The impact of the new races of broomrape (*Orobanche cumana* Wallr.) parasite in sunflower crop in Romania

Maria Pacureanu Joita, Steluta Raranciuc, Emilia Procopovici, Elisabeta Sava, Dumitru Nastase National Agricultural Research and Development Institute Fundulea, N. Titulescu st., no. 1, 915200, Fundulea, Calarasi, Romania, E-mail: mariapacureanu2@yahoo.com, pacurean@ricic.ro

ABSTRACT

The pathogenic composition of broomrape populations has changed over the years, slowly at first, then rapidly in Eastern Europe, Turkey and Spain. In Romania there are three important areas infested with broomrape (*Orobanche cumana* Wallr.), which are different in their infestation degree and presence of different virulence groups. A new highly virulent population of broomrape has attacked sunflower in Romania in 2005. Many commercial hybrids from different companies lost their resistance to this parasite. In the sunflower germplasm collection of Fundulea Institute has been identified a restorer line, AO-548, fully resistant to this new broomrape population. Since this line could be used directly as a parent to produce commercial hybrids, as well as a source of resistance to broomrape in sunflower breeding programs, the inheritance of resistance to the new population of *Orobanche cumana* was studied. Using the cytoplasmic male sterile inbred line AD-66, very susceptible to this population of the parasite, as a female parent, progenies of the cross with AO-548: F₁, F₂ and BC₁ to both parents, as well as the parental lines, were analysed for their reaction to this broomrape population. The results of the observed resistance to broomrape in the line AO-548 is conferred by two independent dominant genes.

Key words: broomrape – inheritance – resistant – sensitive – sunflower – virulence.

INTRODUCTION

Broomrape (*Orobanche cumana* Wallr./*Orobanche cernua* Loefl.) attacks sunflower crop in parts of Europe, the Near East and China (Vrânceanu, 2000). In the last few years the parasite migrated to Western Europe (in 2007 this parasite attacked sunflower crops in France).

Sunflower breeding for resistance to this parasite was started by Pustovoit in 1912 at VNIIMK Institute in Krasnodar, Russia (Sackston, 1992). Using the method of growing all available sunflower material in heavily infested plots, Pustovoit had, by 1927, selected strains with up to 99% of resistant plants (Pustovoit, 1967). By that year, however, previously resistant sunflowers were succumbing to what turned out to be a complex of new races of broomrape. Repeated selection produced lines resistant to the new race complex. Pustovoit did not try to determine the nature of genes controlling this resistance.

Some early Soviet sunflower breeders did study genetic ratios. Meister (1936) reported that resistance to broomrape was inherited as a dominant character, and referred to simple segregation ratios. Later, scientists found that resistance to broomrape races A and B derived from the perennial *H. tuberosus* L. was controlled by a single simply inherited dominant gene (Burlov and Kostyuk, 1976; Pogorletsky and Geshele, 1976).

The virulence of the parasite populations has changed over the years. Vrânceanu et al. (1980) reported on five virulence groups (races or groups of races) of broomrape encountered in Romania, and five types of effective resistance against the respective groups. These investigators set up a series of differentials permitting identification of the five virulence groups, although not the individual races of the pathogen, as each resistance type was effective against a specific race group. The results of complex crossing studies demonstrated a gene-for-gene relationship between virulence groups in the broomrape and resistance in sunflower. They successfully introduced gene *Or5*, which gives resistance to all five race groups, into inbred lines with high combining ability that were the parents of existing or prospective hybrids, and released resistant hybrids.

The different reactions of resistance in varieties of different pathogen sensitivities in sunflower have been reported in recent years. Ciriaev (1987) reported the oligogenic resistance controlled by two genes. Domínguez (1996) has identified the line R-41 having resistance to broomrape controlled by two independent dominant genes. Melero-Vara and Fernández-Martínez (2004) have reported two independent recessive genes for resistance to broomrape.

In Romania, the race F was identified in 1997, as well as the gene (one dominant gene) conferring resistance to this race (Pacureanu-Joita et al., 1998)

Melero-Vara et al. (1989), and other authors quoted works indicating the chemical control of broomrape, but agreed that genetic resistance is the most important method for controlling the parasite. IMI resistance sunflower hybrids may be another way to control it.

This paper presents the results obtained in identifying a new race of *O. cumana* in sunflower crop in Romania, as well as a source of resistance and its inheritance.

MATERIALS AND METHODS

Different sunflower hybrids were tested in fields naturally infested with broomrape, in two important areas in Romania. The investigators set up a series of differentials permitting identification of different virulence groups of the parasite. The different sunflower genotypes (lines and populations) were tested for resistance to the broomrape attack, with a view to identifying new sources of resistance to the most virulent populations of the parasite. Crosses between sensitive lines and new resistant ones were performed in order to establish the inheritance of the genetic resistance. The testing was performed under artificial inoculation using broomrape seeds from two infested areas in Romania. The Panchenko (1975) method was used for testing the artificial infestation conditions.

