OFFICIAL GRADING OF PROPRIETARY FUNGICIDES FOR TREATMENT OF MOULDY ROT DISEASE OF RUBBER TREES

BY

F. BEELEY

The primary object of this article is to place on record a history of the treatment of Mouldy Rot disease of the rubber tree particularly in relation to the establishment of an official approved list of proprietary fungicides which have been investigated and proved to be effective for the treatment of the disease.

The discovery of this disease of the tapping panel of rubber trees in 1916 presented to plant pathologists a special problem in that the tissues affected by the disease are as delicate, if not more so, than the fungus which causes the disease.

It was early discovered that the delicate cambial tissues exposed by the tapping process were particularly sensitive to the contact of minerals or liquid sprays containing dissolved mineral salts. The use of organic compounds, represented by such proprietary tar-acid mixtures as Jodelite, Dougalite, Solignum, Noxo, Brunolinum and Coal tar, normally used for the preservation of structural timber, was investigated and it was found that, when applied neat, they caused considerable damage to the tapped surface of the tree so that weaker mixtures made by emulsifying these products in water with soap became the vogue. Proprietary preparations of emulsified tar oils under various names were soon available on the Malayan market.

The Rubber Growers' Association (Third Malayan Report 1919) issued a report upon tests of a number of fungicides carried out by Sanderson and Sutcliffe. In this report it is pointed out that the strength of fungicide required to kill the Mouldy Rot and Black Stripe fungi in laboratory tests bears no relation to the strength of the same fungicide required for use in the field.

In this report a list of thirteen desirable points in relation to fungicides is given as follows:—

1. Its use should be possible in strength sufficient to kill or prevent the development of the particular organism against which it is used.

2. The planter should be able to tell by a glance that the substance has been applied.

3. It should not have a deleterious effect on the finished rubber.

4. It should not burn the tissue.

5. The penetrative power should be sufficient to be effective but not sufficient to cause wounds. (The thickness of the bark when the application is made is an important factor here.)

6. The application should be simple and rapid, entailing a minimum of expense.

7. The planter should be able to rely on different samples being of constant composition or very nearly so; this applies more particularly to liquids which are used neat or undiluted and which penetrate deeply.

8. The preparation must be easy to handle.

9. Its use must be possible daily (applies especially to liquids used as daily wash) without injurious effect on bark or wood.

10. If used in considerable dilution it is better if miscible with water.

11. It should not coagulate latex.

12. The initial cost must be reasonably low.

13. Re (12) if the initial cost is high this may be counter-balanced if the preparation can be used with effect in more dilute mixture than a cheaper preparation.

Further work on the control of Mouldy Rot is found in the Bulletins of the Department of Agriculture, S.S. and F.M.S.

Sharples, Belgrave, Norris and Ellis (Bulletin 31, 1920) suggest the use of an emulsified form of Brunolinum known as Brunolinum plantarium, while South and Sharples (Bulletin 37 1925) point out the danger of bark damage from use of Jodelite or too strong mixtures of Brunolinum and tar or water-oil emulsions. They also state that "laboratory tests carried out by testing cultures against percentage strength of disinfectants are useless and misleading when used as a basis for field tests". An account is also given of elaborate field tests carried out by the proprietors of the preparation "Agrisol" under the supervision of officers of the Department of Agriculture as a result of which the use of a 20 per cent mixture of Agrisol in water was widely adopted for control of Mouldy Rot in Malaya. It was on the basis of these tests that Sharples, on assuming his appointment as Head of the Pathological Division of the Rubber Research Institute in 1931, formulated a scheme for the testing of proprietary fungicides for use in control of Mouldy Rot with a view to providing a list of preparations which can be used with safety and give a high degree of efficacy.

Owing to the generally unsatisfactory position, particularly in regard to the supply of suitable fungicides to rubber small-holders

a meeting of the Heads of the Pathological Divisions of the Department of Agriculture and the Rubber Research Institute was arranged to discuss the standardization of fungicides for treatment of diseases of the rubber tree. As a result it was agreed that standardization was not sufficient in itself and that controlled field tests were essential. The problem was also referred to the Director of the Imperial Mycological Institute, London, who expressed his agreement with these views; he also mentioned that in Germany there was a scheme operating in different localities for field testing of fungicides which if found satisfactory were included in a list of approved fungicides.

