# Systemic Remissive Property of Metalaxyl Against Downy Mildew in Pearl Millet

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**ABSTRACT**

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Foliar application of metalaxyl effectively suppressed systemic downy mildew (DM) (*Sclerospora graminicola*) symptoms and healthy leaves and earheads were produced (recovery) on two susceptible pearl millet (*Pennisetum americanum*) cultivars, NHB-3 and 7042. A concentration of 31 ppm (a.i.) was effective for recovery of greenhouse plants, whereas for field-grown plants (DM nursery), a higher concentration (>63 ppm) was needed. Plant age at application did not affect recovery (7- to 35-day-old plants recovered), but earhead length for recovered plants was decreased if applications were made after panicle initiation. Recovered plants, both in the greenhouse and in the field, subsequently developed diseased nodal tillers and/or green earheads. The two cultivars showed similar recovery responses to foliar application of metalaxyl in the DM nursery, but seed treatment (2 g a.i./g) was more effective on NHB-3 than on 7042.

The systemic fungicide metalaxyl has been used successfully to control several fungal plant pathogens belonging to the Oomycetes (1). This is the first systemic fungicide reported to be specifically active against downy mildew (DM) pathogens. As a seed treatment, it has provided high levels of protection to several cereals against DM (3,5–7). In recent tests at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India, metalaxyl seed treatment not only satisfactorily controlled DM (*Sclerospora graminicola* (Sacc.) Schroet.) in pearl millet (*Pennisetum americanum* (L.) Leeke) but also gave significant yield increases (5); however, the degree of DM control differed with the level of varietal susceptibility.

The objectives of this study were 1) to gain information on fungicidal action of metalaxyl, 2) to evaluate the efficacy of foliar application of metalaxyl to control DM after its establishment, and 3) to determine varietal responses to metalaxyl application.

## MATERIALS AND METHODS

**Host cultivars and pathogen.** NHB-3, a DM-susceptible $F_1$ hybrid (develops >80% DM under high inoculum pressure in the DM nursery), and 7042, a highly DM-susceptible cultivar (develops >90% DM under relatively low inoculum load), were used. Plants were grown in the greenhouse (25–30 C) in 10-, 15-, or 20-cm-diameter plastic pots (four to 10 plants per pot) filled with a potting mixture (red soil:farmyard manure, 3:1, v/v).

Systemically infected plants obtained by inoculation of 2-day-old potted seedlings with freshly harvested sporangia of the ICRISAT Center isolate of *S. graminicola* (S. D. Singh and R. Gopinath, *unpublished*) were used in all the experiments unless stated otherwise. A drop of inoculum was placed on the tip of each emerging seedling and allowed to flow down to the base. The pots were covered with moist polythene bags, incubated at 20 C for 12 hr, then maintained in a greenhouse.

**Fungicide and its application.** Metalaxyl 25% wettable powder formulation was used. Fungicide suspensions were applied to infected plants with pneumatic hand sprayers. Plants were sprayed to runoff between 1600 and 1700 hours and kept in a greenhouse or, if maintained outside, under a canopy overnight.

**Sporangial germination test.** A suspension of freshly harvested sporangia was mixed with metalaxyl in water to final concentrations of 125, 63, 31, 19, 4, 1.25, and 0.25 ppm active ingredient (a.i.). Sporangial concentration was adjusted to $4 \times 10^5$ sporangia per milliliter in each of the suspensions. A drop of well-shaken metalaxyl-sporangial suspension was placed on a hemacytometer slide to determine the number of sporangia. Sporangia suspended in water were used as checks. Each treatment had two replicates. Slides were incubated at 20 C in the dark for sporangial germination and germination counts were made after 2 hr. The experiment was repeated once.

**Sporangial infectivity test.** Emerging seedlings were individually inoculated with a 20-μl metalaxyl-sporangial suspension of each of the seven metalaxyl-sporangial suspensions. Each treatment consisted of 10 pots with 10–15 seedlings per pot. Check seedlings were similarly treated with water. All pots were incubated for 12 hr under fluorescent light (2,000 lux) at 20 C, then maintained in the greenhouse in a randomized block design. Records on DM incidence were taken 7 and 15 days after treatment (DAT). The experiment was done twice.

**Effect of concentration on recovery.** Metalaxyl at 125, 63, 31, 25, 19, and 13 ppm (a.i.) was sprayed on 7-day-old NHB-3 plants. Each treatment was replicated in four pots. Water-sprayed plants were used as checks. Records were taken 7, 14, and 27 DAT for DM recovery and at maturity for green earhead production and nodal tiller infection.

