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Pot Culture Technique for Study of Hevea Nutrition

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The difficulties involved in carrying out manuring studies on rubber, especially under the conditions now prevailing in Viet-Nam, have led to the search for a simple small-scale method to serve as a basis for the orientation of field experiments. A technique which is relatively easy to apply and which gives certain results similar to those obtained in the field is described. The possibilities of the method are discussed.

The considerable time-lapse necessarily involved in obtaining information about the manurial requirements of *Hevea brasiliensis* and the current difficulties in establishing field experiments in Viet-Nam have inspired the search for new diagnostic methods. This need has made itself particularly felt following the study of eighteen experiments which were in progress in different rubber-growing parts of the country; considerable irregularities have appeared in the growth responses and in 60% of the cases it has become very questionable whether manurial applications will be of any value.

It is worth looking for a solution to these problems in the opportunities offered by the use of pot cultures. The advantages permitted by such scale reduction are obvious: the speed with which results are obtained, ease of observation, possibility of experimenting with numerous soils in a limited area, etc. There are, however, many questions to be answered with respect to the practical application of the results. The immediate consideration is to determine how far greenhouse observations relating to growth, disease resistance and mineral composition can be extended to plants of the same age in the field; beyond this, the relationship between the nutrient requirements of plants of different ages remains to be established. In the end it is also true that the all-important aspect, that of yield, does not normally find a place among the analyses which can be carried out on pot cultures.

The object of the present study is to investigate the possibilities of growing *Hevea* in a limited volume of soil and to determine the level of its response to different nutrient treatments according to the type of soil utilised. The susceptibility of *Hevea* growing in sand culture to the deficiencies induced by subtractive technique has already been demonstrated (BOLLE-JONES, 1954). MIDDLETON (1961) also utilised the principle of subtractive technique to pot cultures in endeavouring to characterise rubber-growing soils with the aid of *Pueraria* as indicator plant. Attempts to confirm this possibility showed, however, that interpretations based on *Pueraria* are to be regarded with caution where the elements S, K, Ca and perhaps N are concerned (POLINIERE AND VAN BRANDT, 1966); in other respects the use of this legume presents no advantage whatever with regard to the provision of a criterion for the mineral status of *Hevea*. The solution to these different limitations accordingly lies in a combination of the respective contributions of Bolle-Jones and Middleton, *i.e.*, in the use of young rubber trees as indicator plants for the status of rubber-growing soils. That the results obtained are comparable with those obtained in the field provides a stimulus for a more vigorous investigation of this small-scale approach.

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MATERIAL AND METHODS

Material (Invariable Factors)

In January 1967, 34 germinated illegitimate seeds of clone PB 86 were transplanted into each of a series of plastic containers (Ø=20–25 cm, × 8 cm high) filled with 1500 g dry earth in the case of the podzolic soils (average depth = 5 cm) or 1300 g in the case of the latosols (average depth = 6 cm); 500 g gravel (average thickness 2 cm) covered the bottom of the pots. The entire collection was placed in a laterally-ventilated greenhouse and covered with sheets of plastic material.

The number of plants per pot was reduced to twenty at the time (March 1967) when the second whorl was developing, and application of nutrient solutions was started six days later.

Dusting with Lindane and Oléoparaphène was carried out to control an attack of scale insects.

Experimental Procedure (Variable Factors)

Eleven soils representing the main rubber-growing regions of Viet-Nam *(Table 1)* were watered with nine solutions, seven of which were respectively deficient in N, P, S, K, Ca, Mg, or trace elements. Each treatment was applied in triplicate.

Measurements

The heights and diameters were measured every two months until July and thereafter every month until the plants were pulled up in October. The dry weights of the various parts of the plants were then determined.

Foliar anomalies were recorded in June and October 1967 *(Table 2)* at the same time as analyses of the mineral composition of the leaves.

