# NUTRITION OF HEVEA BRASILIENSIS II. EFFECT OF NUTRIENT DEFICIENCIES ON GROWTH, CHLOROPHYLL, RUBBER AND MINERAL CONTENTS OF TJIRANDJI 1 SEEDLINGS

*By*

E.W. BOLLE-JONES

*Hevea brasiliensis seedlings were grown in pot sand culture under conditions of varied mineral status. Marked retardation in growth and visual symptoms were observed in plants given restricted supplies of each major nutrient (except calcium) and of iron and manganese. Mineral status affected the concentration of rubber in the stems and petioles during the early part of growth; later, such effects became reduced or reversed. The effects of nutrient deficiency treatments on the chemical composition of the plants are described and the low requirement for calcium and boron pointed out.*

It is important to recognise symptoms of nutrient deficiency in order to assess efficiency of crop production and as a guide to advisory work. The identification of these symptoms and the effect of nutrient deficiencies upon growth and chemical composition of the plant provide a foundation upon which to base manurial and cultural practices. Investigations to this end have been carried out on *Hevea brasiliensis* by E.D.C. Baptist, I. de Haan, and C.E. Rhines, J. McGavack and C.V. Linke.

Baptist carried out early experiments on budded stumps of clòne Tjirandji 1, described in the *Annual Reports* of this institute for 1938, 1939 and 1940; he produced deficiency symptoms for nitrogen, phosphorus, and magnesium, within eighteen months of planting. Symptoms of potassium and calcium deficiencies were not recorded; in fact, the calcium deficient plants produced a heavier weight of shoots and roots than did those supplied with complete nutrient.

De Haan (1950) grew *Hevea brasiliensis* seedlings of unnamed parentage in sand culture and described symptoms or retardation in growth attributed to nitrogen, sulphur, phosphorus, potassium and magnesium deficiencies. He also described symptoms of iron deficiency observed in plants grown in water culture. Values furnished for the weight of the complete plants indicated that potassium deficient plants weighed approximately the same as those grown in the complete nutrient treatment. Although a few of the mineral constituents of the leaves and wood were estimated an attempt to determine the concentration of rubber present in the plant was not made.

RHINES, MCGAVACK and LINKE (1952) grew *Hevea brasiliensis* seedlings of unnamed parentage in sand culture for nine months during which time they observed the occurrence of potassium, magnesium, phosphorus and sulphur deficiency symptoms which were accompanied by a marked decrease in the dry weight of the plants. They found in regard to the nutrient that 'elimination of calcium did not retard growth of these very young rubber trees' and concluded that their 'data indicated benefits in growth rate and increased latex yield from applying critical minerals to balance the tree's nutritional supply'. However, experimental evidence to show the existence of any such relationship between mineral status and rubber content of the plants was not described.

The objectives of the present work were to establish and record symptoms produced by various nutrient deficiency treatments and thus confirm and extend the results of previous workers and to investigate the effect of these treatments on the growth, rubber content and chemical composition of young *Hevea brasiliensis* seedlings.

The experiment, carried out in sand culture, consisted in withholding or providing limited supplies of the major nutrient elements, iron, manganese and boron to the seedlings which were sampled and analysed at intervals during the growth period. Some of the conclusions drawn from this experiment have been described elsewhere (BOLLE-JONES, 1954a).

## EXPERIMENTAL

There were ten treatments: complete nutrient, –N, –S, –P, –K, –Mg, –Ca, –Fe, –Mn, –B; each treatment consisted of two or three pots and was carried out in duplicate. Twelve 'selfed' Tjirandji 1 seeds, obtained from a large monoclone area, were sown in each pot on 6 March 1953. Complete plants were removed from each pot and the total dry weight of shoots and roots recorded on the following dates: 6 July, 17 August, 19 October, 26 January (final sampling). Fully expanded laminae from which the midribs were excised and discarded were taken from different whorl positions and analysed for chlorophyll and mineral contents; the uppermost leaf whorl is always designated as the top whorl while the successive lower one is referred to as the second. The stems and petioles were used for rubber estimation.

This series of samplings was not intended to provide an accurate assessment of seasonal trends (as some of the nutrients were altered in composition during the course of the experiment) but was meant only to give an indication of the relative effect of the different deficiencies on growth and composition.

Details of the sand culture and analytical techniques employed have been described (BOLLE-JONES 1954b). The composition of the complete nutrient was (in milligram equivalents per litre): $NO_3^-$ 7.0, $SO_4^{--}$ 4.0, $PO_4^{---}$ 2.0, $K^+$ 4.0, $Mg^{++}$ 2.0, $Ca^{++}$ 4.0, $Na^+$ 0.66, $Fe^{+++}$ 1.0, $Mn^{++}$ 0.02, $Cu^{++}$ 0.002, $Zn^{++}$ 0.002, B 0.033 (millimols/l), Mo 0.0002 (millimols/l). It became necessary later (19 September) to delete the boron supplement, as the plants appeared to accumulate the element in large amounts, and to increase the $PO_4^{---}$ level to 3.0 meq/l (20 November).

The potassium, calcium, and boron deficient plants were supplied with a nutrient solution similar in composition to that given above but with the respective element deleted. At first the other nutrient solutions were prepared similarly but it was found impossible to grow plants deprived completely of nitrate, sulphate, phosphate or magnesium; it became necessary to supply limited amounts of these ions in order to sustain growth during the experimental period. These amounts were (in meq/l): $NO_3^-$ 1.0 - 2.0, $SO_4^{--}$ 0.5 - 1.0, $PO_4^{---}$ 0.5, $Mg^{++}$ 0.5 - 1.0; they were generally found sufficient to keep the plants alive but insufficient to enable complete recovery to set in. Towards the end of the experiment a limited amount of iron and manganese had to be supplied to the iron and manganese deficient cultures in order that the plants should provide sufficient sampling material.

The iron, manganese and boron deficient nutrient solutions were purified from these elements using procedures described by HEWITT (1952).

## RESULTS

### VISUAL OBSERVATIONS

The complete nutrient plants were the tallest plants in the experiment (TABLE I). From an early stage the effect of decreased nitrogen, sulphur, magnesium or calcium supplies was manifested in decreased height. The subsequent partial recovery in the sulphur deficient plants was due to their being supplied with sulphate to alleviate the severity of the deficiency. Despite the severity of phosphorus deficiency, marked retardation of height elongation did not occur until August but once the effect was obvious further elongation was slight (TABLE I). Both iron and manganese deficient plants, although showing severe symptoms, were not so stunted as the magnesium deficient plants. Boron deficient plants were slightly but consistently shorter than the complete nutrient plants.

The number of leaflets per plant (excluding those of the immature top whorl) may be taken as a guide to the severity of each deficiency. In July the number borne on plants starved of iron, manganese and boron, did not differ from those of the complete nutrient plants. Later (September - October) the defoliating effect of iron or manganese deficiency was manifested in the low number of leaflets (TABLE II). Boron deficiency had no effect at first but towards the end of the experiment produced plants with a larger number of leaflets than those supplied with complete nutrient. The defoliating effect of nitrogen, sulphur, phosphorus and magnesium deficiencies were well marked at an early date and remained so even when plants were supplied with limited amounts of those nutrients. The plants deprived of calcium recovered slightly in January and increased their number of leaflets relative to the complete nutrient plants. Potassium deficiency became sufficiently severe in January to cause almost as much defoliation as was obtained in the phosphorus deficient plants.