**Effect of plant age on recovery.** NHB-3 plants 7, 14, 21, 28, and 35 days old were treated with 31 ppm (a.i.) of metalaxyl. Each treatment was replicated in five pots. Two separate checks—one with healthy untreated and another with diseased untreated plants—were included. Plants were maintained in greenhouse bays in order to prevent reinfection by sporangia that may occur in an open area. Records on recovery were taken 7 and 14 DAT and at maturity, and for earhead length at maturity.

**Field comparison of metalaxyl spray and seed treatment.** The experiment was conducted with two cultivars, 7042 and NHB-3, in the field DM nursery where sporangial inoculum was continually available from heavily sporulating infector rows. Six treatments (including dry seed treatment with wettable powder at 2 g a.i./kg of seed, foliar sprays with 250, 125, 63, and 31 ppm (a.i.), and an untreated check) were arranged in a randomized complete-block design with three replicates. Plots were four 4-m rows. First DM incidence record was taken 20 days after planting (DAP). Twenty-nine DAP, all healthy plants were removed from the plots planted with untreated seed, and only diseased plants in the four spray-treatment plots were sprayed with the respective fungicidal concentration. Observations on recovery in sprayed plots and the increase in DM incidence in seed-treated plots were taken

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10 DAT (39 DAP for seed-treated plots) and at maturity.

## RESULTS

**Sporangial germination and infectivity.** Sporangial germination (zoospore release) ranged from 90 to 100% in all metalaxyl concentrations tested (Fig. 1). Zoospore movement, germination, and germ tube development appeared to be unaffected at all concentrations up to 31 ppm. At concentrations greater than 31 ppm, however, zoospore movement and germination were completely inhibited.

Infectivity of zoospores also varied with metalaxyl concentration. Sporangia suspended in 19 ppm or more failed to cause infection. At lower concentrations, DM infection occurred and incidence was inversely proportional to metalaxyl concentration (Fig. 1).

Probit analysis revealed that percentage of sporangial germination did not differ significantly ($P = 0.05$) among treatments but percentage of infectivity differed. In order to obtain 90% reduction in sporangial infection, at least 3.3 ppm (2.4–5.5 ppm) of metalaxyl is required.

**Effect of concentration.** Percentage of recovery varied with metalaxyl concentration. All plants sprayed with 31 ppm or more showed recovery, whereas the pattern of recovery was inconsistent at lower concentrations. Large variation occurred in production of green earheads and nodal tiller infection in recovered plants (Table 1).

Treated plants produced healthy leaves (recovery from DM) within 7 days. The first leaf to show recovery was only partially symptomless and this area was at the basal end of the leaf blade. Younger leaves showed a progressively smaller area with symptoms until leaves were completely recovered.

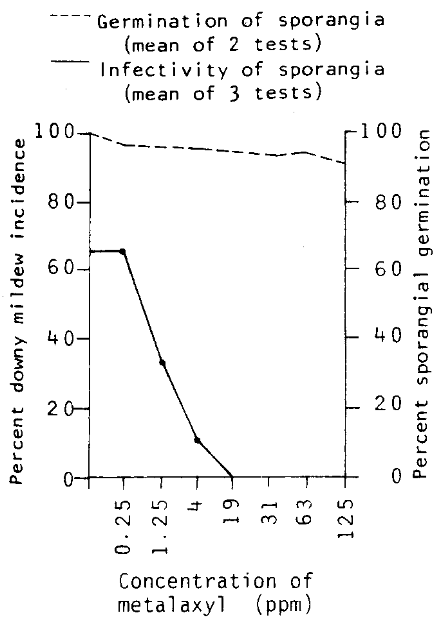


**Fig. 1.** Effect of metalaxyl on germination and infectivity of sporangia of *Sclerospora graminicola*.

Linear regression analysis (weighted) was done to test the effects of fungicide concentration on transformed (arc sine) percentage of foliage recovery, healthy earhead, and nodal infection. The regression equation for percentage of healthy earhead and concentration was $P = 0.751\ (\pm 0.0929) + 0.008\ (\pm 0.0016)\ C\ (r = \pm 0.87)$, where $P$ = healthy earheads, $C$ = concentration (ppm), and $r$ = correlation coefficient between percentage of healthy earheads and concentration. Correlations between transformed percentage of recovery and concentration and between transformed nodal infection and concentration were insignificant.

