

Differences in the growth rates of the wild types and the resistant mutants of *U. maydis* could be observed for other fungicides as well. For other fungicides, the differences between both types approached 100% and the fungicides being tested for concentrations ranged from 4 to 32 µg/ml (Tillman and Sisler 1973).

Out of all the formulations tested, chitosan was least inhibitory to fungal growth. For 1,000 µg/ml, mycelial growth accounted for 35.0% of the control, spore germination for 47.0% of the control, and the germination of *U. maydis* teliospores for 6.6% (Table 1). It was even less inhibitory to the growth of *S. reilianum*: 85.7% (mycelium), 72.5% (sporidia), and 33.5% (teliospores). At low concentrations of 1,10 µg/ml, chitosan stimulated mycelial growth (Table 2). With its inhibitor index amounting to 78.35%, the modified chitosan used in the NCSCA formulation was substantially more effective against *S. reilianum* at a concentration of 1,000 µg/ml (16.8 mm diameter colony, check 77.6 mm). The inhibitor index of the traditional fungicide AMULET under the same conditions was 57.79% (32.5 mm diameter colony, 77.0 mm control) (Zeng *et al.* 2010), (100% of the control = inhibitor index 0%, 0% of the control = inhibitor index 100%).

Test results under field conditions may differ from those observed *in vitro*; corn grains were less affected by head smut when treated with azoxystrobin than with carboxin+thiram (Wrighta *et al.* 2006). Under *in vitro* conditions, growth of *S. reilianum* was more readily inhibited with carboxin and thiram than with azoxystrobin. In other field studies, a better or comparable efficacy was recorded when 2.5 kg of carboxin per ha of soil was applied, compared with 1.6 and 3.2 kg of benomyl per ha of soil used to inhibit head smut (Fullerton *et al.* 1974).

Flusilazole inhibited the germination of *U. maydis* teliospores for all concentrations used, while almost completely inhibiting the germination of sporidia within a range from 100 to 1,000 µg/ml and poorly inhibiting mycelial growth. In other studies, EC₅₀ amounted to 0.18 µg/ml (Markoglou and Ziogas 2000).

For some fungicides, especially at lower concentrations, an increase in fungal growth was observed that was comparable to or slightly higher than the control. These differences were not statistically significant in relation to the control. In other studies assessing the effect of benomyl on *Ustilago striiformis*, it was observed that the fungicide, at concentrations ranging from 2x10⁻⁴ to 2x10⁻⁷, stimulated fungal growth (Robinson and Hodges 1973).

A weak correlation (reflected by the straight lines in the biplot – figure 4) between mycelial growth and the germination of teliospores and sporidia indicates that the effect of the formulations used was different on mycelial growth and different on the germination of teliospores and sporidia for both species of fungi.

REFERENCES

- Aitchison J., Greenacre M. 2002. Biplots for compositional data. *Appl. Stat.* 51 (4): 375–392.
- Gabriel K.R. 1971. The biplot graphic display of matrices with application to principal components analysis. *Biometrics* 58 (3): 453–467.
- Fullerton R.A., Scott D.J., Graham G.J. 1974. Effects of fungicides on the control of head smut of maize, and the relationship between infection level and crop yield. *New Zealand J. Exp. Agric.* 2: 177–179
- Markoglou A.N., Ziogas B.N. 2000. Genetic control of resistance to tridormorph in *Ustilago maydis*. *Phytoparasitica* 28 (4): 349–360.
- Robinson P.W., Hodges C.F. 1973. Benomyl-Induced Growth of *Ustilago striiformis in vitro*. *Phytopathology* 63: 1074–1075.
- Reuveni M., Sheglov D. 2002. Effects of azoxystrobin, difenoconazole, polyoxin B (polar) and trifloxystrobin on germination and growth of *Alternaria alternata* and decay in red delicious apple fruit. *Crop Protect.* 21: 951–955.
- Shurtleff M.C. 1973. *Compendium of Corn Diseases*. The APS Press, St. Paul, MN, USA, 64 pp.
- Slawewski R.A., Ryan E.P., Young D.H. 2002. Novel fungitoxicity assays for inhibition of germination-associated adhesion of *Botrytis cinerea* and *Puccinia recondita* spores. *Appl. Environ. Microbiol.* 68 (2): 597–601.
- Tillman R.W., Sisler H.D. 1973. Effect of chloroneb on the growth and metabolism of *Ustilago maydis*. *Phytopathology* 63: 219–225.
- Wrighta P.J., Fullerton R.A., Koolaard J.P. 2006. Fungicide control of head smut (*Sporisorium reilianum*) of sweetcorn (*Zea mays*). *New Zealand J. Crop Hortic. Sci.* 34 (1): 23–26.
- Zeng D., Mei X., Wu J. 2010. Effects of an environmentally friendly seed coating agent on combating head smut of corn caused by *Sphacelotheca reilianum* and corn growth. *J. Agric. Biotechnol. Sustainable Development* 2 (6): 108–112.
- Ziogas B.N., Markoglou A.N., Tzima A. 2002. A non-Mendelian inheritance of resistance to strobilurin fungicides in *Ustilago maydis*. *Pest Manage. Sci.* 58: 908–916.