

## **Influence of fulvic acid on transport of iron in soils and uptake by paddy seedlings**

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### **Abstract**

The influence of fulvic acid (FA) on the porous system self diffusion coefficient ( $D_p$ ) of Fe in Calciorthent soils of Bihar, India, was determined with the half cell technique. Significantly higher values of  $D_p$  were observed when Fe was applied as Fe-FA to the soil compared to  $FeCl_3$ . The capacity factor of Fe decreased considerably due to its complexation by fulvic acid, resulting in an increase in the  $D_p$  of Fe. The organic carbon content of the soils correlated positively with  $D_p$  of Fe while it showed a negative relationship with active  $CaCO_3$  and the clay content of soils. A soil culture system simulating acquisition of Fe by rice was developed to investigate transport of Fe from the soil solution to the surface of the plant roots through diffusion and mass flow. Mass flow contributed only 5–9% of the total Fe uptake by rice, with the remainder being ascribed to diffusion and root interception. A significant relationship ( $r=0.96^{**}$ ) between  $D_p$ - and Fe-uptake by rice was observed. The uptake of Fe by the crop and the percentage of tissue iron content derived from fertilizer were higher in the case of Fe-FA in comparison with  $FeCl_3$ , indicating the superiority of organically complexed Fe fertilizers over inorganic salts.

### **Introduction**

The ambient concentration of Fe species in calcareous and alkaline soils is usually too low to meet the requirement of the plants (Lindsay, 1974, 1979). In these soils, the presence of humic substances in the soils and the production of complexing substances by the roots have been found to be responsible for the supply of Fe to plants (Bienfait, 1988; Chaney et al., 1972; Landsberg, 1981, 1986; Marschner et al., 1986; Romheld and Marschner, 1978, 1983). The use of chelating substances in alleviating Fe deficiencies in plants grown in calcareous soils has proved extremely effective (Sakal et al., 1982; Singh and Sinha, 1977; Wallace, 1971). The soluble Fe concentration in most calcareous soils, however, remains extremely low, even in the presence of synthetic or natural chelating ligands in soils (Dyanand and Sinha, 1980). The flux of the nutrient to the plant roots by mass flow may not be sufficient to meet the requirement of the crop. Therefore, diffusion may play a significant role in transport of Fe from soil solution to plant roots (Bohn, 1967; Oliver

and Barber, 1966; Sharma and Deb, 1987). Synthetic chelating ligands have been shown to influence the diffusive flux of Fe and other trace metal cations positively (Gupta and Deb, 1985; Milap Chand et al., 1980; O'Connor et al., 1971). However, there is little data in the literature on the diffusive flux of Fe to plant roots as influenced by naturally occurring chelators such as humic substances.

Naturally occurring humic substances enhance Fe availability to plants mainly by two processes (Wilkinson, 1972): (i) increase in the soluble concentration of Fe in the soil solution as complexed Fe species are less prone to precipitation at high pH. Humic substances also solubilize some of the poorly soluble forms of Fe in soils (ii) the transport of iron from bulk soil solution to the plant roots by the processes of diffusion is positively influenced by humic substances.

The objectives of this study were to determine: (i) the influence of a naturally occurring humic substance such as fulvic acid (FA) on the porous system self diffusion coefficient of Fe in soils; (ii) the influence of FA on the flux of Fe to the plant root through diffu-

Table 1. Physical and chemical properties of soils used for different studies

Soil No.	Sand (%)	Silt (%)	Clay (%)	pH Soil: Water (1:2)	EC dSm <sup>-1</sup>	Organic C (%)	Free CaCO <sub>3</sub> (%)	Active CaCO <sub>3</sub> (%)	DTPA extractable Fe (ppm)
1	23.4	54.2	22.4	7.6	0.91	0.26	15	5.0	36.43
2	27.9	39.7	32.4	8.2	0.29	0.21	38	6.5	6.85
3	34.9	50.1	15.0	8.2	0.13	0.33	2	4.2	15.09
4	48.6	39.5	11.9	8.4	0.18	0.87	33	5.7	6.29
5	32.8	62.1	5.1	8.6	0.13	0.47	31	5.0	5.24
6	41.5	44.9	13.6	8.3	0.17	0.37	20	5.7	9.43
7	46.3	45.3	8.4	8.8	1.99	0.55	25	5.4	3.10
8	55.7	42.0	2.3	8.2	0.21	0.71	35	1.2	4.48
9	23.0	67.0	10.2	8.2	1.99	0.29	40	5.7	5.91
10	8.4	82.4	9.2	8.1	0.42	0.65	35	5.2	3.76

sion and mass flow; and (iii) the influence of FA on the uptake of Fe by the plant and the contribution of fertilizer sources in uptake.