TABLE I: AVERAGE HEIGHT OF SHOOT PER PLANT

*Centimetres*

| *Treatment* | *28 April* | *29 May* | *31 July* | *21 Sept* | *20 Nov* | *5 Jan* |
|---|---|---|---|---|---|---|
| *Complete* | 32 | 53 | 76 | 111 | 139 | 161 |
| *-N* | 30 | 38 | 54 | 65 | 75 | 83 |
| *-S* | 30 | 40 | 59 | 84 | 105 | 121 |
| *-P* | 32 | 50 | 68 | 73 | 80 | 91 |
| *-Mg* | 27 | 38 | 49 | 52 | 59 | 68 |
| *-K* | 30 | 44 | 65 | 92 | 117 | 133 |
| *-Ca* | 28 | 44 | 63 | 77 | 90 | 104 |
| *-Fe* | 31 | 52 | 69 | 75 | 96 | 109 |
| *-Mn* | 31 | 51 | 66 | 78 | 90 | 100 |
| *-B* | 32 | 49 | 70 | 91 | 120 | 143 |

TABLE II: AVERAGE NUMBER OF LEAFLETS COUNTED PER PLANT, EXCLUDING THE TOP WHORL, AND THE RATIO OF LEAFLETS TO WHORLS WHICH BORE COUNTED LEAFLETS

| *Treatment* | *Leaflet number* | | | | *Leaflets/whorls* |
|---|---|---|---|---|---|
| | *11 Aug* | *14 Sep* | *14 Oct* | *22 Jan* | *22 Jan* |
| *Complete* | 31 | 44 | 46 | 38 | 27 |
| *N* | 10 | 12 | 11 | 14 | 12 |
| *-S* | 15 | 27 | 39 | 30 | 16 |
| *P* | 9 | 6 | 8 | 20 | 20 |
| *Mg* | 8 | 10 | 15 | 16 | 16 |
| *-K* | 29 | 38 | 45 | 23 | 12 |
| *Ca* | 18 | 18 | 28 | 34 | 20 |
| *-Fe* | 23 | 12 | 11 | 35 | 24 |
| *Mn* | 24 | 24 | 17 | 25 | 19 |
| *B* | 30 | 36 | 50 | 51 | 32 |

The number of leaflets borne per whorl (leaflets/whorls, TABLE II) was greatest in the boron deficient and complete nutrient plants and least in the nitrogen and potassium deficient plants.

The complete nutrient plants were tall and vigorous and became the largest plants in the experiment. The nitrogen and sulphur deficient plants were extremely stunted and yellowish green in appearance. The young expanded laminae, usually of the second whorl, became uniformly yellow and occasionally necrotic at the tip (*Figure 1*) ; these leaves abcissed prematurely.

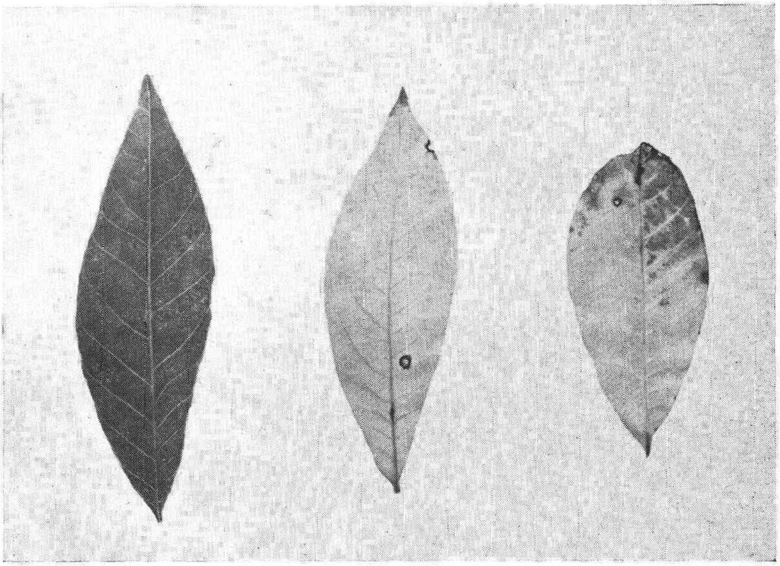


*Figure 1. Nitrogen deficient leaflets (right), complete nutrient leaflet (left); taken from three months old seedlings. Note uniform yellowing of lamina and subsequent necrosis of tip with increasing severity of deficiency.*

Phosphorus deficient plants grew vigorously for the first two months and appeared dark green. Later the plants became olive green and deficiency symptoms (*Figure 2*) appeared in the laminae of the upper or midstem leaves as a yellowish brown or bronze discolouration of the upper lamina surface and a purpling of the underside. The plants became reduced in vigour and 'thin' or spindly (*Figure 3*). When the deficiency was acute the lamina turned upwards at the tip and became scorched.

Magnesium deficiency symptoms usually appeared in the second whorl laminae two months after sowing. In the earlier stages the symptoms were a pale yellow green interveinal mottling which changed to a bright yellow and spread towards the margin (*Figure 4*). These interveinal regions frequently developed brown necrotic patches (*Figure 5*). Later when the plants were larger (four to six months old), these early stages were quickly passed and the main feature was the green band of tissue surrounding the midrib and the intense yellowing of all other regions (*Figure 5*). The earlier stage of interveinal mottling or

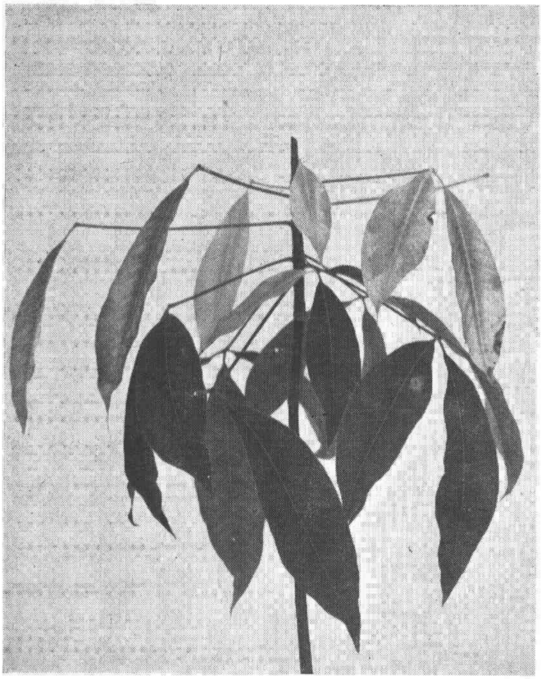


*Figure 2. Acute phosphorus deficiency symptoms in upper leaflets of ten months old seedling. Leaflets at first bronzed but later developed marked purple tints.*

chlorosis has been found, from this and later work, to be distinguishable only with difficulty from the early stages of manganese deficiency; the latter however is not usually accompanied by bright yellow tints neither has it been observed to produce interveinal necrosis.

