THE PHYSIOLOGY OF PARASITISM OF
ROSELLINIA PEPO

by

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# CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>I  INTRODUCTION</td>
<td>1</td>
</tr>
<tr>
<td>II  HISTORICAL</td>
<td>6</td>
</tr>
<tr>
<td>III  MATERIALS AND METHODS</td>
<td>16</td>
</tr>
<tr>
<td>IV  DISCUSSION</td>
<td>71</td>
</tr>
<tr>
<td>V  SUMMARY</td>
<td>75</td>
</tr>
<tr>
<td>VI  REFERENCES</td>
<td>76</td>
</tr>
</tbody>
</table>


(1931) Report on the Department of Agriculture St. Lucia, 1930.

(1932) Report on the Agricultural Department St. Lucia, 1931.

(1934) Report on the Department of Agriculture St. Lucia, 1933.


I. INTRODUCTION.

The object of the following investigation, which is a continuation of the work of Toovey (1935), was to assess the part played by environmental factors, in determining the incidence and spread of the root disease caused by *Rosellinia Pepo*. This disease was studied together with root diseases caused by other species of *Rosellinia* under local conditions on three islands in the Lesser Antilles; - Trinidad, Grenada and St. Vincent, in order to determine any common features present. Nowell (1918), records the following five species in the West Indies, the first three of which he associates with root diseases of cacao and other cultivated plants:


The present investigation deals primarily with *Rosellinia Pepo* Pat., a species originally described by Patouillard (1908) from material on the bark of *Hymenaea Courbaril* collected by Duss in Guadeloupe. The perithecial stage has been recorded in the British Antilles from Dominica, St. Lucia, Grenada and Trinidad and the disease is also established in Jamaica, Porto Rico and Martinique. Cacao is recorded as its most important host although many other crop plants which are planted on land recently cleared from forest can also be susceptible. Temporary shade plants which have been planted to replace cacao trees killed by the fungus, such as dasheen, banana, pigeon pea, horse bean (*Canavalia*) and cassava may all be attacked. Sugar-cane is noted to possess considerable resistance and coconut appears entirely immune.

In the absence of the perfect stage, the fungus is distinguished/
distinguished in the field by the production of fans or stars of white mycelium in the region of the cambium between the bark and the wood of the roots. *Rosellinia paraguayensis* Starb. has a somewhat similar appearance but a much less vigorous growth.

Attention may be drawn here to the difficulty of naming species of Rosellinia by mycelial characters alone in the absence of the perfect stage. The present writer was unable to find the perfect stage of *Rosellinia Pepo* on Cacao and Nutmeg although a search was made in both Trinidad and Grenada. The perfect stage of *Rosellinia bunodes* was however readily found in Grenada on *Hibiscus mutabilis* and on Grapefruit, *Citrus paradisi*, a new host record for this species.

It would appear that a rarely produced perithecial stage is a specific character of *Rosellinia Pepo* and this coupled with its parasitic tendencies should serve to distinguish it from *Rosellinia paraguayensis*. The latter species has been observed by the writer in Grenada fruiting freely and behaving as a saprophyte under environmental conditions identical with those in which *Rosellinia Pepo* in the mycelial stage was found as a parasite.

In the opinion of the writer *Rosellinia Pepo* is the species responsible for *Rosellinia* root diseases of Cacao and Nutmeg in the West Indies. The reasons for this view will be given in a later part of this dissertation.

It will be observed in the following historical summary that our knowledge concerning the parasitism of *Rosellinia Pepo* has in the past been based on field observations and that no isolation of the fungus and experimental proof of its parasitism has been obtained. Proof of parasitism within the genus has however been obtained by Nattrass (1926) using cultures of *Rosellinia necatrix* (Hart.) Berl. the causative agent of white root/
root rot of fruit trees in England, by Gadd (1928) who used pure cultures of *Hosellinia arcuata* to successfully inoculate tea seedlings in Ceylon, and by Thomas, Hansen and Thomas (1934) who demonstrated the pathogenicity of *Hosellinia neoatrix* on small apple and pear trees in California.

Further reference will be made to these species as well as to other parasitic species of the genus *Hosellinia* occurring in both Tropical and Temperate countries as there is evidence that these *Hosellinia* diseases have a common method of spread in the fields by means of mycelial strands. Wilson (1922) emphasises the important bearing of this character on the type of disease produced by these species, in his following description:— "Long strands or filaments made up of a number of longitudinally arranged hyphae are produced which not only penetrate the stem and root of the diseased plant, but spread through the soil and over the substratum and bring about the infection of other plants in the vicinity.

The serious nature of the diseases produced depends largely on this character, for distribution by means of the mycelial strands is rapid under suitable conditions, and it is a difficult matter to exterminate the fungus in the soil when the latter has become thoroughly infected." It was appreciated after a preliminary survey of the literature on *Hosellinia* root diseases that a study of the physiology of parasitism involves three main considerations, the pathogenicity of the parasite, the susceptibility of the host and the environmental factors predisposing towards infection and favouring the progress of the parasite. The relative pathogenicity of the parasite and susceptibility of the host can not readily be distinguished one from the other in disease but are largely influenced by environmental conditions.

In the past, the environment in the opinion of the practical/
practical planter has been considered all important, to the exclusion of the parasite, and according to Butler (1952) has been overstressed. The following is quoted from his article in Nature. "The practical man is often slow to admit that a destructive disease in a plant is due to agencies outside his control. Confronted with such he is inclined to seek for explanations other than the true one. He looks first for some disorder brought about by cultivation or inbreeding or meteorological phenomena. Or he thinks that the soil is unsuitable or has become exhausted or that the plant, if an exotic, has failed to become acclimatised. It is often not until all these have been tested and found wanting, that the true cause is fully realised. Experience has shown that it is unfortunately rare to find the explanation of serious disease in these directions and the dominating factor is usually the presence of a parasite however much its activities may be favoured by secondary causes."

On the other hand, the tendency on the part of pathologists has been to devote all their attention to the parasite with only a passing interest in the environment. This is particularly true of root diseases, which are the least satisfactory of any to investigate. Experimental research is beset with difficulties particularly when the attempt is made to interpret cause and effect.

We thus find two schools of thought with regard to root diseases and their control. The one sponsored by Nowell (1916) regards the parasite as all important and recommends removal or quarantine of the fungus to remove the disease. According to his authority, "in the case of Rosellinia disease of limes the conditions which best suit the tree best suit the fungus also, with the result that the finest trees are most liable to attack and are most frequently killed."
The other advocated by Napper (1932), de Jong (1933) and Briton-Jones (1934) regards the problem as fundamentally an ecological one.

The two different attitudes are of some importance in economic plant pathology where the efficacy of the control measures adopted depends on the degree of accuracy of the interpretation of the disease complex.
According to Howell (1923), the root diseases which occur in the West Indies where forest land has been opened up for agriculture are caused mainly by fungi belonging to the genus Rosellinia. The origin of infection is stated to be from buried roots, logs and other debris remaining from the forest and the disease is said to disappear with the decay of this material when cultivations of an open type, as in those of arable crops, which admit sun and wind to the soil, are adopted.

"The fungi can persist in cultivations such as that of cacao in which the conditions, especially where shade trees are abundant approach to those of the forest in respect of shade and humidity. They also occur in windbreaks and hedges of certain susceptible trees and shrubs in wet or sheltered districts. The causative fungi infect shaded soil rich in decaying vegetable matter and spread slowly through it destroying every plant with which they come in contact".

Two types of infection are recognised, one occurring in new clearings and the other in established plantations. In the former type an association between the diseased trees and forest stumps is recorded, as many as five or six trees being traced to the spreading roots of a single large stump. The author at the same time points out that species of Rosellinia are far from common on dead logs, branches or decaying stumps and that observations show that when the period of heavy losses ensues most of the trees are infected from one another distributing the disease in characteristic patches the number of which is small when compared with the number of stumps and logs in the clearing.

Nowell attributes this to a limited number of species of forest trees of which the stumps can be readily infected by the/
the fungus. In Dominica the largest number of cases are said to occur in connection with Chataigniers (Sloanea spp.), while mahoe cochon (Sterculia caribaea), mahoe piment (Daphnopsis tinifolia) and bois cabrit or goat wood (Algiphila martinicensis) are also recorded as susceptible. The disease does not develop in clearings made on poor and shallow soils or on land which remains in a swampy unaerated condition. The author goes on to state that the disease is liable to occur in cacao fields of any age, depending on the amount of rainfall and the degree of humidity and also the kind and condition of the accessory trees in the cultivation.

"There is usually no evidence of the disease having persisted from the time of the original clearing, which in many cacao estates is a remote one. Its occurrence seems spontaneous and is mostly unexpected. In a few cases it has seemed to start on the cacao trees themselves, but much more usually it has its origin in trees grown for shade or as windbreaks, or shrubs grown as marginal hedges. The most susceptible of the trees commonly grown in or about cacao are avocado, breadfruit, and pois-doux (Inga sp.) Mango and the immortelles are sometimes but less frequently concerned. A year or two after these trees have been cut down, after a large root has been cut in digging a drain, or when they have died from natural causes, the fungus is commonly found to be established on their roots in the same way that it occurs on the stumps in new clearings, and the adjacent cacao trees begin to pick it up. In valley cultivations the disease most frequently appears in situations along the lower courses of ravines or on flats where flood water over-flows, a distribution which suggests that the infective material is often water-carried, probably from the upland forest.

On hill estates the distribution is usually more irregular.
The cases sometimes met with of cacao trees becoming diseased in the absence of stumps can be accounted for by infection of the surface type. The instances noted have been in the wettest districts, where production of organic matter is at a maximum and where cloudy days, the depth of shade produced by luxuriant growth, and the frequent saturation of the soil with water, all reduce the rate of its destruction. The present writer does not agree entirely with the above interpretations of the spread of the disease and these will be criticised when his personal field observations are recorded.

The first account of a Rosellinia root disease in the British West Indies is that of Barber (1893) from Dominica. Although the fungus was not identified his description of the white fan-like network of mycelium between the bark and the roots suggests that the species was *Rosellinia Pepo*. The following plants were noted as liable to attack: Mango, Orange, Bitter Orange, Lime, Pois-doux (*Inga* sp.), Coffee, Cacao, Breadfruit, Eddoes, Sugar-cane, Cassia Fistula and Cassava.

Howard (1901) states in the Grenada Official Gazette that three patches of diseased cacao trees were seen occurring in good well-drained soil and surrounded by healthy vigorous trees and that the disease seemed identical with that described by Barber.

