

Full Length Research Paper

Detection of the parasitic plant, *Orobanche cumana* on sunflower (*Helianthus annuus* L.) in Tunisia

Moez Amri^{1*}, Zouhaier Abbes², Salah Ben Youssef², Meriam Bouhadida², Hamadi Ben Salah² and Mohamed Kharrat²

¹Regional Field Crop Research Center of Beja (CRRGC), Tunisia.

²National Institute of Agricultural Research of Tunisia (INRAT), Tunisia.

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Orobanche cumana broomrape is a parasitic plant described as an agricultural problem for sunflower production in many countries. In Tunisia, this pathogen was found parasitizing sunflower cultivars in some fields in the Béja region, for the first time during the 2009-2010 agricultural season. Clear *O. cumana* attachments were observed in some sunflower fields and the infestation rates ranged from low to very high with an average of 39.4 ± 33.1 , 211.8 ± 164.7 g and 54.9 ± 48.3 g for the total *Orobanche* number (TON/P), the total *Orobanche* fresh weight (TOFW/P) and the total *Orobanche* dry weight per host plant (TODW/P), respectively. Pathogenicity test on several host species (faba bean, rapeseed, sunflower, lentil, pea, tomato, pepper and potatoes) showed that attachments were observed only on sunflower root system. In some fields, *O. cumana* seed bank in the soil was high enough to decrease sunflower yield and to allow fast expansion of the infestation and therefore reduce the sunflower cropping area and production. The early detection of this new parasite in Tunisia and application of quarantine and control measures can be helpful in reducing the speed of its spread.

Key words: Broomrape, *Orobanche cumana*, sunflower, Tunisia.

INTRODUCTION

Broomrapes (*Orobanche* spp. and *Phelipanche* spp.) are holoparasite flowering plants. They completely depend on their host for their nutritional requirements. Under natural conditions, interactions between parasitic plants and their hosts first occur at the underground level by the parasite seed germination in response to specific chemical in root exudates released from the host plant (Bouwmeester et al., 2003; Abbes et al., 2006). These pathogens attack many crop species worldwide. Among all identified species, only five are noxious pests to economically important crops. These species are *Orobanche crenata*, *Orobanche cumana*, *Orobanche minor*, *Phelipanche*

aegyptiaca and *Phelipanche ramosa* (Garcia-Torres, 1994).

O. cumana Wallr., which is characterized by a high propagation coefficient (Shindrova, 2006) is the main sunflower root parasite (Kaya et al., 2004; Eizenberg et al., 2004; Shindrova, 2006; Parker, 2009) causing significant damages to this strategic oilseed crop in many countries such as Turkey, Romania, Ukraine, Bulgaria, China, the Black Sea countries and ex-URSS countries (Sackton, 1992; Garcia-Torres et al., 1994; Parker, 1994, 2009; Melero-Vara et al., 2000; Kaya et al., 2004; Shindrova, 2006; Masirevic and Malidza, 2006; Schneeweiss, 2009; Fernandez-Escobar et al., 2009).

In Tunisia, two major important *Orobanche* species: *Orobanche crenata* Forsk. and *Orobanche foetida* Poiré are known as a serious problem for legume crops mainly faba bean, pea and chickpea (Kharrat and Halila, 1994; Kharrat and Souissi, 2004). *P. ramosa* have been reported in some regions of Tunisia with slight level of infestation (Kharrat et al., 2008). Recently, in June 2010,

*Corresponding author. E-mail: amrimoez@yahoo.fr. Tel/Fax: +216 78 413 077.

Abbreviations: TON/P, Total *Orobanche* number; TOFW/P, total *Orobanche* fresh weight; TODW/P, total *Orobanche* dry weight per host plant.



Figure 1. Emerged *O. cumana* inflorescences parasitizing a sunflower plant (*Helianthus annuus* L.) in a heavy infested field.

a survey on Orobanche took place in the Beja region; *O. cumana* was found parasitizing sunflower cultivars in some fields for the first time in Tunisia. As broomrapes are difficult to be controlled mainly due to their high fecundity and the long-term viability of their seed in the soil, it is imperative to identify the first outbreaks of Orobanche in order to limit the spread of this species to other areas.

This study focuses on reporting this new parasite, *O. cumana* in Tunisia by assessing the degree of *O. cumana* infestation in different sunflower fields, and studying the pathogenicity of the collected *O. cumana* seeds on

several host of different plant families.

MATERIALS AND METHODS

The survey

A survey of sunflower fields at Beja region, the main sunflower cropping area in Tunisia, was carried out during June 2010 at the seed filling stage. The main objective was to assess the degree of infestation with *O. cumana* on sunflower and to estimate the level of infestation in the fields. A total of fourteen sunflower fields were visited, located from East to West (36°43'10.83"N, 9°07'33.36"E; 36°43'23.74"N, 9°04'25.75"E) and from North to South



Figure 2. (a) Attachments of *O. cumana* on sunflower, (b) Magnification 3x.

(36°43'18.12"N, 9°06'29.12"E; 36°41'25.52"N, 9°05'51.68"E).

