T. Pl.-ziekten 67 (1961): 21–24

THE TOXICITY OF SOME FUNGICIDES TO CONIDIA OF *GLOEOSPORIUM FRUCTIGENUM* F. *CHROMOGENUM*[1]

Met een samenvatting: De toxiciteit van enkele fungiciden voor de conidiën van Gloeosporium fructigenum *f.* chromogenum

BY

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INTRODUCTION

Anthracnose of olives is a wide-spread plant disease caused by various species of the *Glomerella* group of fungi (GORTER, 1959). In South Africa it is caused by *Gloeosporium fructigenum* Berk. f. *chromogenum* Gorter (GORTER, 1956, 1959). This fungus can be successfully combated by spraying with home-made Bordeaux mixture (GORTER, 1960).

For treating fruit tree diseases in this country home-made Bordeaux mixture is now rapidly being replaced by factory-made fixed copper sprays and organic fungicides. It was therefore considered desirable to test these fungicides against olive anthracnose.

Comparative field tests would require very large and uniform olive groves. In South Africa there are no such plantings where anthracnose is troublesome. This is because the disease is invariably found only near the coast and here the soil usually slopes and the fertility varies. The fungicides had therefore to be evaluated in spore germination tests only.

METHODS AND MATERIALS

A modification of the slide-germination technique recommended by the American Phytopathological Society (1943, 1947) was used.

Each fungicide was mixed with distilled water and diluted. The dilutions contained respectively 10,000, 1000, 100, 10 and 1 part per million of the fungicide. According to need further dilutions were interpolated to obtain germination percentages between 0 and 100.

Three 3 × 1 inch glass slides, each with two concavities, were cemented with Canada balsam to a 3 × 3 inch glass plate, giving six depressions per plate. This is a modification of the method of HARTSUIJKER (1940). A solid glass rod was used to put one drop of the fungicidal solutions into each of the cavities. The plates were then dried in an oven at 50 °C. Distilled water was used as a control.

A spore suspension was made by shaking distilled water with 2–3 week-old cultures of the test fungus for one minute, and filtering the resulting spore suspension through glass wool to remove mycelium fragments. It was diluted until no more than 10 spores could be found in each micrometer frame of a

[1] Accepted for publication 1 Nov., 1960.

net-oculair at a magnification of 160 ×. The spore suspension was then mixed with an equal volume of a stimulant consisting of 0.1 per cent dextrose and the mineral salts magnesium sulphate, ammonium chloride and potassium dihydrogen phosphate in 2.0 millimol concentrations. Although opinions differ on the advisability of adding a stimulant (TOMKINS, 1932; WILCOXON & MCCALLAN, 1934; HARTSUIJKER, 1940; HORSFALL et al., 1940) there was no choice in this case, because the spores of the olive antracnose fungus germinated very poorly in distilled water alone. This was similar to what LINN (1945) found for *Glomerella cingulata.*

With a graduated pipette 0.08 cc of the spore suspension was put in each concavity of the prepared plates. The cultures were then incubated at 26 °C – about the optimum temperature for growth of the fungus – for 8 hours and then examined for germination of spores. It is usual to incubate such cultures at least 24 hours before examination, but with *Gloeosporium fructigenum* f. *chromogenum* secondary spores are formed within 24 hours, so a shorter period of incubation is necessary. According to WELLMAN & MCCALLAN (1942), the germination results are just as accurate after 6 or 12 hours as after 24 or 48 hours, provided the temperature is kept constant; the importance of the temperature increases as the observation time decreases.

Fifty spores in each culture drop were examined and the number that had germinated was noted. As four replicates of each fungicide concentration had been prepared concurrently, 200 spores were examined for each dilution. This number is adequate for the degree of accuracy aimed at in these experiments (BLUMER & KUNDERT, 1950).

The following copper fungicides were compared: Bordeaux mixture ("Capex" brand), copper oxychloride ("Cupravit") and cuprous oxide ("Perenox"). In addition five organic fungicides were tested viz. ferbam ("Fermate") maneb ("Manzate"), zineb ("Parzate"), captan ("Flit 406") and dichlone ("Phygon XL") of which the first three are dithiocarbamates.

RESULTS AND CONCLUSIONS

For each fungicide the germination figures for the various dilutions were multiplied by a factor $\frac{100}{x}$ in which x was the mean percentage of germinated spores in the controls. The percentage of non-germinated spores obtained in the final dilution series for each fungicide was used for the expression of the response to these fungicides in toxicity curves. These curves were drawn on ordinary graph paper after converting the percentages of non-germinated spores into probits – the probability units devised by BLISS (1935) – and plotting their values against the logarithms of the corresponding dilutions. From these dosage-response curves the E.D. 50 as well as the E.D. 95 were determined for each fungicide. Their values can be found in table 1.

The figures in the table show that there are slight but noticeable differences in the effects of the three copper preparations. Copper oxide was the best and copper oxychloride the worst. Of the three dithiocarbamates ferbam was the most effective and zineb the least. Zineb was even less toxic to the spores than was any of the copper fungicides. This is an interesting result as a similar

TABLE 1. E. D. Values of eight different fungicides for preventing germination of *Gloeosporium fructigenum* f. *chromogenum* conidia.
E.D.-waarden van acht verschillende fungiciden ter voorkoming van ontkieming van conidiën van Gloeosporium fructigenum *f.* chromogenum.

