

## Additive effects of plant oils and fungicides

---

S. PAN

Department of Plant Pathology, Bidhan Chandra Krishi Viswavidyalaya  
Mohanpur 741 252, Nadia

---

'In vitro' additive effects of Neem (*Azadirachta indica* A. Juss.) and Karanja (*Pongamia glabra* Vent.) oils with captan (N-trichloromethylthio-4-cyclohexene 1, 2-dicarboximide) and carbendazim (Methyl-2-benzimidazole carbamate) was assayed by slide germination and growth inhibition techniques using *Drechslera oryzae* as test organism. Either neem or karanja oil when used simultaneously with captan at non toxic levels responded synergistically to the antifungal activity of captan but when used similarly with carbendazim a reduction in fungitoxicity (antagonism) of carbendazim was noted.

Key words: Additive effect, Synergism, Antagonism, *Azadirachta indica*, *Pongamia glabra*, *Drechslera oryzae*, Plant oil, Fungicide

---

The higher plants of tropical countries are known to provide a rich source of natural products with effective antimicrobial and other biological activities (Sharville, 1960; Calpouzos, 1969). Several reports have established the antifungal activity of neem oil (Singh and Singh, 1981; Dwivedi and Dubey, 1986). Pan *et al* (1985) demonstrated the antifungal property of four furanoflavones and one flavone isolated from the plant *Pongamia glabra*. Besides, much are known about the insecticidal synergism and insect control (Metcalf, 1967) but there are no such informations on fungicidal synergism and plant disease control. Although scientists have time and again shown that combined use of more than one fungicides some times gave better control of plant diseases over single ones (Gisi *et al.*, 1985; Cohen, 1986) but both the fungicides were used at their toxic levels. The present work relates to the exploration of additive effects, either synergistic or antagonistic, of two plant

oils, neem (*Azadirachta indica*) and karanja (*Pongamia glabra*), at their non toxic level in simultaneous use with captan and carbendazim.

#### MATERIALS AND METHODS

For the present investigations two plant oils used were those obtained from neem (*Azadirachta indica* A. Juss.) and karanja (*Pongamia glabra* Vent.) and the fungicides, captan (Captaf 75 WP; N-trichloromethylthio-4-cyclohexene 1, 2 dicarboximide) and carbendazim (Bavistin 50 WP; Methyl-2-benzimidazole carbamate).

The assay organism used was *Drechslera oryzae* (Breda de Haan) Subram & Jain (isolated from brown spot infected rice leaves). The organism was maintained and multiplied on PDA medium.

Effect of fungicides, plant oils and their combinations on the spore germination and mycelial growth of *D. oryzae* was assayed by slide germination and poisoned food technique described earlier (Pan and Sen, 1976).

The fungicidal dilutions were prepared, on the basis of formulated product, in sterile distilled water while that of oil emulsions in sterile distilled water with 0.1% sandovit as emulsifier. Sandovit at 0.1% had no inhibitory effect on the test organism both on spore germination and mycelial growth.

#### RESULT AND DISCUSSION

It appears from the results (Table 1) that both neem and karanja oil had little effect on spore germination and mycelial growth of *D. oryzae*. Only 5.5 and 5.8% reduction in germination of spore and 9.2 and 7.0% reduction in growth of mycelia over control was recorded at 1000.0  $\mu\text{g/ml}$  of neem and karanja oil respectively. However, the fungicides, captan and carbendazim were highly inhibitory to both spore germination as well as mycelial growth of the test organism. More than 50.0% reduction in germination of spore over control was obtained at the lowest concentration (7.8  $\mu\text{g/ml}$ ) of captan and carbendazim (Tables 2 and 3). The  $\text{ED}_{50}$  value of captan for mycelial growth was 40.7  $\mu\text{g/ml}$  (Table 4) and that of carbendazim was 28.8  $\mu\text{g/ml}$  (Table 5).

But when the oil, either neem or karanja was used at a fixed dose of its non-toxic level simultaneously with captan or carbendazim there was either increase (synergism) or decrease (antagonism) in the toxicity of the fungicides. The synergistic effect of neem and karanja oil at 500.0  $\mu\text{g/ml}$  was of significance when used simultaneously with captan. Around 7 to 9 times increase in the toxicity of

**Table 1** Effect of different concentrations of neem and karanja oil on the spore germination and mycelial growth of *D. oryzae*

Concentrations ( $\mu\text{g/ml}$ )	Spore germination		Mycelial growth	
	Neem Oil	Karanja Oil	Neem Oil	Karanja Oil
62.5	0.0	0.0	0.0	0.0
125.0	0.0	0.0	0.5	0.3
250.0	0.0	0.0	0.8	0.7
500.0	1.5*	0.8	1.3*	1.0
1000.0	5.5**	5.8	9.2	7.0

\*Per cent reduction in germination/growth over control.

