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# EFFECT OF ZINC UNDECYLENATES ON PLANT PATHOGENIC FUNGI

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## Abstract

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The undecylenic acid (10-undecenoic acid), an eleven-carbon monounsaturated fatty acid is a naturally occurring fatty acid found in the castor oil bean (*Ricinus communis*) and as a product of human sweat glands. It is produced commercially through the vacuum distillation of castor bean oil and pyrolysis of ricinoleic acid (Perkins, 1927). Since 1949, the undecylenate salts have been used as yeast and mold inhibitors in the food industry, and in medicine as topical and systemic antifungals (Bourne, Ireland, Stanberry, Bernstein, 1999). Zinc undecylenate has been shown to be an effective antifungal agent and is the active ingredient in many topical over-the-counter antifungal preparations. Undecylenic acid has been shown to be approximately six times more effective in its antifungal action than caprylic acid. Undecylenate salts have been shown to possess as much as four times the fungicidal effect of undecylenic acid, and may be over 30 times more effective than caprylic acid. These agents have also been shown to be effective in helping to maintain a healthy balance of intestinal and vaginal flora (Chretien, Esswein, Sharpe, 1980).

In the present investigation we conducted *in vitro* and *in vivo* trials with zinc salts of the undecylenic acid with plant pathogenic fungi. The observed results showed that these salts have a strong antifungal effect and can be very effective plant protection remedy.

*Key words:* undecylenic acid, undecylenate salts, zinc undecylenate, plant pathogens, antifungal

## Introduction

Undecylenic acid is a natural fungicide approved in many country of the world as medical remedy for different kind of skin disorders, including infections, itching, burning and irrigations (hemorrhoids). It also is used as remedy for psoriasis. However undecylenic acid and its zinc salts have as fungicidal as antibacterial and antiviral properties – can be used for treatment of herpes simplex virus. As product of human sweat glands the acid and its salts are practically nontoxic for humans and mammalians. Since 1949, the undecylenate salts have been used as yeast and mold inhibitors in the food industry, and in medicine as topical and systemic antifungals (Bourne et al., 1999). Undecylenate salts have been shown to possess as much as four times the fungicidal effect of undecylenic acid, and may be over 30 times more effective than caprylic acid. These agents have also been shown to be effective in helping to maintain a healthy balance of intestinal and vaginal flora (Chretien et al., 1980).

In the current research we conducted *in vitro* trials (germ tube inhibitions tests and radial growth assays) with zinc undecylenate salts against different

phytophathogenic fungi as *Monilia frucigena, Phytophthora capsici, Alternaria solani, Botrytis cinerea, Fusarium oxysporum. In vivo* trials were conducted as two years field tests with grapes variety "Shardone" against downy moldew (*Plasmopara viticola*); apples variety "Golden Delicious" against apple scab (*Venturia inaequalis*) and cucumbers, variety "Gergana" against powdery mildew (*Shphaerotheca fuligineae)*.

## Materials and Methods

Zinc undecylenate salt was received from K.-W.Pfannenschmidt GmbH Company, Hamburg, Germany and tested for antifungal abilities towards economically important for the region of Bulgaria, phytophathigens.

***In vitro tests***

The germ tube inhibition tests were conducted in order to be determining ability of the salts to inhibit a germination of the conidia of the plant pathogenic fungi. The microscopic slides variety "hanging drop" was spayed with water solution with desired concentration. After drying of solution, 20 µl conidial suspensions ($3*10^4$ spores/ml) was added. The slides were incubated for 24-48 h in thermostat under 22-24°C.

Observations with light microscope (10x) were conducted to be determined the germination of the spores (four observation on each slide).

The percent of germination was calculated as follows:

Percent germinated conidia=number of germinated spores*100 / (number of germinated spores + number of non-germinated spores).

According to calculated percents of germination was determined an effectiveness (inhibition) with formula of Abbot (Abbot, 1925)

Radial growth assays *in vitro* trials were conducted according to methods of Thornberry (Thornberry, 1950). In sterile Petri dishes were added 1 ml of the tested solution preliminarily sterilized (1 ml sterile distilled water for control); follow by addition of 9 ml PDA. After vigorously shaking for good mixing of the solution with PDA, inoculation with 10 mm PDA disks with developed mycelium of tested pathogen was conducted (two disks per Petri). The inoculated Petri dished were incubated in thermostat under 22-25°C.

