Q. Jl. Rubb. Res. Inst. Ceylon (1971) 48, 82—91.

# SULPHUR IN THE CONTROL OF WHITE ROOT DISEASE

By

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## INTRODUCTION

Control of White Root disease is a problem in young clearings. Although the importance of the food base and the necessity of good sanitation in the eradication of the root disease is clearly understood by planters, the expenses involved in obtaining a clearing free of food bases often militate against the adoption of this practice.

Peries (1965) suggested the amendment of the surface soil of planting holes with sulphur, for small scale trials in estates, based on encouraging results obtained in pot experiments and a field trial initiated in 1964. An outside estate had reported apparent total cure of White Root disease by incorporating sulphur around the affected roots and a preliminary trial was carried out (Peries, 1956). Although the application of sulphur for treatment of soil in the control of soil-borne diseases had been in practice since 1891, its use in soil treatment for plantation crops in Ceylon or elsewhere had not been reported prior to 1956.

Today a number of estates use sulphur to amend the surface soil of planting holes, at the time of planting, as a precautionary measure to control White Root disease. This is carried out either on a large scale or is confined to areas where *Fomes* had been prevalent in the old stand. However, estates do report the occurrence of *Fomes* patches in treated areas. Therefore, it was important to investigate, in some detail, the effect of sulphur on the control of *Fomes lignosus*. Observations made in the laboratory and in the field, for the past one year, are presented in this paper.

## EXPERIMENTAL

*Effect of sulphur on* F. lignosus: *F. lignosus* failed to grow on the surface of culture media dusted with sulphur but grew within the medium. Sterile soil amended with sulphur failed to support the growth of the fungus. Growth on the surface of amended soil was negligible compared to the growth on the untreated soil (Fig. 1). The fungus failed to grow from artificially inoculated pieces of wood (1·5 cm cubes) buried for a month in unsterile soil, treated with sulphur, when removed and incubated in moist chambers.

These results indicate that sulphur is toxic to the fungus and that it would not grow on substrates treated with sulphur.

*Effect of soil moisture on growth of* F. lignosus: Two types of soil were collected from Dartonfield. One was taken from a clean weeded area (soil A) and the other from an area under leguminous cover crops (soil B). These soils were amended with sulphur and adjusted to 20 and 50% moisture-holding capacities (MHC). Untreated soil served as controls. Artificially inoculated wood cubes (1·5 $cm^3$) were placed at the bottom of bottles and filled with soils given various treatments. Moisture levels were maintained during the period of incubation of 12 weeks.

Growth occurred within a week in the treated and untreated soils adjusted to 50% MHC. Rhizomorphs were thick and white — these turned slightly brownish but remained viable till the end of 12 weeks. Rhizomorphs lysed off in the soils treated with sulphur within three weeks of initiation, dissolution of strands being more rapid and complete in the soil B, taken from an area under cover, than soil A, collected from a clean-weeded area. The former soil had a higher organic matter content than the latter. The difference in pH of treated and untreated dry soil was 0·2 and 0·5 for soil A and soil B, respectively.

The pH of dry soils did not change to any marked degree with the addition of sulphur, but the fungus was inhibited on both soils adjusted to 20% MHC. The fungus grew in the untreated dry soil of the B series, but rhizomorphs were absent and mycelial strands remained viable till the end of the incubation period. The fungus did not grow in the untreated soil of A series, but when the MHC was raised to 50%, growth commenced from half the inoculum pieces and vigorous rhizomorphs were produced.

These results show that the moisture content of the soil is an important factor in the initiation of rhizomorphs. Organic matter content of the soil too would contribute to the lysis of rhizomorphs when soils are amended with sulphur. Inhibition of the fungus in the sulphur-treated dry soils cannot be fully attributed to change in reaction.

*Colonisation of wood pieces by other fungi at the end of 12 weeks:* The wood cubes used as inoculum for the previous experiment were washed, surface-sterilised and plated on malt agar. The percentage of wood pieces colonised by other fungi in each treatment is given in Table 1.

TABLE 1

PERCENTAGE OF WOOD PIECES COLONISED BY OTHER FUNGI

| Treatments | MHC | |
|---|---|---|
| | 50% | 20% |
| Soil A | 65 | 100 |
| Soil B | 57·5 | 92·5 |
| Soil A + sulphur | 30·5 | 76·2 |
| Soil B + sulphur | 35·0 | 100 |

The fungi that colonised the wood pieces were mainly *Trichoderma* spp., followed by *Penicillium* spp., *Aspergillus* spp. and a few unidentified fungi.