\*\*Each insertion is an average of four replications.

**Table 2.** Combined effect of captan and oils (neem and karanja) on the spore germination of *D. oryzae*

Concen- trations of captan ( $\mu\text{g/ml}$ )	Captan alone	Captan+ Neem oil (500.0 $\mu\text{g/ml}$ )	Captan+ Neem oil (250.0 $\mu\text{g/ml}$ )	Captan+ Neem oil (125.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil (500.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil (250.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil (125.0 $\mu\text{g/ml}$ )
7.80	65.2*	100.0**	34.6	29.5	100.0	11.3	7.3
15.60	84.0	100.0	75.0	49.5	100.0	19.9	12.3
31.25	91.2	100.0	97.0	75.0	100.0	94.5	80.8
62.50	95.2	100.0	98.0	93.2	100.0	100.0	100.0
125.00	100.0	100.0	100.0	100.0	100.0	100.0	100.0

\* Per cent reduction in germination over control

\*\* An average of four replications

**Table 3.** Combined effect of carbendazim and oils (neem and karanja) on the spore germination of *D. oryzae*

Concen- trations of carben- dazim ( $\mu\text{g/ml}$ )	Carben- dazim alone	Carben- dazim+ Neem oil (500.0 $\mu\text{g/ml}$ )	Carben- dazim+ Neem oil (250.0 $\mu\text{g/ml}$ )	Carben- dazim+ Neem oil (125.0 $\mu\text{g/ml}$ )	Carben- dazim+ Karanja oil(500.0 $\mu\text{g/ml}$ )	Carben- dazim+ Karanja oil(250.0 $\mu\text{g/ml}$ )	Carben- dazim+ Karanja oil(125.0 $\mu\text{g/ml}$ )
7.80	68.3*	54.5**	5.5	4.5	60.6	4.5	1.9
15.60	88.0	68.4	7.4	6.8	73.0	4.9	3.1
31.25	98.2	73.8	13.0	10.2	85.0	9.2	3.1
62.50	100.0	87.3	18.5	13.4	96.1	10.1	8.7
125.00	100.0	96.1	31.8	15.3	100.0	11.0	10.7

\*Per cent reduction in germination over control

\*\*An average of four replications

**Table 4.** Combined effect of captan and oils (neem and karanja) on the mycelial growth of *D. oryzae*

Concentrations of captan ( $\mu\text{g/ml}$ )	Captan alone	Captan+ Neem oil (500.0 $\mu\text{g/ml}$ )	Captan+ Neem oil (250.0 $\mu\text{g/ml}$ )	Captan+ Neem oil (125.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil(500.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil(250.0 $\mu\text{g/ml}$ )	Captan+ Karanja oil(125.0 $\mu\text{g/ml}$ )
7.80	9.6*	60.7**	45.0	0.0	70.8	18.3	15.0
15.60	15.7	69.1	49.3	16.2	81.5	29.7	34.2
31.25	37.6	74.0	52.7	17.8	83.1	44.7	46.3
62.50	67.6	77.3	75.2	34.7	85.0	72.2	71.8
125.00	79.8	100.0	85.9	70.2	90.5	84.7	85.5
ED <sub>50</sub> ( $\mu\text{g/ml}$ ) <sup>a</sup>							
	40.7	5.5	19.5	83.2	4.2	33.9	31.6

\* Per cent reduction in growth over control

\*\* An average of four replications

a) ED<sub>50</sub> value determined on a log-probit scale**Table 5.** Combined effect of carbendazim and oils (neem and karanja) on the mycelial growth of *D. oryzae*