## Materials and methods

### Soils

Soil samples (0–15 cm depth) were collected from different sites of young alluvium (Calciorthent) on the calcareous belt of North Bihar (India). The air-dried and sieved (2 mm) soils were analysed for their physical and chemical properties. The mechanical analysis of the soil samples was done according to the international pipette method (Piper, 1966). The pH and EC were measured in 1:2 soil suspension. Walkley and Black's rapid titration method as documented by Jackson (1967), was followed for estimation of organic carbon content of soils. Free CaCO<sub>3</sub> was estimated by the method outlined by Puri (1949). The active CaCO<sub>3</sub> in soils was estimated by the method described by Drouineaux (1942). For the estimation of active CaCO<sub>3</sub>, 50 mL of 0.2 M ammonium oxalate was allowed to react with 10 g soil for 2 h. The amount of unused ammonium oxalate was estimated in the filtered solution by back titration of oxalate with 0.2 M KMnO<sub>4</sub> in concentrated H<sub>2</sub>SO<sub>4</sub> at 65–70 °C. Ten samples, differing in their physical and chemical properties, were selected for the laboratory studies. Selected physical and chemical properties of the soils are presented in Table 1.

### Fractionation and purification of fulvic acid from farm yard manure (FYM)

The FYM collected from the dairy farm of Rajendra Agricultural University, Pusa, Bihar contained 48.5% organic carbon. The fractionation of fulvic acid in FYM was carried out according to the method described by Pandeya (1992). The properties and composition of the fulvic acid are reported in Table 2.

### Studies on porous system self-diffusion coefficient of Fe in soils

The porous system self-diffusion coefficient of Fe in soils was measured using the half cell technique as adopted by O'Connor et al. (1971). Twenty-five grams of each soil were weighed into four 100 mL beakers. Two of the samples received an FeCl<sub>3</sub> solution and the other two an Fe–FA complex at the rate of  $8 \times 10^{-8}$  moles g<sup>-1</sup> soil. To one of the sub-samples that received FeCl<sub>3</sub> or Fe–FA, <sup>59</sup>Fe tracer was added at the rate of 0.2 μCi per beaker. The treatments were applied in 50 mL water. The soil samples were moistened and air dried three times over 19 days in order to equilibrate. The equilibrated dried samples were packed in the half cells, maintaining the bulk density of the soil at 1.5 mg m<sup>-3</sup>. A pair of half cells that contained soil sample was equilibrated with <sup>59</sup>Fe along with another half cell without sample, and they were clamped and kept in an incubator at 30 ± 1 °C, 28% moisture and relative humidity of approximately 100% for 19 days to equilibrate. After the diffusion period, the cells were separated and the soils from each half cell were digested

Table 2. The elemental composition and properties of FA extracted from FYM

Elemental composition (%)				Ash content (%)	E4/E6 <sup>a</sup>	Acidic functional groups (cmol kg <sup>-1</sup> )		
C	H	N	O			Carboxyl	Phenolic	Total acidity
32.28	5.34	3.06	59.32	0.95	8.6	902	279	981

<sup>a</sup> Ratio of optical density at 465 and 665 nm wavelength.

by boiling in 50% (v/v) HCl for 2 h. The radioactivity of <sup>59</sup>Fe was measured in digested soil extracts with a gamma-ray spectrometer.

