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IRON AND MANGANESE RELATIONS IN RICE AND BARLEY

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Barley plants grown in standard culture solutions have shown a narrow range of manganese tolerance between deficiency and toxicity levels in the external medium. In the case of Hoagland's solution it has been found that the long-used Mn concentration of 0.5 ppm results in specific toxicity symptoms which appear on the older leaves of barley as small, brown spots. This necrosis, associated with a high Mn-content in the tissues, may be severe enough to cause a considerable lowering of yields. As a consequence, recent investigations have employed lower concentrations of Mn in the composition of Hoagland's solution for some species of plants.

This report contains the results of experiments comparing rice and barley plants grown in Hoagland's solution over a wide range of Mn-concentrations. The study consisted of a comparison of the two grasses with respect to the effects of Mn on yield, symptoms of toxicity or chlorosis, and iron and Mn-content of various plant parts.

Even though the plants were grown in culture solution, it was felt that the results would give an indication of their sensitivity to high Mn-levels. Rice is grown traditionally in submerged soils whereas barley is grown in more or less well-drained soils. Under such divergent conditions of oxidation-reduction, one would expect Fe and Mn to be among the most sensitive of the nutrients present in the soil liable to undergo a change in valence.

METHODS

The plants were grown in painted 5-gallon cans filled with one-fifth strength Hoagland's No. 2 solution. This contains enough NH_4^+ to maintain good pH stability. Iron was supplied at the rate of 0.1 ppm in the form of ferrous sulfate twice weekly. Mn was varied from zero to 5 ppm. The solutions were aerated gently.

Three one-week-old seedlings of barley (*Hordeum vulgare L.*, var. Atlas 46) and rice (*Oryza sativa, japonica* var. Caloro) were placed in separate tanks and allowed to grow for 6 weeks. During this period observations were made on the appearance of the plants after which they were harvested, dried, and weighed. For purposes of analysis the shoots were divided into 3 fractions, young, mature, and old in the ratio of 1 : 2 : 1, except in one case of insufficient plant material. The leaf tissues and the roots were analyzed for Mn and Fe. These were determined as the permanganate, after oxidation with periodate, and as the ortho-phenanthroline complex of iron.

RESULTS

The dry weights of the shoots and roots of both species are given in Fig. 1. Starting from deficient levels of Mn, there is a sharp rise in weight which reaches a peak around 0.1 to 0.2 ppm Mn in the external solution. At higher concentrations there is a gradual

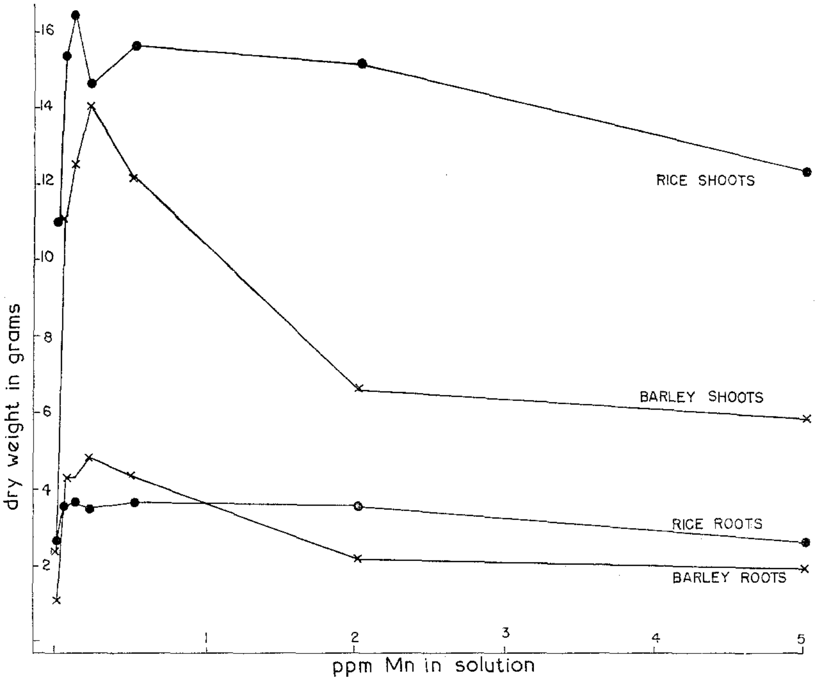

Fig. 1. Dry weights of barley and rice grown in culture solutions with manganese as a variable.