After further discussion at an Inter-Departmental Conference of the Department of Agriculture, the Co-operative Societies Department and the Rubber Research Institute, it was agreed that considerable preliminary investigation was necessary before any recommendations could be made.

A list of the proprietary fungicides available in Malaya for the control of rubber tree diseases was drawn up, and a scheme proposed for the investigation of these in the Rubber Research Institute.

This scheme after approval by the Board of the Institute was submitted by the Director of Agriculture to the Government of the Federated Malay States and approved. As a result, the following Notice was published in the F.M.S. Government Gazette, Vol. 23 No. 7 of the 27th March, 1931, page 713.

NOTICE.

The attention of the Department of Agriculture has been directed to the necessity which exists for the standardization of disinfectants which are approved for use in the treatment of Mouldy Rot on rubber plantations.

2. From the date of this notice therefore in all notices served by the Department of Agriculture under the Plant Disease Prevention Ordinance for the treatment of Mouldy Rot only preparations will be specified which have been subjected to test by an approved authority and proved to be effective.

3. At present the list of such preparations includes:—

1. Agrisol
2. Izal—in certain cases.

4. The Department will be prepared to extend the list to include any other preparations which have similarly been proved to be effective, by approved authorities. It should be pointed out that certain other preparations in common use on estates have also been reported to give satisfactory results but, as these have not yet been subjected to systematic tests, they are not at present included in the list.

5. The approved authority for carrying out such tests for Malaya will be the Rubber Research Institute and the carrying out of such tests will in future be undertaken by the Rubber Research Institute; applications from proprietors of fungicides for the performance of such tests with a view to their inclusion in the list should be addressed to the Director of the Rubber Research Institute accordingly.

6. The tests will be made strictly in accordance with priority of receipt and also in accordance with the capacity of the Institute to deal with them when received.

7. All supplies of disinfectants required for such tests must be furnished without charge by the firms applying for such tests while the actual expenses involved in carrying out the tests must also be borne by the said firms.

8. For further information regarding tests application should be made to the Rubber Research Institute.

9. Proprietors of fungicides and their representatives in Malaya are further notified that preparations which, on samples supplied, have passed the tests prescribed by the Rubber Research Institute will only be admitted to the list provided that the proprietors are willing and able to give a guarantee that the formula for the composition of their preparations will remain unchanged.

10. Should any proprietor of an approved fungicides desire to make any change in the composition of the fungicide he must notify the Director of Agriculture and submit samples of the preparation in its new form for further tests if required to do so. Failure to do this may entail the removal of an approved preparation from the list.

When applications are received from the proprietors of fungicides for the testing of a particular product, the conditions in the above notice are stated in a letter sent to the applicant. In addition, the applicant is informed that the field tests can be carried out only when suitable diseased areas can be found during the rainy seasons, so that no guarantee can be given as to when the results of such tests will be available. The maximum cost of carrying out the tests is $200/- (Straits) which the applicant is requested to deposit with the Institute before the tests are undertaken.

The applicant is also informed that the Institute reserves the right to publish the results, although the usual procedure is only to publish the name of the product, if satisfactory, in the list of fungicides approved for the treatment of the disease.

Since the adoption of this scheme the Institute has tested over forty fungicidal preparations of which only a small proportion have successfully passed the tests and been approved for a place on the White List which at the present date is as follows:—

	Name.	Date gazetted F.M.S.	Concentration to be used.
1.	AGRISOL ...	27. 3.31	15 to 25 per cent. in water
2.	IZAL ...	27. 3.31	3 to 5 per cent. in water
3.	BLACK CYLLIN ...	7. 6.32	5 to 10 per cent. in water
4.	KILLGERM ...	17. 6.32	5 to 10 per cent. in water
5.	KILSOL RED ...	17. 6.32	5 to 10 per cent. in water
6.	BRUNOLINUM PLANTARIUM	17. 6.32	10 to 15 per cent. in water
7.	AGRISOL WHITE ...	6.10.33	10 to 15 per cent. in water
8.	CARGILLINEUM 'B' ...	28. 7.33	To be used neat
9.	DURYCOLIUM ,,,	6. 3.36	To be used neat

It is also of interest to record that a session of the Third Imperial Mycological Conference held in London in 1934 was devoted to a discussion on the "Methods of Standardization of Insecticides and Fungicides". An account of the opening speech quoted from the official report of the Conference reads as follows :--

