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Antifungal Activities of some Phenolics and Related Compounds to three Fungal Plant Pathogens

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Many phenolics have antimicrobial activity in vitro and in vivo (BYRDE 1963, RICH and HORSFALL 1954, BARUAH and CHAKRABARTY 1969, OKU 1958) and have been used for plant disease control (BEAN et al. 1967, KEITT et al. 1939, MARSH and BUTLER 1946). In the present study the relative toxicities of some phenolic compounds to three different plant pathogenic fungi were studied.

Material and Methods

Helminthosporium oryzae, *Alternaria solani* and *Curvularia lunata* (all brown pigment-
ed) were used as test fungi. The PDA medium used contained extract from 200 g of peeled
potatoes, 20 g dextrose and 20 g agar made up to 1000 ml with distilled water. For extra-
cellular enzyme preparations, Sabouraud's broth (MCLEAN and COOKE 1952) was used. Cultures
were maintained in PDA slants. Spore suspensions were prepared from 7—12 day old 9 cm
plate cultures, suspending the spores in 25 ml of cold (5 °C), sterile, distilled water per plate.
Fresh solutions of test compounds were prepared for every experiment and stored if necessary
at 5 °C.

For germination tests cavity slides were used. Solutions of 0.02 m strength were prepared;
addition of one drop of spore suspension to one drop of solution on the slide made the final
concentration 0.01 m. The slides were placed in a petri-dish humidity cabinet and incubated
at 28 ± 1 °C. A solution of malic acid with pH 2.4 was used as low pH control.

To test the effects on growth and sporulation, solutions of test compounds were added
to sterilized PDA media in order to make the final concentration 0.01 m. These petri dishes
were inoculated with spore suspensions (1 ml/plate), incubated at 28 ± 1 °C and observed
after 7 days. Growth was measured and sporulation estimated using a haemocytometer
in comparison with control plates.

Extracellular enzyme extracts were prepared from 10 day old Sabouraud's broth cultures
with usual methods. Effects on tissue macerating enzymes were estimated with potato disc
methods and those on tyrosinase and laccase according to DASGUPTA and VERMA (1961).

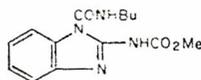
- determination of corrosion rates.
 57 63414b Legislation on the release of physiologically hazardous heavy metals from enamels.
 59 Air Pollution and Industrial Hygiene.
 62 64318k Evaluation of 1,3-butylene glycol as a safe and useful ingredient in cosmetics.
 64 64605b Thermal analysis of some nonprescription acids.

5—AGROCHEMICALS

Available in the computer-readable products *Chemical-Biological Activities (CBAC)* and *Food and Agricultural Chemistry*

RUTH L. BUSBEY AND HAROLD M. SELL

- 69078s Results of fungicide testing. Kobakhidze, D. M.; Batalova, T. S. (USSR). *Zashch. Rast. (Moscow)* 1973, (9), 32-4 (Russ). A review with no refs. The fungicidal effects of 51



prepn., tested in field expts. in various geographic regions of the USSR, are discussed, with special emphasis on benomyl (I) [17804-35-2], Quinolate V-4x, and Vitavax-thiuram [37244-87-4].
 69079t Analysis of herbicide residues by gas chromatography. Cochrane, W. P.; Purkayastha, R. (Plant Prod. Div., Canada Dep. Agric., Ottawa, Ont.). *Toxicol. Environ. Chem. Rev.* 1973, 1(3), 137-268 (Eng). A review with 425 refs.

69080m Bacterial exoenzymes toxic for insects. Proteinase and lecithinase. Lysenko, O. (Inst. Entomol., Czech. Acad. Sci., Prague, Czech.). *J. Hyg., Epidemiol., Microbiol., Immunol.* 1974, 18(3), 347-52 (Eng). A review with 20 refs. on the toxicity of bacterial proteinase [9001-92-7] and lecithinase [9013-93-8], to insects.

69081n Recent trends in pesticides in Japan. Murata, Michio (Chiba Univ., Chiba, Japan). *Kagaku Kogyo* 1974, 25(12), 1329-38 (Japan). A review with 7 refs. on the pesticide prodn. in Japan.

69082p Pesticides and safety. Misato, Tomomasa (Inst. Phys. Chem. Res., Japan). *Kagaku Kogyo* 1974, 25(12), 1339-47 (Japan). A review, with no refs., on pesticides in relation to ecol.

69083q Future insecticides. Fukami, Junichi (Inst. Phys. Chem. Res., Japan). *Kagaku Kogyo* 1974, 25(12), 1348-53 (Japan). The development of new insecticides in relation to ecol. problems was discussed; no refs.

69084r Recent trends in pesticide technology. Kurono, Hitoshi (Nippon Noyaku K.K., Japan). *Kagaku Kogyo* 1974, 25(12), 1354-60 (Japan). A review with 23 refs.