There was no significant difference in species that colonised wood from treated and untreated soils. In the sulphur-treated moist soils, the percentage of pieces colonised was less compared to untreated soils.

*Survival of* F. lignosus *on wood:* The survival of *F. lignosus* was studied in soils adjusted to different moisture-holding capacities and treated either with sulphur or lime. Untreated soil served as the control. Wood pieces (1 × 1 × 5 cm) were artificially inoculated and buried in soil in the different treatments. Moisture levels were maintained throughout the incubation period. At the end of six months all the pieces of wood were removed and incubated in moist chambers.

TABLE 2

PERCENTAGE OF WOOD PIECES IN WHICH *F. LIGNOSUS* REMAINED VIABLE AT THE END OF SIX MONTHS

| MHC | Control | Sulphur | Lime |
|---|---|---|---|
| 20% | 85 | 0 | 100 |
| 50% | 7 | 0 | 80 |
| 70% | 38 | 0 | 63 |

*F. lignosus* was not viable at all three moisture levels tested in sulphur-treated soils. It was viable at all moisture levels, but at varying degrees in soils treated with lime. In untreated soil, it survived better in the dry soil followed by wet, but survival was negligible in moist soils.

This indicates that *F. lignosus* will survive better in wood in dry soils, in water-logged soils and in soils treated with lime. Treatment of soil whether dry, moist or wet, with sulphur appears to kill the inoculum effectively in wood pieces of small volume.

*Survival of* F. lignosus *in naturally infected roots with bark intact:* Naturally infected roots ¼. ½, ¾ and 1 in. diameter, with bark intact were buried in soils treated with sulphur and roots of the same diameters, buried in untreated soil served as controls. Pots in which the roots were buried were left out in the open. At the end of six months the roots were removed and incubated in moist chambers.

Rhizomorphs on the surface of sulphur-treated soil were not viable in most cases. The fungus remained viable within bark covered roots of all diameters and grew from within the wood in sulphur-treated soil. The fungus was viable in the roots of large diameter but not in those of ¼ and ½ in. diameter which were buried in untreated soils.

## DISCUSSION

From the observations of the foregoing experiments, it is evident that sulphur is toxic to *F. lignosus* in pure culture and even in soil it appears to have a fungicidal effect on the fungus. Sulphur, when added to soil, could be toxic *per se*, or it could combine with other substances present in the soil and these newly-formed products could be toxic or micro-organisms such as bacteria, could act on these products to release substances lethal to the pathogen.

The reaction of dry soil did not change markedly when treated with sulphur but the fungus was inhibited in such soils too. Inhibition of *F. lignosus* cannot be attributed in all cases to decreases in pH levels. Muncie *et al.* (1944) observed the same in their studies on potato scab and suspected that free sulphur may be responsible for the reduced infection. Roach *et al.* (1925) and Crowther *et al.* (1927) could not attribute the action of sulphur to the simple function of the final soil reaction and attributed reduction of wart disease to some product of sulphur other than sulphuric acid. Hooker & Peterson (1952) show evidence which suggests that the potato scab organism is inhibited by $H_2S$ gas produced in sulphur-treated soils and this effect is distinct from the influence of H-ion concentration.