Fungicide	E.D. 50 Parts per million *Delen per miljoen*	E.D. 95 Parts per million *Delen per miljoen*
Bordeaux mixture/*Bordeauxse pap* . .	331	2,750
Copper-oxychloride/*Koper-oxychloride*	502	6,760
Cuprous oxide/*Cupro-oxide*	191	1,410
Ferbam	0.22	0.49
Maneb	468	832
Zineb	955	4,680
Captan	32.4	123
Dichlone	66.1	302

relationship between zineb and Bordeaux mixture was noted in spray trials for the control of olive anthracnose in Italy (GRANITI, 1956). The results obtained with maneb in the germination tests were more or less of the same order as those of the copper fungicides. The remaining organic fungicides, captan and dichlone were both more effective than the copper sprays but not as effective as ferbam. Thus the only preparations worthy of further testing under field conditions are ferbam, captan and dichlone.

SUMMARY

The comparative efficacy of a number of fungicides was tested on the conidia of *Gloeosporium fructigenum* Berk. f. *chromogenum* Gorter, the cause of olive anthracnose in South Africa. The tests were based on the slide-germination technique recommended by the American Phytopathological Society and modified to suit local conditions. It was necessary to use a stimulant to encourage germination.

The tests showed that cuprous oxide was slightly better than Bordeaux mixture but that copper oxychloride and zineb were not as good. Captan and dichlone were appreciably more effective than Bordeaux mixture. Ferbam was the most effective of all the fungicides tested.

SAMENVATTING

Wegens de moeilijkheden welke bij praktijkproeven voor het gelijktijdig vergelijken van een aantal fungicide spuitmiddelen in plaatselijke boomgaarden van olijven ondervonden worden, werd de doeltreffendheid van enkele nieuwere koper- en organische schimmeldodende preparaten voor het bestrijden van *Gloeosporium fructigenum* Berk. f *chromogenum* Gorter, de oorzaak van de olijf-anthracnose in Zuid-Afrika, in sporekiemingsproeven bestudeerd.

De gevolgde techniek was met enkele wijzigingen gebaseerd op een door de Amerikaanse Fytopathologische Vereniging aanbevolen methode. In plaats van gewone objectglazen werden echter vierkante glasplaatjes gebruikt, waarop naast elkaar drie objectglaasjes, elk van twee uithollingen voorzien, met

canadabalsem vastgekleefd waren. Om van een goede en gelijkmatige ontkieming van de sporen verzekerd te zijn, was het noodzakelijk een stimulans toe te voegen, waarvoor een oplossing werd gekozen die voor *Glomerella cingulata* doeltreffend was bevonden. Waarnemingen over de ontkieming werden na een tijdsduur van acht uren verricht om te voorkomen dat de vorming van secundaire sporen het kiemingsbeeld vertroebelden.

Drie koperpreparaten en vijf organische fungiciden werden aan de toets onderworpen. De verkregen resultaten zijn af te lezen uit tabel 1. Hoewel het verschil in reactie tussen de drie koperverbindingen gering is, werd er toch een duidelijke aanwijzing gevonden, dat cupro-oxide iets sterker en koper-oxychloride iets zwakker op de sporen inwerkt dan Bordeauxse pap. Bij de dithiocarbamaten deden zich echter opvallender verschillen voor. Zo bleek ferbam een uiterst effectief middel om de sporekieming bij *G. fructigenum* f. *chromogenum* te onderdrukken en is het zelfs het meest toxische van alle onderzochte preparaten. De maneb- en zineb-preparaten waren veel minder effectief; zineb reageerde zelfs zwakker dan Bordeauxse pap. Captan en dichlone daarentegen onderdrukten de kieming weer veel sterker dan Bordeauxse pap, maar niet zo effectief als ferbam.

Van de onderzochte preparaten komen dus alleen ferbam, captan en dichlone in aanmerking voor verdere praktijkproeven om de anthracnose van olijven te bestrijden.

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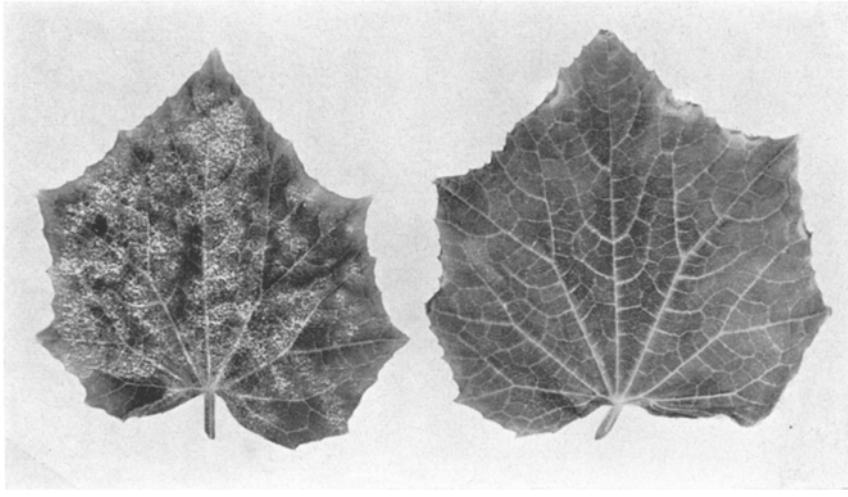

FIG. 1. Powdery mildew on cucumber leaves.
Right: Leaf from plant sprayed with procaine HCl 1000 p.p.m. on first and second day after inoculation.
Left: Leaf from control plant, sprayed with water.

Echte meeldauw op komkommerbladeren.
Rechts: Blad van plant, op de eerste en tweede dag na de inoculatie bespoten met procaïne-HCl (1000 d.p.m.).
Links: Blad van controle-plant, bespoten met water.