Manner of Interpretation and Presentation

Since data relating to growth were based on analyses of several different characteristics, the question of a choice arose when the results differed according to the part under consideration. The following criteria have been applied:

Importance of the part with respect to the practical objectives under consideration;

Reproducibility from one part to another;

Sensitivity to modifying factors, particularly from one soil to another;

Ease of measurement.

Presentation of all the observations which have been made would call for lengthy explanations and a considerable number of tables. It has seemed preferable to select the essential aspects relevant to our purpose and to illustrate them directly in the text by means of a few figures.

EXPERIMENTAL RESULTS

Effect of Different Nutrient Treatments on Different Soils (Interaction Treatment × Soil)

Control. There seems to be a lack of balance in the mineral composition of the leaves: lack of N(2.58%) and excess Mg (0.30%), Mn (80 mg%) and Fe (50 mg%). In addition major irregularities appear according to the soil in question. In some cases these seem to be related to soil fertility (Mg = 0.26% for Quan-Loi 1 compared with 0.40 for Long-Thành and 0.42 for Suzannah; Mn = 31 mg% for Long-Thành compared with 130 for Courtenay) whereas the soil content of other elements has no effect (P = 0.24% for Long-Thành compared with only 0.19 for Courtenay; Fe = 57 mg% for Trang-Bôm against only 47 for Ông-Quê).

Abnormal symptoms appear with average frequency and are distributed in a relatively uniform fashion among the different soils, except that interveinal yellowing WO is obvious only on soils rich in Mn (where it occurs more than ten times as often as elsewhere) and appears to be associated with a high Mn/Fe ratio (> 2).

The podzolic soils show poorer growth than the latosols, especially those latosols with a moderate Mn content. The difference, which for the aerial parts can be in the ratio of one to two (*e.g.*, 47 for Long Thành compared with 107 for Quan-Loi 1) seems to imply that the basic treatment which was chosen is arbitrarily better suited to some soils rather than others.

TABLE 1. PHYSICO-CHEMICAL PROPERTIES OF THE EXPERIMENTAL SOILS

Components \ Soil		Dâu-Tiêng	Long-Thành	Trang-Bôm	Quan-Loi 1	Quan-Loi 2	Tuc-Trung	An-Lôc	Ông-Quê	Binh-Ba	Suzan-nah	Cour-tenay
	Colour (Munsell)	Light brown-ish grey	Grey	Light grey	Yellowish red						Reddish brown	
pH		4.5	4.8	4.7	4.3	4.4	4.4	4.8	4.6	5.3	4.8	4.8
Particle size	Coarse sand*	26	46	38	1	1	1	1	2	1	4	7
	Fine sand*	62	31	28	3	3	4	3	4	4	5	5
	Silt*	5	6	3	9	18	9	16	12	19	18	15
	Clay*	10	19	33	74	58	80	76	74	69	66	57
C (dg)*		7	6	7	27	23	17	12	16	15	14	17
C/N ratio		23	22	14	19	16	13	13	12	14	11	14
Total N (cg)*		3	3	5	14	14	12	10	13	11	13	12
Inorg. N (mg)*		1	1	2	3	3	3	2	4	3	3	3
P (mg)*		1	3	4	24	20	9	39	55	25	103	223
Exchangeable bases at pH 7	K (m-equiv.)*	0.01	0.03	0.02	0.10	0.07	0.08	0.19	0.07	0.21	0.39	0.25
	Ca (m-equiv.)*	0.06	0.32	0.37	0.22	0.16	0.52	0.86	0.57	2.09	1.57	1.22
	Mg (m-equiv.)*	0.01	0.06	0.12	0.16	0.09	0.25	0.48	0.37	0.70	0.86	0.93
	Na (m-equiv.)*	0.02	0.04	0.08	0.08	0.07	0.09	0.10	0.11	0.05	0.19	0.10
Te (m-equiv.)*		2.43	2.84	3.39	9.78	8.89	7.71	8.30	9.48	9.49	9.67	14.26
Total Mn (mg)*		0	4	13	83	85	72	214	198	269	240	306
Total Fe (dg)*		2	6	13	139	145	117	149	136	132	99	125

*% dry soil

Treatment 'no N'. With respect to the mineral composition of the leaves, the absence of N has a relatively depressive effect on N, for all the soils except Quan-Loi 1 (−21*, on average); and on P and Mg, for most of the latosols. It has the opposite effect on P and Mg for Long-Thành; on Ca for the podzolic soils and the latosols with a high Mn content; and on K for all soils.