Stockdale (1908) dealt with the disease in Grenada, Dominica and St. Lucia and referred to the typical star-like web of white mycelium between bark and wood. He regarded as an established fact that the disease spread to cacao from the roots of pois-doux and breadfruit but also recorded its occurrence in fields with no shade trees.

South (1911) reported that *R. bunodes* is very persistent in the soil, reappearing in the fruit crop on a field replanted after remaining in bush fallow for twenty-five years.
South and Brooks (1912) state that the fungus does not appear to grow in decaying humus.

South (1915) described a black root disease of limes and cacao in Dominica and St. Lucia due to *Rosellinia bunodes* and *R. Pepo* and held the opinion that infection with the fungus took place by root contact with previously infested trees or decayed logs. He also recorded cases where infection had taken place with no such sources near.

Fawcett (1915) describes a black root disease of coffee caused by *Rosellinia bunodes* in Porto Rico. Other hosts recorded were *Piper* sp., anamu (*Petiveria alliacea*) and various ornamentals and shrubs - *Graptophyllum pictum*, *Panax plumatum*, *Acalypha mosaica*, rose apple, *Miconia* sp., and *Palicourea* sp. The following appeared immune - *Tradescantia* sp., *Adiantum* sp., guama and mango.

The fungus was observed to spread at a maximum rate of 10-12 feet in one year being retarded only by excessively dry or wet soils, and by natural barriers such as brooks or by the lack of decaying vegetation in the soil. The conditions favouring its growth were found to be those provided by moist shaded soils - conditions also favourable to the coffee trees. It was accordingly found that the best trees suffered most damage and that poor coffee on sun exposed dry slopes remained free from trouble.

The fungus was recorded throughout the coffee-growing regions of the island and its distribution was not found to be limited by climatic factors as in some other diseases.

The author was unable to determine the length of time required to lapse before the ground became fit for replanting in the absence of any treatment of the soil with fungicides. He cites a case of trees replanted in an area where vegetation had/
had been killed two years previously without any reappearance of the disease during a period of 18 months.

Various methods of chemical control were tried on land previously cleared from diseased trees, stumps and decayed vegetable debris. Lime or sulphur applied at the rate of 500 grammes per square metre was found to effectively control the disease over a period of 3 years observation. In plots which received a quarter of the amount of sulphur, 2% or 6 out of 317 trees died. In the control plots which were merely cleaned and ditched 5% or 16 out of 334 trees died. The author does not record the death rate of coffee on untreated plots and so the significance of his figures cannot be determined. It would have been of interest to have known the reduction in the incidence of the disease due to the cleaning and ditching alone. The construction of a sulphur barrier by the formation of a trench 4 inches deep and of equal width, filled at the rate of 15 grammes of sulphur to each metre of trench, was found effective in checking the spread of the fungus.

The most important steps to be taken in controlling the disease were found to be the ditching about of diseased areas and the collection and destruction of diseased trees and vegetable debris.

Matz (1921) records a black root disease of coffee caused by the same species of Rosellinia from a plantation near Porto Rico which had large and crowded shade trees together with a heavy and moist surface mulch. The root disease was found to yield to some extent to rational soil treatment.

Howell (1922) records Rosellinia bunodes and R.Pepo from Trinidad, spreading from forest stumps to living trees in new clearings and from forest material deposited by flooded streams/
streams in older plantations. He notes that it is a slow working disease caused by a fungus which favours damp sheltered situations on wood or other vegetable matter in or on the soil. To check the extension of the disease he recommends chessboard trenching carried below the level of the lateral roots. The mycelium is stated to spread through shaded soil rich in organic matter as well as along the roots of trees. The removal and incineration of diseased trees with as much of their roots as possible is advised.

Wilson (1922) records an outbreak of Rosellinia disease which occurred in 1907 on 2-year old plants of the common spruce, *Picea excelsa*, in a nursery in the south of Scotland. The fungus, which he provisionally assigned to *Rosellinia aquila*, attacked most of the plants in the seed bed. The fungus was successfully eradicated by a thorough soaking of the seed bed with copper sulphate solution and since that date the fungus has not again been recorded. In this case the appearance of the disease may have been due to a chance introduction from the continent and its disappearance might as equally well have been due to adverse climatic conditions as to the nature of the control measures adopted. Diseases attributed to *Rosellinia necatrix* have only been recorded regularly from England, from two main districts, Devon and Cornwall, where the climatic conditions appear particularly suited to this fungus.

Negretti (1923) describes considerable damage done by *Rosellinia Papo* to cacao in the Moca and La Viga districts of the Dominican Republic. According to this investigator the disease originates from dead and rotting stumps in forest clearings and infects living plants chiefly through wounds. The fungus is also found on stumps of cacao plants cut down during/
during thinning operations. The preventative measures recommended are the removal and incineration of all stumps and dead plant parts, to drain the soil well and to leave plenty of room between the trees for air and ventilation. Further measures are a thorough trenching of the diseased area coupled with "disinfection" with quicklime, slaked lime or sulphate of iron.

Nattrass (1926) investigated in England a white root rot of fruit trees caused by *Kosellinia necatrix* (Hart) Berl., and observed that fine attenuated hyphae, which he described as "exploration" hyphae travelled through the soil, filling up minute cavities especially worm burrows. His inoculation experiments showed that only the diffuse mycelium permeating the soil was able directly to infect the root system of the host plant. He doubted whether the fungus could directly attack any part of the plant other than the fine rootlets.

According to Gadd (1928) the opposite state of affairs holds for the parasitism of *Kosellinia arcuata* on tea seedlings in Ceylon. Seedlings inoculated with pure cultures of this fungus were observed to become attacked after two months, at the tap root near the collar and the fungus to have extended down along it and into some of the lateral roots. The finer feeding roots however were mostly uninjured.

Walters (1928) states that *Rosellinia* root disease is prevalent in St. Lucia and is favoured by waterlogged soil.

Beaumont and Hodson (1929) record their field observations on a basal bulb rot of *Narcissus* in Cornwall, England, which they provisionally regard as being caused by *Rosellinia necatrix*. The effect of the disease in the field is to cause large patches to appear which are bare of plants. The plants on the outside of the circle are easily pulled up and exhibit a chocolate/
chocolate brown decay of the base and scales. The bulbs in the centre may be completely decayed. The disease is usually met with at the end of the autumn rains, as the fungus is stated to require large quantities of moisture. It was found that bulbs could safely be replanted when the affected patch of soil was turned over and thoroughly dried out in the summer. Bulbs only slightly attacked could be dried off and planted the next season. It was also recommended that an isolation trench be constructed to prevent the fungus spreading when the patch was first observed.

Stell (1929) observes that the Rosellinia diseases of cacao in Trinidad affect some of the most productive trees as these grow on soil rich in organic matter where the fungus is said to thrive.

Walters (1931) further states that the Rosellinia root diseases of cacao cause more loss of yield in St. Lucia than any other disease or pest of this host. He advises the replacement of the very susceptible shade tree 'Immortelle' (Erythrina spp.) by the highly resistant Gliricidia. In the following year (1932) he observes that the disease becomes prominent as the shade trees are removed and recommends accordingly a gradual shade reduction by heavy pruning. The same writer reports (1934) that the disease spreads rapidly in fields abandoned owing to the depressed condition of the market and these have been replanted with limes, grapefruits or coconuts. There is no well authenticated instance of Rosellinia invading the uninjured roots of sour orange in St. Lucia, but the fungus is known to invade the wounded roots of this host.

Briant (1933) in a survey of the incidence of "burning" disease of arrowroot in St Vincent, attributed to Rosellinia bunodes.
bunodes, observed that the affected patches were often in hollows where the subsoil was wet and sandy. These wet subsoils were found to occur even on slopes. He considered that the presence of *Rosellinia* in a field did not necessarily result in parasitism even if a suitable host was present. In his opinion the fungus is a common saprophytic inhabitant of soils especially those rich in organic material, the pathogenic capacity of which depends entirely on the presence of certain conditions of which excessive moisture is the chief. Prevention consists in adopting improved draining methods and in planting disease free material.

During a short visit to St Vincent by the present writer certain features of the disease were recorded which were different to those observed by Briant. These will be discussed in the next section of this dissertation.

Thomas, Hansen and Thomas (1954) state that much damage has been caused during the five previous years by *Rosellinia necatrix* in apple and apricot orchards in California. The pathogenicity of the fungus was demonstrated but no mention was made of control measures adopted.

Briton-Jones (1954) has criticised the orthodox views of the past with regard to *Rosellinia* root diseases. In connection with infection from root stumps he has pointed out that *Rosellinia* sp., rarely if ever establish themselves on tree stumps and that in the British West Indies, the fungi which establish themselves on tree stumps and assist in their disintegration do not belong to the parasitic group. Outbreaks in the absence of tree stumps are observed to be extremely common and the explanation that these arise from infection of the living trees by spores falling on exposed or injured lateral roots is discredited in favour of the supposition that
the fungi are already present in the soil growing saprophytically on fragments of decaying vegetable matter and that the fungi only become parasitic when certain soil conditions are favourable. He further suggests that the elimination of factors favouring the fungi is a method of controlling the disease and is fundamentally sounder than efforts to eliminate the sources of infection which so often only prove futile.

Toovey (1935) describes experiments which he performed to test the relation between soil conditions and the infection of *Hibiscus mutabilis* by *Rosellinia Pepo*. The results he obtained, although inconclusive, suggested that a limited range of soil moisture existed in which Rosellinia could function as a parasite. The use of diseased fragments of cacao roots as a source of the fungus in these experiments partly explains the inconclusive nature of his results. The writer has found this method very unsatisfactory.

Further experiments are described which showed that there did not appear to be any relation between the major soil nutrients, nitrogen, potash and phosphate, and susceptibility towards infection. Field observations did not show any relationship between soil reaction and the distribution of Rosellinia root disease of cacao.
The present investigation is limited to a study of the effect of environmental factors on the incidence and spread of Rosellinia root disease already established in the field. No attempt was made to investigate the origin of the disease on newly cleared forest as it was felt that this was a separate problem.

The changes which take place in the environment when forest is cleared are very great and the attention of research workers has only recently been drawn to this subject. Corbet (1935) stresses the necessity for a biological investigation into this matter and gives a review of the literature.

Nature of the investigation.

The nature of the following investigation was determined by a general consideration of the factors concerned in the study of the physiology of parasitism which included the following:

1. The amount of infestation present.

   This factor was recorded in field observations together with information with regard to age and nature of the infective material.