decrease in yield for rice and a very steep drop for barley. In Fig. 2 are shown the values of Mn-content in the 3 fractions of barley

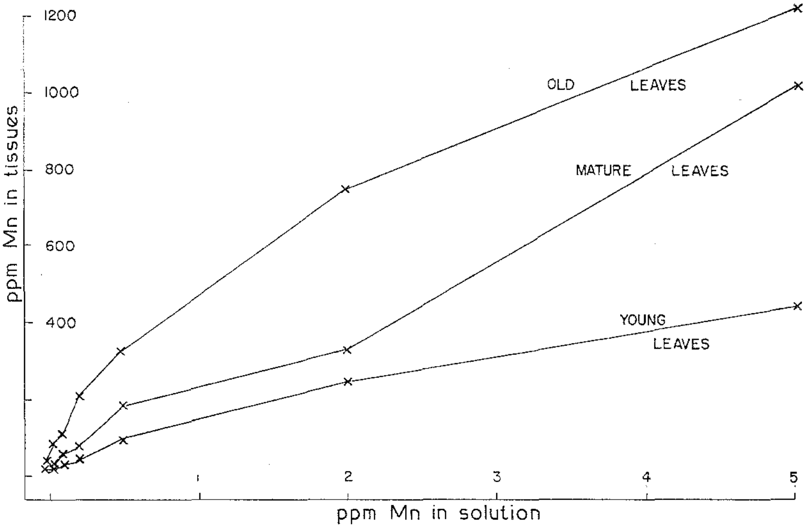

Fig. 2. Manganese content of barley leaves as a function of manganese in solution.

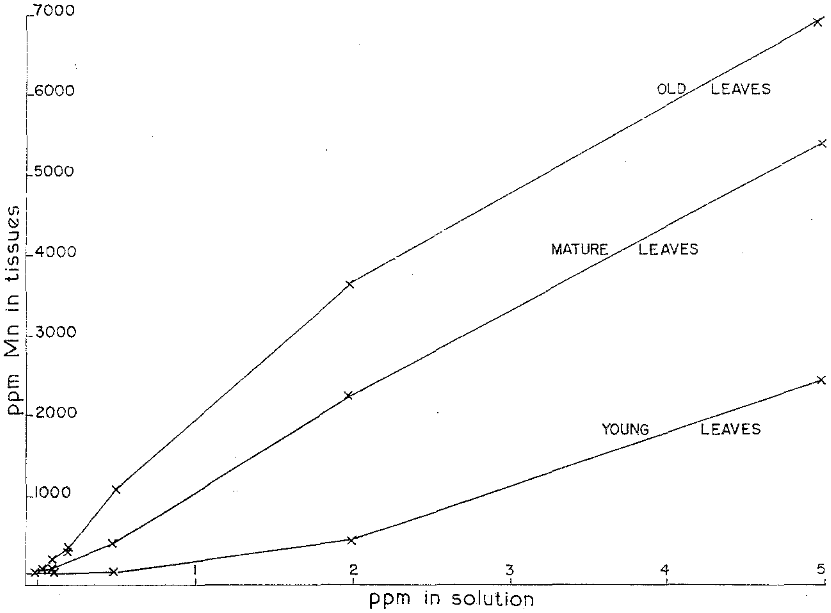

Fig. 3. Manganese content of rice leaves versus manganese in solution.

leaves plotted against Mn-concentration in the solution. The tissue Mn rises with increasing Mn in the external medium, with the young leaves reaching a value of 400 ppm, the mature leaves 1000 ppm, and the old leaves just over 1200 ppm. The trends are similar for rice leaves as shown in Fig. 3, except that the Mn-values are much higher. The young leaves reach a value of over 2000 ppm Mn, the mature leaves more than 5000 ppm, and the old leaves around 7000 ppm. These figures are 5 to 6 times higher than the quantities found in barley leaves at the same solution concentrations. There is a reversal of this situation where the roots are concerned. In Fig. 4 it will be seen that barley roots reach a Mn-content of 8000 ppm while rice roots have about 5000 ppm at a Mn-concentration of 5.0 ppm in solution.