The observations were conducted on 3, 7, 10, and 14 days after inoculation with ruler, measuring the mycelium zone around inoculated disks. On base of this results Area Under Disease Progressive Curve (AUDPC) (Cooke, Gareth, Kaye, 2006) was calculated by Python program language script created by Donyo Ganchev (ActiveState Code, Recipe 576545: AUDPC Calculation). Effectiveness was calculated on base of the values of AUDPC with formula of Abbot. One-way ANOVA analysis was conducted for determination of statistically proved differences between control and tested solutions with R Program Language for Statistical Computing. The starting concentration of zinc undecylenate salt in both conidial and Thornberry tests were 0.01 %.

***In vivo tests***

Apple trees 6 years old, variety "Golden Delicious" ware treated with tested solutions on every 7 to 10 days from pheno-phase "1cm green" to pheno-phase "6 cm fruit" (Agrios, 2004). After first observed visual signs of the apple scab on leaves, were conducted observations on 100 leaves from each variant before each treatment according to five ranks scale with calculation of Index of Mc. Kynney with Python Program Language script created by Donyo Ganchev (ActiveState Code, Recipe 576628: Index of Mc. Kynney). On base of calculated values of the Index of Mc. Kynney were calculated Area under Disease Progressive Curve (AUDPC) and relative AUDPC. On base of values of AUDPC was calculated the effectiveness with formula of Abbot. One-way ANOVA was conducted for determination of statistically proved differences between control and tested solutions with R Program Language for Statistical Computing. The same methods was applied to filed trials with grapes, 6 years old, variety Shardone for examination of the activity of water solution of zinc undecylenates salts on downy mildew (*Plasmopara viticola*).

Cucumbers, variety Gergana were growing in

greenhouse conditions. They were treated with conidial suspension (3*10$^4$ spores/ml) of *Spherotheca fuligineae* received from infected cucumber leaves with shaking in distilled water with shutel apparatus. After observation of first visual manifestations of powdery mildew on leaves, regular treatments with tested solutions on every 7 to 10 days were conducted. The math manipulation of received results was the same as in case of field tests with grapes and apple trees.

## Results and Discussion

***In vitro tests***

The conducted germ tube inhibition tests with conidiospores of *Monilia fructigena* showed the strong antifungal ability of the zinc undecylenates salts (Figure 1). The minimal effective concentration was 0.00005 %. The inhibition of the germination of conidia of *Alternaria solani* was achieved even in lower concentration – 0.00001 % (Figure 2). According to conidispores of *Fusarium oxysporum*, the minimal effective concentration was 0.3 % (Figure 3); for *Botrytis cinerea* – 0.001 % (Figure 4).

The conducted radial growth assays test with *Monilia fructigena* showed the potential of the undecylenates to inhibit the growth of mycelium at 0.01 % (Figure 5) and at 0.5 % for mycelium of *Alternaria solani* (Figure 6). The tests with *Phytophthora capsici* (Figure 8) showed the excellent inhibitory effect of tested substance at 0.01% concentration. According to mycelium of *Fusarium oxysporum*, the minimal effective concentration was 0.5% (Figure 7).

***In vivo tests***

In the conducted *in vivo* tests with cucumber plants variety Gergana zinc indecylenates were tested in two concentrations – 0.1 and 0.2 % against powdery mildew. As standard was used fungicide Timorex 66 EC on base of oil from *Malaleuca alternifolia* at 0.5 %. The results showed that there is no difference between effectiveness of the Timorex and zinc undecylenate at 0.1% (91.2 % for zinc undecylenate and 90.7 % for Timorex 66 EC – $p>0.05$). In higher concentration - 0.2%, it was even better than standard – 97.5 %. The conducted ANOVA proved that there were no statistical differences in the effectiveness in