2. The percentage of soil moisture.

   The importance of soil moisture has been stressed by research workers on Rosellinia diseases both in Temperate countries and in the Tropics. The effect of this factor on the incidence of the disease is not however simple to analyse. Howard (1901) for example records diseased cacao in well-drained soil and Walters (1928) states that the disease is favoured by waterlogged soil. The problem is further complicated by the fact that the fungi belonging to the genus Rosellinia not only/
only travel over the surface of the soil but through the soil. This necessitates the interpretation of soil moisture in terms of the physical structure of the soil.

3. Temperature.

The effect of this factor was not investigated in the field as it was believed that variations of this factor would be too small to make their effect felt. Experiments were performed in the laboratory on the relationship between temperature and the growth of the fungus as this factor plays an important part in the control of the fungus in the field.

4. Soil Reaction.

The Hydrogen Ion Concentration was recorded in the field from diseased and healthy areas. It soon became apparent that this factor gave no indication that it would be of assistance in analysing the disease complex. The factor was however studied in the laboratory in relation to the growth of the fungus.


Soil samples were taken in the field from diseased and healthy areas and data on the above factors recorded.


This was also recorded from soil samples taken in the field.

7. State of the Cultivation.

This was also recorded in field notes.

An account will first be given of field observations on the above factors and this will be followed by descriptions of infection experiments with one factor under control. As any one of these factors may act singly or in conjunction with any of the others in the disease complex the selection of any one factor for experimental investigation is a matter for some deliberation. The two factors chosen in this investigation were soil moisture and available chemical fertility.
Field Observations.

Field observations were carried out on different estates in the following three islands in the Lesser Antilles: Trinidad, Grenada and St. Vincent, with the object of obtaining information regarding environmental conditions and the incidence of Rosellinia. Notes were recorded on the previous history of the estates, the rainfall, and general topography. A detailed soil analysis was performed by the Department of Chemistry of the Imperial College of Tropical Agriculture, through the kindness of Professor Hardy. The methods used in the soil analysis are recorded in a paper by McDonald, Hardy and Rodriguez (1933).

The following estates were visited in Trinidad:

- Diego Martin, River Estate; Caura Valley, La Veronica and San Carlos Estates; Montserrat, La Vega and Montana Estates; Moruga, Henry Estate; Macqueripe, Tucker Valley Estate; Arima, Verdant Vale Estate.

In Grenada, estates in the following parishes were visited:

- St. John's Belvidere and Woodford Estates; St. David's, Petite Etang Estate; St. Andrew's, Tuileries Estate; St. George's, Annandale Estate. In St. Vincent, Sion Hill Estate was visited in connection with the burning disease of arrowroot.

Field Observations in Trinidad.

River Estate.

The previous history of this estate is of interest. Records show that the valley in which it is situated has been under cultivation for at least 100 years. The last crop of sugar, which had been grown for over 50 years was taken in 1893. Up to 1900 the land supported ground provisions or was left abandoned. Cacao was then planted for the government, which purchased the estate in 1897, under the contract system. This was performed badly with little or no proper supervision, with/
with the result that cultivations were poor, there were no drains and grass was allowed to be removed for fodder purposes. This latter act coupled with charcoal burning robbed the land of much organic matter.

The presence of the disease in this area, low in organic matter, free from forest stumps and also from the forest type of plant association for many years is interesting from the ecological view. The cultivation of sugarcane introduced a crop which was resistant to Rosellinia root disease and demanded quite a different type of cultivation to that of a crop produced under forest conditions which receives practically nothing at all. One may speculate whether the fungus was able to persist in the soil as a saprophyte during the years of sugar cane cultivation, to resume its parasitic powers with the planting of cocoa, when forest conditions were restored. Evidence of a similar nature has already been noted from the observations of South (1911) who recorded that *Rosellinia bunodes* remained in fallow land for twenty-five years, to reappear with replanting of fruit trees.

Or did the sugar cane cultivations so upset the ecological conditions as to cause the disappearance of the fungus, to be reintroduced by the means of a new agency such as flood water? Caution however must be exercised when flooding is postulated as a factor since two distinct effects are present. There is first the mechanical transference of infected root fragments from diseased to healthy areas and secondly the alteration of local environmental conditions which may affect the pathogenicity of a fungus already present in the soil. Examples will be mentioned of observations at other estates where flooding may occur without the appearance of Rosellinia disease. At River estate however there appears to be evidence of/
of an association between the amount of flooding and the incidence of root disease. Diseased patches were found most commonly at the bends of river banks or at narrow culverts where the flood water overflowed during the rainy season. The patches were observed to follow the tracks taken by the flood water. Improvements conducted by the present manager of the estate, Mr O' Connor, in 1922-23 by deepening the main drains and redesigning culverts have according to his observations, materially reduced flooding and the incidence of the disease.

Observations within a diseased area.

A diseased patch situated in field 20b., in a flat area of alluvial silt, and with an annual rainfall of 60-70 inches, was selected for a detailed study of soil factors. This field was 35 years old and the diseased patch 20 years old in 1935. In 1925, when the diseased patch was 10 years old, the shade trees in this field were cut out. No increase in the incidence of Rosellinia root disease was observed, in fact the opposite was recorded. In the same year cacao supplies were given permannure at the rate of 15 tons per acre, but this treatment it was found, did not prevent an attack by the fungus.

The control measures adopted, consisted in uprooting diseased trees and burning them. No lime was added to the soil. Field 20 b. is at present in good condition and is intensively worked. The diseased area is roughly oval in shape and lies North West to South East (Fig.1.)
The incidence of the disease is summarised in Table I below:

**TABLE I.**

Analysis of trees in the Rosellinia Patch on the No Shade Block on River Estate (Field 20b.) December 13th, 1935.

<table>
<thead>
<tr>
<th>Area</th>
<th>Healthy Trees</th>
<th>Infected Trees</th>
<th>Dead Trees</th>
<th>TOTAL</th>
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<tr>
<td></td>
<td>O. s. S.</td>
<td>O. s. S.</td>
<td>O. s. S.</td>
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<tr>
<td>Central</td>
<td>13 21</td>
<td>-</td>
<td>34</td>
<td></td>
</tr>
<tr>
<td>Annular</td>
<td>19 34</td>
<td>8</td>
<td>65</td>
<td></td>
</tr>
<tr>
<td>Outer</td>
<td>36 6</td>
<td>1</td>
<td>44</td>
<td>143</td>
</tr>
</tbody>
</table>

0 = Original tree  
S = Supply up to 3 years  
S = Supply established for 7 - 8 years

*First two rows.

Three distinct areas can be recognised within the patch, which is shown in figure 2.
Figure 2.

(1) A central area where the disease originated. This is surrounded by the first isolation trench which failed to prevent the spread of the disease. There are no original trees left in this area but twenty-one out of thirty-four supplies are over six years old which suggests that the area is recovering from the onslaught of Rosellinia. No evidence was obtained that the fungus was in an active parasitic state in this region.

(2) An annular area formed between the first isolation trench and one constructed at a later date. In this region we have a high proportion of supplies between the ages of one to three years, in a very poor state of health. Evidence was obtained that the fungus was in an active parasitic state in this region both cacao (Fig 3) and cassava (Fig 4) supplies being found attacked. (M 7 and M 8 in diagram).
Bananas, dasheen and sugar cane growing in this region for shade were not observed to be attacked. On the outer margin of this area a few original trees were found dying or recently killed by the fungus. (Fig 5.)
(3) an outer area beyond the second isolation trench comprises the rest of the field in which all the trees are healthy save two in the first row on the North West corner. The presence of supplies in this part of the field as well as in the South East indicates that the second isolation trench has also proved ineffective in preventing the spread of the disease. In figure 6 is shown part of the isolation trench south of the patch showing trees M 8, M 9 and M 10.

Figure 6.

Observations on the rate of spread of the disease from tree to tree spaced 12 x 12 feet, confirmed the observations of previous workers that the time taken is roughly two years. The fungus was observed to travel from tree to tree by root contacts, disease avoidance being correlated with absence of root contacts with diseased roots. Tree number M 10 in the annular area is a case in point. On dissecting out the root systems of trees numbered L 10, M 10 and M 9 it was observed that the roots of the last named curved away from M 10 (Fig 7) and...
and had established contact with L 10 from which it received infection. The photograph which was taken facing East shows trees M 9 (infected) and M 10 (healthy). A faint yellowing of the leaves, characteristic of Rosellinia infection, was to be observed on M 9 when the fungus had travelled in the lateral roots to within thirty-six inches from the collar. Mycelial fans of the fungus were also found in the soil at a depth of six inches to one foot below the surface but always originating from a portion of diseased root. The mycelium was also observed in earth worm burrows in a similar manner to that described by Nattrass (1926) for Rosellinia necatrix.

Analysis of soil factors.

Bulk soil samples for analysis were obtained by means of auger borings from seven spots in each of the following areas:

A. Healthy area six rows distant from the second isolation trench.
B. Annular area of fungal activity.
C. Area of origin and recovery from disease.

Four sampling depths were taken, three, six, twelve and twenty-four inches being considered sufficient as the majority of the cacao feeding roots occur within these extremes.

The results obtained on analysis are given in Table II.

Soil Texture.

The figures show definite evidence that the area where the disease originated is more sandy and that the soil becomes stiffer as the healthy area is approached.
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**Analysis of Soil Samples from Diseased and Healthy Cacao Areas**
Hydrogen Ion Concentration.

The disease has spread from a more alkaline to an acidic area. This quite possibly may be due to lime which has been added as a part of the control measures in the early stages of the disease and therefore of no significance.

Nitrogen and Organic Matter.

There are no significant differences in organic matter content or in total nitrogen in the three areas, nor are there any definite trends in the C/N ratio.

Available Potash and Phosphate.

In the area of origin and recovery from the disease the Potash and Phosphate are adequate. Phosphate deficiency is higher in the healthy area and is very low indeed in the annular patch. The same is true for Potash.

Rate of Nitrification.

The rate of nitrification is very much slower in the annular area the available nitrogen being reduced owing to increased exposure.

Discussion.