The potassium deficient plants became noticeably yellower in October. The midstem laminae developed symptoms which originated as a bright yellow or orange brown mottle near the tip and spread marginally downward; this stage was followed by an almost white marginal border which was later replaced by a scorch. The veins generally remained green and the appearance

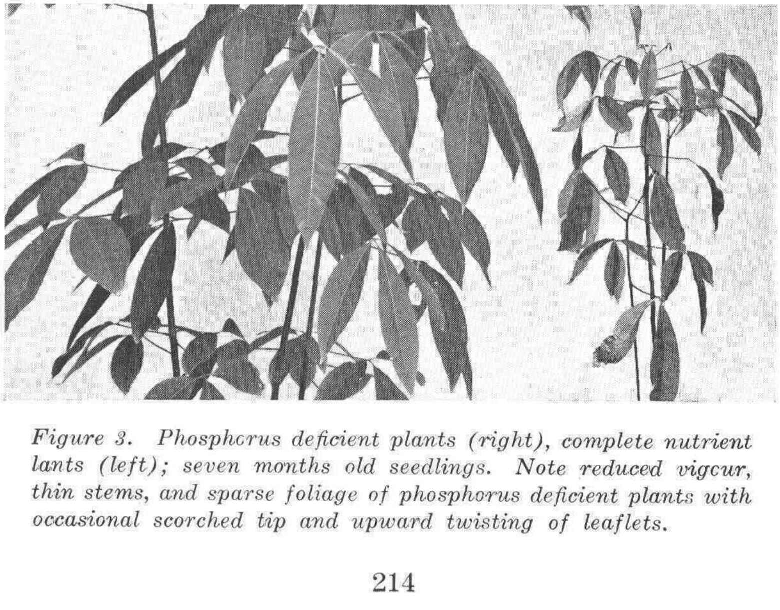


*Figure 3. Phosphorus deficient plants (right), complete nutrient lants (left); seven months old seedlings. Note reduced vigour, thin stems, and sparse foliage of phosphorus deficient plants with occasional scorched tip and upward twisting of leaflets.*

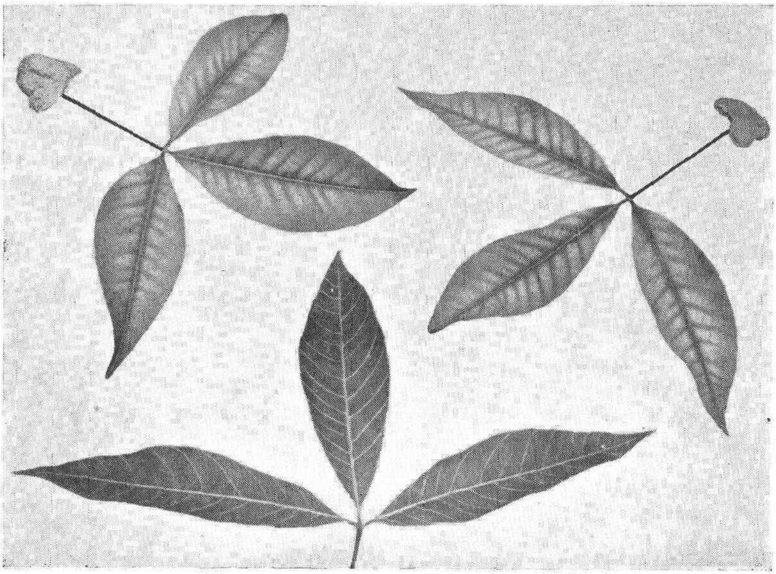


*Figure 4. Magnesium deficient leaves (top), complete nutrient leaf (bottom); taken from three months old seedlings. Note pale yellow green, later becoming yellow, interveinal mottling commencing near midrib.*

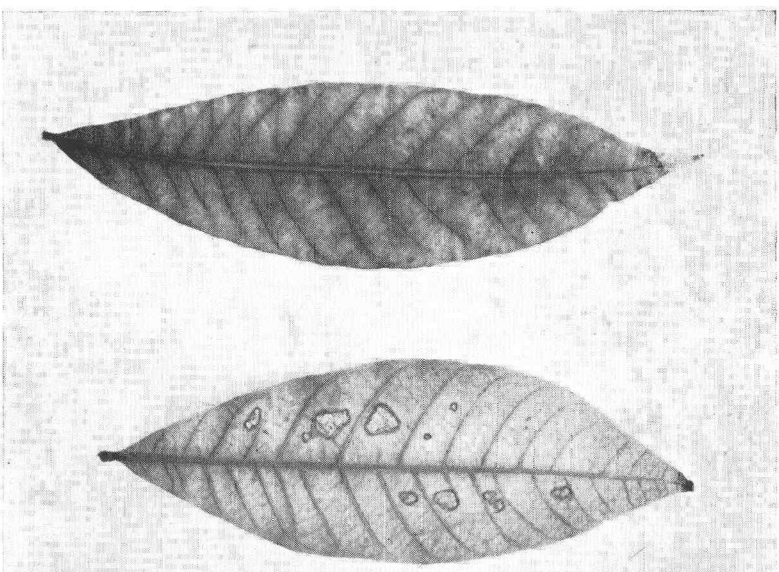
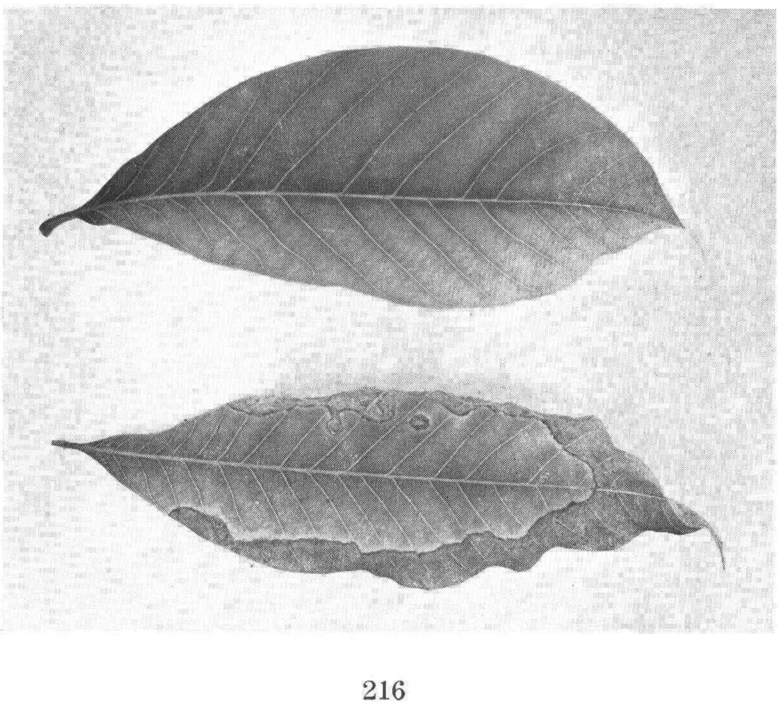


*Figure 5. Magnesium deficient leaflets. Note: (top) block of green tissue surrounding midrib and outer bright yellow region of a leaflet of six months old plant; (bottom) interveinal necrotic regions in a severely affected leaflet of a three months old plant.*

*Figure 6. Potassium deficient plant (left), complete nutrient plant (right); ten months old seedlings. Note defoliated condition and paler colour of potassium deficient plant. Each scale division represents three inches.*

*Figure 7. Potassium deficient leaflets taken from plant shown in Figure 6. Note marginal paling followed by necrotic scorch.*



was one of iron deficiency except for the sequence of symptoms and the presence of marginal scorching (see BOLLE-JONES 1954b, *Figures 3 & 4*). As a result of these symptoms considerable leaf fall ensued (*Figure 6*). Subsequent symptoms appeared in the uppermost expanded laminae; they appeared as a yellowish marginal border which surrounded a dull diffuse greyish green region. The yellow border was later replaced by a very marked severe marginal scorch (*Figure 7*).