#### Determination of capacity factor (b)

The capacity factor for labile Fe in soils was determined by shaking 5 g of soil, previously equilibrated with a known activity of <sup>59</sup>Fe, with 10 mL of deionized water. The equilibration time by intermittent shaking corresponded to the time allowed for the diffusion of Fe through the half cells. All the samples were incubated for 19 days to equilibrate in an incubator-cum-shaker at 30±1 °C. After equilibration, the soil suspension was centrifuged for an hour. After centrifugation, a 2 mL aliquot was taken for the measurement of radioactivity of <sup>59</sup>Fe with a gamma-ray spectrometer. The capacity factor was calculated from the amount of <sup>59</sup>Fe originally added to the soil and the radioactivity of <sup>59</sup>Fe remaining in the filtered extract, using an equation described by (O'Connor et al., 1971):

$$b = \frac{\theta (\text{^{59}Fe adsorbed/cm}^3 \text{ soil} + \text{^{59}Fe in solution/cm}^3 \text{ solution})}{\text{^{59}Fe in solution/cm}^3 \text{ solution}}$$

where  $\theta$  = the volumetric moisture content (cm<sup>3</sup> H<sub>2</sub>O/cm<sup>3</sup> soil).

The transient state equation as described below was used for calculation of Dp in soils (O'Connor et al., 1971).

$$Dp = \frac{\pi(q/q_0)^2 \times L^2(b + \theta)}{t}$$

where Dp = the porous system self-diffusion coefficient (cm<sup>2</sup> sec<sup>-1</sup>); q = the amount of <sup>59</sup>Fe that diffuses from one half cell of soil to the other in time t (cpm); q<sub>0</sub> = the total amount of labelled <sup>59</sup>Fe in the system (cpm);  $\pi$  = constant equal to 3.14; L = the length of the half-cell (cm); t = the time in seconds during which the two half cells of soil remain in contact; b = the capacity factor, defined as the ratio of the total diffusible component to the amount of that component in solution;  $\theta$  = volumetric water content of the soils.

#### Studies on the influence of FA on transport and uptake of Fe by paddy seedlings grown in soils

Rice seeds (*Oryza sativa*) were sown in acid-washed fine quartz sand. Seedlings were irrigated with half strength Hoagland's nutrient solution (Arnon and Hoagland, 1940) devoid of Fe, twice in fifteen days. Plants were watered with distilled water as required. Pots were kept under diffuse light to avoid etiolation.

The paddy seedlings were uprooted carefully so that the root system of the plants remained intact. Roots were thoroughly washed with distilled water. Fifty grams of each of the four selected soils were used for growing paddy seedlings in beakers with treatments of Fe as follows: (i) No Fe (ii) FeCl<sub>3</sub> at 8×10<sup>-3</sup> mol g<sup>-1</sup> soil and (iii) Fe-FA at 8×10<sup>-3</sup> mol g<sup>-1</sup> soil. The Fe sources were labelled with carrier-free <sup>59</sup>Fe at 5  $\mu$ Ci per 50 g soil. All the treatments were replicated three times. Ten paddy seedlings were transplanted in each beaker by placing the roots in soil layers. One set of the beakers with the above-mentioned treatments was kept in a growth chamber having approximately 100% relative humidity while, the other set was kept in a growth chamber having 60–75% relative humidity.

A constant water content of 28% of the soil was maintained during 48 h of growth. At the end of this period, the plants were carefully removed from the soil to minimize root damage. Plants were washed with 0.1 M HCl and then with distilled water. Plant samples were dried at 80 °C. After recording dry weights, they were transferred into 100 mL conical flasks for digestion in a tri-acid mixture (Piper, 1966). The digested samples were used to determine the radioactivity of <sup>59</sup>Fe.

The data were computed according to the following equations:

Specific radioactivity of Fe in plants =

$$\frac{\text{Counts of } ^{59}\text{Fe (counts per sec)}}{\text{Total Fe in plants (mmoles)}}$$

Table 3. Porous system self diffusion coefficient,  $D_p$  ( $\text{cm}^2/\text{sec}$ ) of iron in soils as influenced by sources of iron