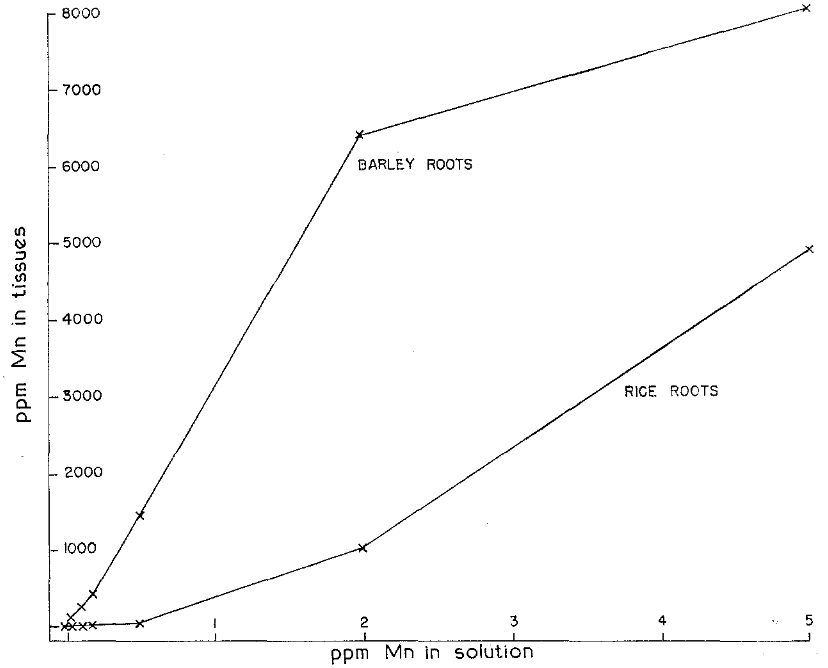

Fig. 4. Manganese content of barley and rice roots of plants grown in solutions at various manganese levels.

The tissues were also analyzed for Fe and in Fig. 5 there appear the results obtained for barley leaves. There is a steep drop in Fe content in all 3 groups of tissues going from zero to 0.2 ppm Mn in

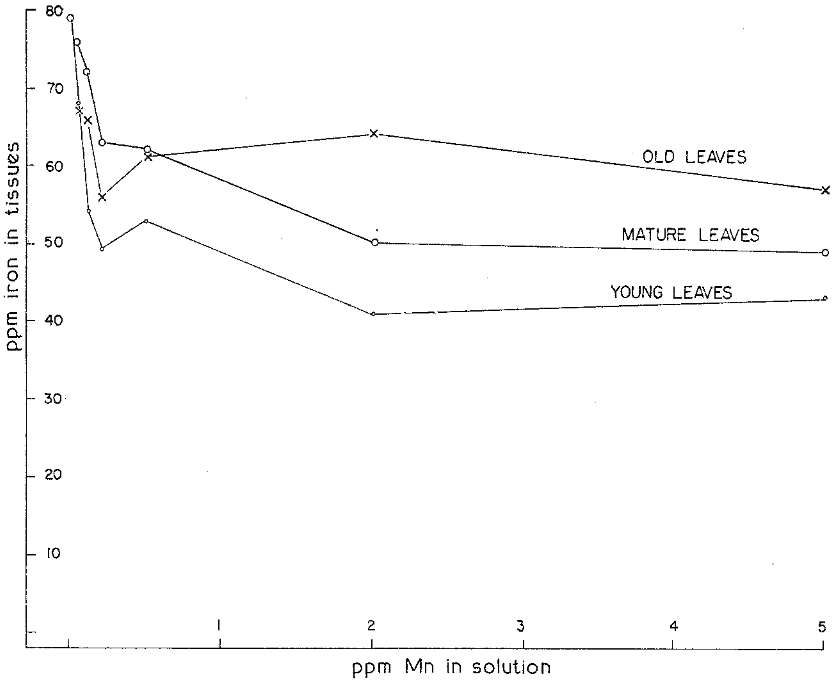

Fig. 5. Iron in leaves of barley plants grown at several levels of manganese.