**Effect of plant age on recovery.** There was no effect of plant age on recovery even up to 35 days; however, plant age at treatment greatly influenced the number of healthy earheads and earhead length, which was largest on 14-day-old plants and progressively decreased with increased age (Table 2). Nodal tiller infection ranging from 5 to 10% was recorded in all treatments.

Quadratic regression analysis revealed that 62% variation in earhead length could be accounted for by plant age at the time of metalaxyl treatment. Differences in the percentage of healthy earheads produced by plants treated at 28 and 35 days were significant ($P = 0.05$) by the $Z$ test (based on normal values).

**Recovery in the field DM nursery.** All sprayed and seed-treated plots of the two cultivars showed significantly ($P = 0.05$) less DM than unsprayed plots. However, the seed treatment that effectively controlled DM on NHB-3 failed to give an acceptable level of DM control on 7042 (Table 3).

## DISCUSSION

Metalaxyl sprays were effective in suppressing DM symptoms in systemically infected pearl millet plants. A single 31-ppm metalaxyl spray was the lowest concentration for an acceptable DM control in greenhouse plants, whereas under conditions of continual sporangial supply as obtained in the DM nursery, a concentration of 63 ppm or more was required.

The two cultivars treated with equal fungicide concentration (>63 ppm) in the field showed similar recovery responses. However, the rate of seed treatment that was effective for protection of NHB-3 in the field was not as effective for the more susceptible 7042. This is in agreement with earlier findings that more DM-susceptible cultivars require more fungicide for acceptable protection (5).

Production of nodal infection and/or green earheads on recovered plants was observed in the greenhouse as well as in the field. Nodal tiller infection could occur either from mycelium present in the recovered plants or from sporangial

**Table 1.** Percentage of recovery, healthy earhead production, and appearance of nodal tiller infection on plants recovered from *Sclerospora graminicola* infection after spraying with metalaxyl in the greenhouse

| | | | Plants with | |
|---|---|---|---|---|
| Concentration of metalaxyl (ppm) | No. of plants sprayed | Recovery (%) | Healthy earhead (%) | Nodal infection[a] (%) |
| 125 | 21 | 100 | 100 | 14 |
| 63 | 20 | 100 | 85 | 25 |
| 31 | 23 | 100 | 78 | 14 |
| 25 | 30 | 83 | 53 | 57 |
| 19 | 23 | 100 | 78 | 48 |
| 13 | 29 | 79 | 41 | 65 |
| 0 | 24 | ... | 0 | 100 |
| Water check | ... | ... | ... | ... |

[a]Nodal tiller infection was noted 20–25 days after planting.

**Table 2.** Percentage of recovery, healthy earhead production, and mean head length produced by plants recovered from *Sclerospora graminicola* infection after treatment of foliage with 31 ppm of metalaxyl in the greenhouse

| Plant age at treatment (days) | No. of plants treated | Recovery (%) | Healthy earhead (%) | Earhead length (cm) (mean ± *SE*) |
|---|---|---|---|---|
| 7 | 60 | 100 | 100 a[z] | 9.5 ± 0.37 |
| 14 | 37 | 97 | 100 a | 10.6 ± 0.62 |
| 21 | 47 | 100 | 100 a | 9.3 ± 0.35 |
| 28 | 37 | 97 | 95 a | 5.0 ± 0.34 |
| 35 | 17 | 100 | 58 b | 4.8 ± 0.34 |
| Healthy check | 66 | ... | 100 | 12.08 |
| Diseased check | 50 | 0 | 0 | 0 |

[z]Figures followed by the same letter are not significantly ($P = 0.05$) different based on statistics[z] (standard normal deviate) used for testing the equality of two binomial proportions ($z = \hat{P}_1 - \hat{P}_2 / \sqrt{\hat{p}\hat{q}\,(1/n_1\ 1/n_2)}$), where $\hat{P}_1$ and $\hat{P}_2$ are observed proportions in two cases based on samples of sizes $n_1$ and $n_2$, respectively.