Soil No.	FeCl <sub>3</sub>			Fe-FA		
	$b^a$	$Da^b$	$D_p=Da/(b+\theta)^c$	$b$	$Da$	$D_p=Da/(b+\theta)$
1	839.54	$1.371 \times 10^{-7}$	$1.632 \times 10^{-10}$	128.68	$4.690 \times 10^{-8}$	$3.633 \times 10^{-10}$
2	901.48	$7.782 \times 10^{-8}$	$8.629 \times 10^{-11}$	87.88	$6.537 \times 10^{-8}$	$7.404 \times 10^{-10}$
3	709.53	$1.096 \times 10^{-7}$	$1.544 \times 10^{-10}$	226.35	$1.050 \times 10^{-7}$	$4.631 \times 10^{-10}$
4	723.53	$8.919 \times 10^{-8}$	$1.232 \times 10^{-10}$	150.76	$1.072 \times 10^{-7}$	$7.093 \times 10^{-10}$
5	678.78	$2.063 \times 10^{-7}$	$3.037 \times 10^{-10}$	91.69	$3.367 \times 10^{-7}$	$3.655 \times 10^{-9}$
6	723.63	$1.517 \times 10^{-7}$	$2.696 \times 10^{-10}$	44.85	$1.064 \times 10^{-7}$	$2.352 \times 10^{-9}$
7	678.78	$1.934 \times 10^{-7}$	$2.847 \times 10^{-10}$	178.82	$2.796 \times 10^{-7}$	$1.560 \times 10^{-9}$
8	518.17	$1.687 \times 10^{-7}$	$3.253 \times 10^{-10}$	36.22	$1.530 \times 10^{-7}$	$4.199 \times 10^{-9}$
9	779.88	$1.469 \times 10^{-7}$	$1.883 \times 10^{-10}$	84.64	$1.007 \times 10^{-7}$	$1.184 \times 10^{-9}$
10	690.00	$9.856 \times 10^{-8}$	$1.427 \times 10^{-10}$	48.62	$6.571 \times 10^{-8}$	$1.340 \times 10^{-9}$

<sup>a</sup> Capacity factor.

<sup>b</sup> Apparent diffusion coefficient.

<sup>c</sup> Porous system self diffusion coefficient.

Specific radioactivity of Fe in fertilizer =

$$\frac{\text{Counts of } ^{59}\text{Fe (counts per sec)}}{\text{Total Fe in fertilizer (mmoles)}}$$

Per cent Fe derived from the fertilizer =

$$\frac{\text{Specific radioactivity of samples at 0 h}}{\text{Specific activity of added source at 0 h}}$$

## Results and discussion

The distribution of Fe between the solid and solution phases was used to estimate the capacity factor which represented the number of units of diffusible Fe present per  $\text{cm}^3$  of soil to increase or decrease its solution concentration by one unit. The release of Fe from the labile pool of the soil to the solution is referred to as the capacity factor. The data in Table 3 indicate wide variations in the capacity factor for Fe, when any of the sources were applied to soils. Table 4 lists the correlation coefficients between different physical-chemical properties of the soils and the capacity factor. The organic carbon was significantly but negatively correlated with the capacity factor ( $r = -0.877^{**}$ ), whereas, clay and active  $\text{CaCO}_3$  contents exhibited significant positive correlation ( $r = 0.888^{**}$  and  $0.796^{**}$ ), respectively, when  $\text{FeCl}_3$  was applied to the soils. The soil properties, however, had no influence in determining the capacity factor of soils if Fe-FA was applied. A multiple regression equation, with statistically signifi-

cant coefficients to predict the capacity factor of soils for inorganically applied Fe, is as follows:

$$\text{Capacity factor} = 470 + 7.6 (\text{Clay}) + 31.3 (\text{Active CaCO}_3) \dots R^2 = 0.902^{**}$$

The clay content of the soil had a profound bearing on the capacity factor of Fe in soils. Iron is present in the soil solution in di- or tri-valent forms. Due to its positive charge, Fe ions are strongly attracted by the negatively charged clay particles. Therefore the clay content of the soil immobilises the inorganic Fe ions from the solution phase to the solid phase. Such surface adsorption of ions might be responsible for an increase in the capacity factor of Fe in soils. In fact, a very small fraction of  $\text{Fe}^{2+}$  or  $\text{Fe}^{3+}$  remains mobile in solution for a limited period (Olsen and Camper, 1968; Rattan and Deb, 1980; Singh et al., 1980).