"Dr. Gussow (Canada) in introducing the subject gave an outline of the "procedure adopted in Canada, the United States and Germany. In Canada "an Act for the regulation of the sale of fungicides was passed in 1927, the "primary object of which was to prevent the sale of fraudulent products. All "products have to be registered, the well-known and tested substances are "granted registration immediately; those tested to a limited extent but known "to be effective are allowed registration temporarily; for those untested, ex- "perimental data are requested and tests arranged, whilst the worthless materials "are refused. Applications for registration require to be accompanied by a "fee of $20 gold, and the name and percentage of all ingredients have to be "stated. Every registration expires on 31st December. In the United States "similar control is established. In Germany the procedure involves duplicated "tests the data from which are referred to an official body, which rejects or "permits the use of the material. The ingredients are either disclosed or com- "municated confidentially. The testing of fungicides by field tests gives the most "satisfactory results, but it requires money and time, and it is very desirable "that research should be undertaken in connection with the laboratory testing "of such materials."

Mr. Sharples, who represented the Rubber Research Institute, gave an account of the testing of fungicides in Malaya which was greatly appreciated by other delegates.

Below, an account is given of the laboratory work which led to the present method of field testing of fungicides.

PART II

Proposed Tests for Fungicides

MICROSCOPICAL

If in emulsion, globule size is of importance and it is found that the smaller the size of globule the greater the fungicidal effects, the more permanent the emulsion and the greater the ease of mixing with water. Large globules tend to coalesce easily to give an oily film detrimental to living plant tissues.

If in oil or paste form the physical properties are noted for ease of use and covering power.

If in powder form, fineness and free-running non-aggregating properties are noted.

Globule size for an emulsion is usually 1—2 μ and not greater than 3 μ—*i.e.* globules in continuous Brownian movement when in a water film. Fine powders should have the following particle sizes—Min 3—6μ; Bulk 12—18μ; Max 30 μ. Larger particles, like

grit, are considered unsatisfactory though a few large particles often assist in preventing aggregates and in maintaining free-running powder form, as all fine powders tend to aggregate on standing in bulk.

Chemical

The product is examined to determine the nature of the fungicide and its concentration from which the approximate strength for use in the field can be determined *e.g.* a fluid which on treatment with 20 per cent sulphuric acid shows a content of 40 to 50 per cent of tar acids will probably be successful in the field at a concentration of 7 to 15 per cent in water. If 90 per cent of tar acid is found then a strength of about 3 to 5 per cent will be necessary in the field.

Only one paste fungicide the composition of which was supplied by the makers has been tested by us.

If the product is too alkaline or acid it is likely to be detrimental to plant tissues.

Penetration Tests on living Bark

The bark of a rubber tree is excised to imitate light, medium and deep tapping over an area of at least 6 x 2 ins. and the fungicide is applied at various predetermined strengths to effect (a) slight penetration of the bark (b) medium and (c) deep or harmful penetration and killing of the bark. The lethal strength is noted. Two to four days are usually sufficient to determine this factor.

Any damage done to the bark by volatile components or invisible fluids is often difficult to detect, but a good test is to make a knife-edge cut into the remaining bark across the excised surface and to note the condition of the latex or juice which exudes. A thin, almost colourless fluid indicates danger and it is often found that the cambium beneath is discoloured—being more sensitive than the bark (cortex) to the gas or fluid—and slowly dies. A thin, milky fluid shows that the maximum concentration has been reached, while a thick, creamy latex indicates a healthy condition of the bark.

Surface effects on the bark are noted, since a fluid which kills, shrivels and blackens the outer cells is hardly desirable whereas some fungicides appear to improve the condition of the outer cells and to promote bark renewal.

It should be noted that cells near the cambium are far more sensitive to fungicides than cells further out in the cortex. Therefore the test of a fungicide to be applied to the panel after tapping is that it should only penetrate lightly on excisions made close to the cambial region.

On the outer hard bark most of the fungicides may be used at higher concentrations without danger to the inner bark.

The visual penetration four days after application of the fungicide should be not greater than one millimetre, preferably less than half a millimetre.

The tests are carried out under reasonable shade as direct sunlight on the exposed tissue may cause considerable damage which may be erroneously attributed to the fungicide. Dark mixtures particularly appear to penetrate more deeply into bark in sunlight than they do under shade, owing to the heat absorbing properties of the black surface.