Certain important features are apparent from an examination of the above data. There is first the sandy nature of the soil, greater at the centre of origin of the disease. Interpreted in the light of water relationships this means greater fluctuations in water supply to the roots. Flooding, to which this field was subject, would accentuate those fluctuations, and further weaken those cacao trees growing in that area. The evidence appears suggestive that such a weakening predisposes the tree towards infection. The profound changes in the soil due to an altered environment caused by the death of the trees needs no comment. It should be noted that the statement that the fungus cannot exist in a poor soil/
soil is erroneous as an examination of the date for the annular area of fungal activity will show. It is of interest to note that the organic matter content is not significantly higher in the diseased than in the healthy area. There is in addition to be noted the extraordinary poor level of fertility of the soil in the annular area B, brought about by the exposure of the soil as a result from the death of the original trees. It is not to be wondered that the supplies failed to become established, especially when it is known that no manurial treatment was given at the time of planting. Recent work at River Estate has shown that with careful cultivation and manuring, young cacao seedlings may commence bearing when two years and four months old. The following manurial experiment was carried out with supplies in order to determine whether manuring would enable them to "grow away" from the disease. Six pairs of supplies were planted in February 5th, 1936, in the annular area B, near six diseased trees; K 11, K 4, L 4, M 7, M 8, and M 9 shown in the diagram. A supply was planted on opposite sides of each tree at a distance of three feet from the trunk, one receiving manurial treatment and the other serving as a control (Fig 8)
Good supplies were used. The manurial treatment consisted of an application at monthly intervals of half a bucket of liquid manure and three pounds of sulphate of potash. The controls received plain water. An examination four months later showed that 10 out of the 12 supplies had been killed by Rosellinia, one manured and one control supply remaining healthy. These two supplies were found to be free from infection as a result of disease avoidance. This experiment proved the futility of planting supplies in soil which contained diseased root fragments, without some previous attempt being made to ameliorate the soil conditions.

Finally brief comment may be made on the "wave of life" of the fungus. It was observed that the centre of origin of the diseased area C became also the area of recovery. As the supplies are now 7 - 8 years old it has taken nearly ten years of supplying to reestablish the crop. During this period the fungus has ceased its activities in this area and has travelled outwards at approximately the rate of one tree every two years. This rate of spread has been modified in some parts of the field, due to the position of the drains, as on the West side and also no doubt to the position of the roots of the individual trees themselves since the spread of the fungus by root contact is an important feature of the disease.

A criticism may be made here on the manner in which the isolation trenches have been dug, as it illustrates a common fault when carrying out quarantine measures. It will be observed that the isolation trench merely runs round the infected area and does in fact isolate a large island of land which can remain a hot-bed of infection in spite of the slow ecological changes which follow in the wake of the wave of fungal activity. A glance at the diagram will show that isolation/
isolation trenches which are merely circumsambulatory, fail entirely to stop the spread of the fungus and that the shape of the diseased area in no way conforms with the plan of the isolation trench.

The area of greatest fungal activity is in the South East corner of the patch which has just reached the boundary of the second isolation trench. Further observations in this area will be of interest.

It has been demonstrated at La Veronica Estate in the Caura valley that a thorough cultivation of the soil coupled with the construction of drains running across the infected area will effectively control the disease.

It is suggested that the diseased patch in field 20 b. is now too large to attempt the reestablishment of cacao with any economic success. As an alternative, the planting up of an acre of Cacao Oranges might be tried. This tree should prove resistant to the fungus and at the same time will provide a crop of economic value.
La Vega Estate.

A diseased patch of cacao was investigated in Goodwa Field, at the above estate in Montserrat which was of unusual topographical interest. The area, on Brasso Sand soil with an annual rainfall of 98 inches, was situated on a miniature plateau on a hill slope near to a ravine. This is represented in the accompanying photograph. (Fig 9.)

The diseased patch did not involve more than one dozen trees and had not spread any further in extent during the twenty years in which it had been under investigation. During this period the trees had been re-supplied in this area. There appeared to be some factor or set of factors in operation limiting the spread of the fungus.

Soil samples were taken from the diseased area and from a healthy area five rows distant under similar conditions as those previously described. The results of analysis are shown in Table II where the diseased area D, is compared with the healthy area E.

**Soil Texture.**

Both the samples are very sandy and there is nothing to choose/
choose between the two as far as this factor is concerned.

Hydrogen Ion Concentration.

Both samples are very acid, the diseased area being very much more so than the healthy and is too low for the normal production of cacao.

Nitrogen and Organic Matter.

The organic matter is much lower in the diseased area than in the healthy area. The C/N ratio is lower in the diseased area and unfavourable for normal cacao production.

Available Potash and Phosphate.

The available Phosphate is low in both samples and is very deficient. The available Potash is also low in both but is 30% higher in the diseased area.

Rate of Nitrification.

The available nitrogen is about the same for both samples.

Discussion.

One important factor would appear to emerge from a survey of the above analysis, namely that neither available Phosphate nor Potash are connected with the incidence of the disease. No one particular factor of those analysed can be selected to distinguish the diseased from the healthy area and so it remains again to consider the importance of topography and the water relationships of the soil.

Although at first the impression might be obtained that the land in this area is well drained owing to the sandy nature of the soil and the close proximity of a ravine, the writer feels that this might not be the case during the rainy season. Rain water will collect on the hill slope immediately above the plateau and in the absence of contour drains will collect on the projecting shelf of the hillside. Thus will be obtained a condition analogous to that described by/
by Briant (1933) of a wet subsoil on a hill-slope defining the limits of Rosellinia root infection.

At the same estate it was observed in Philip Field, on Brasso Clay soil, that although certain portions of the field were subject to periodic floodings which caused a reduced yield from the trees, the land was quite free from Rosellinia root disease. From this observation it would appear that some nice balance of soil moisture relationships is required before Rosellinia root disease can appear.

Montana Estate.

Another diseased patch of cacao was investigated in Montserrat in Field 2 on the above estate. The area was of similar topography to Goodwa Field at La Vega estate, and was situated near the summit of a hill ridge, as shown in diagram. (Fig 10)
The cacao was interplanted with bananas and coffee, and bushes of the latter were also found killed by Rosellinia Pепo.

The soil analysis of the diseased area F, is given in Table II.

**Soil Texture.**

Can best be described as nondescript.

**Hydrogen Ion Concentration.**

This is normally acid.

**Organic Matter and Nitrogen.**

The Organic Matter is very high when compared with the other samples which have already been dealt with. The C/N ratio is normal.

**Available Potash and Phosphate.**

The available Phosphate is extraordinary low. The potash is low but not markedly so.

**Rate of Nitrification.**

The rate of production of Nitrates is adequate.

**Discussion.**

The disease was observed to have spread from the plateau up the side of the ridge to the summit leaving a trail of dead and dying cacao trees in its wake. A similar interpretation to that put forward to explain the presence of the disease at La Vega is also offered here. The spread of the disease from the plateau can be understood when interpreted in the light of the increased conditions of moisture which obtain in this field due to its high elevation and the presence of bananas which aid in maintaining humid conditions.
La Veronica Estate.

A visit was made to Field 5 on this estate in the Caura valley where an outbreak of the disease had occurred on cacao ten years previously. The field was on a steep slope and was bounded on its lower end by a small stream. The disease was observed to have spread from a collection of debris at the bend of the stream shown in (fig. 11), in an upward direction against the gradient of the field. Successful control of the diseased area, which was similar in extent to the one investigated at River estate, was obtained by following the treatment advised by Nowell, with a few modifications. The method used was the "complete" eradication of diseased tree stumps and the large pit formed by the excavation was left exposed to the air for several months before replanting. No lime was used.
The successful establishment of new cacao supplies was attributed to the clearing and replanting of the whole patch in one operation. Manuring of the supplies was found to be essential for establishment at the first attempt. The shade given to the supplies was not excessive and was provided by *Gliricidia maculata*, shown in figure 12.

The success attendant on these measures is to be expected when it is appreciated that such operations have produced through cultivation, manuring and drainage, completely new soil conditions.

Whether the measures are such as to admit general acceptance by the economic plant pathologist is another question with cacao at its present price.

With regard to the origin of the disease, the planter held the opinion, which is often stated in connection with this disease, that infection had been introduced from his neighbour's estate situated higher up the valley.
San Carlos Estate.

This estate in the Caura valley was visited in order to determine what truth there was in the statement that infection at La Veronica Estate was being received from material transported by the river from this area. In spite of the fact that the estate had been neglected for several years owing to the economic depression, three cacao trees only were found diseased due to *Kosellinia Peps* after an extensive search, and no case was seen where the diseased area was as large as that on the lower estate. There is clearly no justification for putting the blame for the appearance of the disease in an estate on to one's neighbour.

Siparia Estate.

Although unable to visit this estate personally, the writer has information from the Manager concerning the incidence of the disease on cacao which is in agreement with his own findings. The disease does not appear to cause any serious damage and when it does appear on the sandy soils in this area it is on similar hill-side plateaux near ravines such as have already been described at La Vega. Flooding which occurs in some areas is not observed to be followed by the appearance of the disease.
Henry Estate.

A visit was paid to the cacao in McDougal Field, 45 years old, situated on flat-lying land composed of light sandy soil. To the north the field sloped upwards to McDougal Hill. (fig. 13)

The annual rainfall in the area was from 60 - 80 inches. In 1931, two rows of cacao trees started dying as a result of infection by *Rosellinia Pepo*. Ten trees were involved and these were treated by the complete eradication of the stumps, and a pit 5 feet deep was left open for one year. Lime was then sprinkled over the area and covered in. No further sign of the disease was observed on the supplies. The felling of immortelle shade trees on this estate caused no increase in the incidence of *Rosellinia* root disease nor did the hurricane of 1933.

Two trees only, which showed evidence of the fungus in an active state, were found during the visit in 1936. Both were near McDougal Hill. The occurrence of the disease at/
at the base of the hill suggests a comparison with La Vega and Montana estates. It was observed in Smart Field, adjacent to McDougal Field and north of McDougal Hill, that no Rosellinia was present. This the writer believes can be accounted for by the heavier nature of the soil in this field. Soil samples from the diseased area in McDougal field were taken, but the results of analysis are not yet to hand.

**Tucker Valley Estate.**

The present crop in Backadire Field, on this estate, consists of 10 year old nutmegs. Previous crops have been sugar-cane, coconuts and cacao, the latter being grown for over 50 years. The nutmegs were quite healthy when visited in June, 1956, in spite of the fact that Rosellinia has been found on cacao in this valley. Six months previous to the writer's visit to the estate the manager started a programme of cutting out some of his shade trees which consist mainly of Wild Chataignier and Bread Fruit. In view of the tradition that this practice gives rise to outbreaks of Rosellinia root disease, the writer feels that it is worth while, for the next two years, to keep this and similar estates under close observation.

**Verdant Vale Estate.**

This estate was taken over in 1925 by the present manager and planted entirely with nutmegs. The main constituent of the soil is shale, there are no drains and manuring is not practised. Cultivations consist in rough cutlassing. Immortelle trees on the estate were cut out at the time when it was taken over, with no subsequent appearance of Rosellinia root disease. The writer again suggests that the heavy nature of the soil was not conducive to the presence of Rosellinia.
Field Observations in Grenada.