The next most significant soil property determining the capacity factor of soil was active  $\text{CaCO}_3$ . The  $\text{CaCO}_3$  content of young alluvium calcareous soils can be as high as 60% of the solid matrix. In the present study it ranged between 2–40%. However, most of the  $\text{CaCO}_3$  is not reactive due to low solubility. It has been observed that  $\text{CaCO}_3$  in the size range of the clay and fine silt are reactive due to their relatively higher solubility (Russel, 1966). Total  $\text{CaCO}_3$  of the soil was not correlated with the capacity factor but a significant positive correlation with active  $\text{CaCO}_3$  indicated the influence of this parameter on the capacity factor of Fe in soils (Table 4). An increase in the values of the

Table 4. Correlation matrices of capacity factor (b) and Dp with soil properties

Soil parameters	FeCl <sub>3</sub>		Fe-FA	
	b	Dp	b	Dp
pH	-0.379	0.461	0.179	0.375
EC	0.159	0.162	0.187	-0.236
Organic carbon	-0.877**	0.636*	-0.335	0.656*
Clay	0.888**	-0.757*	-0.167	-0.671*
CaCO <sub>3</sub>	-0.072	0.148	-0.529	0.298
Active CaCO <sub>3</sub>	0.796**	-0.606	0.164	-0.613
Av. Fe	0.437	-0.265	0.273	-0.441

\* Significant at 5% level.

\*\* Significant at 1% level.

capacity factor due to the increase in the content of active CaCO<sub>3</sub> of soils may be attributed to the reaction of soluble Fe species and formation of insoluble hydroxides, carbonates and bi-carbonates of Fe (Gupta and Deb, 1981; Jurinak and Baver, 1956; Lindsay, 1979).

The organic matter present in the soil provides chelating agents which function as a sink for micronutrient cations and heavy metals. They also complex the poorly soluble Fe compounds and bring them into solution. The chelating ligands also protect the metal cations from precipitating at higher pH levels. They neutralise the charges on the metal ion which reduces the likelihood of their adsorption on the solid phase matrix of the soils as exchangeable ions (Flaig et al., 1975; Hodgson, 1968; Milap Chand et al., 1980; Prasad et al., 1982; Sinha and Prasad, 1977). All these factors contribute to the decrease in the capacity factor of Fe in soils with an increasing organic carbon content in soil.

The physical and chemical properties of the soils studied failed to influence the capacity factor when Fe was applied in complexed form (Table 4). This indicates that soluble chelated Fe molecules are less prone to precipitation due to the high pH and high CaCO<sub>3</sub> content of the soils.

#### *Porous system self-diffusion coefficient of Fe in soils*

The porous system self diffusion coefficient is an index of the amount of the metal that diffuses per unit time through a soil-water system. The values for Fe applied to the soil as FeCl<sub>3</sub> and as the Fe-FA complex are presented in Table 3. The organically complexed Fe had higher values of porous system self diffusion coeffi-

cient than the inorganic Fe. The reduction in the capacity factor due to complexation of Fe by fulvic acid and the subsequent maintenance of higher concentrations of Fe in soil solution are probably the reasons for this result (Gupta and Deb, 1985; O'Connor et al., 1971; Rattan and Deb, 1981; Sinha and Prasad, 1977). The poor efficiency of FeCl<sub>3</sub> in the process of diffusion in calcareous soils may be attributed to its precipitation as hydroxide due to an alkaline pH and surface chemical adsorption of Fe in the solid matrix of the soil (Wilkinson, 1972).

It is interesting to note that the apparent diffusion coefficient (Da) of Fe measured in seven out of ten soils was higher in the case of FeCl<sub>3</sub> than for the Fe-FA complex. The fulvic acids are large sized molecules and form still larger molecules on forming complexes with metal ions. Such molecules will require higher energies for their thermal motion in soil solution. Their movement is also expected to be slowed down considerably due to the presence of colloidal particles and other ionic species in the path of the movement. It is likely that the apparent coefficient of such complexed Fe macromolecules will be lower in comparison with Fe<sup>2+</sup> or Fe<sup>3+</sup> which are both considerably smaller (Wilkinson, 1972). The porous system self diffusion coefficient of Fe-FA complexes observed to be higher in comparison with inorganic Fe species may, therefore, solely be attributed to a decrease in the capacity factor due to a reduction in its reactivity with the soils and the ability to remain in soil solution.