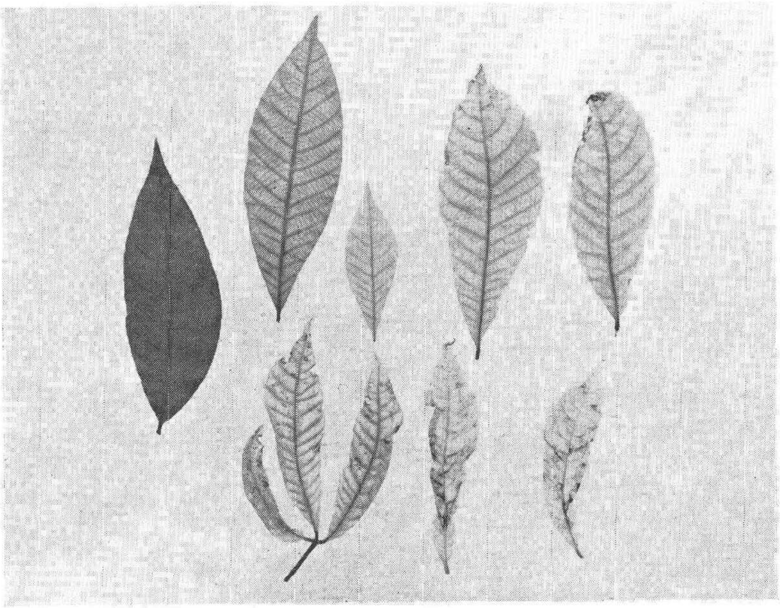
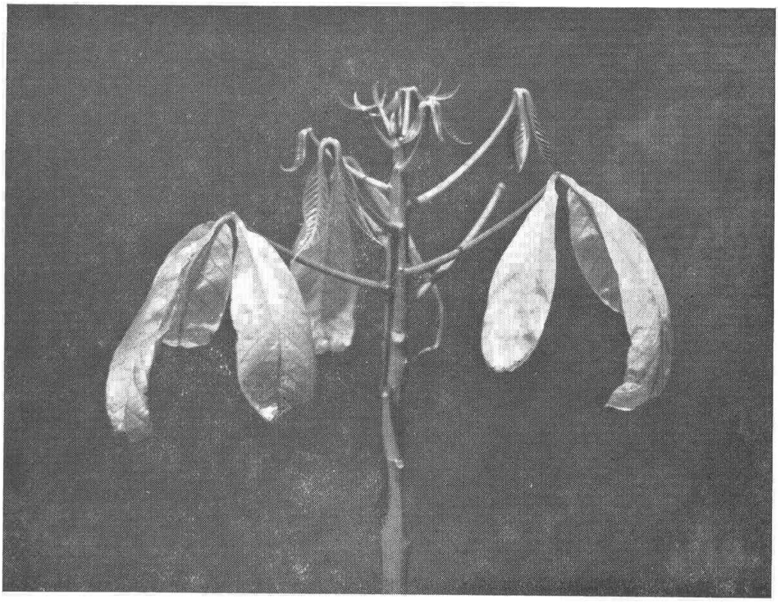


*Figure 8. Iron deficient cholorotic leaflets (right), complete nutrient leaflet (left); taken from three months old plants. Note intense interveinal chlorosis of affected leaflets, their extremely tender condition and subsquent distortion of margins accompanied by a slight necrosis.*

Although the calcium deficient plants were smaller than those of the complete nutrient no diagnostic visual symptoms were apparent.

Iron deficient plants developed acute interveinal chlorosis in the youngest laminae (*Figure 8*). In severe instances the lamina became completely yellow or white (*Figure 9*), except for the green veins, and the stem growing point was killed; growth was continued by means of new sideshoots (*Figure 10*).

Growth of the manganese deficient plants was retarded (*Figure 11*). Laminae of the midstem and uppermost whorls developed diffuse pale yellow green interveinal regions (*Figure 12*). The veins remained markedly white but were surrounded by green strips of tissue and thus stood out in contrast to the uniformly pale green interveinal regions (*Figure 13*).



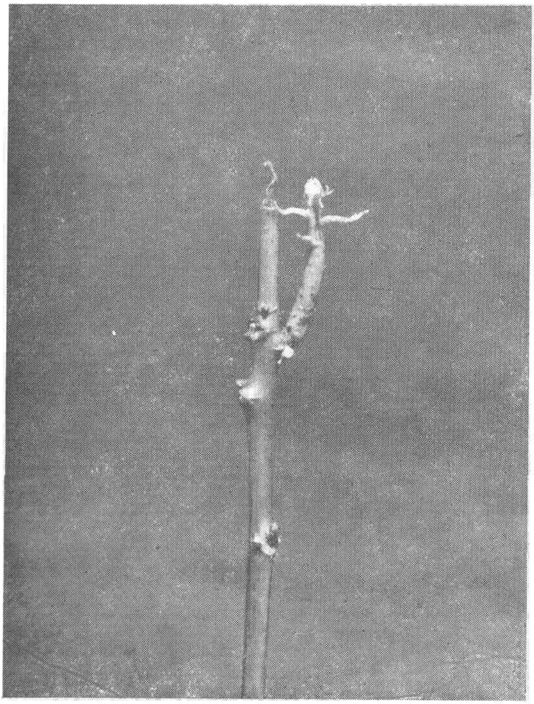
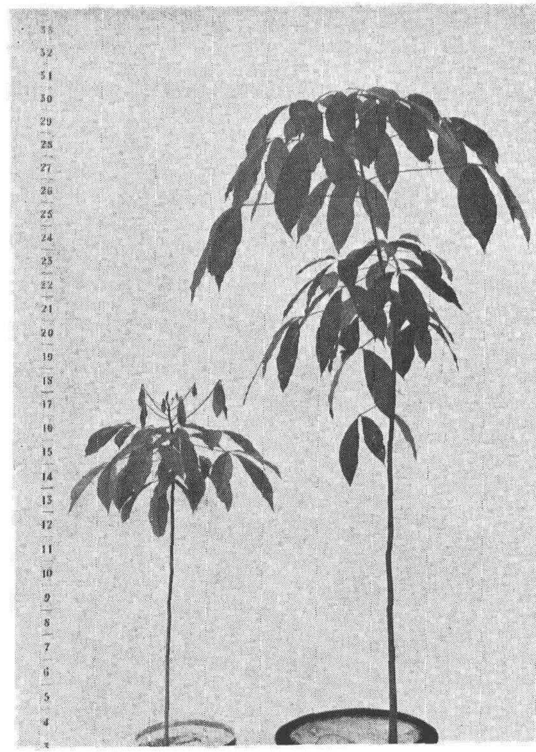


*Figure 9. Terminal shoot of iron deficient plant (six months old seedling). Note distorted reflexed appearance of much reduced youngest leaflets and the whitish yellow appearance of the older leaflets.*

The boron deficient plants were characterised for the first part of the experiment by their dark green foliage.

*Figure 10. Tip of iron deficient plant (six months old seedling). Note death of original growing point and continuation of growth by a new, but still chlorotic, sideshoot.*

*Figure 11. Manganese deficient plant (left), complete nutrient plant (right); ten months old seedlings.*



Growth continued to be vigorous and no leaf symptoms were observed until January. The young leaves surrounding the growing point of one plant died after turning black at the tips (*Figure 14*); another plant shed its youngest laminae before they expanded and this was

*Figure 12. Manganese deficient plant; close-up of plant shown in Figure 11. Note the well defined veins and diffusely pale green interveinal regions of the expanded laminae.*

