

# PHD THESIS - CĂTĂLINA-IOANA IACOB

## SUMMARY

**Key words:** ornamental plants, *Phytophthora infestans*, *Phytophthora parasitica*, quarantine pathogens, quarantine pests

The economic importance of ornamental plants has been increasing in many countries and the international demand has expanded rapidly. Cut flowers represent the largest segment of the industry, followed by potted plants, trees and shrubs, flower bulbs and other organs of breeding. Cut flowers market in Western Europe has grossed over US \$ 12 billion, followed by United States with 6.9 billion US dollars. The Netherlands holds a 60% of exports of ornamental plants followed by Colombia, Italy and Israel (Malter A.J., 1996).

Global production centers of flower crops are changing, for example Ecuador is starting to replace Colombia as the new cut flower production center in South America. Other emerging centers of production of cut flowers are India and Africa. A crucial factor in floriculture is quality, which includes interaction with pathogenic microorganisms and plant longevity (Lawson R. H., 1996).

The PhD thesis „**Research regarding the main pathogen agents and pests that attack a series of ornamental plants in Moldova**” was developed over three years of study 2013-2016 and is structured into eight chapters. To achieve this thesis samples were collected from different locations in the studied area, further processed and then analyzed in the laboratory.

Three experiments were conducted and numerous observations outside these experiences. The first experience consisted in the organization of a bifactorial experiences set in randomized blocks in the premises of the Botanical Garden of Iasi. The factors of this experience were represented by *Dahlia variabilis* Cav. cultivars and the treatments applied to them prior to planting. After the picketing of the land was done, the bulbs of *Dahlia variabilis* Cav. were planted at a depth of 10 cm. The planting material was kept in Dithane M 45 solution 0.2% concentration and Captan 80WDG solution 0.3% concentration for four hours.

The aim of the second experiment was to determine the influence and effect of the culture media on the development of two *Phytophthora* species and then the testing of eleven different fungicides on the two species *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. The two species were inoculated on Petri plates prepared before with four different culture media, PDA, CMA, TA and V8.

After selecting the most favorable media for the development of colonies of the two *Phytophthora* species, we tested eleven fungicides each with a different active substance or a different active complex through the “Including the substance in nutrient media method” (Drobotă I., 2008).

The concentration of the fungicide was the one recommended by the manufacturer for the pathogens of the *Phytophthora* genus, which was included in the culture media at a temperature that did not destroy the active ingredients of the fungicides.

Five repetitions in Petri dishes were carried out for each of tested products and after the solidification of the media in each dish, inseminations were performed with the aid of the inoculation loop in the center of each dish.

After culturing, the dishes were placed in an incubator at a temperature of 27°C and observed daily. All of the values recorded for each of the eleven product were compared with the values of the untreated control.

The development and colony appearance were observed for ten days and the number of spores for ten weeks starting with the 18<sup>th</sup> of April 2016. While in order to determine the colony diameter observations were made daily, for the number of spores observations were made every two weeks.

Culture media was poured into Petri plates (Gosselin 3 vents) with a diameter of 9 cm, and then seeded with both *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. After seeding Petri dishes were kept in an incubator at a temperature of 27°C.

In determining the number of spores, 63.63 cm<sup>2</sup> of mycelium were collected from the surface of the culture medium and mixed with 250 ml of sterile water. The containers with sterile water and *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary., were placed in a refrigerator at a temperature of 7° C, to make sure that zoospores are released. The number of spores per milliliter was determined for each of the fungi using the hemocytometer.

The biological material used to achieve the last experience is represented by both *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. culture, as well as the four floral species *Petunia x hybrida* Vilm., *Catharanthus roseus* L., *Verbena hybrida* L. and *Nicotiana sanderae* Wat. The experiment was carried out in the ICAM greenhouses, where optimum conditions of temperature and humidity were maintained constantly for the development of all four flower species.

Seedlings of *Petunia x hybrida* Vilm., *Catharanthus roseus* L., *Verbena hybrida* L. and *Nicotiana sanderae* Wat. were inoculated with a suspension of *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. in two ways and then the results were compared to the uninfected control.

After determining the values of the number of spores per milliliter of solution we confirmed a minimum of 240000 spores/ml in order to achieve infection (Fraedrich S. W. și colab., 1989). Then using a spray dispersion we assured the suspension with *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. on the foliar surface of the four flower species.

The second method consisted trimming the seedlings roots and placing them in a germ suspension for 5 minutes. This inoculation was performed at the time of the seedlings transplantation.

Part I has three chapters which lists bibliographic data. The first chapter is a synthesis of the history of using ornamental plants, a description of the importance of the establishment and maintenance works and some data on the economic impact of ornamental species. Chapter II concerns the current state of knowledge both nationally as well as internationally of the pathogens that attack ornamental species.

The third chapter consists in a description of the main quarantine pathogens and pests that attack ornamental plants and the phytosanitary risk according to the organization EPPO (European Plant Protection Organization).