The values of porous system self diffusion coefficients of Fe in soils treated with FeCl<sub>3</sub> were positively correlated with organic carbon ( $r = 0.636^*$ ) and were negatively correlated with clay content ( $r = -0.757^*$ ) and active CaCO<sub>3</sub> ( $r = -0.606$ ) (Table 4). The porous

Table 5. Effect of iron sources on uptake of Fe ( $\mu\text{moles pot}^{-1}$ ) by plants grown in different soils under controlled growth conditions at 100% relative humidity

Soil No.	Control	FeCl <sub>3</sub>	Fe-FA	Mean
1	1.596	1.790	2.200	1.862
2	1.509	1.660	2.422	1.867
5	1.297	1.983	2.831	2.037
10	1.527	1.797	2.760	2.028
Mean	1.482	1.807	2.553	—

CD<sub>0.05</sub> for source of Fe = 0.230\*.

Table 6. Effect of iron sources on uptake of Fe ( $\mu\text{moles pot}^{-1}$ ) by plants grown in different soils under normal growth condition at a relative humidity between 60–75%

Soil No.	Control	FeCl <sub>3</sub>	Fe-FA	Mean
1	1.634	1.949	2.446	2.009
2	1.545	1.755	2.481	1.927
5	1.456	2.369	3.096	2.307
10	1.632	1.913	2.885	2.143
Mean	1.566	1.996	2.727	—

CD<sub>0.05</sub> for source of Fe = 0.245\*; for soils = 0.283\*.

system self diffusion coefficient of Fe when the Fe-FA complex was the source, also exhibited a positive correlation with the organic carbon content of the soils, whereas, the clay content was negatively correlated. The active CaCO<sub>3</sub> plays a significant role in determining the porous system self diffusion coefficient of Fe, irrespective of its source of application. The diffusion of Fe decreased with an increase in active CaCO<sub>3</sub>. Such an effect may be due to a considerable increase in the capacity factor as noted in the investigation (Table 3). The carbonate system operating in calcareous soils buffers the system to an alkaline pH. Iron present in the soil solution in inorganic ion species is precipitated as the hydroxide. A high Ca<sup>2+</sup> concentration in solution is also likely to compete for a site on fulvic acid ligands which were added as an Fe-FA complex. The Fe may be displaced by Ca<sup>2+</sup> from the fulvic acid and be found in the soil solution as an inorganic ionic species. Such displaced Fe in the soil solution may eventually be precipitated. This is probably the reason why active CaCO<sub>3</sub> exhibited a negative correlation with the porous system self diffusion coefficient of Fe in soils.

Table 7. Amount of Fe ( $\mu\text{moles pot}^{-1}$ ) transported to the plant roots by mass flow

Soil No.	Control	FeCl <sub>3</sub>	Fe-FA	Mean
1	0.038 (2.32)	0.159 (8.20)	0.246 (10.05)	0.147 (6.85)
2	0.036 (2.33)	0.095 (5.44)	0.059 (2.37)	0.063 (3.38)
5	0.159 (10.93)	0.386 (16.29)	0.265 (8.55)	0.270 (11.90)
10	0.105 (6.43)	0.116 (6.06)	0.125 (4.33)	0.115 (5.60)
Mean	0.084 (5.50)	0.189 (9.00)	0.173 (6.32)	

Figures in parentheses = % of the total uptake.

#### Relative availability of inorganic FeCl<sub>3</sub> and Fe-FA complex as sources of Fe to plants

##### Iron uptake

The uptake of Fe by paddy seedlings, as influenced by source of Fe, is reported in Tables 5 and 6. The former represents a condition which provided nearly 100% humidity in the growth chamber, whereas, in the later case, conditions provided nearly 60–75% relative humidity. Statistical analysis of the data indicated a significant difference in the uptake of Fe under 100% relative humidity due to its sources of application (Table 5). There was an approximately 41% increase in the uptake of Fe with application of Fe-FA over that of application of FeCl<sub>3</sub>. Similar results were observed in the studies on uptake under normal growth conditions having nearly 60–75% humidity in the growth chamber (Table 6). The mean effects of sources of Fe were observed in the order: Fe-FA > FeCl<sub>3</sub> > Control.