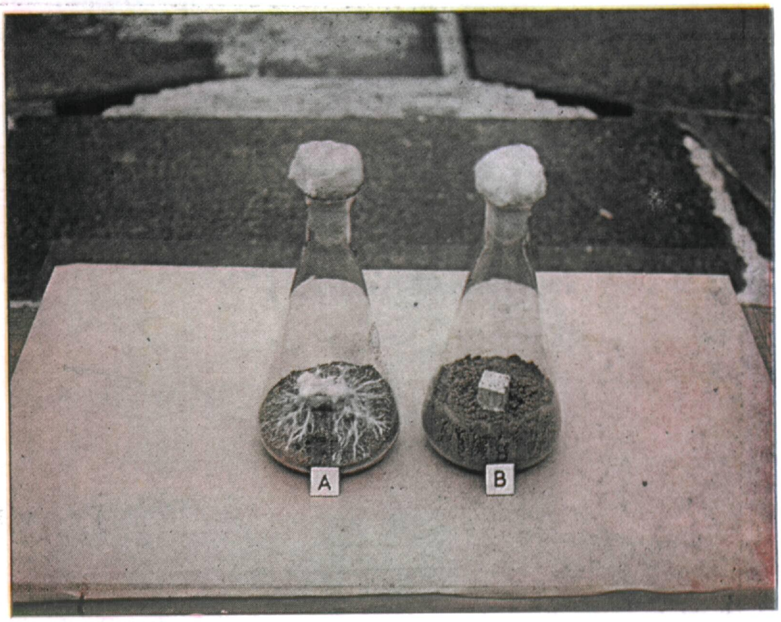


Fig. 1. Growth of the fungus *Fomes lignosus* in sterile unamended soil (A) and sterile soil amended with sulphur (B).

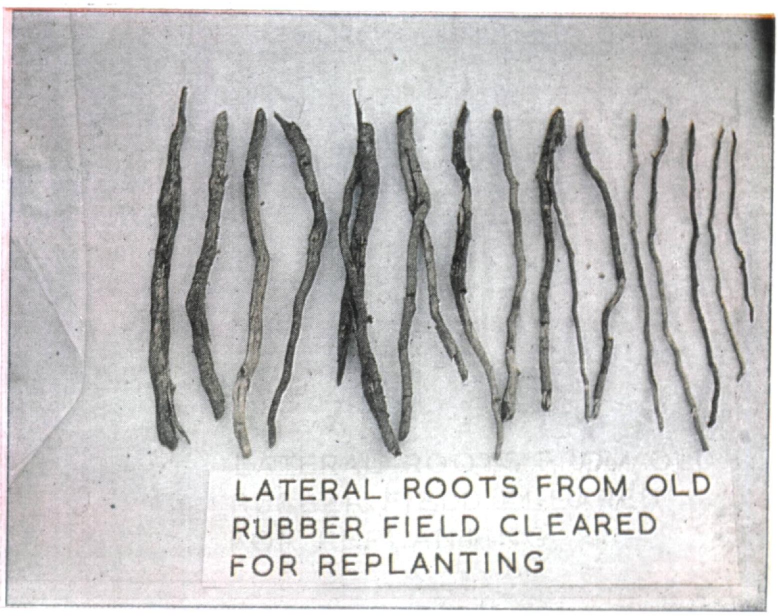


Fig. 2.

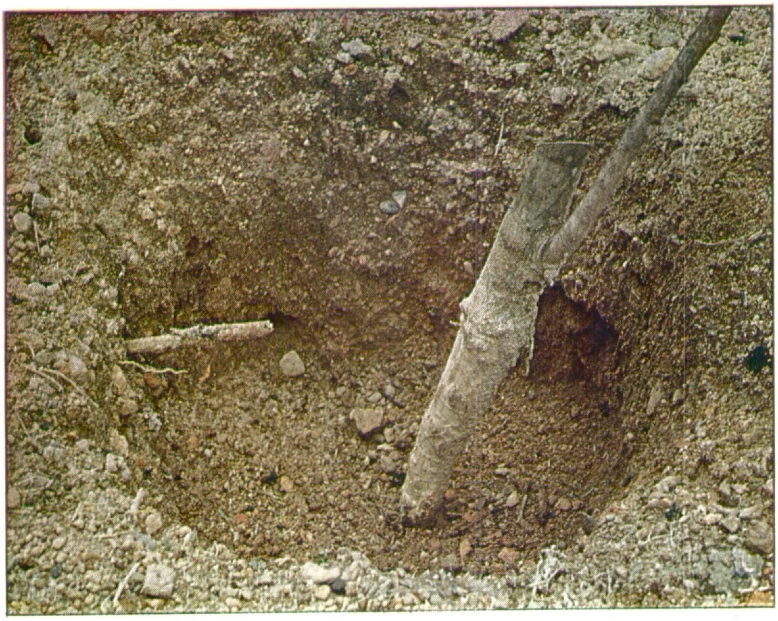

Fig. 3. Size of the food base that had caused infection of a one-year old budding by being in contact with it in the planting hole.


Fig. 4. Food base from three-year clearing

Lysis of rhizomorphs in sulphur-treated soil could be due to the toxic substances produced in them or due to the action of micro-organisms other than fungi. No correlation could be found between the occurrence of antagonistic fungi and inhibition of *F. lignosus* in the treated soils. Lysis was rapid and complete in soils of higher organic matter content and these had a higher population of bacteria than soil A.