Observations on root disease were carried out in Grenada under a similar range of conditions as in Trinidad. Examples of successful treatment of the disease were recorded as well as those which had been neglected. It may be emphasised in passing, that Rosellinia root diseases have been overrated in their importance in Grenada. Many cases were observed where cacao trees were dying in patches due to physiological causes such as old age, exposure to wind, flooding by sea water and neglected drains. In one case Nutmeg canker was confused with Rosellinia root disease.

Tuileries Estate.

On this estate a successful example of the control of root disease on cacao was found in Munich Field. The field which is 45 years old is situated at 600 feet on a hill slope facing east. The soil contains many boulders and is composed of red soil derived from olivine basalt. The annual rainfall is 80 inches.

The disease first appeared in 1915, when the field was 25 years old, in the middle of the slope and spread in an upward direction. There were no shade trees in the vicinity of the diseased patch although the field was planted with Bread Nut, Bread Fruit and Tendre acailoux which have the reputation of being most susceptible to Rosellinia. No evidence could be obtained at the time of waterlogged conditions in the soil nor were there any signs of underground springs. The treatment given in 1915 consisted of a very thorough excavation of the diseased trees and the complete removal of infected debris. The operation was performed in the dry season and the soil in the area was passed through a builder's/
builder's sieve. A circular isolation trench was also dug. About 20 trees were involved, and the total cost of the operations which employed 5 men for 14 days was about £3.0.0.

Nutmeg and cacao replanted after this treatment remained healthy and the result of the operations was a complete success. When the field was visited in June 1936, it was not possible to find any trace of Rosellinia.

Further observations will be required on Field No.1 on this estate where nutmegs, 40 years old, are being thinned out. The thinning operations started in April 1936, but up to the present time there has been no appearance of Rosellinia root disease.

Belvidere Estate

Nutmegs are the main crop grown on this estate. In Wilson Field, attack by Rosellinia *Pepo* was observed on trees which varied in age from 7 to 45 years. The field was situated on a hill slope of red soil, 3-4 feet deep, in an area which has an annual rainfall of over 120 inches.

Part of the field had in previous years grown cacao. It was observed that where nutmegs had been planted on the site of old Rosellinia patches they too became infected by the fungus. Such a case is shown in figure 14. A 9 year old nutmeg tree.
is illustrated on a hill slope facing west, which has become infected from the stump of a 25 year old cacao tree, previously killed by the fungus, 3½ feet above.

In another case the infection of a 45 year old nutmeg tree from a breadfruit tree, killed by the fungus was observed. The disease was controlled in this instance by using tree surgical methods. The dead breadfruit tree was removed, all infected parts of the nutmeg roots cut off and the healthy cut ends treated with tar. This method of dealing with the disease has proved successful on this estate.

**Annandale Estate.**

A Rosellinia patch of long standing, which had slowly spread uphill, was investigated on this estate in Ross Field. The field, 50 years old, was situated on a steep hill slope at a height of 1,200 feet on boulder strewn red soil, with an annual rainfall of over 120 inches. Cacao, nutmeg and avocado pear trees were all found to be attacked but bananas and tannias were found growing free from the disease lower down the hill side. Figure 15 shows a view of the patch looking
down the hill in a South West direction. From right to left can be seen the stumps of cacao, nutmeg and avocado pear, all killed by the fungus. In the background are the bananas and the ground provisions garden, occupying the place of 5 rows of cacao trees down the side of the hill slope. The presence of such gardens in old cacao plantations suggests previous sites of Rosellinia patches which the planter in olden times, finding unprofitable had turned to other uses. Figure 16

shows the same patch viewed in a South East direction. In the foreground is the stump of a diseased nutmeg tree which was burnt as part of the control measures adopted. On examining the roots of the stump in the ground it was found that these were untouched by the fire and that the fungus was still viable. It was clear that the attempt made merely to burn the stumps without their excavation was of no practical value whatsoever. On account of the stony nature of the ground the excavation of the roots presents great practical difficulties. The cost of the labour involved alone would prohibit this measure. In the same illustration on the right will be observed a cacao tree/
tree infected from a nutmeg, the stump of which still remains 4 feet away and in direct line with the observer. Soil samples were taken here for comparison with those from a healthy area on the same contour level six rows away. The results of analysis are not yet to hand. On further excavation of the root system of the cacao tree it was found that the fungus had travelled down the main root for a distance over 4 feet. It will be appreciated that under these conditions the use of shallow isolation trenches alone would be of little use in attempting to control the spread of the disease.

Woodford Estate.

This estate was visited in order to investigate the case recorded by Nowell (1923) of Rosellinia paraguayensis parasitic on cacao. In 1919, cacao trees were observed in Blayen Field which was then 40 years old, to be dying round the large shade trees of Tendre acailloux (Pithecolobium Berterianum) in the dry weather. The planter cut down these shade trees and this act was followed by a severe infestation of thrips. The estate was then visited by Prof. Ballou and Newell who found Rosellinia paraguayensis on the roots of the dying cacao. The field was then limed, drained and replanted with cacao together with branches of Immortelle for shade trees. The new planting of cacao grew away well and gave no further trouble. It should be noted that no attempt was made to remove the old dead cacao stumps nor was there any sanitary precautions taken. The history of this field does not conform with that of others suffering from Rosellinia root diseases and in the opinion of the writer should be regarded with suspicion. It is felt that the sudden loss of shade and the attack by thrips would in themselves prove sufficient to kill the cacao which was in any case.
case suffering from old age. In the experience of the writer, *Rosellinia Pepo* appears to be the species responsible for root diseases of cacao and nutmeg in the West Indies. This is confirmed by practical experience in isolating the same fungus from different *Rosellinia* patches in Trinidad and Grenada.

Petite Etang.

As there appeared to be some doubt as to whether *Rosellinia bunodes* could attack cacao and thus further complicate the problem, the behaviour of this species was also investigated. The first case examined was a hedge of *Hibiscus mutabilis* which bordered the vegetable garden near the works. The area composed of red soil had been under cultivation for a period of 15 years, and experienced an annual rainfall of 100 - 120 inches. The disease first appeared in the hedge one year after planting. When these observations were made, two years later, the fungus had killed out 30 yards of the hedge. At one place the fungus was observed to pass a cacao tree, the roots of which passed directly underneath. No infection could be found on the cacao roots, although these were surrounded by diseased *Hibiscus* roots thickly covered with the perfect stage of the fungus.

The other case examined was in Field 2, where a 5 year old grapefruit tree was found which had died as a result of an attack by *Rosellinia bunodes*. At the base of the tree which had been dead for one year, were obtained specimens of the perfect stage of the fungus. The tree was surrounded by two cacao trees and one cacao stump. None on examination showed any trace of the fungus.

The result of these observations throws some doubt on the possibility of *Rosellinia bunodes* attacking cacao.
The object of the visit to St. Vincent was to observe the relationship between the soil moisture factor and the incidence of *Rosellinia bunodes* on arrowroot. Previous inoculation experiments performed by the writer in Trinidad proved that *Rosellinia pepo* was capable of producing symptoms of "burning disease" of arrowroot, identical with those caused by *Rosellinia bunodes* and described by Briant (1953) in St. Vincent.

**Sion Hill Estate.**

A fresh outbreak of "burning disease" was investigated on Spring Piece Field, on land which had only recently been planted with healthy arrowroot. The soil was medium-light in texture and the annual rainfall was 110 inches. The field had been planted in March 1935 for the provision of supplies and the plants started to quail 9 months later. The symptoms of quailing consist in the upward rolling of the leaves which takes place under both wet and dry conditions of the weather. The disease appeared where arrowroot had not been grown before, on the highest and driest part of the field adjacent to a road cutting. (fig. 17).
When a search was made for some factor responsible for the presence of the disease, it became evident that neither a waterlogged nor an excessively damp soil could be responsible. There were no underground springs in the area, nor was there any disease in the hollow parts of the field. The disease under these conditions differed from previous accounts in its rate of spread. In an adjacent 5 year old field, the following record of the rate of spread of a diseased patch was kept:

<table>
<thead>
<tr>
<th>Year</th>
<th>Number of Baskets</th>
<th>Weight in lbs.</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>Diseased Rhizomes</strong></td>
<td><strong>Diseased Rhizomes</strong></td>
<td></td>
</tr>
<tr>
<td>1934</td>
<td>1/2</td>
<td>25</td>
<td>11.5</td>
</tr>
<tr>
<td>1935</td>
<td>7</td>
<td>700</td>
<td>$3.0</td>
</tr>
<tr>
<td>1936</td>
<td>15</td>
<td>1500</td>
<td>$6.75</td>
</tr>
</tbody>
</table>

Several such patches may form in a field and join together, causing serious economic losses.

Some factor which was limiting the spread of the disease in the higher regions of the island was clearly lacking here. The writer suggests that this factor was not so much soil moisture as soil texture. Past experience has shown that Rosellinia disease is favoured by soils of a sandy texture and is absent in stiff heavy soils. Soil moisture then, is to be interpreted in terms of the physical structure of the soil, rather than as a purely quantitative factor. Evidence was obtained that the disease could be transferred by means of labourer's implements from diseased to healthy areas. The wind was also found to be a factor in the dissemination of portions of diseased rhizome.

Cassava and sweet-potato were observed to be immune to the fungus on this estate, while yams, tannia and Tous-les-mois were susceptible. The latter was planted in a Rosellinia patch in the attempt to find an alternative crop plant, but succumbed within/
within a month from planting, the old tuber becoming infected first by the mycelial strands of the fungus.

Control measures suggested, were the removal of all diseased tubers from the field in sacks and the exposure of the patch to the sun in the dry season.

Summary of Field Observations.

Briefly summarising the field observations it may be noted that of the soil factors examined, the Hydrogen Ion Concentration, amount of Nitrogen, Organic Matter, Available Potash and Phosphate and Rate of Nitrification seem to play no part in determining the incidence of Rosellinia root disease. There is some evidence however that Soil Texture when considered in relation to Topography and the Soil Moisture factor may play an important part.

Cultural Studies of the Parasitic Species of Rosellinia.

