 2.659^{**}\ \text{Fe}_\text{p}$	0.69
51	$\text{M-3} = 44.576 + 3.155^{**}\ \text{Fe}_\text{p} + 1.801^{*}\ \text{Fe}_\text{o} - 0.180^{(0.12)}\ \text{Fe}_\text{d}$	0.61
52	$\text{M-3} = 8.808 + 2.234^{*}\ \text{Fe}_\text{p} + 2.456^{\circ}\ \text{Fe}_\text{o} + 16.987^{**}\ \text{Fe-CC} - 0.510^{**}\ \text{Fe}_\text{d}$	0.57

* and **: significant at 5 and 1 %, respectively, by the F test

nevertheless, it was significant in various equations generated by multiple regression (equations 29, 30, 32, 35, 36, 41, 42, 44, 51, and 52). That means that individually this property was not able to explain the variations in the Fe concentrations by the extractants, but in combination it has good correlation with both methods.

In addition to enabling analysis of the effect of properties in combination on extractants, multiple regression enables the correlation between extractants to be refined (Table 5). Simply including the SB property ensured significant improvements in the correlation between M-1 and M-3, increasing the R^2 from 0.75 to 0.89 (Equation 34). Other properties, such as clay content, pH, m, and Al^{3+}, were inserted in the correlation, ensuring improvements in the correlation between extractants (Equations 31, 33, 43, and 46) and increasing the coefficient of determination to values as high as 0.91 for M-1 and 0.86 for M-3.

The concentration of Fe in the plants ranged from 56.5 to 122.9 mg kg^{-1} (Table 6), similar to the values obtained by Abreu et al. (2004) and Silva et al. (2009). The mean concentrations of the micronutrient were within the adequate range according to Raij et al. (1997) and Epstein and Bloom (2005), who affirmed that the leaf concentrations in dry matter in corn should be between 30 and 250 mg kg^{-1}.

Table 6. Dry matter, concentration, and content of Fe in plants cultivated in samples of 13 soils collected at two depths

$\text{Soil}_{\text{layer}}$	Dry matter	Fe concentration	Fe content
m	g per pot	mg kg^{-1}	mg per plant
$1_{0.00-0.20}$	24.23	63.58	1.52
$1_{0.20-0.40}$	26.08	56.51	1.34
$2_{0.00-0.20}$	25.16	70.57	1.75
$2_{0.20-0.40}$	19.18	94.74	1.84
$3_{0.00-0.20}$	23.30	60.16	1.37
$3_{0.20-0.40}$	16.63	70.16	1.17
$4_{0.00-0.20}$	20.71	68.48	1.44
$4_{0.20-0.40}$	19.42	70.37	1.37
$5_{0.00-0.20}$	23.77	71.04	1.63
$5_{0.20-0.40}$	16.14	68.82	1.15
$6_{0.00-0.20}$	18.26	68.20	1.28
$6_{0.20-0.40}$	16.71	58.26	0.98
$7_{0.00-0.20}$	23.44	65.46	1.53
$7_{0.20-0.40}$	16.95	74.41	1.2
$8_{0.00-0.20}$	19.64	68.04	1.33
$8_{0.20-0.40}$	25.47	122.98	3.15
$9_{0.00-0.20}$	17.80	77.22	1.36
$9_{0.20-0.40}$	19.50	56.98	1.11
$10_{0.00-0.20}$	22.85	75.48	1.74
$10_{0.20-0.40}$	19.96	57.52	1.18
$11_{0.00-0.20}$	14.04	58.92	0.92
$11_{0.20-0.40}$	23.38	88.16	2.11
$12_{0.00-0.20}$	28.13	89.50	2.47
$12_{0.20-0.40}$	22.32	98.27	2.36
$13_{0.00-0.20}$	14.75	108.84	1.50
$13_{0.20-0.40}$	41.64	118.09	4.89
Mean	21.52	76.18	1.68
CV (%)	23.2	28.1	17.7

Table 7. Correlation between dry matter in the shoots (DM), concentration and content of Fe in the plant, and concentration of Fe-pyrophosphate (Fe_p) and of clay

No.	Equation	R^2
53	$DM = 8.813 + 0.173^{**}$ Fe concentration	0.32
54	$DM = 16.42 (Fe_p)^{0.149^{**}}$	0.36
55	Fe concentration $= 50.217 + 5.942^{**} Fe_p$	0.43
56	Fe content $= 1.238 + 0.057^{*} Fe_p$	0.24

* and **: significant at 5 and 1 %, respectively, by the F test.

There was positive correlation between the variables of dry matter and Fe concentration in the plant (Table 7), indicating that plants with higher concentrations of the element in the shoots tend to be more productive (Equation 53). Thus, there is still potential for plant response to Fe within the range of leaf values considered adequate, indicating that this wide range of suitability, between 30 and 250 mg kg^{-1}, should really be narrower. Dry matter production and leaf concentration and content of Fe correlated positively with Fe_p content (Equations 54, 55, and 56), indicating that the Fe fraction bound to the organic components of the soil was what most contributed to availability of the element to the plant, as found by Wei et al. (2010).

The M-1 and M-3 methods were inefficient in evaluating Fe availability to the plants. There was no correlation between the Fe concentration in the soil by the extractants and its concentration in the plant (0.02^{ns} for M-1 and 0.07^{ns} for M-3) or Fe accumulation in the shoots (0.01^{ns} for M-1, and 0.02^{ns} for M-3). This lack of correlation has already been observed by Abreu et al. (2004), who worked with corn and soybean and the extractants Mehlich-1, Mehlich-3, Resin, DTPA, and AB-DTPA, and they did not obtain significant correlations between concentrations and contents of Fe with any of the extractants for either species.

CONCLUSIONS

Most of the soil Fe content in *Latossolos* occurs in the form of high crystallinity and low solubility pedogenetic oxyhydroxides; therefore, most of it is not available to plants. In contrast, the Fe bound to the organic components is the fraction that most contributes to availability of the element to plants, and plants with greater leaf contents of Fe produce a greater amount of biomass.

The extractants Mehlich-1 and Mehlich-3 were inefficient in evaluating Fe availability to plants. The extraction capacity of Fe of the two methods is similar and the methods have high mutual correlation; the concentration of Fe extracted by Mehlich-1 is more influenced by pH, m, Al^{3+}, and Fe_d, whereas Mehlich-3 is more influenced by P-rem, H+Al, Fe_p, and OM.

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