Soil treatment with sulphur for the control of *F. lignosus* appears to be effective at all moisture levels tested.

As shown earlier, whatever the action of sulphur, it does not penetrate the bark and the fungus would survive longer within wood protected by bark in treated soils. This would appear to be a disadvantage but if the fungus is kept in check, the substrate may gradually disintegrate and the plant due to its growth would gradually become more resistant and the inoculum potential would at the same time become smaller, so that the plant would survive. *F. lignosus* in its relation to *Hevea* is essentially a weak parasite (Fox, 1961) and with increase in age of the tree, massive inocula would be required to cause serious infection of it.

The results of the field experiment initiated in 1964 (Peries, 1965) clearly indicate that sulphur amendment of surface soil of planting holes significantly reduces the incidence of White Root rot in the first two years and from the third year onwards the incidence remains more or less the same in sulphur-treated and control plots. In order to understand these results and the occurrence of *Fomes* patches in sulphur-treated fields it is necessary to look into the methods of clearing practised in Ceylon.

*Method 1:* Felling and clearing by hand or with the help of an elephant. In a field cleared by this method the tap root and the laterals will be left behind in the soil. This would mean that in areas with heavy incidence of *Fomes* the diseased roots will remain in the soil and it is not advisable to adopt this method to clear such areas.

*Method 2:* Felling by the use of a monkey grubber and clearing by hand or with the help of an elephant. When this method is adopted to clear an old stand the stumps are grubbed along with the tap root and most of the laterals, and are burnt. The material left buried in the undisturbed soil would be the small lateral roots of varying lengths (Fig. 2). Large laterals and tap roots of most diseased trees may not come out intact when grubbed and the chances are that some of these too would remain buried in the soil.

The food base is an important factor in root diseases. Any piece of plant material that harbours the fungus and is of a size that could cause an infection of a living root would be a likely food base. The size of the food base that is required to cause an infection is dependent on the age of the plant or tree. John (1966) reported that the volume of inoculum required to cause a successful infection on a young plant is smaller than that required to infect a mature tree. Then in the case of one and two-year old plants succumbing to *Fomes* root rot, the infection would be mostly caused by food bases of small volume which are abundant in the field and as disease initiation is by contact of healthy roots with the food bases, those food bases in and around the planting holes would be the cause of infection of rubber plants in the early years of a clearing (Fig. 3).

*Fomes lignosus* is capable of growing through the soil for short distances from food bases. Bancroft (1912) observed that under damp conditions *F. lignosus* could spread out freely into heavy soils for nearly a foot from infected roots. An infection could be initiated from food bases lying close to the tap root of very young plants. By treating the surface soil of planting holes with sulphur, at the time of planting, and again when the plants are about an year old, such infections are obviated by inhibiting the growth of the fungus in treated soils; if the fungus did grow the rhizomorphs would lyse off and hence the causal agent of White Root rot is held in check.

John (1960) reported that the fungus lost its viability in roots of 2 in. diameter and less in two years. In a new clearing most of the available food bases would be of this category and by the third year these would have completely disintegrated. Thus treatment of soil of planting holes with sulphur holds the fungus in check in food bases of small volume during the critical period after which these inocula loose their potential for infection, by the disintegration of the substrate.

When *Hevea* plants are about three years old the roots would have grown out of the planting holes and at this time (*i.e.* after three years) what would remain in the field as food bases would be roots of larger volume (Fig. 4). After the second year, the addition of sulphur to the planting holes or surface application of sulphur to the field will not be of any advantage; excess sulphur may affect the nutrition of plants, delay the decomposition of the larger food bases scattered in the field, and will not have the desired effect on the incidence of disease. In addition it would increase the costs of disease control.

From the third year onwards regular inspection of the field for foliar symptoms, correct treatment of *Fomes* infected trees *i.e.* excision of infected roots and tissues, removal of food bases and disposal of all diseased plant material by burning and collar inspection of trees adjacent to diseased trees should be carefully carried out to prevent *Fomes* patches getting established in young clearings and mature stands.