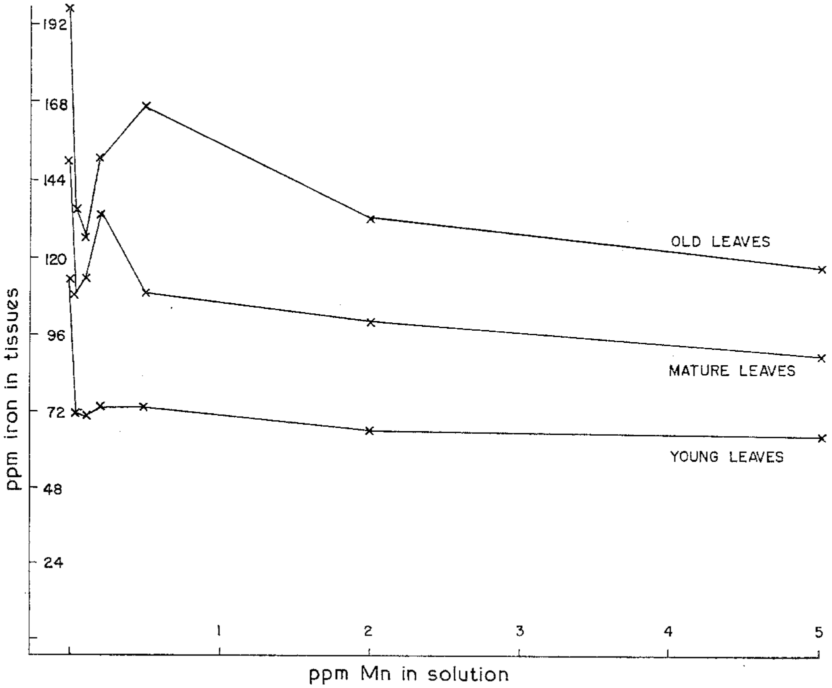

Fig. 6. Iron content of rice leaves of plants grown in solutions with manganese as a variable.

the external solution, but from there on the values are relatively stable. In Fig. 6 the curves for rice also show a rapid decrease in Fe-content as Mn is increased at the lower end of the scale, with a slight tendency to decrease at the higher concentrations of Mn. It will also be noted that the Fe content tends to be about twice as high in rice as it is in barley.

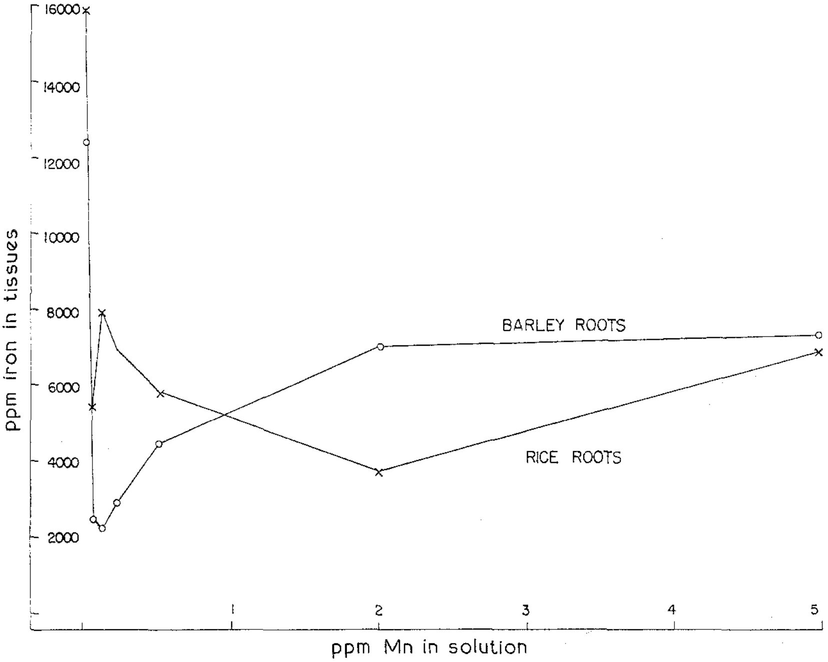

Fig. 7. Iron content of barley and rice roots of plants grown in culture solutions with different manganese concentrations.