##### Amount of Fe transported to the plant roots by mass flow process

The uptake of Fe by the plants, grown in a growth chamber with 100% relative humidity, was attributed to that pool of Fe in the soil solution which was intercepted by roots and the Fe that was transported from the bulk of the soil solution to plant roots by means of diffusion. In the case of plants grown the under 60–75% relative humidity, however, Fe moved freely in soils by means of mass flow due to the suction gradient created by roots, root interception and diffusion. The difference in uptake of Fe in these two cases may be

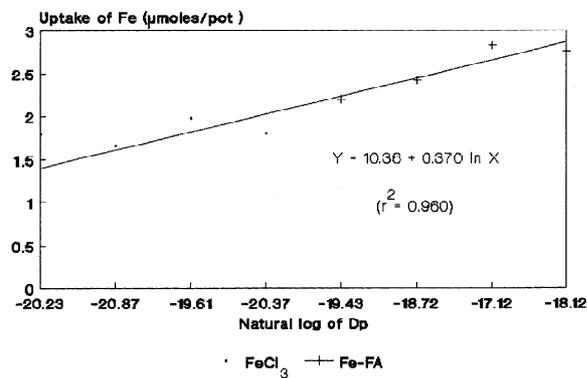


Figure 1. Relationship between DP and uptake of Fe ( $\mu\text{moles pot}^{-1}$ ) by plants under normal growth conditions (60–70% relative humidity).

attributed to the Fe that had been transported to the root surface by the process of mass flow only. Table 7 demonstrates the difference in uptake under two sets of conditions and the percentage of total uptake of Fe due to the movement by mass flow of nutrient in soil solution.

On average, 5.5 to 9.0% of the total uptake of Fe could be attributed to the mass flow occurring in different soils. This signifies that the transport of Fe in soil solution is largely governed by the process of diffusion and root interception. This may be due to the fact that the ambient concentration of Fe in soil solution is extremely low and movement of iron is hardly sufficient to meet the requirement of the plants (Bohn, 1967; O'Connor et al., 1971). It was also interesting to note in the present studies that on average, 9% of the total uptake was due to mass flow in the  $\text{FeCl}_3$  treatment, whereas, it was only 6.32% and 5.50% in Fe-FA and control treatments, respectively. The values obtained in the control treatment signifies a very low solubility of endogenous Fe sources resulting in low concentrations in soil solution.

#### *Relationship of porous system self diffusion coefficient (Dp) of Fe in soils with Fe uptake by paddy seedlings*

Relationships between porous system self diffusion coefficient of Fe in soils and iron uptake by paddy seedlings exposed to radioactive-labelled Fe sources in different soils are depicted in Figures 1 and 2. The relationships between these two parameters under two sets of conditions of the experiment were statistically significant. The relationships may be described by the following straight line equations:

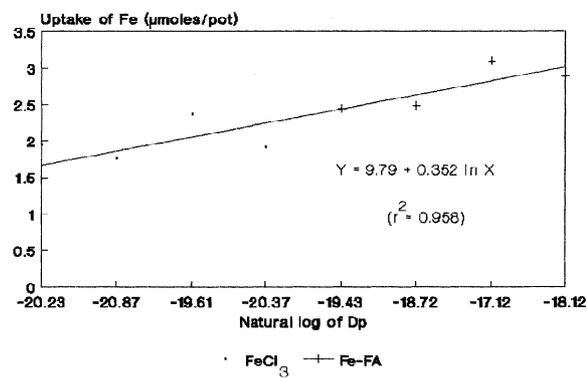


Figure 2. Relationship between Dp and uptake of Fe ( $\mu\text{moles pot}^{-1}$ ) by plants under controlled growth conditions (100% relative humidity).

(i) Normal 60–75% humidity conditions (Figure 1)

$$Y = 10.36 + 0.370 \ln X, (r = 0.960^{**})$$

(ii) Controlled 100% humidity conditions (Figure 2)

$$Y = 9.79 + 0.352 \ln X, (r = 0.958^{**})$$

where  $Y$  = uptake of Fe by seedlings ( $\mu\text{moles/pot}$ ) and  $X$  = porous system self diffusion coefficient of Fe (Dp) in soils ( $\text{cm}^2/\text{s}$ ).

The two equations, computed from the data of uptake of Fe by seedlings grown in different calcareous soils and their corresponding porous system self diffusion coefficient had a predictability of nearly 96%. These two relationships clearly indicate that the diffusive flux of Fe in the soils has a positive and significant bearing on the uptake of this nutrient by seedlings. The results also suggest that diffusion of Fe is a major factor that influences the availability of Fe and its uptake by plants.