Concentrations of carbendazim ( $\mu\text{g/ml}$ )	Carbendazim alone	Carbendazim+ Neem oil (500.0 $\mu\text{g/ml}$ )	Carbendazim+ Neem oil (250.0 $\mu\text{g/ml}$ )	Carbendazim+ Neem oil (125.0 $\mu\text{g/ml}$ )	Carbendazim+ Karanja oil(500.0 $\mu\text{g/ml}$ )	Carbendazim+ Karanja oil(250.0 $\mu\text{g/ml}$ )	Carbendazim+ Karanja oil(125.0 $\mu\text{g/ml}$ )
7.80	19.6*	16.5**	0.0	0.0	27.3	0.0	0.0
15.60	21.5	16.5	0.0	0.0	37.5	0.0	0.0
31.25	58.8	16.8	0.0	0.0	38.0	3.2	0.0
62.50	68.0	18.5	0.0	0.0	38.6	5.0	0.0
125.00	81.1	29.5	14.7	0.0	40.1	9.3	10.3
ED <sub>50</sub> ( $\mu\text{g/ml}$ ) <sup>a</sup>							
	28.8	—	—	—	—	—	—

\* Per cent reduction in growth over control

\*\* An average of four replications

a) ED<sub>50</sub> value determined on a log-probit scale.'—' Signifies ED<sub>50</sub> value could not be determined.

captan, particularly with respect to the mycelial growth of test organism was envisaged (Table 4). On the other hand when used with carbendazim the oils responded antagonistically irrespective of the dosages of oils (Tables 3 and 5).

It has been suggested that a synergist component may accumulate in the fungal cell membrane and stimulate the passive uptake of other components, thus

increases the toxicity of the second or it may act by inhibiting the energy-dependent efflux of the second component that consequently accumulate in the mycelium and enhance fungitoxicity (DeWaard and VanNistelrooy, 1982).

Alternatively, a synergist component may increase the interference of a fungicide with the membrane functions and thus increase the toxicity of the fungicides over the initial (Davidse, 1980). Although what has happened in the present investigations is not known but it is clear that both neem oil and karanja oil, when used simultaneously with captan, have resulted a synergism in the fungitoxicity of captan either through increased migration of captan in sufficient quantity of its fungitoxic level to the actual site of action or in a different way in a non-specific manner as suggested by Dewaard and VanNistelrooy (1982).

This perhaps appears to be a first report on synergistic effect of neem and karanja oil on the toxicity of captan to *D. oryzae*. Further studies in this area are underway.

#### REFERENCES

- Calpouzos L. (1969). Oils,—In 'Fungicides Vol II' (ed.) Torgeson, D. C., Academic Press, Inc. Ltd. (London).
- Cohen, Y. (1986). Fungicides mixtures controlling late blight of Potatoes. Medelingen van de Faculteit Landbouwweten-Schappen Rijsuniversiteit Gent. **51**, 715-718.
- Davidse, L. C. (1984). Antifungal activity of acylalanine fungicides and related chloroacetanilide herbicides. In 'Mode of Action of Antifungal Agents' (ed.) A.P.J. Trinci and J. F. Ryley, Cambridge Univ. Press, P. 239-255.
- Dewaard, M. A. and VanNistelrooy, J. G. M. (1982). Antagonistic and Synergistic activities of various chemicals on the toxicity of fenarimol to *Aspergillus nidulans*. *Pesticide Science* **13**, 279-286.
- Dwivedi, R. S. and Dubey, R. C. (1986). Effects of volatile and nonvolatile fractions of two medicinal plants on germination. *Trans. Brit. Mycol. Soc.* **87**, 326-328.
- Gisi, U., Binder, H. and Rimbach, E. (1985). Synergistic interactions of fungicides with different modes of action. *Trans Brit. Mycol. Soc.* **85**, 299-306.
- Metcalf, R. L. (1967). Mode of action of insecticide synergists. *Ann. Rev. Entomology*, **12**, 229-256.
- Pan, S. and Sen, C. (1976). Fungitoxic properties of four new fungicides. *Z. Pflkrankh. Pflschutz.* **83**, 577-583.
- Pan, S., Mukherjee, B., Ganguly, A., Mitra, S. R. and Bhattacharjee, A. (1985). Antifungal activity of some naturally occurring flavonoids. *Z. Pflkrankh. Pflschutz.* **92**, 392-395.
- Sharvelle, E. G. (1960). The nature and use of modern fungicides, Burgees Publishing Co., Minnesota, U. S. A., P. 308.
- Singh, H. B. and Singh, U. P. (1981). Effects of volatiles of some plant extracts and their oils on conidia of *Erysiphe polygoni*. *Australian Plant Pathology*, **10**, 66-67.

( Received for publication 30 January 1990 )