**Table 3.** Incidence and severity[a] of downy mildew on two pearl millet cultivars treated with metalaxyl

| | Percent downy mildew | | | | | |
|---|---|---|---|---|---|---|
| | 7042 | | | NHB-3 | | |
| **Treatments** | **Total plants** | **Incidence** | **Severity** | **Total plants** | **Incidence** | **Severity** |
| Foliar sprays | | | | | | |
| 250 ppm | 141 | 10.7 | 4.9 | 167 | 1.1 | 0.6 |
| 125 ppm | 135 | 12.2 | 6.1 | 135 | 2.0 | 1.0 |
| 63 ppm | 135 | 8.4 | 4.4 | 154 | 10.0 | 6.2 |
| 31 ppm | 159 | 24.0 | 15.0 | 144 | 28.6 | 18.9 |
| Seed treatment[b] | 339 | 48.0 | 35.5 | 276 | 16.8 | 10.6 |
| Check | 339 | 86.0 | 79.9 | 189 | 87.0 | 79.9 |
| LSD ($P = 0.05$) | | 7.2 | 6.3 | | 14.2 | 10.9 |

[a] Derived from the data on the number of plants in each of the five reaction categories (1 = no downy mildew, 2 = only aerial tillers infected, 3 = fewer than 50% of the basal tillers infected, 4 = more than 50% of the basal tillers infected, and 5 = no productive earhead produced) at the final scoring by the following formula: $[(X(2) + 2X(3) + 3X(4) + 4X(5)]/4N \times 100)$, where $X(2) - X(5)$ = number of plants in disease categories 2–5, and $N$ = total number of plants.

[b] Seed was treated at 2 g a.i./kg seed.

inoculum from other infected plants. Presumably, nodal infection on field-recovered plants was attributable to new infections, whereas nodal infection in greenhouse-grown recovered plants, where sporangial inoculum was absent, was due to latent mycelial infection. This clearly demonstrates that metalaxyl was not lethal to the fungus. Perhaps with decrease in fungicide concentration as a result of plant growth, the pathogen resumed activity and produced symptoms on nodal tillers. The inability of metalaxyl to inhibit sporangial germination in concentrations up to 125 ppm, contrasted with its ability to inhibit movement and germination of zoospores at concentrations >63 ppm and to prevent sporangial infection at concentrations >19 ppm, are supportive evidences of its fungistatic action at lower concentrations. The fungicide is reported to be highly inhibitory to in vitro mycelial growth of *Phytophthora parasitica* and *P. citrophthora* (4) and inhibitory to sporangia and zoospores of *P. infestans* at >50 μg/ml (2).

Production of green earheads by recovered plants, however, is related to plant age at the time of spray treatment and the stage of panicle initiation, which begins about 20 DAP in early-maturing pearl millet varieties. In systemically infected plants, panicle malformation occurs at the time of panicle initiation (S. D. Singh, *unpublished*). Once the process of malformation is complete, metalaxyl application in any concentration is unable to reverse the process. If sprays are undertaken before panicle initiation (as in this study about 21 days after planting), recovered plants would produce healthy earheads of normal length. Delayed sprays would result in the production of correspondingly shorter earheads. The ability of metalaxyl to provide good protection as a seed treatment (5,7) and to permit recovery of systemically infected plants by foliar application makes it an ideal fungicide for DM control in pearl millet.


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## LITERATURE CITED


1. Bien, W. L., Young, T., Dill, T. R., Snow, J., and Seifried, E. B. 1978. CGA 48988 a unique fungicide for the control of Oomycetes. (Abstr. 67) Phytopathol. News 12:142.
2. Cohen, Y., Reuveni, M., and Eyal, H. 1979. The systemic antifungal activity of Ridomil against *Phytophthora infestans* on tomato plants. Phytopathology 69:645-649.
3. Exconde, O. R., and Molina, A. B., Jr. 1978. Note: RIDOMIL (Ciba-Geigy), a seed dressing fungicide for the control of Philippine corn downy mildew. Philipp. J. Crop Sci. 3:1-5.
4. Farih, A., Tsao, P. H., and Menge, J. A. 1981. In vitro effects of metalaxyl on growth, sporulation, and germination of *Phytopathora parasitica* and *P. citrophthora*. Plant Dis. 65:651-654.
5. Singh, S. D. 1983. Variable cultivar response to metalaxyl treatment in pearl millet. Plant Dis. 67:1013-1015.
6. Venugopal, M. N., and Safeeulla, K. M. 1987. Chemical control of downy mildew of pearl millet, sorghum and maize. Indian J. Agric. Sci. 48:537-539.
7. Williams, R. J., and Singh, S. D. 1981. Control of pearl millet downy mildew by seed treatment with metalaxyl. Ann. Appl. Biol. 97:263-268.