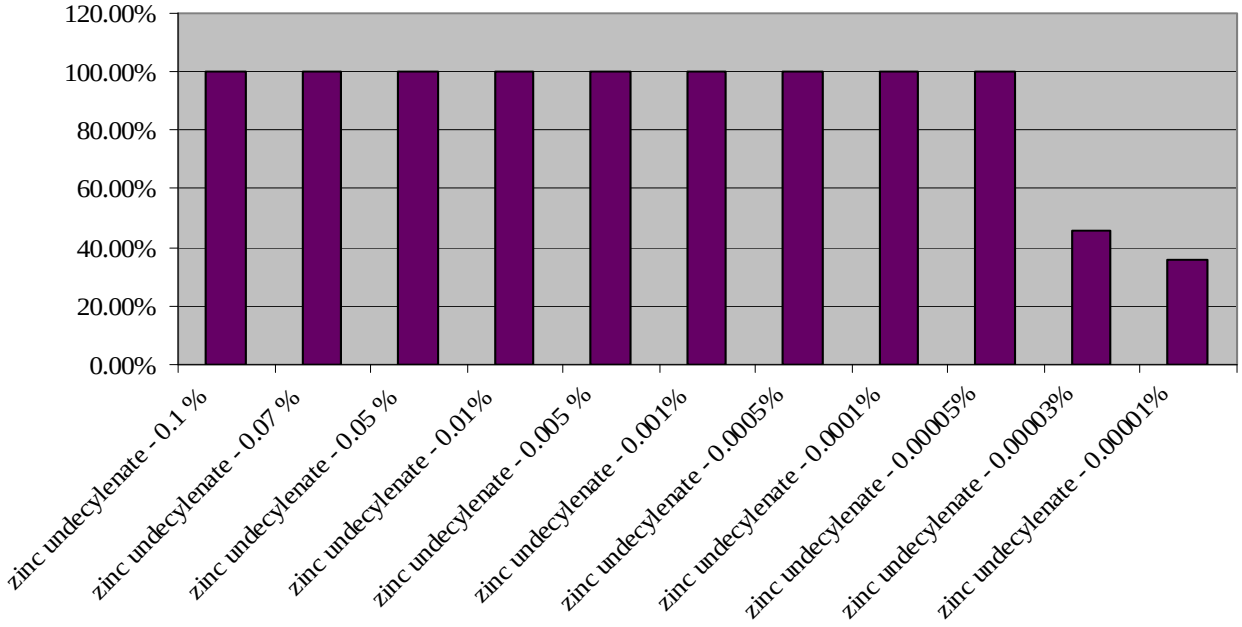


**Fig. 1 *In vitro* conidial tests with *Monillia fructigena***

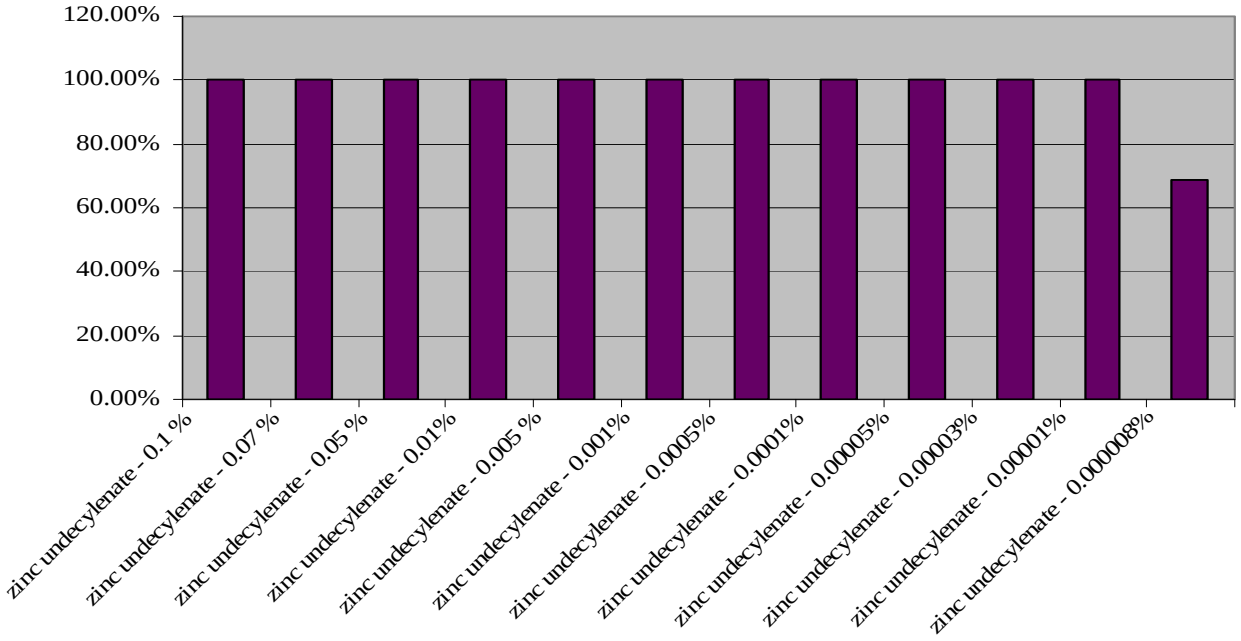


**Fig. 2 *In vitro* conidial tests with *Alternaria solani***

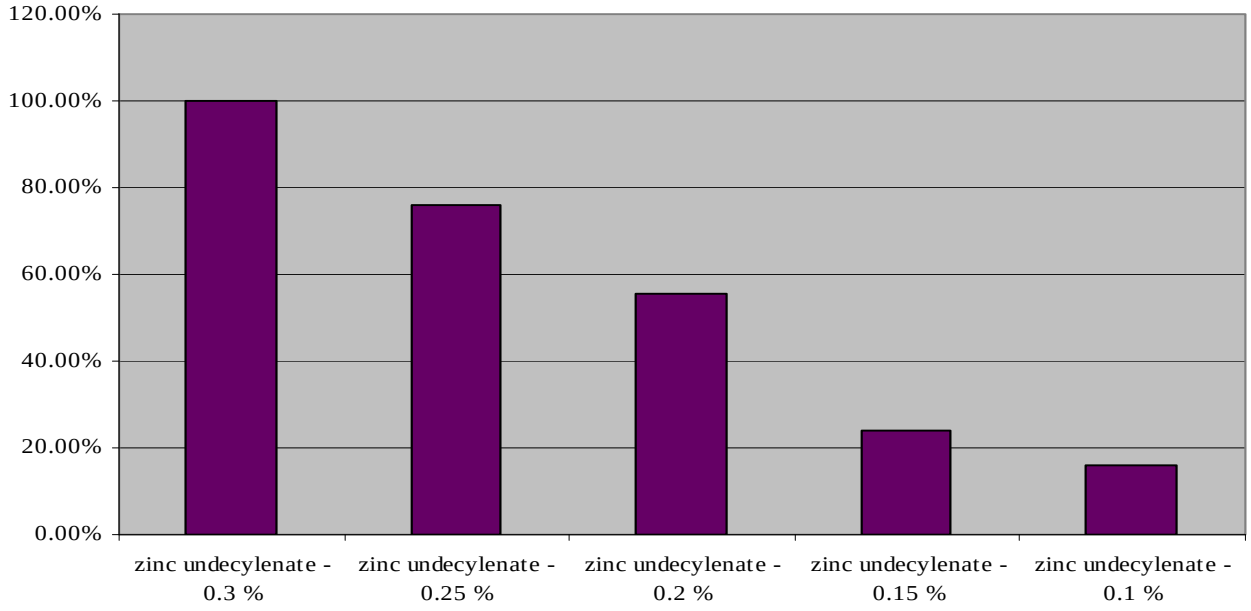


**Fig. 3. *In vitro* conidial tests with *Fusarium oxysporum***

the two years of examinations in both tested concentrations.

In field trials with apples, variety "Golden Delicious", zinc undecylenate was tested at 0.8 % concentration against apple scab. As standard was used fungicide Dithane M-45 on base of Mancozeb. Results showed that also as tests with cucumber, tested solution had better antifungal activity than standard (90.9 % effectiveness calculated by formula of Abbot, 86.2 % for Dithan M 45, $p<0.05$). The con-

