

# WAGENINGENUR

# Baseline sensitivity of *Phytophthora infestans* lifecycle components to NC 224 20SC

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# Introduction

The aim of this study was to determine  $EC_{50}$  values of NC224 20 SC for four stages in the lifecycle of *Phytophthora infestans*. The four selected stages were:

- Zoospores release
- Zoospore motility
- Cystospore germination
- Oospore formation in planta

This research was carried out for Nissan Chemical industries.

### Materials and methods

*In vitro:* NC224 20SC was added to aliquots of a *P. infestans* sporangial suspension (50000 sporangia/ml, isolate IPO82001) at three points in time during zoospore release and germination:

- From the start (targeting zoospore release)
- After 2 hours (targeting zoospore motility)
- After 4 hours (targeting cystospore germination)

*In planta:* Potted potato plants (c.v. Bintje) were spray inoculated with a 1:1 mixture of *P. infestans* A1/A2. Symptoms were allowed to develop for eight days before treatment with NC224 20SC at 5 different dose rates, Ranman (0.2 l/ha + adjuvant at 0.15 l/ha) or Tattoo C (2.7 l/ha). After three weeks additional incubation, oospores were extracted and quantified, including "live and dead" differentiation.

### Zoospore release

Zoospore release (% sporangia releasing zoospores) was determined after 2 hours incubation in the presence of NC224 20SC at 10°C. Results are given in Figure 1. The EC<sub>50</sub> value of NC224 20SC for zoospore release, as determined using logistic regression analysis, was found to be 0.016 ppm.

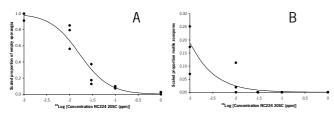


Figure 1. A: Observations (•) and fitted sigmoid curve describing the relationship between the scaled fraction of empty sporangia (zoospores released) and the <sup>10</sup>Log of the concentration of NC224 20SC (ppm).

*B:* Observations (•) and fitted sigmoid curve describing the relationship between the scaled fraction of motile zoospores and the <sup>10</sup>Log of the concentration of NC224 20SC (ppm).

# Zoospore motility

Zoospore motility was assessed after 45 minutes incubation in the presence of NC224 20SC at 10°C. Results are given in Figure 1B. The EC<sub>50</sub> value of NC224 20SC for zoospore release falls in the 0 – 0.001 ppm range and, using logistic regression analysis, was calculated to be 0.0002 ppm (0.2 ppb).

# Cystospore germination

A zoospore suspension was plated on 1.5% water agar plates 15 minutes after adding NC224 20 SC. Germination was microscopically assessed after 20 hours incubation in the presence of NC224 20SC at 10°C. Results are give in Figure 3.

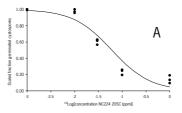


Figure 3. A: Observations (•) and fitted sigmoid curve (solid line, described by equation 3) describing the relationship between the scaled fraction of germinated cystospores and the 10Log of the concentration of NC224 20SC (ppm).

### Oospore formation in planta

Oospore formation responds sensitive to exposure to NC224 20SC. Both, the total number of oospores and the number of viable oospores formed are reduced. The  $EC_{50}$  value for the fraction of viable oospores was determined to be 35% of the recommended dose rate. When compared to the standard treatments included in the experiment, NC224 20SC performed at least equal to Tattoo C and Ranman.



Figure 3: Potato plants (c.v. Bintje) from the oospore experiment 8 days after inoculation and just prior to treatment with fungicides. Conclusions

NC224 20SC was found to be very effective against the stages of the *P. infestans* life cycle tested. The sensitivity of sporangia, zoospores and cystospores to NC224 20SC give reason to believe that this compound could be developed into a fungicide highly lethal to spores of *P. infestans.* Protection against foliar and tuber infection are likely to be two of the strong points of this new fungicide.

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