The *Oidium* symptoms BO and interveinal yellowings FO and WO occur to a very slight extent in this treatment: by contrast the number of completely yellow or fallen leaves is extremely high.

Although the dry weight of the aerial parts and above all of the leaves is much diminished by lack of N, that of the roots is largely unaffected (+3) and only on latosols with a low Mn content is an appreciable effect to be

*As Vi of the value for the control (%−100 with respect to a reference value, where the lower value is always expressed in terms of the one which is greater).

detected. These are, moreover, the soils which by comparison with the control show the greatest sensitivity to N deficiency (−43 for the dry weight of the whole plant on Quan-Loi 1, against −1 on Binh-Ba).

Treatment 'no P'. The lowering of the P content of the leaves is clearly marked only in the case of the podzolic soils, precisely those which had given a relatively high uptake under the control treatment; in absolute terms the levels corresponding to the different soils tend to be about the same (0.17%).

The interveinal yellowings FO and WO are particularly pronounced on the soils with a high Mn content (An-Lôc, Suzannah, Ông-Quê, Courtenay, Binh-Ba).

The rather positive growth reaction resulting from the omission of P seems to indicate that the amounts being applied were too great, at least for the podzolic soils where the uptake was very pronounced. Among the latosols poor growth is found on the Quan-Loi series, the soil P content of which is relatively low

TABLE 2. DEFINITION OF FOLIAR AND BRANCH SYMPTOMS

Designation of symptom	Location	Symptom: anomalies of				
		Dimension	Form	Texture	Colour of living tissues	Dead tissues
BO	Leaves of the uppermost whorl	0	0, or shrivelled	0, or hard	Oily patches+white powdery circles	0
CO	Leaves	0	0, or shrivelled	0	Diffuse yellow to brown spot	0, or brown tip
DO	Leaves	0	Wavy margins	Hard	Normal	0
EO	Leaves of the uppermost whorl*	Large	Shrivelled	0	Ground uniform green to yellow	Brown spots (3 mm)
FO	Leaves of the uppermost whorl	0	0	0	Interveinal bleaching, ground pale green to yellow	0
G1	Leaves	0	Asym-metrical	0	0	Greyish-white patches (10–50 mm)
G2	Leaves	0	0	0	0	General pale brown scorching
JO	Leaves	0 to small	0	0 to soft	Pale yellowish green	0
J1	Leaves	0 to small	0	0	Clear yellow to whitish yellow	0
SO	Branches	Dieback				
WO	Leaves of the uppermost whorl†	Small	Elongated	Soft	Pale yellow between veins, later entirely so	0
XO	Branches	Leaflets and leaves falling or fallen				

*In certain cases, followed by J1 †Followed by XO

(2.5% Te); no such effect appears in the case of Tuc-Trung (soil P = 1.2% Te).

Treatment 'no S'. The lack of S is clearly reflected by a considerable decrease in the S content of the leaves. Mg shows marked fluctuations in both directions according to the soil (−32 for Quan-Loi 2, +25 for Long Thành), and N, Fe and above all P show a positive response.

The foliar symptoms already noted in connection with N deficiency (yellowing JO and abscission XO of the leaves) reappear here.

All of the soils under consideration were very sensitive to the omission of S.