The use of mixtures of fungicidal coal-tar oils, in an asphalt-kerosene medium, on the tapping panel has been abandoned on account of the great absorption of heat on the black surface particularly during wintering, which follows so quickly on the rainy Mouldy Rot season.

Laboratory Tests

Much useful work has been done by various research institutions throughout the world to find a satisfactory laboratory means of determining the fungicidal, bactericidal or germicidal value of a preparation. None of these have yet been considered very satisfactory. Though the Rideal-Walker test has found considerable popularity as a means of determining bactericidal value it does not follow that the product tested will give the same efficacy when used upon living plant or animal material. Recently a compound of high "R.W." coefficient proved useless as a fungicide on rubber trees whereas one of comparatively low "R.W." coefficient proved very efficient in control of Mouldy Rot.

It has been found that, with cultures of the Mouldy Rot fungus, *Ceratostomella fimbriata,* in agar medium, the strength of say a tar-acid emulsion required to kill the fungus was so low compared with that required in the field, that no indication could be obtained from such culture tests which would enable the value of a specific compound, for the purpose of control of Mouldy Rot or indeed any other disease of the tapping panel, to be judged. It could be considered as giving merely a comparison of the lethal efficiency of the various compounds in that medium.

An alternative laboratory method was also investigated. This consists of inoculating clean strips of living bark 6 × ¼ in. in sterile tubes with the Mouldy Rot fungus. The fungus is allowed to develop on the cambial side of the bark strip for seven days, after which the thick-walled resting spores are found to be present in great numbers. The strips of bark are then immersed in beakers containing various strengths of the fungicide under test. Scrapings

of the fungus and tissue are then taken from the strips of bark at short intervals of time, usually after immersion for 2, 5, 10, 15, 20, 25 and 30 minutes, and the scrapings are washed and inoculated into a tube or plate of nutrient agar.

This time-immersion method of test gives results considerably more comparable to the field tests than the Rideal-Walker test, but is still so inferior to the field-test method that it has been decided that the latter should be considered the only suitable final test of the efficacy of a fungicide for the control of the disease.

Field Tests

In carrying out field tests the following practice is adopted.

Choice of Site

An area of rubber carrying a heavy infection of Mouldy Rot is selected at the most convenient distance from the laboratories.

Lay-out of Plots

Trees showing mouldy tapping panels are numbered by stencil and divided into plots of 25 trees, each having 100 per cent infection. The condition of neighbouring fields of trees serve as controls to indicate the normal progress of the disease when no treatment is given.

In the case of a tar-acid emulsion the preliminary laboratory tests will have given some indication of the probable strength of the fungicide required for the field tests—say 10 per cent. Four plots of 25 trees are then treated with a different concentration around this figure say 5, 10 and 15 per cent in water. Treatment by spraying each tapping day is given and a close watch is kept for the appearance of symptoms of bark burning or too deep penetration of the fluid, together with a daily record of the number of trees showing mould. Within two weeks it can usually be discovered that the lowest strength is useless for killing the fungus while possibly the highest strength, though killing the fungus within the first few days, also shows signs of over-penetration and consequent bark burning. Thus the early field tests give the probable minimum strength required to kill the fungus and the maximum safe strength at which the fungicide can be applied to the bark without injury.

The plot receiving only a 5 per cent strength of fungicide will then be treated with a 7.5 per cent solution and the " 15 per cent " plot with a 12.5 per cent solution, while a further main plot of 100 trees will be treated as in estate practice with an average

strength of fungicide, say in this case a 10 per cent concentration, for one month, records being taken each day of the number of trees mouldy and conditions of the renewing bark before, during and after experiment.

During these tests no special effort is made to prevent infection from neighbouring untreated trees, and each tapper may tap a number of treated and untreated trees at will so that the test is as thorough as possible.

When treatment is stopped, a note is usually made of the time required for the reappearance of mould on the treated trees.

Any preparation which fails to control the disease during a period of one month's treatment is considered as unsuitable for a place on the White List of fungicides for control of the disease.

On the other hand, fungicides which cause bark burning at or near the strength required for killing the mould are also regarded as unsuitable. In fact this has been the chief reason for rejection of many proprietary products.