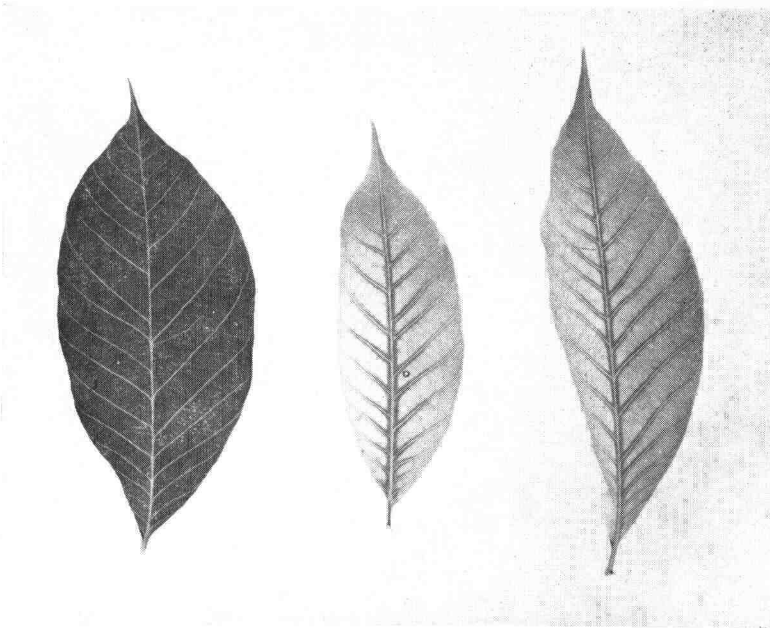
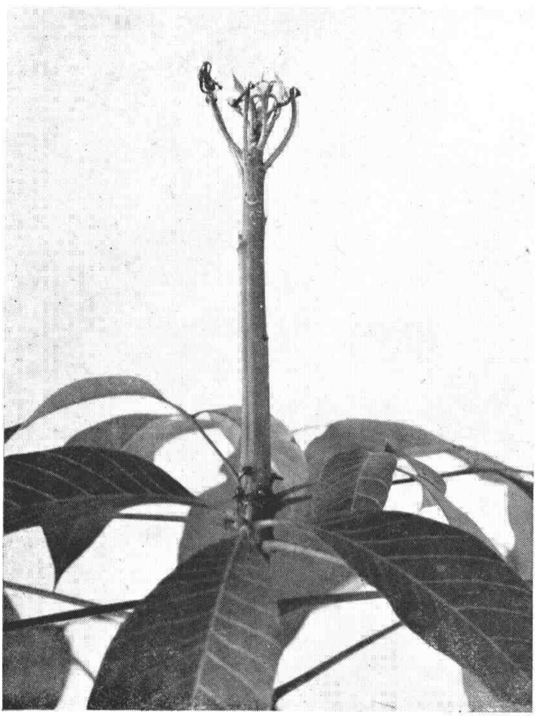


*Figure 13. Manganese deficient leaflets (right); complete nutrient leaflet (left). Note well defined veining accompanied by green piping and pale green or yellowish interveinal regions.*



accompanied by a pale yellow diffuse chlorosis of the topmost expanded laminae. Whether these symptoms were characteristic of boron deficiency remains to be confirmed. Death of the growing point was not observed to occur but apical growth was markedly retarded and the plants declined in vigour.

*Figure 14. Suspected boron deficiency; ten months old seedling. Note blackening and premature death of youngest laminae.*

### TOTAL DRY WEIGHT OF COMPLETE PLANT

The reductions in dry weight caused by nitrogen, sulphur, magnesium and calcium deficiencies were evident even at the July sampling. The effects of phosphorus, iron and manganese deficiencies were not marked until August, five months after sowing. Boron deficiency did not significantly affect yield until the January sampling when a depression was observed (TABLE III). The reduced values obtained for many of the deficient plants reflected the efficiency of the sand culture technique employed.

### RUBBER CONTENT

STEMS AND PETIOLES TABLE IV—The stems and petioles of the plants grown in a replicate were bulked together prior to drying, milling and extraction. Striking and consistent reductions in rubber concentration as a result of mineral deficiencies were obtained at the July sampling. The complete nutrient plants contained a significantly higher concentration of rubber than plants receiving other nutrients. The decrease obtained in the boron deficient plants was marked even though they did not exhibit leaf symptoms and appeared visually to be the most healthy and vigorous plants in the experiment.

TABLE III: TOTAL DRY WEIGHT OF COMPLETE PLANT

*Shoot plus root in grams*

| *Treatment* | *July* | *August* | *October* | *Januyar* |
|---|---|---|---|---|
| *Complete* | 13 | 31 | 92 | 241 |
| *-N* | 4 | 8 | 17 | 33 |
| *-S* | 5 | 13 | 64 | 119 |
| *-P* | 11 | 14 | 17 | 41 |
| *-Mg* | 4 | 7 | 11 | 34 |
| *-K* | 9 | 21 | 52 | 126 |
| *-Ca* | 7 | 16 | 27 | 108 |
| *-Fe* | 10 | 17 | 20 | 89 |
| *-Mn* | 11 | 17 | 18 | 51 |
| *B* | 12 | 28 | 77 | 190 |
| *Min. 5% sig diff* | 3.4 | 7.2 | 22.7 | 36.3 |
| *Min. 1% sig diff.* | 4.9 | 10.3 | 32.7 | 52.1 |

At the August sampling, when the plants were five months old, the rubber concentration of the complete nutrient plants was still significantly higher than for the plants deficient in nitrogen, iron or boron. The concentration was particularly low in the boron deficient plants which did not, at this time, differ significantly from the complete nutrient plants in respect of dry weight per plant.

At the seven month stage the concentration of rubber in the stems and petioles of nutrient deficient plants generally exceeded that of the complete nutrient plants except for the boron deficient plants. It was apparent that whenever the dry weight yield per plant was low (TABLE III), the rubber concentration tended to be high.

At the January sampling stems and petioles of sulphur deficient plants contained a notably smaller concentration of rubber than the complete nutrient plants but, as for the other deficiencies this difference was not significant.

ROOTS, TABLE IV—The concentration of rubber found in the roots was less than that of the stems and petioles but whereas the concentration values obtained for the latter decreased with time those of the roots remained fairly constant. No relation between mineral status and rubber concentration of the roots was distinguished.

TABLE IV: RUBBER IN DRY STEMS AND PETIOLES (STEM + PETIOLE AND ROOTS)

*Percentages*

| *Treatment* | *July* (stem + petiole) | *August* stem + petiole) | *October* (stem + petiole) | *October* (roots) | *January* (stem + petiole) | *January* (roots) |
|---|---|---|---|---|---|---|
| *Complete* | 2.37 | 1.26 | 0.73 | 0.51 | 0.77 | 0.47 |
| -*N* | 2.02 | 1.09 | 1.09 | 0.51 | 0.88 | 0.67 |
| *S* | 1.70 | 1.22 | 0.92 | 0.35 | 0.54 | 0.56 |
| -*P* | 1.27 | 1.16 | 0.86 | 0.41 | 0.76 | 0.59 |
| -*Mg* | 1.83 | 1.28 | 1.12 | 0.53 | 0.90 | 0.56 |
| -*K* | 1.57 | 1 26 | 0.99 | 0.47 | 0.60 | 0.48 |
| *Ca* | 1.58 | 1.34 | 0.94 | 0.48 | 0.73 | 0.45 |
| -*Fe* | 1.58 | 1.01 | 1.25 | 0.55 | 0.84 | 0.55 |
| -*Mn* | 1.61 | 1.27 | 1.01 | 0.50 | 0.83 | 0.44 |
| *B* | 1.48 | 0.94 | 0.72 | 0.49 | 0.68 | 0.47 |
| *Min 5% sig diff* | 0.26 | 0.17 | 0.37 | 0.20 | 0.28 | 0.36 |
| *Min 1% sig diff.* | 0.37 | 0.25 | 0.54 | 0.29 | 0.41 | 0.52 |

## CHLOROPHYLL AND MINERAL CONTENTS OF THE LAMINAE

TABLES V, VI & VII—Expanded laminae were taken from the top and second whorl positions; these were analysed for chlorophyll and as many of the mineral nutrients as the amount of material permitted. The results for July, October and January values were separately analysed statistically; this analysis should reveal any interaction between whorl and treatment which might exist. In the event of such inter-action achieving significance this is indicated in the tables.