Treatment 'no K'. The decrease in the K content of the leaves is particularly obvious on the soils containing little Mn (Quan-Loi, Tuc-Trung); the K content of the soil seems to have no direct control over this effect since the decrease noted for Ông-Quê (the soil K content of which is very close to Quan-Loi 2) is of the same order as that for soils with a high K content (*e.g.*, Suzannah).

K deficiency seems to promote the appearance of foliar symptoms of bad drainage (GI) and also dieback (SO), but only on Dâu-Tiêng are these phenomena obvious.

Growth is affected by lack of K on most of the soils under consideration. It is curious to note that Quan-Loi 1 shows one of the clearest responses to K deficiency (dry weight of the aerial parts = −59) while Quan-Loi 2 shows no reaction whatever; this is not explicable either on the basis of the mineral composition of the soils or of that of the leaves. A further point deserving mention is that in this treatment growth of the roots is inhibited to a somewhat greater extent than that of the leaves (−17 against −14).

Treatment 'no Ca'. The decrease in the Ca content of the leaves is particularly obvious on the podzolic and Quan-Loi soils. Mg shows a variable response while on most soils the N and K levels increase; the same is true for P on three podzolic soils.

The symptoms CO (diffuse yellow to brown spotting of the leaves) are clearly obvious on the podzolic soils where the decrease in foliar Ca is accompanied by an increase in P.

In the absence of Ca growth seems to be retarded to the same extent on both the podzolic soils and the latosols. Here again, there is a lack of correlation between the degree of Ca deficiency in the leaves and the inhibition of growth: thus, for example, Binh-Ba gives aerial parts with a dry weight of 49 g (−23 by comparison with the control) and a Ca content of 0.36% (−34), whereas Courtenay yields a dry weight of 72 g (−5) at a Ca content of 0.28% (−44). It is to be noted that the N content for Courtenay is much lower than that for Binh-Ba (2.82% against 3.42).

Treatment 'no Mg'. The decrease in the Mg content of the leaves (−40) is generally accompanied by a decrease in the Ca level (−18). There is a considerable increase in K (+28 on average) irrespective of the soil.

The foliar symptoms of a general yellowing accompanied by small brown spots (EO) are evident only on the latosols: it is noteworthy that among the soils with a high Mn content it is indeed the one which contains the least Mg where the incidence is greatest (3.9% Te for Ông Quê compared with 5.7 for the next soil in the list). Interveinal yellowing (WO) is also more pronounced on the Ông-Quê soil.

In most cases omission of Mg had a beneficial influence on growth, which seems to imply that the amounts being applied were too great; only on Quan-Loi 1 is there a depressive tendency in the absence of Mg. The dry weights of the roots are affected to the same extent as those of the aerial parts.

Treatment 'no trace elements'. The leaves show a marked decrease in Mn and an increase in Fe, and the ratios K/Ca and Mg/Ca are on average lower.

Foliar symptoms of *Oidium* attack (BO) are especially pronounced, 33% of the trees being affected compared with 27% for the control.

There is a very obvious increase in the growth of the aerial parts on all soils except Quan-Loi 2.

Treatment 'no nutrients' (no manuring). The leaf contents of all the elements are diminished with the exception of Fe (+25) on all soils and K (+10) on the soils with a low Mn content.

Foliar symptoms of damage from the defoliants scattered in accordance with military demands (DO) are here clearly evident; the same is true of the yellowing (JO) preceding leaf abscission. By contrast, interveinal yellowing (FO and WO) occurs to a negligible extent. Only on the Quan-Loi soils is growth obviously less than for the control; the effect is, however, scarcely apparent in the dry weight of the roots.

Summary, with Respect to Type of Treatment

The treatments leading to the most pronounced disturbances in mineral composition are the omissions of: all nutrients; trace elements; Ca; Mg; K; S; N; P, in order of decreasing importance.

When unaccompanied by associated parasitic infections, foliar symptoms of deficiency appear mainly as follows:

High incidence of interveinal yellowing (FO and WO) on omission of P, K or Mg, and low incidence on omission of N or all nutrients.