As a point of interest a record, published in the Annual Report of the Rubber Research Institute 1934, p. 94 of an experiment to demonstrate the efficiency of the various types of fungicides used for the control of the disease in heavily infected small-holdings rubber areas is given below. The materials used were Killgerm (a proprietary fungicide) and "Linsocresyl" (prepared in the Pathological Division), as representatives of the tar-acid emulsions; Cargilineum 'B' mixture and Agrisol Paste (both proprietary fungicides), as representatives of the greasy waterproof pastes; Singapore Tar and Asphalt-Solar Oil plus 10 per cent Brunolinum, as representatives of the black water-proof paints which find a fairly wide use. A resin-spirit mixture in common use in Netherlands India is also included but, during the wet weather, was found seriously to damage the bark, although on the advent of dry weather early in December further applications proved to have no ill-effects on the renewed bark. The results are given in tabular and graphical form below. The initial degree of infection in each plot was 100 per cent. The condition of the renewed bark was noted weekly and the flow of latex was judged by pricking the renewing bark with a knife point. It has frequently been noticed that, if the fungicide has damaged the bark, even only slightly, the latex flow is poor and the latex produced is of a thin watery nature. The results indicate the superiority of the tar acid emulsions.

Guarantee

A proprietor whose preparation gives satisfactory results in the above tests is required to give a guarantee that the composition

Fungicide Tests in Control of Mouldy Rot

Fungicide	Application	When clear of disease	Condition of bark	Latex
1. Killgerm 10 per cent in water	Daily	After 16 paintings	Very good condition	Good
2. Linsocresyl 10 per cent in water	Daily	After 17 paintings	Good condition	Good
3. Cargilineum applied neat	Every 4th day	After 6 paintings	Fair to good	Good
4. Resin Spirit applied neat	Every 3rd day	After 6 paintings	Hard crust formed. Bark damaged in wet weather. Renewal of bark good in dry weather.	Fair to Good
5. Singapore Tar applied neat	Every 4th day	After 7 paintings	Burning where the bark is tapped deeply; otherwise good bark renewal.	Fair
6. Asphalt-Solar Oil applied neat	Every 3rd day	After 7 paintings	-do-	Fair
7. Agrisol Paste applied neat	Every 4th day	After 6 paintings	-do-	Fair

and formula of the preparation will remain constant and, should any alteration take place, he is required to notify the Director of Agriculture at once, so that, if necessary, further tests can be made. If these conditions are complied with the preparation is gazetted by Government and placed on the official list of fungicides recommended for use in control of the disease.