The following methods have been suggested for the eradication of *Fomes lignosus* in young clearings :-

1. Removal of all roots large and small manually;

2. To allow the planting site to lie fallow for a period of two years in order that the small laterals may decompose within that period;

3. Mechanical means of clearing by disc ploughing with a tractor and removal of all the small roots;

4. Tree poisoning as practised in Malaya;

5. Sulphur treatment of surface soil of planting holes at the time of planting and again when the plants are about an year old.

The first method is expensive and would add immensely to the cost of replanting. The second adds two years to the normal six-year immature period of rubber, making it eight years before the field is brought into tapping. It is reported that root disease is negligible when an old stand is cleared by mechanical means (Newsam, 1967). This method cannot be practised on all terrain in Ceylon. Young (1964) discouraged the practice of tree poisoning with sodium arsenite in Ceylon as it was not an effective method. However, the Rubber Research Institute of Malaya reports that tree poisoning with 2, 4, 5 - trichlorophenoxy acetic acid (2, 4, 5 - T) is effective in reducing the incidence of White Root disease.

Compared to all the above methods it would appear that the use of sulphur in the control of *Fomes* root disease in young clearings is cheap, effective and could be practised in all terrain, provided the old stand in which *Fomes* root rot is prevalent is cleared by the method advocated and discussed earlier in this paper.

ACKNOWLEDGEMENT

We thank Dr. R. Satchuthananthavale, Botanist, for the helpful suggestions and for the preparation of the slides presented with this paper.

REFERENCES

Bancroft, K. (1912). A root disease of para rubber tree (*Fomes semitostus* Berk). *Dept. Agr. Federated Malay States Bull.* **13,** 30; cited from Fox, 1965.

Crowther, E. M., Glynne, M. D. and Roach, W. A. (1927). Sulphur treatment of soil and control of wart disease of potatoes in pot experiments. *Ann. appl. Biol.* **14,** 422—427.

Fox, R. A. (1961). White Root disease of *Hevea brasiliensis*—recent developments in control techniques. *Rep. 6th Commonw. Mycol. Conf. 1960,* 97—100.

Fox, R. A. (1965). The role of biological eradication in root disease control in replantings of *Hevea brasiliensis.* In *Ecology of Soil-borne Plant Pathogens.* ed. K. F. Baker and W. C. Snyder, London, John Murray, 348—362.

Hooker, W. J. and Peterson, L. E. (1952). Sulphur soil treatment for control of sweet potato soil rot incited by *Streptomyces ipomoea. Phytopathology* **42,** 583—591.

John, K. P. (1960). Loss of viability of three parasites in infected root sections buried in soil. *J. Rubb. Res. Inst. Malaya* **16,** 173—177.

John, K. P. (1966). Effect of inoculum size and age of trees on root disease infection of *Hevea brasiliensis. J. Rubb. Res. Inst. Malaya* **19,** 226—230.

Muncie, J. H., Morre, H. C., Tyson, J. and Wheeler, E. J. (1944). The effect of sulphur and acid fertilizer on incidence of potato scab. *Amer. Potato Jour.* **21,** 293—304.

Roach, W. A., Elynne, M. D., Brierly, W. B. and Crowther, E. M. (1925). Experiments on control of wart disease of potatoes by soil treatment with particular reference to the use of sulphur. *Ann. appl. Biol.* **12,** 152—190.

Newsam, A. (1967). Clearing methods and root disease control. *Plrs' Bull. Rubb. Res. Inst. Malaya* No. 92, 176—182.

Peries, O. S. (1956). Report of the Mycology Department for the year 1955. *Rep. Rubb. Res. Bd. Ceylon* 1955, 80—81.

Peries, O. S. (1965). Review of the Plant Pathology Division. *A. Rev. Rubb. Res. Inst. Ceylon* 1964, 60—61.

QUESTIONS AND ANSWERS

Question: Will not sulphur affect the acid status of the soil? (Mr. M. N. Sadanandan).

Answer: Acidity increases with the addition of sulphur to the soil. This does not remain constant. Gradually the reaction changes to the original pH.

Question: (*a*) Has not the addition of sulphur to the planting hole been found to cause subsequently a malformation and dieback of the apical shoot. (*b*) Is this condition not caused by irregularities of nutrient uptake which results in toxic levels of certain elements being built up in the plant? (Mr. M. Forster).