69085s Technical development of herbicides in Japan. Takita, Kiyoshi (Kumiai Chem. Ind. Co., Ltd., Tokyo, Japan). *Kagaku Kogyo* 1974, 25(12), 1369-73 (Japan). A review with 26 refs.

69086t Plant-growth regulators. Takahashi, Nobutaka (Fac. Agric., Univ. Tokyo, Tokyo, Japan). *Kagaku Kogyo* 1974, 25(12), 1374-9 (Japan). A review with 6 refs.

69087u Control of storage container pests by gassing. Baeumert, Klaus (Heerd-Lingler G.m.b.H., Frankfurt/Main, Ger.). *Chem. Rundsch.* 1974, 27(46), 17-19 (Ger). A review with no refs.

69088v Rodents as pests in the food industry. Hermann, Guenther (Biol. Forsch., Bayer A.-G., Leverkusen, Ger.). *Chem. Rundsch.* 1974, 27(46), 21-2, 24, 29 (Ger). A review with many refs.

69089w System for automatic sampling in gas chromatography. Ranft, K. (Bayer. Hauptversuchsanst. Landwirtschaft., Freising-Weihenstephan, Ger.). *Landwirtsch. Forsch., Sonderh.* 1974, 30(1), 231-7 (Ger). Samples (2.3 µl.) were sucked into calibrated capillaries. The solvent was removed by evapn. in a N stream. The capillaries were then introduced into a rotating drum, controlled by a clock, which sequentially dropped the capillaries into the injector block of a gas chromatograph.

69090q Optimization of the sulfur phosphorus emission detector and comparison with the electrolytic conductivity detector to some sulfur-containing insecticides and herbicides. Greenhalgh, R.; Cochrane, W. P. (Chem. Biol. Res. Inst., Agric. Canada, Ottawa, Ont.). *Int. J. Environ. Anal. Chem.* 1974, 3(3), 213-28 (Eng). The flame parameters of the sulfur phosphorus emission detector (SPED) were examd. for max. response to a thiocarbamate, a phosphate, and a phosphorothioate, under a given set of gas chromatog. conditions. An O flame was preferred for the S compd. with an O/H ratio of 0.2-0.25 and a low H flow of 100-150 mg/min, whereas for P compds. an air flame gave the best response with an O/H ratio of 0.1-0.15 and a higher H flow of 200 ml/min. The response of the SPED was similar to that of other flame photometric detectors in that it gave a linear response in the P mode, a squared relationship in the S mode for S compds., and a quadratic response, for

79 67748m Determination of trace level quantities of arsenic via a novel kinetic method.

For patents of related interest see also Section:

57 63501c Test for lead.

63 64477m Modified snake neurotoxin against degenerative neurologic diseases.

compsd. that contd. both P and S. With the latter, the amt. of cross-channel interference in the S made from P was greatly decreased when an air flame was employed. A comparative study of the response of 31 S-contg. compds. with the SPED and Coulson electrolytic conductivity detector (CCD) indicated the latter to be more sensitive by a factor of 3; however, it was not specific. Of all the compds. studied, which included triazines, thiocarbamates, phosphorothioates, and phosphorodithioates, Supracide and its metabolite were the only compds. which gave anomalous responses. The thiodiazoliny S atom did not appear to give S₂ on combustion, whereas it could be detected by the CCD in the pyrolytic mode.

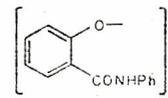
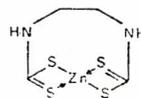
69091r Direct determination of N'-phenylurea derivatives in herbicide technical products and formulations, using gas-liquid chromatography. Buser, Hansueli; Grolimund, Karl (Ciba-Geigy Ltd., Basel, Switz.). *J. Assoc. Off. Anal. Chem.* 1974, 57(6), 1294-9 (Eng). Direct gas-liq. chromatog. of a no. of N'-phenyl-N,N-dimethylureas and N'-phenyl-N-methoxy-N-methylureas was investigated. Representatives of both groups were studied in detail, calibration curves using an internal std. were established, and herbicide tech. products and formulations were analyzed quant. A 2 or 3% silicone SE-30 column was used in conjunction with a flame ionization detector. Operating temps. were chosen on the basis of thermal stability data obtained by differential scanning calorimetry. Gas-liq. chromatog.-mass spectroscopy coupling confirmed that intact urea compds. elute from the chromatog. column.

69092s Evaluation of bactericides and bactericide combinations for the control of bacterial leaf spot and tip burn of *Philodendron oxycardium* incited by *Xanthomonas dieffenbachiae*. Knauss, J. F.; Waters, W. E.; Poole, R. T. (Agric. Res. Cent., Inst. Food Agric. Sci., Apopka, Fla.). *Proc. Fla. State Hort. Soc.* 1971 (Pub. 1972), 84, 423-8 (Eng). As shown in expts. carried out by artificial infection, weekly sprayings with *Kocide 101* [20427-59-2] (formulation contg. cupric hydroxide in combination with *Dithane M45* [8065-67-6]) control the leaf spot and tip burn of *P. oxycardium* caused by *X. dieffenbachiae*. The sprayings were initiated at least 3 weeks prior to inoculation. Sprayings with the *Kocide 101*-*Dithane M45* mixt. alternating weekly with *Agri Strep* (streptomycin sulfate) [3810-74-0] also provided good control. The level of N fertilization had little effect on the bactericidal efficiency.