CHLOROPHYLL CONTENT—At the earliest sampling magnesium deficiency reduced the chlorophyll content of the laminae, particularly those of the second whorl. This effect was not apparent in October as some magnesium had had to be supplied to the plants.

TABLE V: CHLOROPHYLL AND MINERAL COMPOSITION OF TOP AND SECOND WHORL LAMINAE. JULY SAMPLING

*Presented as mg chlorophyll/gm dry lamina; % N, S, P, Mg, K, Ca or p.p.m. Fe, Mn and B of dry lamina*

| Treatment | Chlorophyll | | N | | S | | P | | Mg | | K | | Ca | | Fe | | Mn | | B | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | | Whorl | |
| | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd | top | 2nd |
| Complete | 5.9 | 6.8 | 3.5 | 3.5 | .24 | .26 | .28 | .23 | .26 | .24 | 1.6 | 1.4 | .38 | .76 | 85 | 80 | 35 | 38 | 68 | 181 |
| -N | 6.4 | 6.7 | 3.5 | 3.4 | — | — | .36 | .24 | .24 | .19 | 1.8 | 1.5 | .52 | .51 | — | — | 145 | 129 | 159 | 64 |
| -S | 6.5 | 5.0 | 3.6 | 3.3 | .40 | .40 | .32 | .24 | .27 | .14 | 1.5 | 1.5 | .51 | .68 | — | — | 74 | 54 | 64 | 197 |
| -P | 6.3 | 6.0 | 3.0 | 2.9 | .17 | .21 | .16 | .12 | .19 | .16 | 1.6 | 1.4 | .26 | .50 | — | — | 20 | 21 | 68 | 147 |
| -Mg | 5.0 | 4.3 | 3.8 | 2.7 | .18 | .24 | .26 | 20 | .05 | .02 | 2.8 | 2.4 | .76 | 1.34 | — | — | 82 | 105 | 136 | 184 |
| -K | 5.6 | 6.4 | 3.4 | 3.6 | .28 | .28 | .30 | .22 | .26 | .32 | 1.2 | 1.1 | .38 | .78 | 46 | 72 | 20 | 24 | 122 | 180 |
| -Ca | 6.0 | 6.2 | 3.6 | 3.5 | .19 | .18 | .26 | .22 | .40 | .35 | 2.2 | 2.2 | .14 | .21 | 60 | 98 | 31 | 27 | 112 | 122 |
| -Fe | 6.4 | 5.9 | 3.7 | 3.5 | .20 | .24 | .31 | .22 | .21 | .25 | 1.6 | 1.4 | .44 | .90 | 58 | 104 | 24 | 30 | 65 | 156 |
| -Mn | 5.7 | 6.4 | 3.6 | 3.7 | .22 | .24 | .32 | .25 | .24 | .22 | 1.6 | 1.5 | .42 | .70 | 49 | 86 | 12 | 16 | 62 | 183 |
| -B | 6.4 | 7.3 | 3.3 | 3.7 | .20 | .24 | .26 | .21 | .20 | .18 | 1.6 | 1.6 | .39 | .74 | 62 | 98 | 32 | 40 | 4 | 5 |
| Min. 5% sig. diff. | 1.48 | | 0.45 | | 0.064 | | 0.076 | | 0.078 | | 0.57 | | 0.33 | | 36.2 | | 25.9 | | 58.0 | |
| Min. 1% sig. diff. | 2.06 | | 0.61 | | 0.089 | | 0.105 | | 0.107 | | 0.78 | | 0.45 | | 51.1 | | 35.6 | | 80.3 | |
| Treatment x Whorl | n.s. | | Sig. 5% | | n.s. | | n.s. | | n.s. | | n.s. | | n.s. | | n.s. | | n.s. | | Sig. 0.1% | |

TABLE VI: CHLOROPHYLL AND MINERAL COMPOSITION OF TOP AND SECOND WHORL LAMINAE. OCTOBER SAMPLING

*Presented as mg chlorophyll/gm dry lamina; % N, S, P, Mg, K, Ca or p.p.m. Fe, Mn and B of dry lamina*

| Treatment | Chlorophyll | | N | | S | | P | | Mg | | K | | Ca | | Fe | | Mn | | B | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd | Whorl top | Whorl 2nd |
| Complete | 5.3 | 6.0 | 2.9 | 2.8 | .14 | .19 | .20 | .16 | .24 | .23 | 1.2 | 1.0 | .40 | .70 | 82 | 86 | 11 | 16 | 20 | 44 |
| -N | 4.7 | 4.4 | 2.8 | 2.6 | — | — | .26 | .31 | .20 | .17 | 1.7 | 1.7 | .47 | .66 | 68 | 90 | 22 | 48 | 68 | 172 |
| -S | 5.3 | 5.6 | 3.2 | 3.0 | .12 | .16 | .22 | .18 | .19 | .19 | 1.3 | 1.0 | .52 | .78 | 66 | 76 | 10 | 14 | 30 | 78 |
| -P | 5.6 | 6.4 | 3.1 | 2.5 | — | — | .17 | .13 | .15 | .14 | 1.4 | 1.1 | .41 | .66 | 62 | 80 | 31 | 44 | 79 | 235 |
| -Mg | 6.1 | 5.6 | 3.4 | 3.2 | — | — | .24 | .21 | .16 | .10 | 1.8 | 1.7 | .86 | 1.06 | 87 | 96 | 24 | 22 | 73 | 181 |
| -K | 3.6 | 3.0 | 3.1 | 2.9 | .20 | .20 | .25 | .24 | .40 | .56 | 0.5 | 0.4 | .52 | 1.08 | 54 | 61 | 7 | 12 | 52 | 152 |
| -Ca | 4.7 | 4.2 | 3.1 | 2.9 | .19 | .22 | .23 | .20 | .42 | .40 | 2.2 | 1.8 | .20 | .16 | 64 | 78 | 8 | 12 | 76 | 164 |
| -Fe | 4.5 | 4.4 | 3.5 | 2.9 | — | — | .24 | .21 | .30 | .31 | 1.5 | 1.2 | .48 | 1.06 | 53 | 84 | 12 | 16 | 42 | 131 |
| -Mn | 4.4 | 3.6 | 3.9 | 3.4 | — | — | .32 | .24 | .24 | .16 | 1.6 | 1.0 | .48 | 1.22 | 59 | 87 | 6 | 14 | 70 | 173 |
| -B | 5.6 | 7.5 | 3.0 | 3.0 | .16 | .17 | .22 | .18 | .22 | .22 | 1.3 | 0.9 | .50 | .88 | 72 | 92 | 17 | 34 | 12 | 14 |
| Min. 5% sig. diff. | 0.82 | | 0.48 | | 0.060 | | 0.059 | | 0.087 | | 0.37 | | 0.260 | | 26.7 | | 11.7 | | 39.2 | |
| Min. 1% sig. diff. | 1.14 | | 0.66 | | 0.087 | | 0.082 | | 0.119 | | 0.51 | | 0.358 | | 36.9 | | 16.2 | | 54.2 | |
| Treatment x Whorl | Sig. 1% | | n.s. | | n.s. | | n.s. | | Sig. 5% | | n.s. | | Sig. 5% | | n.s. | | n.s. | | Sig. 1% | |