The results of the Fe-analysis of the roots of both rice and barley are shown in Fig. 7. Again, as in the shoots, there is a sharp drop in Fe-content of the roots as the Mn-concentration in solution is raised from zero, but in the toxic range of Mn-concentration the curves do not show consistent trends.

The Fe/Mn ratios by tissues are given in Table 1. In both species the ratios decrease from the young to the old tissues, reflecting a greater tendency for Mn to accumulate in the older tissues than for Fe.

Examination of the plants before harvesting revealed a severe incidence of brown spotting on the old barley leaves at the two highest Mn-concentrations. These necrotic spots were less severe on the rice leaves but were otherwise similar in aspect to the ones on barley. In neither species did chlorosis make an appearance under the influence of high Mn-uptake. There were no visible signs of Mn toxicity on the roots of barley or rice.

TABLE 1

Iron-manganese ratios in barley and rice tissues

Mn in solution, ppm	Barley				Rice			
	Young leaves	Mature leaves	Old leaves	Roots	Young leaves	Mature leaves	Old leaves	Roots
0.00	3.0	—	—	—	6.3	6.3	4.7	1300
.05	2.7	1.8	.69	19.3	6.9	1.7	1.2	350
.10	1.3	1.1	.54	9.0	3.2	1.2	.67	334
.2	.98	.76	.26	6.1	4.3	.46	.42	216
.5	.41	.33	.18	3.0	2.9	.28	.15	60
2.0	.16	.15	.08	1.1	.15	.04	.04	3.6
5.0	.11	.05	.04	.9	.03	.02	.02	1.4

DISCUSSION

These experiments show barley to have a greater sensitivity to high Mn than does rice as measured by the intensity of necrotic spotting on the leaves and decrease in growth. What is even more remarkable, this occurs in spite of a much higher accumulation of Mn in rice leaves by a margin five or six times greater than that found in barley tissues under comparable conditions. The lowered yield under high Mn may reflect the influence of necrotic spots in the leaves decreasing the area of active photosynthesis. In addition to this, the high Mn content of the leaves may also produce some biochemical disorders which would require a different kind of study to demonstrate.

The ability of rice plants to tolerate higher Mn-concentrations than can barley may be part of an adaptive mechanism acquired in the course of evolution that enables this species to grow under submerged soil conditions with enhanced Mn-availability. Barley is adapted to a well-drained soil environment and may be unable to cope with a high Mn-regime as well as rice. It must be added, however, that in these experiments rice tissues had roughly twice as high an Fe-content as barley and this may contribute to its greater tolerance of high Mn, in accordance with the relationships obtained by Somers and Shive[15].

The findings reported here of a great tolerance for Mn by rice plants do not support a theory propounded recently by Senewiratne *et al.* [14] as to why rice shows superior growth in submerged versus drained soils. This theory claims that, under drained conditions, featuring a predominantly nitrate source of nitrogen, there is greater Mn-uptake by rice and this is supposed to affect the indoleacetic acid oxidase mechanism and accelerate the destruction of auxin. In the text of that paper it is stated that as much as 0.40 per cent Mn (4000 ppm) was found in rice plants grown under unflooded conditions. In their table 2, however, the data shows only .0375 per cent Mn in the soluble fraction and 0.0414 per cent in the insoluble fraction for a total of 0.0789 per cent. This total is only one-fifth the value given in the text. These amounts are well below those found necessary in our experiments to produce toxicity effects sufficient to lower yields. In other words, our evidence does not fit into the suggestion that high Mn-uptake in well-drained soils might be a decisive factor in the inferior growth of rice compared with that grown in submerged soils.