The diffusion coefficient of Fe in soils, when applied as the Fe-FA complex, was higher in comparison with those with  $\text{FeCl}_3$  since a higher Fe concentration was maintained in the former case due to a lower capacity factor. Mass flow may be of little significance in influencing uptake of Fe by plants with a low Fe concentration. At a relatively higher concentration of Fe due to application of Fe-FA in soils, however, the influence of mass flow may become more important. At higher Fe concentrations due to its complexation by fulvic acid, there was also an increase in the diffusive flux. The uptake of Fe, however, did not increase linearly with an increase in the porous system self diffusion coefficient of Fe in soils but took a hyperbolic shape. This is because of the fact that the rate of uptake of complexed iron species is lower than the inorganic Fe species rate (Kannan and Wittwer, 1967).

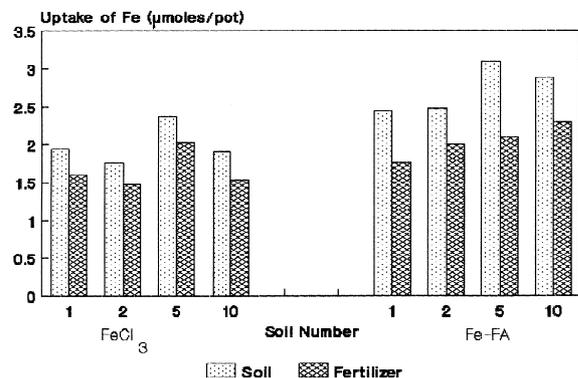


Figure 3. Contribution of soils and fertilizers to uptake of Fe by paddy seedlings under normal growth conditions (60–70% relative humidity).

A significant positive correlation between concentration of Fe in soils and Fe uptake by plants was reported by O'Connor et al. (1971). They applied inorganic Fe and Fe complexed with synthetic chelates. They attributed the higher uptake of Fe by plants to an increase in the diffusion coefficient of Fe in soils. Similar relationships are reported by Gupta and Deb (1985) for the case of Zn. Their was a conclusion similar to ours except that the equation of the straight line was different.

#### *Proportion of Fe in paddy seedlings derived from applied sources*

The fractions of Fe in paddy seedlings derived from fertilizer sources and soils are represented graphically in Figures 3 and 4. It was observed that Fe as a fulvic acid chelate generally supplied Fe to the plants at least twice as effectively as FeCl<sub>3</sub>. The influence of sources of Fe on the fraction of Fe derived from fertilizer was statistically significant ( $P=0.05$ ), whereas the influence of soil and interaction effects of soil by sources of Fe were statistically non-significant in controlled conditions experiments (Tables 5 and 6). All these three factors, however, were statistically significant in normal growth conditions. The increase in the efficiency of Fe-FA as a fertilizer in comparison with FeCl<sub>3</sub> might have resulted from composite effects of the chelating agents in increasing the solubility of the native Fe of soil, keeping Fe in soil solution by protecting it from surface chemical reactions and by increasing the diffusive and convective flow of Fe from the bulk of the soil solution to the plant roots.

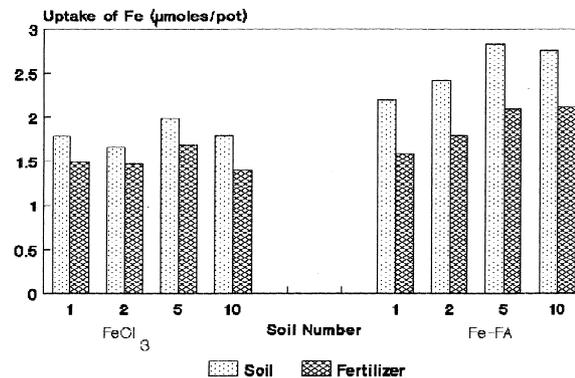


Figure 4. Contribution of soils and fertilizers to uptake of Fe by paddy seedlings under controlled growth conditions (100% relative humidity).

## Conclusions

Naturally occurring humic substances such as fulvic acid were found to overcome the rate-limiting step of transport of Fe from soil solution to plant roots by diffusion. As the diffusive flux of Fe in soil solution serves as the main source of its supply to plant roots, the role of humic substances in alleviating Fe deficiency in calcareous soils is major. The observation that the efficiency of Fe-FA as a fertilizer is much greater than that of FeCl<sub>3</sub>, has important implications for the control of Fe deficiency in crop plants.

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