ABSTRACT

Rapeseed (*Brassica napus* L.) has great economic importance due to the oil used in food and for biodiesel production.

One of the main pathogens causing significant yield decreases of this crop is *Sclerotinia sclerotiorum* (Lib.) de Bary. Yield losses due to this pathogen may be, under certain conditions, up to 80%.

The doctoral thesis entitled "***Use of molecular markers to identify germplasm sources in rapeseed (Brassica napus L.) with genetic resistance to Sclerotinia sclerotiorum (Lib.) de Bary***" is divided in two parts and comprises seven chapters. The first part describes the species *Brassica napus* L. and *Sclerotinia sclerotiorum* (Lib.) de Bary and the evaluation methods for rapeseed resistance against the attack of this pathogen. The second part includes the presentation of the biological material and research methods, natural and institutional environment as well as the results and conclusions.

Chapter I - ***Description of the Brassica napus L. species*** - contains a comprehensive documentary on the area of cultivation, history, origin, evolution, biology and technological peculiarities of rapeseed.

The Brassicas have always been important as vegetables, seeds, fodder, green manure and spices and played an important role in human history, being part, in some form or another, of his food.

Rapeseed is a herbaceous plant with pivot root, straight stem, more or less branched, with alternate and waxed leaves; the inflorescence is a yellow raceme and the fruit is a dehiscent silique, that contains 10-30 seeds .

Chapter II - ***Characterization of the pathogen Sclerotinia sclerotiorum (Lib.) de Bary*** intends to present the economic importance, host plants, reproductive structures, morphological and physiological characteristics, symptoms, prevention and control measures for the disease caused by it.

In Chapter III - ***The state of researches regarding the resistance of rapeseed (Brassica napus L.) to the attack of Sclerotinia sclerotiorum (Lib.) de Bary*** presents the methods for assessing the plant resistance to the pathogen attack.

Chapter IV contains a description of the objectives of the thesis, the material and methods used at each stage of research.

Thesis objectives aimed to:

• study the morphological traits and resistance to the attack of white rot as a result of artificial infection on rapeseed plants;

• apply the techniques based on RAPD and SSR molecular markers in order to assess the biological material used;

• assess the genetic similarity of the rapeseed cultivars by generating a dendrogram;

• correlate the genetic structure with the phenotypic behaviour after the infection with the pathogen *Sclerotinia sclerotiorum* (Lib.) de Bary.

The biological material consisted of 130 rapeseed cultivars, from the Centre for Genetic Resources of Netherlands. For the evaluation of the morphological traits, were investigated:

- plant height;
- number of branches;
- number of siliques per plant;
- silique length;
- number of seeds per silique;
- number of seeds per plant;
- weight of the seeds per plant;
-thousand-grain mass.

This chapter describes the methods of inoculation on rapeseed cotyledons and leaves, and also the techniques based on molecular markers.

Chapter V - ***Natural and institutional conditions of the researches*** includes information on the topography, vegetation and soil of the Ezăreni farm, the L.E.C.O.M. structure and climatic conditions of experimentation in 2010-2011.

Chapter VI - ***Results and discussion*** - presents the results of the researches. It is divided into several chapters and presents the results of the assessment of morphological traits, of the artificial inoculation on rapeseed cotyledons and leaves, and application of techniques based on molecular markers.

Following the evaluation of the morphological traits:

- the plant height ranged between 60.33 and 152, 33 cm with an average of 113.01 cm;

- the number of branches ranged between 4.33 and 15.33, with a mean value of 8.37;

- the number of siliques per plant ranged between 110.66 and 1270 with an average of 393.72;

- the average silique length was 6.93 cm, with a minimum of 4.04 cm and a maximum of 9.09 cm;

- the number of seeds per silique varied between 13.13 and 36.9;

- the number of seeds per plant ranged between 2442.96 and 39198.03, with an average of 9576.79;

- the seed weight per plant ranged from 11.73 g and 195.21 g;

- the thousand-grain mass had values between 3.07 and 7.06 g, with an average of 4.90 g.

Following the artificial infection on rapeseed cotyledons and leaves, the cultivars behaved differently depending on the isolate and the method of inoculation used.

To identify cultivars with a higher level of tolerance to the disease, the cultivar *Elena* was used as control.

Thus, for the artificial infection with the *Giessen* isolate on cotyledons, 38 cultivars had the size of lesions lower than the control. When using the same isolate on leaves, were found 19 cultivars more tolerant than the control. For the artificial infection with the *Ezăreni* isolate on cotyledons, 47 cultivars had a better response than the control.

As a result of the infection on leaves, were identified 26 cultivars with lesions smaller than the control.

The genetic diversity of the biological material has been assessed using the RAPD method. A dendrogram has been generated, that grouped the cultivars in six clusters based on the similarity coefficient.

215 polymorphic fragments were obtained from a total of 301 amplified. The genetic similarity was calculated from the data obtained using the UPGMA method and using as variable the genetic similarity coefficient. It ranged between 0.61 and 0.89, indicating a high variability for the biological material used.

As a result of the application of SSR techniques, were identified 10 markers associated with rapeseed resistance to *Sclerotinia sclerotiorum* (Lib.) de Bary for the *Giessen* isolate used on cotyledons and 8 for the *Ezăreni* isolate.

For the artificial infection on leaves, were identified 5 markers associated with resistance to white rot isolate using the Giessen isolate and 4 for *Ezăreni*.

At the end of the thesis, in Chapter VII are presented the conclusions that summarize the contributions for identifying the cultivars with higher tolerance to white rot attack.

The markers identified in this research are, along with the others previously mentioned in the literature, another step towards identifying rapeseed genes, for resistance to the attack *of Sclerotinia sclerotiorum* (Lib.) de Bary.

Keywords: *Brassica napus* L., *Sclerotinia sclerotiorum* (Lib.) de Bary, markeri moleculari