On this same point it should be added that considerable work can be cited to show that Mn-availability and absorption are actually enhanced by reducing conditions in poorly aerated or submerged soils. In the first place, Schollenberger, Metzger, Robinson, and Ponnemperume, among others, have shown that submergence increases the amount of Mn extractable from soil. In the second place, several workers have demonstrated that Mn uptake is increased when soils are treated so as to favor reducing conditions. Godden and Grimmett[6] found 50 to 60 per cent more Mn in oats and mustard grown in undrained compared to drained soils. Piper[10] was able to increase the uptake of Mn in oats by water-logging soils a week before seeding. He also showed more Mn was absorbed by oats grown in soil kept at 90 per cent water saturation compared to soils at lower moisture tensions. In experiments conducted by Clark *et al.*[3], the Mn-content of rice plants grown under submerged conditions was tenfold greater than that found in plants grown without submergence. It is difficult to reconcile all of these results with the theory that rice plants grow better in submerged soils because they absorb less Mn.

The analysis of the tissues for Fe shows that increasing Mn in the low range causes a steep drop in the Fe-content of both barley and rice. This would suggest a strong interaction but it may, instead, be a dilution effect since the curves in that region of supplied Mn are fairly good inverse images of the growth curves. In the higher range of Mn there is no appreciable effect on the iron content of the tissues.

It is interesting that none of the plants exhibited symptoms of chlorosis, in the presence of such large amounts of Mn in the tissues, as has been reported by many workers in the past. In reviewing this subject some time ago[17], Twyman made the observation that "at this stage it is not possible to differentiate between iron deficiency and manganese toxicity." It wasn't long after this that Wallace and Hewitt[18], Berger and Gerloff[1], and Löhnis[8] produced evidence to distinguish between the two conditions. The toxicity symptoms in our experiments were confined to the shoots and this is in accord with Bortner's work[2] in which he reported that the roots were unaffected by the high Mn-treatments. It is quite likely, though, that at still higher Mn-concentrations the roots would also show symptoms.

The roots of barley and rice are high in iron as suggested by the chemical analysis. Much of this iron may be deposited on the external surface of the roots judging from their bright, red color. This was noticed long ago by Gile and Carrero[5] who found a large amount of iron in the ash of rice roots. They assumed it was due to the presence of finely divided iron oxide on the roots and called it selective contamination from the soil. Since our plants were grown in nutrient solutions, the question of soil contamination is ruled out and we must be dealing with something more like selective non-absorption. A more positive way of looking at it might be to consider the electrostatically negative character of the root surface as an attractive force for the highly charged, positive ferric ions.

Gericke[4] also reported this heavy iron coating on the roots of rice plants and was one of the first to recognize the pre-eminence of iron in the nutrition of rice. In testing the then known essential minerals, he found that iron, above all other nutrients, had to be supplied throughout the greater part of the growth period to maintain satisfactory growth. He interpreted this to mean that either the rice plant could not absorp sufficient iron in its early stages to sustain later growth or the iron was so immobilized in the older tissue that it was unavailable for new growth.

SUMMARY

The growth of rice and barley was studied in nutrient solutions with Mn as a variable over the range from zero to 5 ppm. The optimum growth in these solutions was obtained when the Mn-concentration was at 0.1 and 0.2 ppm. Below this level the yields dropped rapidly for both species. At higher levels of Mn the yield of barley fell rapidly while that of rice did not show a significant decrease until the 5-ppm treatment was reached and even then the drop was slight compared to barley.

Severe symptoms of Mn toxicity appeared on the old leaves of barley but only slight symptoms showed on rice. Neither species developed chlorosis. The Mn-content of barley leaves reached a value of 1200 ppm, while that of rice went as high as 7000 ppm. In the roots the situation was reversed with barley having 8000 ppm Mn against 5000 for rice.

Starting from zero Mn supplied, the iron content of rice and barley fell rapidly in the low range of Mn but this appeared to be a dilution effect. At the higher range of Mn in solution, the tissue iron leveled off. The level of Fe in rice leaves tended to be roughly double that in barley. Substantial amounts of iron were found in the roots but much of this was visible on the surface, probably in the form of iron oxide.

The results were discussed in the light of a recent theory concerning the possible role of Mn in accounting for the inferior growth of rice in drained soils compared to submerged soils.

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