Several isolations of Rosellinia Pepo and Rosellinia bunodes were made during this investigation, which are recorded below.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Host</th>
<th>Species</th>
</tr>
</thead>
<tbody>
<tr>
<td>River Estate, Trinidad</td>
<td>Cacao</td>
<td>Rosellinia Pepo</td>
</tr>
<tr>
<td>Belvidere Estate, Grenada</td>
<td>Nutmeg</td>
<td>Rosellinia Pepo</td>
</tr>
<tr>
<td>Annandale Estate, Grenada</td>
<td>Cacao</td>
<td>Rosellinia Pepo</td>
</tr>
<tr>
<td>Petite Etang Estate, Grenada</td>
<td>Hibiscus</td>
<td>Rosellinia bunodes</td>
</tr>
<tr>
<td>Petite Etang Estate, Grenada</td>
<td>Grapefruit</td>
<td>Rosellinia bunodes</td>
</tr>
<tr>
<td>Sion Hill Estate, St.Vincent</td>
<td>Arrowroot</td>
<td>Rosellinia bunodes</td>
</tr>
</tbody>
</table>

It was not possible to study these fungi in Trinidad in view of the quarantine regulations. It is hoped in Britain to make a comparative study of all these isolations. The present account deals only with Rosellinia Pepo isolated in Trinidad.
Isolation and Cultural Characteristics of Rosellinia Pepo.

The fungus was isolated from the roots of a diseased tree obtained from River Estate in October 1955. The method of isolation consisted in separating the bark of the root from the wood and inoculating Dox's and 2% Dextrose-Potato media with small pieces of tissue bearing the white mycelial fans. No sterilising agents were employed, the operation being performed in an inoculation chamber, due care being taken to prevent contamination from the surface of the roots. Six out of the twenty-four primary isolations yielded cultures of Rosellinia Pepo.

The fungus was cultured on a variety of media, when it was found that growth was best on natural media containing organic material and that growth was poor on synthetic media.

Three types of mycelial growth were observed to occur, similar to the types described by Nattrass (1926) for Rosellinia necatrix:

1. the greenish-grey to white mycelial fans (Fig. 18)

![Figure 18.](image)

2. mycelial strands which are white at first and later become black with a white core and vary from 5 to 15 millimetres in length and

3. the "exploration hyphae" of Nattrass — a very diffuse white mycelium composed of fine attenuated hyphae.

The mycelial strands were formed only in the host tissues and below the surface of the media.

The/
The following is a description of the growth of the fungus on various media.

(1) 2% Malt Agar.

Growth at room temperature (25-28°C.) is at first white. After 3 days the centre becomes green tinged and a faint zonation appears. By the sixth day dark brown striae have appeared and three zoned rings formed. After 14 days the culture presents a dark green-brown shiny appearance with a white margin and is covered with a light aerial growth closely applied to the surface of the medium in the form of a flattened mycelial fan. Mycelial strands are not produced abundantly on this medium. They appear within 3 weeks from the time of inoculation and are white at first then black with a white centre. The graphium stage is only produced on this medium, after 4-5 months.

(2) 10% Malt Agar.

Growth is slower than on 2% Malt Agar, with a denser aerial growth and more marked zonation. The initiation of the graphium stage becomes apparent after 10 to 14 days, in the zones of darker green which alternate with zones of lighter green. No spores are produced. The growth of the fungus throws the medium into large wrinkles. Black mycelial strands with a white centre are formed abundantly on this medium within 14 days. The final colour of the culture is a dark brown.

(3) 2% Malt and Sterile Sand.

Growth white at first becoming dark green with a white margin after 21 days. Abundant aerial mycelium is produced which turns dark brown after 6 weeks. During this period, greyish-brown mycelial fans travelled up the sides of the flask to a height of 6 centimetres.

(4) Leonian's Medium.

Growth on this medium was indistinguishable from 2% Malt Agar.

(5)
(5) Dextrose-Potato.

Growth was slower and there was less aerial mycelium produced on this medium compared with 2% Malt Agar. A dark brown growth was obtained and black mycelial strands were produced in 3 weeks. A sterile graphium stage was formed after 3 months.

(6) Oatmeal Agar.

Growth at first white, then tinged with green and finally turning dark brown. Mycelial strands are formed in 3 weeks but the graphium stage is not developed.

(7) Lima Bean Agar.

Growth was similar to that on the above media with a more luxuriant formation of aerial hyphae. Mycelial strands were formed abundantly.

(8) Sterilised Potato Plugs.

Growth on Irish Potato plugs was slow, a dense mass of compact purplish-black hyphae being formed after 3 weeks with no graphium stage. On Sweet Potato plugs a dark brown to black growth was obtained and the graphium stage was well developed and produced spores. Mycelial strands were formed inside the plug and grew out into the cotton wool at the bottom of the test tube.

(9) Sterilised Cacao Branches and Roots.

The fungus grew readily on sterilised Cacao branches, 1/2" in diameter. After 17 days the initiation of the graphium stage was observed in cracks on the drier parts of the bark and on the cut ends. (Fig. 19).
The sterilised roots showed no initiation of the graphium stage, due possibly to the moister conditions prevailing in that series.

Examination of an inoculated root after 4 weeks showed the presence of white lines under the bark. These developed into white stars 10 weeks after inoculation. (Fig. 20).

The fungus also grew into the cotton wool plug at the base of the test tube with the formation of black lines containing "bladder" cells. (See Fig. 19.) The graphium stage bearing spores was formed after 3 months.

(10) **Campbell's C Medium.**

A mixture of Sawdust, Quaker Oats and Maize Meal moistened with 10% Malt Extract solution was used as a medium with the hope that such a rich organic mixture would encourage the formation of the perithecial stage of the fungus. Growth was slow at first, dark grey-green in colour with a white margin. There was no sign of the perfect stage of the fungus after 2 months, although by this time the white mycelial strands had penetrated the medium in all directions.

(11)/
Growth was very poor on this medium, white, superficial and scanty.

(12) Dox's Agar.

Growth was slow, submerged and dark brown in colour on this medium with little aerial development of the mycelium. Mycelial strands were formed late. The graphium stage appeared on the 2% Malt agar inoculum after 50 days and produced spores. They were also formed on pieces of diseased root material used as an inoculum but never on the agar itself.

(13) Richard's solution agar.

Growth was poor, submerged and lighter in colour than on Dox's agar. The graphium stage appeared on the 2% Malt inoculum after 30 days but produced no spores.

(14) Brown's medium.

Similar growth was obtained on this medium as on Richard's solution.

(15) Mixed media.

The following mixture of media was made up to test the observation on the production of the graphium stage on the 2% Malt inoculum on Dox's agar. A glass ring was placed in the centre of a Petri dish and the whole sterilised. The ring was then filled with 2% Malt agar and when this had set, the outside space was filled with Dox's agar. The malt was then inoculated with the fungus. The graphium stage was produced abundantly after a period of 40 days on the Dox's agar just outside the glass ring. These can be seen in Fig. 21 in the petri-dish containing the light coloured strain. The dark coloured strain produced the graphium stage just as abundantly but is not visible to the camera.
In one plate they were produced on the malt inside the ring. Spore production was well developed 50 days from the date of inoculation.

Effect of Temperature on the Rate of Growth.

The rate of growth of the fungus was measured by obtaining the mean of five plates of 2% Malt extract agar. Both the depth of the agar and the size of the inoculum, 0.5 cm. were standardised. The following series of temperature were used: -

7.2°C., 14.4°C., 25°C., 27°C., and 33°C. The relation between temperature and the rate of growth is shown in Fig. 22.
Growth was observed to be inhibited at temperatures of 7.2°C, and 33°C. When removed from these temperatures it was found that one out of the five cultures held at 33°C for five days started growth at room temperature and that apparently the other inoculated plates were dead. On the other hand all the plates held at 7.2°C for fourteen days started growth at room temperature.

The fungus was observed to grow best at a temperature ranging between 25°C and 27°C. At 25°C, the average rate of growth per day was 0.5 centimetres. It is of interest to compare this rate of growth in culture with the rate of spread of the disease in the field. Both the present writer and other observers have noted that the time taken for the disease to travel from one tree to the next is approximately two years. With a planting distance of 12 feet this means a rate of spread of \( \frac{4 \times 91.5}{365.2} \) centimetres per year, (1 yard = 91.5 cm.)

This gives a rate of spread per day of \( \frac{366}{365.2} \approx 0.5 \) centimetres. This agreement may be a pure coincidence and is merely recorded as a point of interest.

It was further found that cultures of the fungus were killed when exposed to the heat of the afternoon sun for \( \frac{1}{2} \) hours. This is of importance in connection with the control measures adopted in checking the disease in the field.

**Effect of Hydrogen Ion Concentration on the Growth of the Fungus.**

The effect of the Hydrogen Ion Concentration on the growth of the fungus was studied on broad lines only. Three series of plates of 3% Bacto Nutrient agar with the pH adjusted to 4.5, 6.3 and 8.5 respectively were inoculated with the fungus. It was found that there was nothing to choose between the three series as far as rate of growth was concerned. *Rosellinia Pepo* would thus appear to be tolerant of a wide range of Hydrogen Ion Concentration and this is confirmed by the field observations.
Inoculation Experiments in the Laboratory.

The pathogenicity of *Rosellinia Pepo* was tested by inoculation experiments with Cacao seedlings grown on agar media under aseptic conditions. The Cacao seeds germinated within 14 - 21 days from sowing on 2% Dextrose-Potato agar - twice as long as the normal time on damp sand. The seedlings were inoculated when 3 weeks old and wilting and death took place within 14 days. This is also the normal period for infection for plants growing in soil and sand. Examination showed that infection had originated at the collar of the hypocotyl from the region of one of the lateral roots.

Small white mycelial stars, characteristic of this fungus, were formed between the stele and the outer cortex and mycelial strands were very abundant in the host tissues and in the media. In some of the seedlings these had penetrated the stele. The fungus was re-isolated from infected seedlings. Control seedlings in flasks of agar (a) without the fungus and (b) inoculated with *Penicillium glaucum*, *Mucor* sp., and *Bacillus mesentericus* remained healthy although some of the latter were stunted. These recovered when transferred to pots containing damp sand.

Cacao seedlings have been grown for six to eight weeks in flasks containing 2% Dextrose-Potato agar, 2% Malt agar and Knudson's Solution B agar inoculated with *Penicillium* sp. and have still remained healthy whilst seedlings inoculated with *Rosellinia Pepo* died within two weeks. This is shown in Fig. 23. The flask in the centre is inoculated with *R. Pepo* and on either side are the *Penicillium* controls.

![Figure 23](image-url)
These observations show that the experiments were fair tests of the pathogenicity of the fungus and that *Hoaellinia Pepo* can be regarded as a primary parasite.