69093t Antifungal activities of some phenolics and related compounds to three fungal plant pathogens. Mukherjee, N.; Kundu, B. (Dep. Plant Pathol., Univ. Kalyani, Kalyani, India). *Phytopathol. Z.* 1973, 78(1), 89-92 (Eng). Of 18 compds. (mostly phenolic) tested for their fungicidal activity against *Helminthosporium oryzae*, *Alternaria solani*, and *Curvularia lunata*, tannic acid, pentachlorophenol, picric acid and pyrogallol were the most effective.

69094u Prevention of cotton wilt. Babirov, G. G. (USSR). *Zashch. Rast. (Moscow)* 1973, (11), 26 (Russ). *Editon* [3773-49-7], at 6 kg/ton cotton seeds, prevented cotton wilt better than did antibiotic 1/a at 8 kg/ton, or an *Editon*-antibiotic 1/a mixt. Included in the compn. of the antibiotic were phytoncides: 70% pecan juice and 30% cauln (cauliflower ext.) (sic). Seed treatment with 6 kg *Editon*/ton increased the cotton yield from 19.3 (control) to 26.8 centner/ha.

69095v Adaptation of *Botrytis cinerea* to zineb, zinc salicylanilide, and their mixture which exhibits synergistic effects. Golyshin, N. M.; Abelentsev, V. I. (Vses. Nauchno-Issled. Inst. Khim. Sredstv Zashch. Rast., USSR). *Mikol. Fitopatol.* 1973, 7(6), 498-501 (Russ). After 30 reinoculations in the



Zn²⁺
II

presence of zineb (I) [12122-67-7], zinc salicylanilide (II) [5789-39-9], or a 1:1 I-II mixt. which exhibits synergism, *B. cinerea* cultures had increased their resistance to the fungicides 10-, 3-, and 2-fold, resp., relative to controls. The adaptation of *B. cinerea* to I, II, and their synergistic mixt. had a non-hereditary

character
high spc
69096w
bluegras
Hardison
Sci. 197

BAY ME
-triazol-
possibly
and fla;
bluegras
slight ph
69097
germina
L. K. (I
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C-90 [5
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Several compounds tested affected growth, sporulation and spore germination differently. HORSFALL et al. (1955) reported such differences in response in different stages of growth of a fungus.

Table 2 shows the effects of ten phenolics on enzyme preparations of two fungi. All compounds inhibited macerating enzyme activities of *H. oryzae* totally and those of *A. solani* to various degrees. The effects on tyrosinase and laccase are difficult to correlate with the effects on germination, growth, and sporulation. Qualitative differences of the same enzyme might explain the different reactions of the two fungi studied.

Table 2

Effects of some phenolics (0.01 M) on enzyme activities of *Helminthosporium oryzae* and *Alternaria solani*. 0 no action, 1 slightly inhibited, 2 moderately inhibited, 3 highly inhibited, 4 completely inhibited, S used as substrate. Average of 4 replicates. Tm = tissue macerating enzyme, Ty = tyrosinase, L = laccase

Compound	<i>Helminthosporium oryzae</i>			<i>Alternaria solani</i>		
	Tm	Ty	L	Tm	Ty	L
Benzoic acid	4	0	4	3	0	2
Sulfanilic acid	4	0	3	3	0	3
Pyrogallol	4	S	4	3	S	2
Tannic acid	4	S	4	3	4	3
2,4 - Dinitrophenol	4	4	4	4	3	3
Picric acid	4	4	4	4	1	3
Gallic acid	4	S	4	2	S	3
Cinnamic acid	4	0	4	2	0	3
Pentachlorophenol	4	4	4	4	4	1
Rosolic acid	4			4		
Control (Catechol)		S	3		S	2

Summary

Ten out of 18 compounds (mostly of phenolic nature but with widely different structures) have been found highly antifungal against *Helminthosporium oryzae*, *Alternaria solani*, and *Curvularia lunata*. Tannic acid, pentachlorophenol, picric acid and pyrogallol were the most promising inhibitors. Several compounds affected growth, sporulation, and spore germination to a different degree. Tyrosinase, laccase, and the macerating enzyme of *H. oryzae* were generally more inhibited than those of *A. solani*, corresponding to the comparatively less affected growth and sporulation of *A. solani*.

Zusammenfassung

Antifungische Wirkungen verschiedener Phenole und verwandter Verbindungen

Von 18 geprüften Verbindungen zeigten zehn eine starke Wirkung gegen *Helminthosporium oryzae*, *Alternaria solani* und *Curvularia lunata*. Verschie-