RESULTS

In Romania, more than 55% of the sunflower cultivated area is infested with broomrape. There are three important areas with a high infestation degree and the presence of different virulence groups (Fig. 1). The high infestation degree in the first area, situated near the Black Sea, is given by race F, and race G has also been identified in this area. In the second area, situated in Ialomita-Braila, race F is well represented.



Fig. 1. The main areas of broomrape infestation in sunflower crop in Romania.

In recent years, the parasite *Orobanche cumana* has developed new races in a short time in sunflower crops in Romania, compared to the first period (Fig. 2). So, if 15 years have passed since the identification of races A and B until race E appeared, as well as from race E to race F, the races G and, may be, H, have appeared in a shorter time and spread quickly over a large area.



In 2006, in a sunflower crop cultivated in Tulcea area, near the Black Sea, some of the hybrids resistant to the race F lost their resistance, being infested at a high percentage (Fig. 3). The hybrids having resistance to the races G or H, were fully resistant.



Fig. 3. The behaviour of some sunflower cultivated hybrids in an infested area with broomrape, Romania, 2006

Using broomrape seeds collected from this area, different inbred lines used as differentials for the races E, F and G have been tested, as well as some resistant hybrids, under artificial infestation conditions in the greenhouse. At the same time, the same genotypes were tested, using the broomrape seeds collected from Ialomita-Braila area. The results (Table 1) showed that the differential for the race F, the inbred line LC 1093, lost its resistance in Tulcea area, having full resistance in the Braila-Ialomita area. The inbred line AO-548 is resistant in both cases. The same behaviour was shown by the hybrids having as parents these two lines (Favorit and Daniel).

| Sunflower genotype | Reaction to the broomrape races | Source of broomrape | | | | |
|-----------------------|--|--|---------------------------|--|---------------------------|--|
| | | Ialomita -Braila | | Tulcea - Constanta | | |
| | | Number of infested sunfl. plants | Infestation degree (%) | Number of infested sunfl. plants | Infestation degree (%) | |
| P-1380-2 | E - A | 10/10 | 41.7 | 10/10 | 77.4 | |
| LC-1093 | F - A | 0/10 | 0.0 | 3/10 | 1.9 | |
| Kd-3-2 | F - A | 0/10 | 0.0 | 2/10 | 1.2 | |
| AO-548 | G - A | 0/10 | 0.0 | 0/10 | 0.0 | |
| Od-832-2b | F - A | 0/10 | 0.0 | 3/10 | 1.8 | |
| Favorit | F - A | 0/10 | 0.0 | 5/10 | 2.3 | |
| F-225 | F - A | 0/10 | 0.0 | 7/10 | 2.9 | |
| PR64A83 | (E)F - A | 8/10 | 19.7 | 10/10 | 73.1 | |
| PR64A71 | (G)H | 0/10 | 0.0 | 2/10 | 0.9 | |
| Daniel | G - A | 0/10 | 0.0 | 0/10 | 0.0 | |

Table 1. The reaction to the broomrape attack in different sunflower genotypes (Fundulea, 2006-2007)

The test conducted under natural infestation conditions in Braila area, using some differentials for the races E, F and G, confirmed that race G was still not present in this area (Table 2).

Table 2. The reaction of the broomrape attack to sunflower under natural infestation conditions – Braila, Romania, 2007

| Sunflower genotypes | Reaction to the broomrape races | Infestation degree (%) | |
|---------------------|---------------------------------|------------------------|--|
| P-1380-2 | E - A | 49.7 | |
| LC-1093 | F - A | 0.0 | |
| O-7455 | E - A | 8.7 | |
| Sel-10481 | E - A | 19.7 | |
| Kd-3-2 | F - A | 0.0 | |
| AO-548 | G - A | 0.0 | |
| AD-66 | Sensitive | 69.7 | |

All tests for resistance to broomrape, which were performed in 2006 and 2007, have shown that, in all the infested areas, the restorer inbred line AO-548 was fully resistant. This line was crossed with AD-66, a CMS line, in order to establish the inheritance of resistance in this restorer line. The F_1 generation was obtained in the field, after which, the crosses and selfings to obtain BC₁ and F_2 generations were carried out under artificial infestation conditions, in pots, in the greenhouse. The plants were kept in pots until maturity, after that they were uprooted and their roots carefully washed to observe any established broomrape nodules. The plants free of nodules or stalks in the roots were considered resistant. The observed ratio of resistant and susceptible plants, in each generation, as well as the goodness of fit of observed – expected ratios are shown in Table 3. The F_2 progeny segregated at a ratio of 15:1 (resistant:susceptible), whereas the BC1 on the susceptible parent, AD-66, segregated according to 3:1