High incidence of complete yellowing and leaf abscission (JO, XO) on omission of N, all nutrients, or S, and low incidence on omission of P.

The other manifestations are typical of the deficiency in question.

It has been seen that the incidence of *Oidium* was very obvious in the absence of trace elements, whereas the extent to which it occurred in the treatment 'no N' was less than half that for the control. The severity of defoliant damage in the unmanured treatment (nearly twice that for the control) is also to be noted.

On the whole, the omission of N; Ca or S; K; or all nutrients produced (in order of decreasing importance) the most-pronounced ill-effects. The treatments in which Mg or trace elements were omitted have a clearly beneficial effect.

Summary, with Respect to Type of Soil

The mineral composition of the leaves generally shows no particular response to any one type of soil more so than to another.

In the absence of associated parasitic attacks, foliar deficiency symptoms arise mainly as follows:

High incidence of interveinal yellowing (FO and WO) and complete yellowing (JO) on soils rich in Mn; and

High incidence of scorched patches (GI), localised diffuse yellow to brown discoloration (CO) and leaf fall (XO) on podzolic soils.

When parasitic infections occur at the same time there is:

Widespread yellowing of the leaves and brown spot formation (EO) and *Oidium* attack (BO) on soils rich in Mn; and

High incidence of dieback on podzolic soils.

Damage due to defoliants was clearly greater on the latosols than on the podzolic soils.

Growth on the podzolic soils was in general poor, whereas the latosols with a low Mn content gave excellent results.

Summary, with Respect to Different Criteria

Arranged in order of decreasing significance, the mineral constituents of the leaves which have the greatest diagnostic value are:

Mn, Ca, K, Mg, Fe, S, N, P, for differences between the treatments; and

Mn, Mg, Ca, K, Fe, S, P, N, for differences between the soils.

The foliar symptoms which allow the most satisfactory distinctions are (in order of diminishing importance):

Leaf abscission (XO), complete yellowing (JO), interveinal yellowing (FO and WO), yellowing with brown spots (EO), etc., for differences between the treatments; and

Interveinal yellowing (WO), yellowing with brown spots (EO), dieback (SO), etc., for differences between the soils.

With respect to growth, the dry weight of the aerial parts and above all that of the fraction 'leaves' seems to comply well with the criteria listed earlier (see *page 251*) for distinguishing between treatments as well as between soils. It is noteworthy that variation in the dry weight of the roots does not always correspond to changes in the aerial parts; the roots re-

act to K deficiency to a much greater extent than to lack of N, whereas the aerial parts show the opposite effect. There is accordingly an absence of distinctive response on the part of the dry weight of the whole plant.

It is to be noted that measurements of height and especially diameter are generally of low sensitivity (−20 and −10, respectively, for the most serious single deficiency, lack of N). A worsening of the effects of deficiency between July and October is furthermore reflected in the height measurements only for the treatment 'no Mg', whereas the dry weights of leaf samples taken for foliar analysis indicate a worsening of the position in the majority of the treatments.

DISCUSSION AND CONCLUSIONS

Present Position

Experimental design. The method which has been employed (about 1500 g soil per pot, selection of twenty illegitimate seedlings, etc.) seems to assure very satisfactory sensitivity and reproducibility. The few irregularities noted may be largely attributed to:

Inadequate success in the control of scale insects; and

Unsatisfactory choice of manurial treatment.

The later conclusion is based on the following considerations:

Lack of balance in the control (complete manurial treatment) with respect to N, Mg, Mn, Fe and perhaps P;

Inequalities in the growth of the controls on different soils: preliminary experiments suggest that uniformity (at optimal growth levels) should be obtainable with a really effective treatment; and

Absence of a response or even a positive reaction when P, Mg or trace elements are omitted.

A study of the treatment best suited to individual soils is clearly necessary before the small-scale trials can be pursued further.