TABLE VII: CHLOROPHYLL AND MINERAL COMPOSITION OF TOP AND SECOND WHORL LAMINAE. JANUARY SAMPLING

*Presented as mg chlorophyll/gm dry lamina; % N, S, P, Mg, K, Ca or p.p.m. Fe, Mn, and B of dry lamina*

| Treatment | Chlorophyll | | N | | S | | P | | Mg | | K | | Ca | | Fe | | Mn | | B | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd | Whorl top | 2nd |
| Complete | 6.4 | 6.9 | 3.1 | 2.6 | .17 | .18 | .22 | .18 | .24 | .17 | 1.1 | 1.1 | .51 | .82 | 66 | 86 | 18 | 26 | 15 | 7 |
| -N | 5.9 | 5.8 | 3.0 | 2.7 | .24 | .30 | .28 | .30 | .16 | .11 | 1.8 | 1.3 | .56 | .75 | 86 | 68 | 32 | 31 | 8 | 12 |
| -S | 5.8 | 5.7 | 3.0 | 2.9 | .12 | .12 | .22 | .18 | .12 | .06 | 1.5 | 1.4 | .57 | .82 | 68 | 74 | 15 | 16 | 5 | 10 |
| -P | 5.6 | 6.3 | 2.8 | 2.9 | .15 | .22 | .12 | .14 | .08 | .06 | 1.5 | 1.1 | .40 | .62 | 60 | 72 | 26 | 30 | 13 | 19 |
| -Mg | 6.2 | 5.6 | 3.3 | 2.9 | .18 | .22 | .22 | .20 | .10 | .02 | 2.5 | 1.7 | .76 | 1.32 | 72 | 78 | 14 | 24 | 8 | 8 |
| -K | 5.1 | 3.2 | 3.0 | 2.1 | .14 | .16 | .27 | .19 | .33 | .59 | 0.4 | 0.3 | .63 | 1.10 | 50 | 58 | 16 | 22 | 9 | 12 |
| -Ca | 5.1 | 5.5 | 3.1 | 2.8 | .15 | .18 | .23 | .20 | .33 | .22 | 2.3 | 1.8 | .11 | .14 | 61 | 76 | 11 | 12 | 5 | 8 |
| -Fe | 4.8 | 5.6 | 3.0 | 2.8 | .16 | .18 | .24 | .22 | .20 | .14 | 1.4 | 1.0 | .63 | 1.01 | 58 | 65 | 16 | 22 | 5 | 2 |
| -Mn | 4.6 | 5.2 | 3.4 | 3.4 | .18 | .18 | .31 | .28 | .18 | .16 | 1.7 | 1.4 | .46 | .79 | 72 | 96 | 8 | 9 | 12 | 9 |
| -B | 7.0 | 6.6 | 3.2 | 2.7 | .14 | .16 | .22 | .18 | .21 | .17 | 1.4 | 1.2 | .58 | 1.00 | 74 | 83 | 24 | 34 | 1.2 | 0.4 |
| Min. 5% sig. diff. | 1.39 | | 0.35 | | 0.050 | | 0.048 | | 0.075 | | 0.52 | | 0.236 | | 26.1 | | 5.7 | | 5.3 | |
| Min. 1% sig. diff. | 1.90 | | 0.48 | | 0.068 | | 0.065 | | 0.102 | | 0.71 | | 0.323 | | 35.7 | | 7.8 | | 7.2 | |
| Treatment x Whorl | n.s. | | Sig. 5% | | n.s. | | n.s. | | Sig. 1% | | n.s. | | n.s. | | n.s. | | n.s. | | Sig. 5% | |

Iron and manganese deficiencies resulted in a reduced chlorophyll content at the August sampling (not presented here); this effect became more marked later. Although the calcium deficient plants had no distinct symptoms it was noteworthy that the laminae contained less chlorophyll than those of the complete nutrient plants at the October and January samplings.

When potassium deficiency symptoms became severe in October chlorophyll content was sharply depressed; this depletion was more obvious in the second whorl laminae.

Boron deficient laminae generally contained a higher chlorophyll content than those taken from complete nutrient plants.

NITROGEN CONTENT—The total nitrogen content was higher in the top whorl laminae and decreased as the leaves became older. Nitrogen starved laminae contained slightly less nitrogen than those of the complete nutrient plants but this difference was not significant. Manganese deficiency increased the nitrogen content of the laminae, presumably due to nitrate accumulation.

SULPHUR CONTENT—More sulphur was found in the second than in the top whorl laminae. At the July and August samplings the concentration of sulphur in the laminae of sulphur deficient plants was higher than for those supplied with complete nutrient. The October sampling however revealed a slight depletion in the sulphur content of laminae taken from sulphur deficient plants; this reduction became marked in January. At the last sampling nitrogen deficient laminae were found to accumulate more sulphur than those of the complete nutrient plants.

PHOSPHORUS CONTENT—The phosphorus content of the laminae was highest in the youngest leaves. Reduced supplies of phosphorus decreased markedly the phosphorus content of the laminae. Reduced supplies of manganese consistently increased the phosphorus content; postassium deficiency produced the same effect in the later samplings.

MAGNESIUM CONTENT—The magnesium content of the laminae was generally higher in the younger laminae. The laminae of either potassium or calcium deficient plants accumulated more magnesium than those of the complete nutrient plants whereas restricted supplies of magnesium, nitrogen, sulphur or phosphorus depleted the magnesium content of the laminae.

The analysis of variance indicated that the treatment × whorl interaction was significant at the later samplings. This may have been partially due to the effect of calcium deficiency being more marked in the top whorl laminae and of potassium deficiency in the second whorl.

POTASSIUM CONTENT—The concentration of potassium in the lamina was highest in the youngest leaves. The laminae of nitrogen, magnesium, or calcium deficient plants consistently accumulated more potassium than those of the complete nutrient plants; these effects were obvious even at the July sampling. As the plants grew older the potassium content of the potassium deficient plants diminished markedly in accord with the severe visual symptoms then apparent.

CALCIUM CONTENT—The concentration of calcium in the lamina increased with age of leaf. Magnesium deficient laminae consistently contained, from the earliest sampling, markedly higher concentrations of calcium than found in the complete nutrient plants. This was also true for potassium deficient plants in the later samplings and to some degree for those deficient in boron. Even though the calcium content of calcium deficient top whorl laminae was reduced to 0.11 per cent at the January sampling, diagnostic deficiency symptoms were not recorded.

IRON CONTENT—The iron content of the top laminae was lower than that of the second whorl. At the earlier samplings, when iron deficiency was quite marked, the iron content of the second whorl laminae of plants deprived of iron exceeded or was equal to that of the complete nutrient plants. Later the iron content of the iron deficient laminae became less than that of the complete nutrient plants. It was noticeable throughout that laminae deprived of potassium possessed a lower iron content than the complete nutrient plants.

MANGANESE CONTENT—The concentration of manganese in the laminae increased with age of leaf. The reduced amount of manganese available to the manganese deficient plants was reflected at all samplings in the low amount of manganese found in the lamina. Laminae taken from plants supplied with limited amounts of nitrogen accumulated manganese in quantities exceeding those found in complete nutrient laminae. There is a suggestion that calcium and potassium deficiencies caused reductions in the manganese contents of the laminae.

BORON CONTENT—The concentration of boron in the laminae increased with age of leaf. The third whorl laminae (not presented here) of the complete nutrient plants contained 215 p.p.m. of boron at the July sampling and 204 p.p.m. at the August sampling. Similar laminae taken from boron deficient plants contained 9 p.p.m. (July) or 10 p.p.m. (August) of boron. The levels attained in the complete nutrient laminae were high and it was therefore decided to omit boron from the micronutrient supplement supplied to all treatments; the October sampling accordingly showed a drop in boron content to a level of 116 p.p.m.