The second part of the thesis contains personal contributions structured in five chapters. Research materials and methods used to develop the thesis but also the objectives and activities proposed are described in the fourth chapter. The main objectives of the thesis were:

- Description of the biology, ecology and epidemiology of the pathogens *Phytophthora infestans* de Bary. and *Phytophthora parasitica* Dast.;
- Identification of the nutritious substrate influence on the growth and development of the pathogens *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary.;
- Comparative resistance study of the species *Petunia x hybrida* Vilm., *Catharanthus roseus* L., *Verbena hybrida* L. și *Nicotiana sanderae* Wat. to the attack of *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary.;
- Establishing of curative methods for combating the pathogen agents *Phytophthora infestans* de Bary. and *Phytophthora parasitica* Dast.;
- Comparative resistance study of the *Dahlia variabilis* Cav. cultivars - Spike, Magic Moment, Figurine, Moray Linda and Witteman's best, to the pathogen agents attack under culture conditions in central Moldova;
- Identification of new pathogens and pests of ornamental plants;
- Developing methods of integrated combat for the main pests and pathogen agents attacking ornamental plants.

The characterization of the natural frame were carried out in Chapter V and includes six subchapters on geographical location, geological and geomorphological aspects of the area, hydrography of the area, climate, soil description overlapping over this area and spontaneous flora

One of main proposed objectives for the development thesis was linked to integrated control of pathogens and pests that attack ornamental species. This objective is the subject of chapter VI, which presents mechanical, agrophytotechnical and biological methods of control, use of resistant varieties and enumeration of pathogens and pests flagged as quarantine for our country. In the same chapter a list of fungicides and preferred time of their use for a range of pathogens and pests is presented.

The results are presented in chapter VII, which describes the pathogens determined on both flower species as well as on ornamental woody species. On the flower species six optional parasitic fungi were determined: *Trichothecium roseum* Pers., *Rhizopus* spp., *Penicillium* spp., *Aspergillus* spp. and *Pleospora oblongata* Niess. A total of twelve species of parasitic pathogen agents were determined on floral species: *Pythium de Baryanum* Halle., *Peronospora* spp., *Sphaerotheca pannosa* var. *rosae* de Bary, *Microsphaera begoniae* f.c. *Oidium begoniae* Putt., *Uromyces dianthi* Pers., *Puccinia horiana* Henn., *Phragmidium mucronatum* Pers., *Diplocarpon rosae* Wolf., *Capnodium salicinum* Mont., *Botrytis* spp., *Sclerotinia sclerotiorum* de Bary. and *Fusarium* spp.

In order to determine the incidence of pathogens and pests, samples were taken from four different locations: the courtyard of the University of Agricultural Sciences and Veterinary Medicine, Cotu Morii, Botanical Garden "Anastasiu Fatu" from Iasi and Titu Maiorescu park. These samples were collected for 90 days, every decade of the month. Five coniferous species were studied of which three species were trees *Pinus nigra* J. F. Arnold., *Picea abies* L. and *Abies concolor* Lind. ex Hild. and two arbustifere species *Juniperus horizontalis* Moench. and *Thuja occidentalis* L.

*Lophodermium pinastri* Schrad., *Diplodia pinea* Desm., *Dothistroma septosporum* Dorog., *Pestalotia hartegii* Tubeuf., *Verticillium* spp., *Alternaria* spp., *Trichoderma* spp., *Cladosporium* spp. and *Fusarium* spp. were determined on *Pinus nigra* J. F. Arnold. and also the pests *Cinara pini* L., *Lepidosaphes ulmi* L., *Tetranychus urticae* Koch., a species from the *Pseudococcidae* family and a species from the *Thripidae* family.

On *Picea abies* L. saprofitic pathogen agents from the genera *Cladosporium* and *Alternaria* were determined, which were most likely installed on tissues previously attacked by parasitic fungi or other pests. The only parasite genus was *Fusarium*. Regarding the pests on this species there have been observed *Physokermes piceae* Schrank., *Cinara pini* L., *Tetranychus urticae* Koch. and *Bryobia rubrioculus* Scheuten.

On *Abies concolor* Lind. ex Hild. saprofitic pathogen agents from the genera *Cladosporium* and *Alternaria* were determined. Both genera were observed on samples from two locations, the inner courtyard of the University of Agricultural Sciences and Veterinary Medicine and the Botanical Garden. The pests observed on the further mentioned species were *Tetranychus urticae* Koch., a species from the *Pseudococcidae* family and a species from the *Thripidae* family.

Most of the pathogen agents determined on *Juniperus horizontalis* Moench. were saprofitic pathogen agents from the genera *Cladosporium* and *Alternaria*, the only parasite present genus was *Fusarium*. *Aspidiotus nerii* registered the highest values in all four locations while *Tetranychus urticae* Koch. was determined in only three out of the four locations with very low values.

This chapter also illustrates the results regarding the influence of the nutrient substrate on the development and growth of the pathogen agents *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. but also the results of the testing of the eleven fungicides of both *Phytophthora* species. The chapter ends with the statistic data of the ANOVA test results.

This paper brings real contributions concerning the pathogen agents and the pests determined on a number of ornamental plants, but also the morphology of the *Phytophthora parasitica* Dast. and *Phytophthora infestans* de Bary. species.