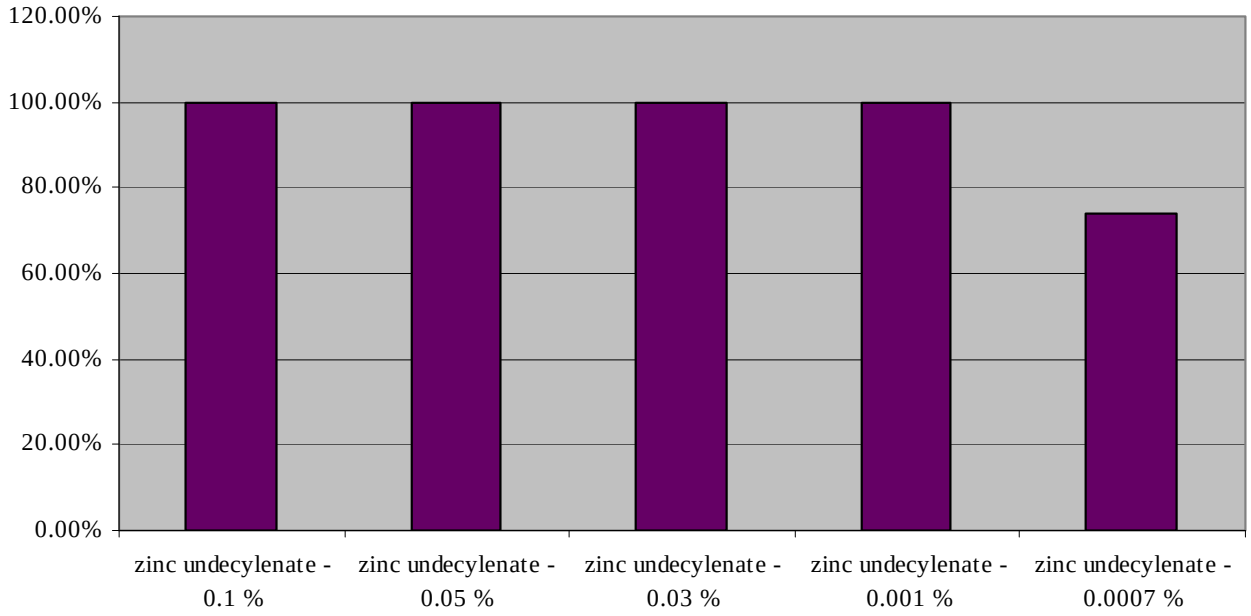


**Fig. 4. *In vitro* conidial tests with *Botrytis cinereae***


**Fig. 5. *In vitro* Thornberry tests with *Monillia fructigena***

ducted ANOVA proved that there were no statistical differences in the effectiveness in the two years of examinations.

Field trials with grapes were conducted with variety "Shardone" against downy mildew. Zinc undecylenate was tested at 0.05 and 0.1 % concentration. As standard was used Bravo 500 on base of chlorotalonil. The received results show that at 0.05 % concentration tested solution had lower fungicidal activity than standard (65.8 % for zinc undecylenate, 85.5 % for Bravo 500, $p<0.05$), but at 0.1% analogically with Bravo 500 – 90.1 % for zinc unde-