The pathogenicity of the fungus was further tested on arrowroot. Rhizomes were inoculated with 14 day old cultures of the fungus grown on sweet-potato slices, half being wounded and the remainder left uninjured, the whole being planted in sand which was kept moist. One month later, distinct signs of quailing of the leaves, a characteristic symptom of "burning disease" were recorded from all the inoculated plants, the controls remaining healthy. On examination it was found that both wounded and uninjured rhizomes were penetrated by the fungus, the infected areas appearing light brown in colour in contrast to the white of the healthy tissues. A longitudinal section through the infected rhizome revealed a mass of ramifying mycelial strands, white in colour and thus distinct from the black mycelial strands formed by *Rosellinia bunodes*. Apart from this one character the two species are indistinguishable in their parasitism of arrowroot.

**Method of infection.**

An experiment was carried out in order to determine whether the fungus itself or its staling products were instrumental in predisposing the host root system to penetration. Roots of cacao seedlings were inoculated with an 8 day old culture of the fungus on 2% Malt agar. The seedlings were then planted in damp sand. On examination 5 days later it was found that no browning of the roots had occurred on which the inoculum had been placed but that "exploration hyphae" had travelled through air spaces in the sand in an upward direction to other roots, and there caused browning in advance of penetration wherever contact between hyphae and host cells was established. (Fig. 24)
When seedlings were inoculated at the collar, all the roots near this point were penetrated and the mycelium spread rapidly down the outside of the main root penetrating other laterals in its path. (Fig. 26.)
From these observations it appeared that aeration was an important factor promoting the rapid growth of the mycelium and that death of the roots could be brought about by contact with the mycelium alone.

An attempt was made to infect cacao seedlings by inoculating the hypocotyl above the collar. Small pieces of a 14 day old culture on 2% Malt agar were applied without injury to the hypocotyl and kept moist with cotton wool wrapped in tinfoil. No infection was obtained after one month.

The Influence of the Substratum on the Pathogenicity of the Fungus

Quite frequently in the literature on Rosellinia root diseases one meets references to suggestions that the nature of the substratum on which the fungus grows affects its pathogenicity. Thus Fawcett (1915) writes— "As the fungus is known to live on dead vegetable matter, it is probable that these stumps furnish such abundance of food material that it becomes strong enough to attack living plants, whereas ordinarily it merely makes use of the usual decaying material covering the soil in well-shaded places".

The effect of the substratum was tested experimentally by growing cacao seedlings under sterile conditions and inoculated with Rosellinia Pepo on both organic and inorganic media. No evidence however was obtained that the fungus was less parasitic when grown on an inorganic medium - Knudson's medium, when compared with growth on organic media - 2% Dextrose-potato and 2% Malt agars. Death of the seedlings was brought about by the fungus in exactly the same time.

It would be dangerous to apply deductions based on observations on cacao seedlings in glass bottles to the mature tree in the field but at the same time the writer feels that here we may have an indication that the rôle played by organic matter in/
influencing pathogenicity may be merely a quantitative and
not a qualitative one. The parasitism of this fungus would
thus appear to be in a different category from that of Botrytis
where the spore germ tube is said to be unable to penetrate the
tissues of certain plants until the spore has been supplied with
organic nutriment.

**Viability of Spores.**

All attempts to germinate the spores obtained from both
natural sources and from culture have up to the present failed.
The spores were put up in hanging drops of sterile water, 1% Malt
and 2% Malt agar.

Attempts to produce infection of young seedlings of cacao
by inoculation of a spore suspension at the region of the collar
were also negative. There may be several reasons for this
apparent inactivity of the spores. They may lose their viability
rapidly or some set of factors may be necessary before germina-
tion can take place. It may be noted in connection with this
subject that Carruthers (1903) working with a species of
Rosellinia causing root disease of tea in Ceylon failed in an
attempt to infect a tea plant growing in a pot of sterilised
soil by sowing the spores of the fungus. He did succeed however
in obtaining a vigorous growth of the fungus by the same means
on buried tea branches which had previously been sterilised.

Viala (1891) referring to Rosellinia necatrix on the vine
states that in his experience both ascospores and pyenospores
failed to germinate.

**Infection Experiments with One Factor under Control.**

Most of the soil factors with the exception of soil moisture
have already been considered in the field and have been shown to
play little part in determining the incidence of the disease.

The/
The object of the following experiments was to attempt to determine experimentally the part played by soil moisture and the available chemical fertility, in predisposing the parasitism of cacao seedlings by *Kosellinia Pepo*.

The numerous difficulties which beset the investigator on attempting to establish a series of soil moistures have been well described by Toovey (1935). Following his recommendations the method of constant weight was used in the following experiments.

**Effect of Variation in Soil Moisture Content.**

**Experiment No. I.**

The first experiment was set up on November 26th 1935. Forastero cacao seedlings, germinated and grown in sand for 20 days, were planted in pairs in tin cans 8" x 6" containing soil obtained from River Estate. Five series of different soil moisture contents were established as follows:

<table>
<thead>
<tr>
<th>Series</th>
<th>Moisture content (% Saturation)</th>
<th>Cacao Seedlings</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>90</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>B</td>
<td>75</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>C</td>
<td>60</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>D</td>
<td>45</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>E</td>
<td>30</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td><strong>Total</strong></td>
<td><strong>120</strong></td>
<td><strong>60</strong></td>
</tr>
</tbody>
</table>

In each series 12 tins were inoculated with fragments of cacao root infected with *Kosellinia Pepo* in a viable condition, the fragments being placed in a layer 5 centimetres below the surface. The control consisted of 6 tins inoculated with sterilised diseased root fragments. The soil moisture in each series was maintained by the method of constant weight.

After six weeks, no signs of wilting were apparent in any/
any of the series. A kind of mesophyll chlorosis was observed on about 20% of the plants in both the inoculated and uninoculated series which proved to be of no statistical significance. An examination of the plants removed from the pots showed that although the seedlings had produced between 4 to 5 leaves, there had been no further development of the root system since transplanting. This state of affairs still existed three months after the establishment of the experiment. It was also observed that no growth of the fungus on the diseased fragments had taken place. A careful microscopical examination of the roots of all the plants in one series (C) failed to reveal any association with hyphae of the Rosellinia type.

The material chosen for the inoculum was apparently too old and had lost its viability. There was also the possibility that lack of aeration, had adversely affected the growth of the fungus. It must be stressed that pot experiments of this nature are very artificial and that the environmental conditions produced in the first three inches of soil in the pot does not even approximate the same three inches in the field. It was decided to abandon the method of using naturally infected root material and to use pure cultures of *Rosellinia Pepo* on sterilised sweet potato slices instead. This it was felt would give greater control of the inoculation experiments.

Experiment No. II.

A second experiment to test the effect of soil moisture on the incidence of Rosellinia attack on cacao seedlings was set up on February 29th 1936. The experimental procedure differed from the first experiment in that seedlings 14 days old were used. Pure cultures of *Rosellinia Pepo* on sweet potato slices, 14 days old replaced naturally infected root fragments as a source of inoculum. Four different soil moisture series were established as follows:
In each series 8 tins were inoculated with two sweet potato slices of *Kosellinia Pepo* placed vertically near the root systems of the seedlings. Two pots served as controls in each series.

The following results were obtained over a period of one month and have been summarised in the graph, Fig. 26.

**Figure 26.**

**Series A, 100% Saturation.**

All the inoculated plants in this series failed to develop and these seedlings which were of any size eventually wilted. Examination of the root systems which were blackened and foul smelling failed to reveal the presence of the fungus. Death of the seedlings had evidently been caused by either diffusion of/
of toxins from the inoculated potato slice or due to the products of anaerobic decomposition of the inoculum as the control plants remained 'healthy' although stunted. This series confirmed the observation made by several workers that the fungus cannot develop in waterlogged soil.

**Series B, 85% Saturation.**

Infection of seedlings was observed to be slow in this series in comparison with the two drier series C and D, which follow. The first seedling wilted 16 days after inoculation and only 6/16 of the inoculated seedlings died during the period of observation. The fungus was reisolated from the diseased seedlings in this series.

**Series C, 65% and D 35% Saturation.**

There was little to choose between these two series as the rate of infection was practically the same. The majority of the seedlings killed were, as shown in the graph observed after 10 to 15 days from the time of inoculation. This was before any signs of infection had occurred in series B. In each series 13/16 and 12/16 were killed respectively. The fungus was reisolated from both series.

Representative samples from the four series are illustrated in fig. 27.

The series in order of ascension are A, B, C and D. One control pot is shown on the left of each series.
Experiment No. III.

The following experiment was set up on March 3rd 1936, in order to test the effect of varying sand moisture on the incidence of Rosellinia attack on cacao seedlings. Four different sand moisture series were established as follows:

<table>
<thead>
<tr>
<th>Series</th>
<th>Moisture content % Saturation</th>
<th>Cacao seedlings Inoculated</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>95</td>
<td>16</td>
<td>4</td>
</tr>
<tr>
<td>B</td>
<td>75</td>
<td>16</td>
<td>4</td>
</tr>
<tr>
<td>C</td>
<td>50</td>
<td>16</td>
<td>4</td>
</tr>
<tr>
<td>D</td>
<td>25</td>
<td>16</td>
<td>4</td>
</tr>
</tbody>
</table>

In each series 8 pots were inoculated in a similar manner as in the previous experiment. Two uninoculated pots served as controls. The pots were watered on alternate days with complete nutrient solution and rain water. Sand moistures were kept constant by the method of constant weight.

The results obtained from this experiment were confusing as at a first sight they contradicted those obtained in Experiment II.

An examination one month after inoculation revealed the following information.

Series A, 95% Saturation.

Infection was of normal rapidity in this series and 11/16 of the inoculated seedlings were killed.

Series B, 75% and C, 50%.

Infection was not so rapid in these series. In B, 6/16 of the seedlings were killed and in C, 2/16 seedlings.

Series D, 25% Saturation.

No infection by Rosellinia was obtained in this series but half the number of the seedlings died from physiological wilt.

It was observed that the fluctuations in moisture content in the sand cultures were very great and as a result it cannot be claimed.
claimed that the sand moisture contents were maintained. This rapid fluctuation in moisture content may explain the different results obtained in this experiment compared with No. II. The greater incidence of the disease in the drier series of Experiment No. II was found to be correlated with greater quantities of the fungus, which filled the air spaces with dense masses of woolly mycelium after the fashion observed in worm burrows in the field. The greater rate of mortality in the drier series may be attributed to the fact that the fungus mycelium was able to travel faster through the soil, better aerated than in the wetter series, which showed less evidence of mycelial activity.