The values presented in TABLES V, VI and VII showed that the boron deficient plants, although apparently healthy in appearance, had a very low boron content. There was evidence that the effect of treatment on boron content varied with whorl position.

## DISCUSSION

All the nutrient deficiencies investigated produced a reduced concentration of rubber in the stems and petioles during the earlier part of the experiment. Later, when the differences between the dry weight of the complete nutrient plants and the deficient plants became pronounced (TABLE III), the concentration of rubber in the deficient plants frequently exceeded that of the complete nutrient plants (TABLE IV). Whenever the dry weight of the plant was low, the rubber concentration appeared to be high.

It was inferred that mineral status would only have a direct effect on rubber concentration in the earlier stages of growth when the differences in dry weight between complete nutrient and deficient plants were relatively small. The total rubber content of the plant was however governed by the dry weight production which, in turn, was dependent on mineral status. Accordingly, under field conditions where nutrient deficiencies are sufficiently severe to cause an appreciable decrease in the vigour and dry weight production of the plant, a smaller yield of rubber per tree can be expected.

The failure to obtain a maintained positive correlation, as was recorded for July, between mineral status and rubber concentration may reflect the limitation of the procedure employed for obtaining a representative value for this concentration. The choice of bulked stems and petioles as material for rubber extraction was largely governed by the weight of dried rubber-rich material required; petioles alone were unlikely to furnish this weight in an experiment which employed two or three pots per replicate only. If the rubber content of the laminae could be determined it might become possible to detect a relationship between mineral status and rubber concentration even in the younger laminae of mature trees. The inclusion of stems, in the material extracted in the present experiment, increased the proportion of non rubber bearing tissues in the material extracted as the plants grew older; this reduced the rubber concentration values obtained. In later experiments the green portion of the stems was separated from the brown and used for analysis; this maintained higher concentration values. It was noteworthy that the concentration of rubber in the roots did not diminish with time so markedly as that of the stems and petioles (TABLE IV).

The retardation of growth as a result of nutrient deficiency was usually accompanied by visual leaf symptoms. The calcium deficient plants did not exhibit any diagnostic symptoms despite the retarded growth and much depleted calcium content (TABLE VII); if the experiment had continued with the plants strictly deprived of calcium, deficiency symptoms would probably have appeared despite the low requirement *Hevea brasiliensis* possesses for this nutrient.

The concentration of boron found in boron deficient plants was exceedingly low (TABLES V, VII) and would have been expected if found in a temperate climate crop, to be accompanied by severe deficiency symptoms. The boron requirement of the plant was therefore low—growth was not appreciably diminished until the last sampling—but its ability to accumulate boron was also great, as was found for the complete nutrient plants (TABLE V) which were supplied with only 0.37 p.p.m. of boron in the nutrient. The ability to withstand high internal concentrations of boron, and to survive on low amounts of calcium, may represent an adaptation of *Hevea brasiliensis* to the strongly acid soil conditions found in Malaya.

The general efficiency of the cultural technique employed compared favourably with that of previous workers (*loc. cit.*). It is not however claimed that nitrogen and sulphur deficiencies can be clearly recognised from one another except by analysis. *Hevea brasiliensis* appears to be extremely susceptible to the effects of iron and manganese deficiencies; the absence of other nutrients, with the exception of boron, rapidly caused growth retardation.

The chemical analyses gave information, on the effects of nutrient deficiencies on the composition of the plant, which was of value in providing a basis for future investigations and as a source of guidance in the interpretation of analytical results obtained in the course of advisory work.

Deficiencies of potassium or manganese were found to increase the phosphorus content of the laminae. Deficiencies of nitrogen, sulphur, or phosphorus decreased the magnesium content of the laminae which was increased by deficiencies of potassium and calcium. Deficiencies of nitrogen, magnesium, or calcium increased the potassium content of the laminae. Restricted nutrient supplies of magnesium or potassium increased the calcium content of the laminae.

The effects of calcium and potassium deficiencies on the magnesium, potassium and calcium contents and potassium deficiency on phosphorus content confirm those reported by RHINES, MCGAVACK and LINKE (1950).

Many of the above relationships are well known as a result of investigations on other crops. The increased manganese content of the lamina under conditions of nitrogen deficiency and the increased nitrogen content obtained with manganese deficiency, are of special interest. The depressive effect of restricted potassium supply on iron content confirmed similar results obtained with potato and which have been recently reported elsewhere (BOLLE-JONES 1953); this effect was accompanied by a much reduced chlorophyll content in the later samplings (TABLES VI, VII).

For most of the nutrients determined, with the exception of nitrogen, magnesium and boron, the choice of top or second whorl laminae for analysis was probably immaterial as there was no consistent, highly significant, whorl × treatment interaction recorded. However, the concentration of the nutrient elements vary with whorl position and the preferred choice, when leaf material is limited, is usually that whorl which contains most of the element being examined: such laminae are expected to show a larger variation of concentration as a result of treatment. In investigations on the effect of treatment on nitrogen, magnesium and boron contents it is better to discriminate clearly between laminae taken from different whorls, as the results revealed that the effect of treatment on the content of these three elements varied with whorl position.

## SUMMARY

Seedlings of *Hevea brasiliensis*, clone Tjirandji 1, were grown in pot sand cultures and were supplied with none or limited amounts of nitrogen, sulphur, phosphorus, magnesium, potassium, calcium, iron, manganese and boron. Each such treatment was found to retard or limit growth; visual symptoms for all the elements but calcium and boron were observed and described.

The concentration of rubber found in the stems and petioles increased with improved mineral status during the early part of the experiment. Later, when the effects of treatment on dry weight yield became pronounced, this effect was reduced and sometimes reversed.

The low requirement of *Hevea brasiliensis* for calcium and boron, and the accumulation of the latter in large concentrations in the lamina were shown. A deficiency of potassium or manganese increased the phosphorus content of the lamina. Nitrogen, sulphur or phosphorus deficiencies decreased the magnesium content of the lamina which was increased by potassium and calcium deficiencies. Restricted nutrient supplies of nitrogen, magnesium or calcium increased the potassium content; deficiencies of magnesium or potassium increased the calcium content. Potassium deficient laminae contained a lower iron content than those of the complete nutrient.

Evidence for an interrelationship between the nitrogen and manganese in the plant was adduced.

*I thank Mr C.G. Akhurst and Mr G. Owen for facilitating the progress of the work described above and I acknowledge the able assistance of Mr K. Ratnasingam and others of the Soils Division in the experimental operations.*

Soils Division
Rubber Research Institute of Malaya
Kuala Lumpur

June 1954

## REFERENCES

BOLLE-JONES, E.W. (1953) A study of the interrelationships of iron and potassium in the potato. *Int. Congr. Soil Sci.* (Transactions vol. II) 186

BOLLE-JONES, E.W. (1954a) Effect of mineral status on the concentration of rubber in *Hevea brasiliensis*. *Nature, Lond. 173*, 690

BOLLE-JONES, E.W. (1954b) Nutrition of *Hevea brasiliensis*. 1. Experimental methods. *J. Rubb. Res. Inst. Malaya 14*, 183

DE HAAN, I. (1950) Deficiency symptoms in *Hevea brasiliensis*. *Arch. Rubbercult. 27*, 107

HEWITT, E.J. *Sand and Water Culture Methods Used in the Study of Plant Nutrition.* Commonwealth Agricultural Bureaux, Farnham Royal, 1952

RHINES, C.E., MCGAVACK, J., and LINKE, C.J. (1952) Mineral Nutrition of *Hevea brasiliensis*. *Rubb. Age, N.Y. 70*, 467