(resistant:susceptible), indicating that resistance to *Orobanche cumana* in AO-548 line is conferred by two single genes with independent action.

| expected ratios | | | | |
|------------------------|-----------|-------------|-----------------|-------|
| Material (generations) | Plants | | Europeted ratio | D0/ |
| Wateriai (generations) | Resistant | Susceptible | | P 70 |
| AD-66 (P1) | - | 15 | - | |
| AO-548 (P2) | 15 | - | - | |
| F1 | 15 | - | - | |
| F2 | 189 | 14 | 15:1 | 80-90 |
| BC1 (AD-66) | 50 | 15 | 3:1 | 50-70 |
| BC1 (AO-548) | 65 | - | - | |
| | | | | |

Table 3. Broomrape resistant and susceptible sunflower plants in the parental, F1, F2 and BC1 generations of crosses between AD-66 (cms) and AO-548, and the goodness of fit of observed (vs) expected ratios

DISCUSSION

The parasite *Orobanche cumana* has became more and more dangerous for the sunflower crop in Romania. In 2006, most resistant sunflower hybrids cultivated in an infested area with this parasite were attacked, some of them at a high attack degree (80%).

The behaviour of some sunflower genotypes regarding resistance to broomrape, under natural and artificial infestation conditions, has shown that the parasite virulence is increasing. The inbred line AO-548 was fully resistant.

The inheritance of resistance to broomrape in AO-548 line is conferred by two independent dominant genes. This line has a good combining ability, being used directly in the obtention of commercial hybrids.

REFERENCES

- Burlov, V.V., and S.V. Kostyuk. 1976. Development of counterparts restoring pollen fertility and resistant to broomrape (*Orobanche cumana* Wallr.) and downy mildew (*Plasmopara helianthi* Novot.). p. 322-326 (vol. 1). In: Proc. 7th Int. Sunflower Conf., Krasnodar, USSR.
- Ciriaev, P.V. 1987. Ustoicivosti samoopâlennĩh linii podsolnocinika k zarazihe. Naucino-tehn.Biulleteni VNIIMK, Krasnodar, 11(97):3-5.
- Dominguez, J. 1996. R-41 a sunflower line, carrying two genes for resistance against a highly virulent Spanish population of *Orobanche cernua*. Plant Breed. 115:203-204.
- Meister, G.K. 1936. Selektia I Semenovodstvo, No.7
- Melero-Vara, J.M., J. Dominguez, and J.M. Fernández-Martínez. 1989. Evaluation of differential lines and a collection of sunflower parental lines for resistance to broomrape (*Orobanche cernua*) in Spain. Plant Breed. 102:322-326.
- Melero-Vara, JM., and J.M. Fernández-Martinez. 2004. Genetic resistance to broomrape in sunflower crop in Spain. Abstract COST Meeting in Bucharest, Romania, 4-6 November, 2004.
- Panchenko, A.I. 1975. A screening method of evaluating the breeding materials for resistance to broomrape. Vestnik selkohoz, nauki, No.2.
- Păcureanu-Joița, M., A.V. Vrânceanu, G. Soare, A. Marinescu, and I. Sandu. 1998. The evaluation of the parasite-host interaction in the system *Helianthus annuus* L. – *Orobanche cumana* Wallr. In Romania. p. 153-158 (vol. 1). In: Proc. 2nd Balkan Symposium in Field Crops.
- Pogorletsky, B.K., and E.E. Geshele. 1976. Sunflower's immunity to broomrape, downy mildew and rust. p. 238-243 (vol. 1). In: Proc. 7th Int. Sunflower Conf., Krasnodar, USSR.
- Pustovoit, V.S. 1967. p. 1-35. In: Handbook of Selection and Seed Growing of Oil Plants (In Russian) Transl. 1973, Isr. Progr. Sci. Transl., Jerusalem.
- Sackston, W.E. 1992. On a treadmill: Breeding sunflowers for resistance to disease. Ann. Rev. Phytopathology 30:529-551.
- Vrânceanu, A.V. 2000. Floarea soarelui hybrida. Editura Ceres, B.
- Vrânceanu, A.V., V.A. Tudor, F.M. Stoenescu, and N. Pârvu. 1980. Virulence groups of Orobanche cumana Wallr., differential hosts and resistance sources and genes in Sunflower. p. 74-82 (vol. 1). In: Proc. 9th Int. Sunflower Conf. Torremolinos, Spain.