It is of interest to observe that Toovey (1935) in his Experiment No. 2, also found a greater mortality among his Hibiscus cuttings in the series of low soil moisture content.

Effect of Available Chemical Fertility.
Experiment No. IV.

The following experiment was set up on January 17th 1936 with the object of testing the effect of the lack of essential elements on the incidence of attack by Rosellinia Pepo on cacao seedlings. The experiment had to be abandoned 14 days after the seeds had germinated owing to the number of deaths caused by the fungus.

Two cacao seeds were sown in a 6" flower pot with sterile sand whose moisture content fluctuated from 50 - 75% Saturation. Five series were set up as follows:

Series.  
A Complete Nutrients  
B Lacking Nitrogen  
C Lacking Potassium  
D Lacking Phosphate  
E Lacking Calcium

In each series 12 pots were inoculated with a 14 day old culture of Rosellinia Pepo on a sweet potato slice, placed horizontally with/
with a seed at either end. Six uninoculated pots served as controls. The pots were watered on alternate days, with culture solution and rain water. The following is an analysis of the experiment one month after establishment.

![Figure 28](image)

**Table VII.**

<table>
<thead>
<tr>
<th>Series</th>
<th>UNINOCULATED</th>
<th>INOCULATED</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Failed to germinate</td>
<td>Dead</td>
</tr>
<tr>
<td>A</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>B</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>C</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>D</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>E</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Percentage</td>
<td>3.3</td>
<td>0</td>
</tr>
</tbody>
</table>

It was found that failure of infection in the inoculated series could be traced either to the drying out of the fungus inoculum or to failure of an establishment of contact between fungus and host. In no instance did a microscopic examination reveal any association between fungus and host in the roots of the plants which remained healthy in the inoculated series.

Fairs of seedlings taken from the same pot are shown in figure 28.
No mycelium of *K. Pepo* was found on the healthy seedling shown in the centre, although the other seedling from the same pot became infected. This evidence gave indication that the environmental factor was secondary in importance and that the presence of the fungus was all important. Fig. 29 shows that the seedlings could be attacked at any time after germination.

Several cases were found in the inoculated series, shown in Table VII, where the seeds failed to germinate due to attack by the fungus.

Although it was impossible to run the experiment long enough to test the effect of the lack of essential elements on the susceptibility of the host, it has at least been demonstrated that under the conditions of this experiment *Rosellinia Pepo* can behave as a serious parasite.

The experiment was continued on the control pots of the series at a later date when mineral deficiencies were expected to have a greater effect on susceptibility of the older plants to fungal invasion.

Four of the control pots were inoculated in the usual way with cultures of the fungus and two pots were left as a control on March 25th 1936. One month later examination showed that series/
series B (minus Ng) and D (minus P) were still healthy but that series A (Complete), C (minus K) and E (minus Ca) all showed marked leaf symptoms. This was not however connected with infection by the fungus as the controls were all equally affected.

The conclusion was reached similar to that by Toovey (1935), namely that the available chemical fertility plays no part in determining the incidence of infection. This conclusion is further supported by field observations on the disease.

**Inoculation Experiments in the Field.**

A successful inoculation experiment was performed on February the 14th 1936, on a tree in Field 7a at River Estate, an area in which normally no Rosellinia is found. Two large roots were carefully excavated in order to avoid damage and covered for a length of 18 inches with 12 cultures of the fungus grown for one month on Oatmeal agar. The inoculum was covered with damp cotton wool before replacing the soil. No further moisture was added in this experiment which was carried out during the dry season. On examination one month later the roots were observed to be covered with smoky grey mycelium. Microscopic examination of root sections failed to reveal any hyphae beyond the cork layer. Further roots, which had not previously been disturbed were examined three months later when it was found that successful penetration had taken place with the formation of white mycelial stars below the bark, characteristic of *Rosellinia Pepe*. In this respect the fungus differs from *Rosellinia necatrix* which according to Nattress (1926) only attacks the root system of its host by means of the fine roots.
IV. DISCUSSION.

The evidence submitted in the previous sections of this dissertation leaves little doubt that *Rosellinia Pepo* is a primary parasite. Observations in both the field and the laboratory have shown that whenever contact between parasite and host is established a parasitic invasion has followed, whether the host has been in the seedling stage or as a mature tree.

It is all the more puzzling therefore to find in the field that the parasite is confined in its activities to restricted areas. A study of the general distribution of the fungus in Trinidad and Grenada does not show any limitation due to climatic factors. Records have been obtained of estates situated from a few hundred feet above sea level up to over 1,200 feet with an annual rainfall which varied from 60 inches to over 120 inches.

With regard to soil texture, the disease has so far been observed only on alluvial silts and red soil, but not on clay. This restriction of soil types does not however solve the problem of the restriction of the disease to a patch within the soil type.

The soil moisture factor can act in several different ways according to whether the fungus is travelling over the surface or through the soil. In the former case excessively moist conditions with an abundance of undecomposed ground litter will undoubtedly favour the presence of the mycelial fans of the fungus, described by many recorders of this disease. South and Brooks (1912) state however that the fungus does not appear to grow in decaying humus. It therefore follows that if steps are taken to ventilate the cacao plantation both to reduce the relative humidity of the atmosphere and to encourage decomp-
decomposition of ground litter that the superficial spread of the fungus will be checked.

The second method of spread used by the fungus is through the soil which the writer has so far observed only where there is a sandy texture. Soils of this nature offer the optimum conditions of humidity and aeration for the spread of the fungus. The rate of spread through such soils has been shown experimentally to depend on the moisture content, an excessive amount checking the rate of progress.

The Rosellinia patches may be classified into two categories with regard to the biotic factor. We have one in which the patches are neglected, where the disease is allowed to run its full course unchecked save by natural barriers, and the other where the planter has made some attempt at eradication.

An example in the first category was described from observations made at River Estate in Trinidad. These showed that the fungus had a growth phase which covered approximately 10 years under these local conditions. The wave of fungal infection continued to pass on centrifugally and was still active at the margin in 1935, 10 years after the growth phase had ceased at the centre of the Rosellinia patch.

Examples of the second category may be taken from La Vega and La Veronica estates in Trinidad and from Tuileries estate in Grenada. The diseased patch at La Vega is of the same age as that at River estate. The number of infected trees however is only 20 compared with over 100 at River estate.

This difference is to be explained by the fact that at La Vega the original diseased trees had been uprooted and the area resupplied. The sandy nature of the soil enabled drying out to take place and most of the fungus infected material was killed through exposure. The presence of a few infected trees may be regarded as evidence that this process of drying out/
out of the fungus had not been performed thoroughly. La Veronica and Tuileries estates are examples where a thorough eradication of infected material was attempted with very successful results. Under these conditions, the planting of supplies was made possible within a year of the operations. This period should be compared with the 10 years required under natural conditions, where the fungus is left undisturbed.

It was not found possible to obtain any evidence that *Rosellinia Pepo* could exist under field conditions in the presence of a suitable host, without becoming parasitic. The success attendant on the use of tree root surgical methods, where the removal of infected roots also removes the disease emphasises the importance of the fungus as a factor in the disease complex. Further evidence of this was obtained from a field experiment where the roots of a healthy cacao tree were successfully inoculated with the fungus in a locality where the root disease was not normally present.

The result of these observations appears to show that the *Rosellinia* root diseases are of quite a different nature compared with those caused by other genera of fungi such as by *Fomes*.

Napper (1932) has investigated the incidence of root disease of rubber caused by *Fomes lignosus* under clean-cleared and uncleared conditions. His results showed that the amount of disease was less under a cover than on a clean weeded area. The presence of woody Crotalaria plants in the cover, susceptible to *Fomes* attack, was found to reduce the incidence of the disease on the rubber tree. Fawcett (1915) records *Rosellinia bunodes* on several shrubby cover plants in diseased coffee plantations in Porto Rico, but makes no statements as to whether the incidence of the disease was thereby reduced. *Rosellinia Pepo* appears to have a more restricted host range and/
and apart from cacao and nutmeg, cassava is the only plant which the writer has observed to become parasitised. The observations of others, that bananas, dasheen, pigeon pea and horse bean are attacked has not been confirmed.

De Jong (1933) describes experiments which appear to indicate that *Fomes lignosus* is only a weak parasite on rubber except under special environmental conditions which stimulate the growth of the fungus and impair that of the trees. A large number of trees were inoculated with pure mycelial cultures of *Fomes lignosus* and these developed a profuse growth of mycelium but in no case did decay follow and the rhizomorphs finally disintegrated. Even the wounding of inoculated roots did not promote infection. It was also observed that the decay caused by this fungus often stops on its own accord, the infected areas becoming surrounded by callus and finally healing completely.

In one respect only did the Fomes disease of rubber resemble that caused by *Rosellinia* of cacao. This was that the nature of the soil was found to act as a predisposing environmental condition, red soil and quartz sand being recorded as particularly favourable to the development of the fungus.

It would thus appear that the root diseases caused by species of the following different genera, *Armillaria*, *Fomes* and *Rosellinia* must each be studied as separate entities. It does not follow that the knowledge acquired after the study of any one member of this group will be of any assistance in elucidating problems connected with the physiology of parasitism of the others.
V SUMMARY.

1. An account is given of an attempt to gauge the factors, both in the field and in the laboratory, which control the physiology of parasitism of *Rosellinia Pepo*.

2. A description of the fungus in pure culture and proof of its parasitism is given.

3. An analysis of the soil showed no relation between the available chemical fertility and the incidence of the disease. This was confirmed by controlled inoculation experiments.

4. Similar negative information was obtained with regard to the percentage of organic matter and the C/N ratio. Laboratory experiments proved that the presence or absence of organic matter did not influence the pathogenicity of the fungus.

5. No relation was found between the soil reaction and the distribution of the disease in the field.

6. The disease was only observed on soils of light texture and this is stressed as being of importance in the study of the water relationships of the soil.

7. It was established in experiments conducted to test the relation between soil moisture and the incidence of infection of cacao seedlings by the fungus, that the rate of infection varied under different conditions of moisture content. No infection occurred in the water-logged soil series nor in the very dry sand series (25% Saturation).

8. A comparison is made between *Rosellinia* root diseases and those caused by species of other genera of fungi. The necessity for treating them as separate problems is stressed.
VI REFERENCES.


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A Scheme of Treatment for the Control of Fomes lignosus in Young Rubber Trees. Ibid, 4, No. 1, p. 34-38.


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(1913) Some root diseases of permanent crops in the West Indies. West Indian Bull., 12, p.479.