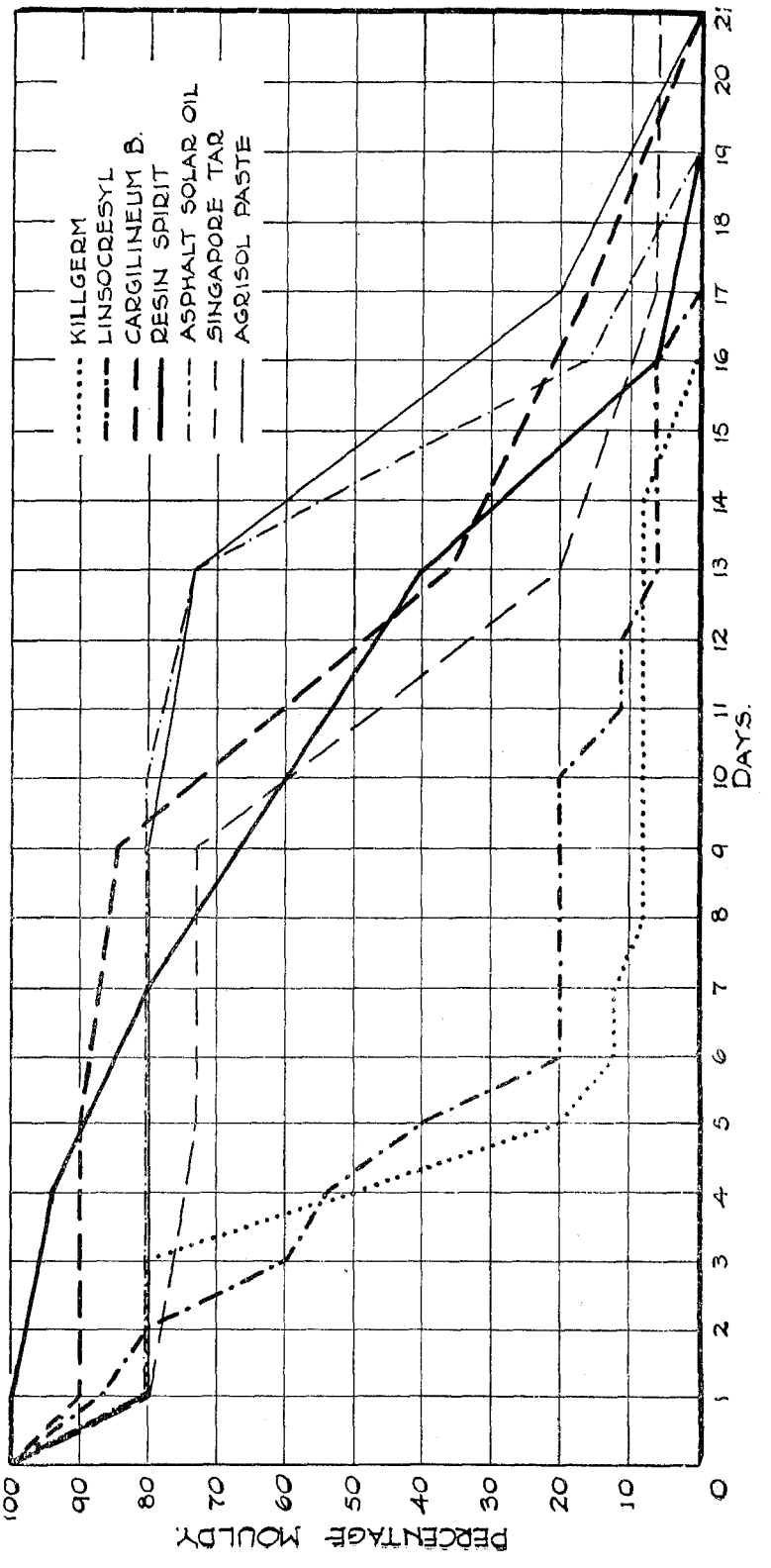
Nomenclature

In conclusion a few words on the terminology used in this article may serve to clear up misunderstandings. In Webster's International Dictionary the following meanings of the words Fungicide, Antiseptic and Disinfectant may be found.

"Fungicide (n)—Any substance which destroys fungi"

"Antiseptic (n)—An antiseptic substance. That which may be used to destroy bacteria with little or no harmful effect on the living body."

"Disinfectant (n)—A substance adopted for destroying the bacteria in, and rendering harmless and inoffensive, objects, places


FUNGICIDE TESTS - SETAPAK.
NOV. - DEC. 1934.
KILLGERM
LINSOCRESYL
CARGILINEUM B.
RESIN SPIRIT
ASPHALT SOLAR OIL
SINGAPORE TAR
AGRISOL PASTE
PERCENTAGE MOULDY
100
90
80
70
60
50
40
30
20
10
0
1
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3
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5
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DAYS.

or materials containing putrefactive or pathogenic bacteria. A disinfectant differs from an antiseptic in not being intended for use upon the living body; hence many substances not applicable as antiseptics may serve as disinfectants or stronger solutions of antiseptic substances may be employed."

If, in the above meanings, the word "rubber tree" is regarded as the living body, the correct use of the above terms in plant therapeutics can better be understood.

A fungicide is a substance which will destroy fungi without injury to plants upon which the fungi are growing.

A disinfectant is a substance which will destroy fungi and bacteria irrespective of its action upon living plants. As a rule disinfectants are of concentrated form and would almost certainly be fatal to the plant.

In general a substance used in low concentrations may be used as a fungicide while the same substance in higher concentration becomes a disinfectant and plant destroyer. The margin between these two strengths may be regarded as the margin of safety and the greater this margin the more safe the material for use as a fungicide. This is of importance when it is remembered that in wet weather it is more difficult to control Mouldy Rot and there is a great tendency to use stronger fungicides; if there is only a small safety margin this is impossible without having considerable trouble from bark burning.

In the White List of fungicides given previously the normal strength for use of the fluid, and the maximum strength which it is reasonably safe to use under abnormal conditions, *i.e.* during wet weather, are given.

Summary

A brief history is given showing the reasons which led to the official testing of substances used for the control of Mouldy Rot and the provision of an official list of fungicides for use for such purpose in Malaya.

A description is given of laboratory and field tests formulated for the purpose of testing these substances.