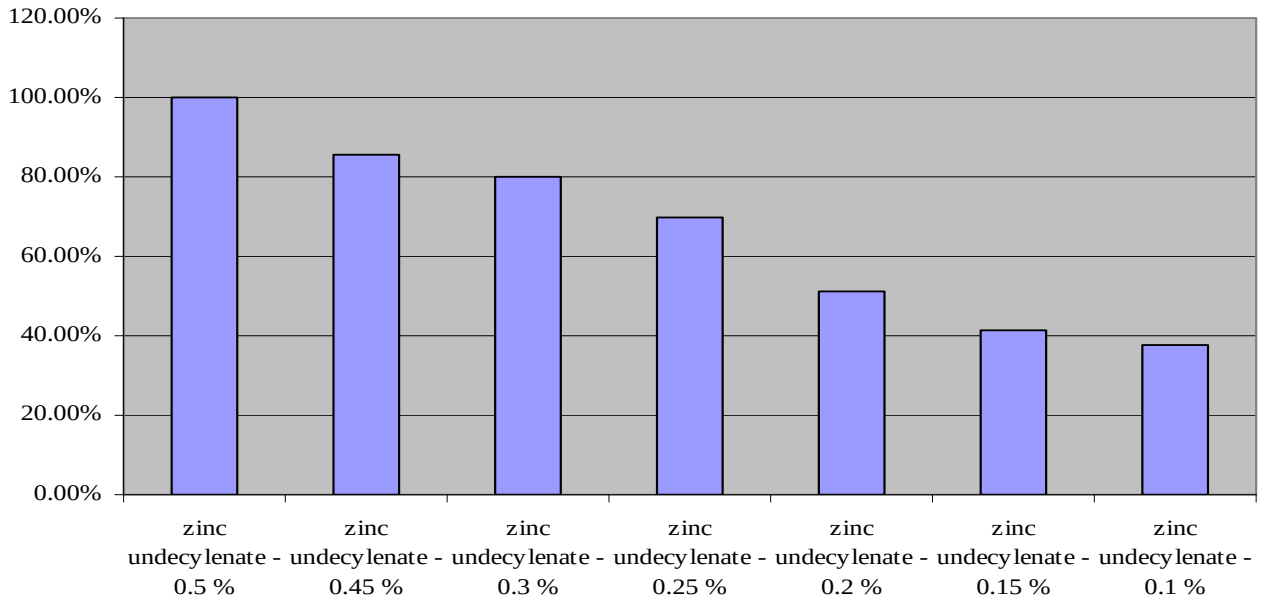


**Fig. 6. *In vitro* Thornberry tests with *Alternaria solani***

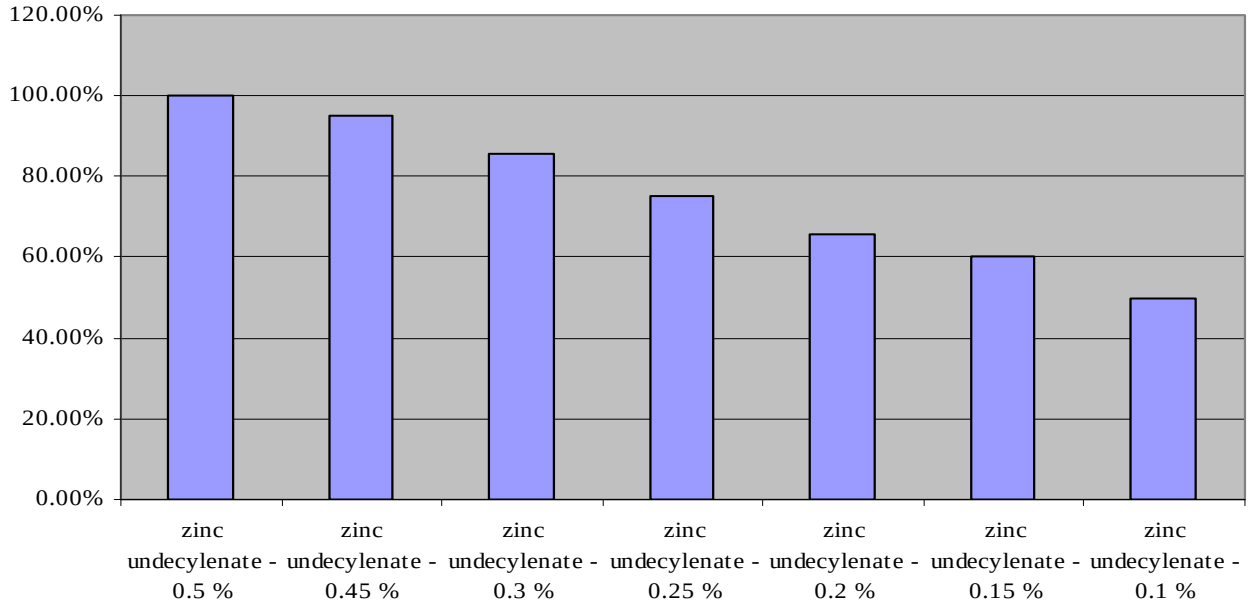


**Fig. 7. *In vitro* Thornberry tests with *Fusarium oxysporum***

cylenate, $p>0.05$. The conducted ANOVA proved that there were no statistical differences in the effectiveness in the two years of examinations in both tested concentrations.