Answer: We have not noticed any such effects in our experimental areas. The correct method of amendment of soil is to sprinkle ½ lb of sulphur on the surface soil of the planting hole (2 ft square) and lightly fork it in. (*b*) If sulphur is put into the alavangoe hole and the stump planted in it then the plant would be scorched by being in direct contact with sulphur.

Question: In 1966 clearing of RRIC 45 and 1967 clearing of RRIM 623 the uprooting was done by a monkey grubber. Any cases where *Fomes* was observed on the old stand was cleaned out as far as practicable and funds permitted. In the 1966 clearing a ½ lb of sulphur was mixed with the soil, and filled into the planting hole, whilst in the 1967 clearing the ½ lb of sulphur was merely sprinkled around the graft. In addition to this with each successive dusting season during wintering, one round of dusting to the ground at 12 lb/acre was given. Now in their 4th year and 3rd years respectively there are about two to three cases of *Fomes* per acre in the 1966 clearing and about eight cases per acre in the 1967 clearing. What do I do now ? (Anon).

Answer: Sulphur amendment of surface soil of planting holes does not protect the plants once the roots start to grow out of the planting holes. Any infection that results after the second year of planting would be caused by roots coming in contact with larger food bases lying outside the planting holes. When a tree is detected by foliage symptoms to be infected by *Fomes lignosus*, it is important to uproot the dead tree and collar examine the trees, on either side along the row until a healthy tree is detected. All the diseased trees should be treated and infected plant material burnt. This will prevent the establishment of *Fomes* patches. PCNB-based collar protectants could be used to paint the collar and 9 - 12″ of the lateral roots of all the treated trees and a few healthy trees along the row so that infection would not recur or new infections set in for another two years. Dusting the ground with sulphur or addition of sulphur to the planting holes after the first year will not give any control of White Root disease. This practice is uneconomical and should not be resorted to.

Question: Would it not be a step in the correct direction to direct experiments with a view to finding a fungicide toxic to the fungus *Fomes lignosus* and to use same to soak the affected soil in the area round the affected plant to exterminate the fungus. This should immensely reduce costs of treatment and be more convenient in addition. Single treatment of one plant sometimes costs as much as Rs. 10/- presently. (Mr. C. N. M. Rodrigo).

Answer: Most of the fungicides are expensive and in treatment of a root disease, it is important to eradicate the food base.

Question: You mention the application of sulphur to two-year old plants. What is the best method of its application? (Anon).

Answer: We never mentioned any application of sulphur to two-year old plants. Sulphur should not be used for the control of *Fomes* root disease after the second application, that is when the plants are about an year old.

Question: Would adequate *Fomes* control be achieved in the mature area, if the food base and main root is destroyed and sulphur broadcast, leaving all the laterals in the ground? (Anon).

Answer: This will not give adequate control. Large laterals too should be removed. Trees on either side of the diseased trees along the rows should be collar examined and treated, if rhizomorphs are present on the roots, and all diseased plant material should be burnt. Addition of sulphur in any manner after the first year of planting is of no consequence in the control of White Root disease.

Question: Is the use of sulphur advantageous in the treatment of *Fomes* in mature plants? Also (*a*) would you advocate that the treated area be kept clear of covers etc.? (*b*) rate of application of sulphur per sq ft, (*c*) frequency of application of sulphur. (Anon).

Answer: No, not necessarily; but the covers should generally be kept well away from the base of the plant.

Question: (*a*) What do you mean by the treatment of soil with sulphur (*b*) Is it just mixing the soil with sulphur, (*c*) How much sulphur would you suggest for a hole of size 2′ × 2′ × 3′? (Anon).

Answer: (*a*) Addition of sulphur to the surface soil of planting holes.

(*b*) Sprinkle sulphur on the surface and lightly fork it in. There is no need to mix the entire soil in the planting hole thoroughly with sulphur. (*c*) 4 oz of sulphur.

Question: Won't there be any sulphur contamination in the latex if sulphur is used in the control of root diseases? (Anon).

Answer: Sulphur is used only in the very early years of the plant.

Question: Comment on the use of Tillex for treatment of *Fomes* affected trees? (Anon).

Answer: No comment is necessary as there is just no place for Tillex in the treatment of *Fomes*. This matter was discussed fully at the conference held in 1965. (Dr. O. S. Peries).

The question asked regarding Tillex was asked by me, as I did not know that this question was discussed in 1965 as I was attending school at that time. (Mr. M. Hapugoda).