Measurements. Examination of the mineral composition of the leaves has contributed to the understanding of the observed phenomena to only a very limited extent. By contrast, the study of foliar symptoms proved to be the best way of characterising the different soils. Where growth is concerned, measurements of height and diameter seem to be of little value: the dry weights of the aerial parts and above all of the leaves are much to be preferred, although it is accepted that other parts are at least equally responsive to specific deficiencies.

In future it will be appropriate to consider particular parts according to the nature of the deficiency, *e.g.*, the roots in the event of cationic deficiencies. The problem of studying the progressive development of a deficient condition could be approached by periodic uprooting of plants (when the plants have to be thinned because of overcrowding) or by taking samples of the entire foliage.

Significance of the small-scale approach. There are a number of aspects which seem to be in agreement with what is already known from fundamental research, field experiments, and plantation observations.

(a) The small-scale studies show a good reaction to the treatments applied to the Quan-Loi soils. It happens that in our field experiments the soils of this region were also found to give the best responses with respect to both growth and yield.

(b) The improved resistance to *Oidium* noted for the N-deficient plants is in accordance with general information concerning the behaviour of the fungus. The very great susceptibility on the Mn-rich soils from the Long-Khanh region moreover appears to confirm previous plantation experience.

(c) The difficulty of identifying mineral disorders merely on the basis of the mineral composition of the leaves applies here also. On the other hand, the damaging effects of Mn so frequently noted in the Long-Khanh district are confirmed by this investigation: there even appear to be possibilities of remedying it, in view of the associated diagnostic feature of an Fe imbalance detectable, in particular, by visual symptoms on the foliage.

Among the results apparently at variance with available information it is necessary to note the low P level in leaves from the Cour-

tenay and Ông-Quê soils, whereas analyses of leaves from mature plantings in these regions show a P content in excess of 0.3%. This aspect requires investigation to establish, in the first instance, if it has to be attributed to the factor 'pots' or to the factor 'age of plant'.

Incidental Observations

The object of the study being to explore the possibilities of scale-reduction rather than exploitation of the results obtained it hardly seems worthwhile to return in detail to the many observations already noted. A few points of interest can however be recalled in passing:

(a) Contrary to what has been implied elsewhere (INSTITUT DES RECHERCHES SUR LE CAOUTCHOUC AU VIET-NAM, 1968) the recommended complete treatment was under certain conditions less suitable than the subtractive treatments.

(b) It remains to be verified in the field whether modified applications of the different elements will permit establishment of a minor N-deficiency and a consequent improvement in the resistance to *Oidium*, without there being any undesirable effect on growth.

(c) The availability of S seems to play an important part in the growth of young rubber trees.

(d) The extent to which a given deficiency is reflected within the plant is correlated to only a slight extent with either visual symptoms on the foliage or with growth. Consideration of the proportions of different elements (N/Ca, N/Fe, Mn/Fe etc.), already studied by BEAUFILS (1961), sometimes permits a better appreciation of the function 'soil'.

(e) Use of the same mineral standard for different soils leads to serious difficulties in interpretation. It will accordingly be appropriate to consider, for each of the major soil groups of Viet-Nam, the definition of specific reference systems to which the leaf analyses correspond.

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DISCUSSION

Chairman: Dr. S. K. Ng

Dr. Poliniere, in replying to points raised by Mr. E. Bellis and Dr. M.M. Guha on the relationship between the results of small-scale experiments, foliar and soil analyses and field practice, confirmed that the soil used in several of the pot tests had come from the sites of manurial field experiments. Pot and field experiments gave similar indications of nutrient requirements, except for calcium and magnesium. In one area the leaves showed symptoms of acute iron deficiency despite the high foliar content of iron found on analysis. This was traced eventually to excess manganese in the leaf as a result of applying a fertiliser of high manganese content to a manganese-rich soil. It was such application which had induced iron-deficiency symptoms, illustrating the need for caution in interpretation of data from foliar analysis.