## Conclusion

The results from *in vitro* and *iv vivo* tests proved the strong antifungal activity of zinc undecylenate salts

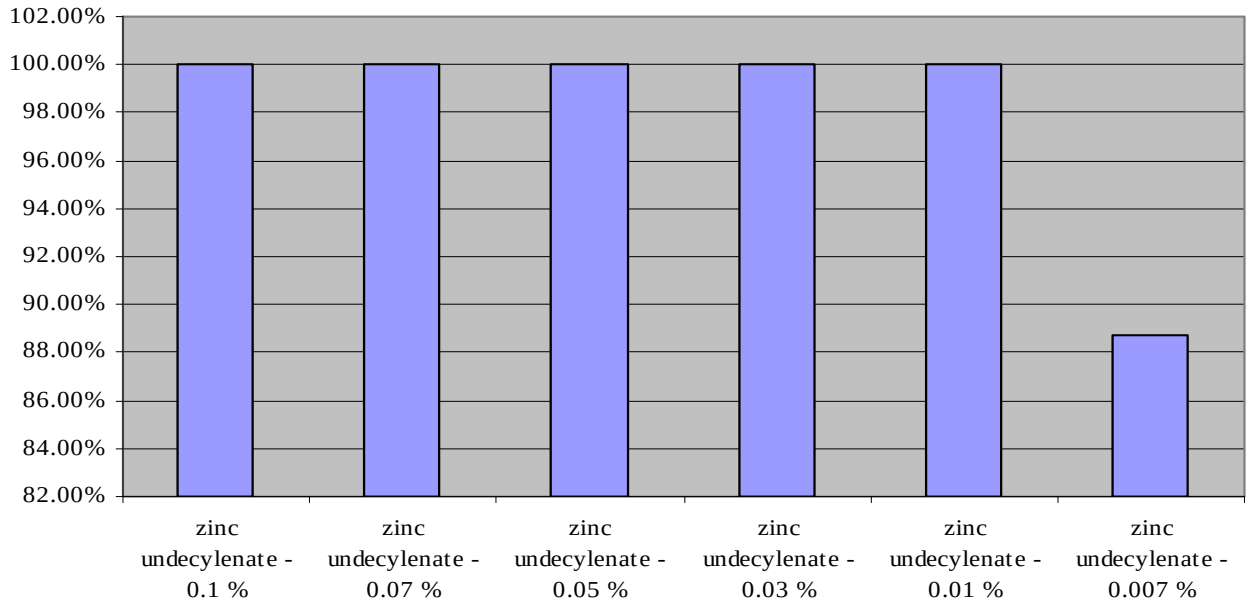


**Fig. 8. *In vitro* Thornberry tests with *Phytophthora capsici***

against plant pathogenic fungi and potential of these salts to be developed as commercial fungicide plant protection products, applicable in organic agriculture and integrated pest management systems. Python and R-program languages meted all requirements for statistical manipulation of data received from antifungal in vitro and in vivo tests. These computer languages are much more effective, powerful and flexible than typically used GUI statistical software products.

## References

**Abbot, W. S.,** 1925. A method for computing the effectiveness of an insecticide. *Journal of Economic Enthomology,* **18**: 267-271.

**Agrios, G. N.,** 2004. Plant Pathology. Fifth Edition, *Elsevier Academic Press.* USA.

**Bourne, N., J. Ireland, L. R. Stanberry and D. I. Bernstein,** 1999. Effect of undecylenic acid as a topical microbicide against genital herpes infection in mice and guinea pigs. *Antiviral Res.*, **40:** 139-144.

**Chretien, J. H., J. G. Esswein and L. M. Sharpe,** 1980. Efficacy of undecylenic acid-zinc undecylenate powder in culture positive tinea pedic. *Int. J. Dermatol.*, **19:** 51-54.

**Cooke, B. M., J. D. Gareth and B. Kaye,** 2006. The epidemiology of plant diseases. *Springer - Verlag,* Netherlands.

**Perkins, C.,** 1927. Preparation of undecylenic acid from castor oil. *J. Am. Chem. Soc.*, **49:** 1073.

**Thornberry, H.,** 1950. A paper-disk plate method for the quantitative evaluation of fungicides and bactericides. *Phytophatology,* **40:** 419-429.

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