Estimation of infestation level

The infestation degree was assessed by determining the Orobanche total number (OTN/P), the total Orobanche fresh weight (TOFW/P) and the total Orobanche dry weight per host plant (TODW/P) of 5 samples. Plants were gently dug up from each infested field and the root systems were washed. The Orobanche shoots were dried for 24 h at 90°C before weighing.

Pathogenicity test

A pot experiment was undertaken in the Agricultural Experimental Unit of Oued-Beja (36°44'N; 9°13'E) in order to study the pathogenicity of the collected *O. cumana* seeds on several host species belonging to four different families: *Fabaceae* (faba bean, lentil and pea), *Solanaceae* (tomato, pepper and potatoes), *Brassicaceae* (rapeseed) and *Asteraceae* (sunflower). The different species were sown in 3 L-pot filled with sterilized soil. An amount of 20 mg of *O. cumana* seeds per kg of soil was added to inoculate each pot. For each species, four replications were applied with one plant/pot. The trial was grown under natural conditions. After five months of growing, the plants were removed and roots were gently washed. The total Orobanche tubercles number (TON/P) and total Orobanche fresh weights (TOFW/P) per host plant species were recorded.

Statistical analysis

The analysis of variance (ANOVA) was determined using the SPSS statistical program v.13 and the differences among treatments for

all measurements were compared at 0.05 significance level using Duncan multiple range test.

RESULTS AND DISCUSSION

During an exploratory visit to fourteen oilseed crop fields in the Beja region in June 2010, *O. cumana* was found parasitizing sunflower in six fields. Clear attachments between the roots of sunflower and Orobanche were observed (Figures 1 and 2). The broomrape morphology and biology were typical of *O. cumana*: Pubescent-glandular plant with usually simple and thin stem; bract and bracteoles with a lanceolated division calyx; corolla blue-purple with curved tube; bilobed and ciliated upper lip and lower lip with three spreading lobes and hairy external face; four hairy stamens in the basis with glabrous anthers or short hair; unilocular capsule with 2 valves adherent at the basis (Figure 3).

These results were confirmed by a pathogenicity test which revealed that among the different species studied only sunflower developed Orobanche tubercles on root system with a total number of 46.8 attachments per plant. This test allowed discrimination between *O. cumana* and *O. cernua*. Earlier publications included *O. cumana* under *O. cernua* species; the latter is known to be a parasite of *Solanaceae* such as tomato, tobacco, pepper and eggplant (Parker, 2009).

The infection rates in infested fields varied from low (1.4 ± 1.95 attachments per plant) to very high ($74.6 \pm$



Figure 3. (a): Lateral view of *O. cumana* blue-purple flower showing the corolla and the calyx, (b): The *O. cumana* reproductive organs (the pistil and the 4 stamens).

Table 1. Total Orobanche number (TON), total Orobanche fresh weight (TOFW) and Orobanche dry weight per plant (ODW) in six sunflower infested fields.

Sunflower field	Geographic coordinate	Total Orobanche number per plant (TON/P)	Total Orobanche fresh weight per plant (TOFW/P) (g)	Total Orobanche dry weight per plant (ODW/P) (g)
<i>Hamrounia 1</i>	36°43'04.34"N 9°07'17.90"E	74.6±16.9 ^d	351.6±45.1 ^b	100.2±21.8 ^d
<i>Hamrounia 2</i>	36°42'59.19"N 9°07'22.23"E	30.4±31.4 ^{bc}	215.8±220.2 ^b	51.8±64.5 ^{bc}
<i>Hamrounia 3</i>	36°42'57.14"N 9°06'31.08"E	52.2±18.7 ^{cd}	288.3±94.0 ^b	71.9±32.1 ^{cd}
<i>Msayia 1</i>	36°41'53.88"N 9°06'09.38"E	69.4±16.6 ^d	344.78±45.6 ^b	92.9±22.5 ^{cd}
<i>Msayia 2</i>	36°41'40.38"N 9°06'24.98"E	8.2±4.6 ^{ab}	58.7±38.5 ^a	10.6±6.95 ^{ab}
<i>Khmayisia</i>	36°43'15.79"N 9°04'46.15"E	1.4±1.95 ^a	11.4±15.9 ^a	2.1±2.9 ^a

Data are means ± SD, values with the same letter per colon are non significantly different at $P = 0.05$ (Duncan test).

16.9 attachments per plant) with an average of 39.4 ± 33.1 , 211.8 ± 164.7 g and 54.9 ± 48.3 g for TON/P,

TOFW/P and TODW/P, respectively (Table 1). These data indicate that the soil seed bank of *O. cumana* in

some fields is high enough to seriously affect the yield and limit the development of the crop in the Beja region. Sunflower was reported to be specifically infected by *O. cumana* in Spain, Balkan region, Turkey, ex-URSS countries, China, Moldavia, Romania, Bulgaria, Hungary, Serbia, eastern Europe, eastern Mediterranean, Syria and Egypt and North African coast (Schneeweiss, 2009; Masirevic and Malidza, 2006; Sauerborn, 1991; Parker, 1994). Eizenberg et al. (2004) reported that *P. aegyptiaca* and *P. ramosa* can parasitize sunflower in the field and the greenhouse. However, this is the first report of *O. cumana* infecting sunflower in Tunisia. This could be a further threat for this oilseed crop in Beja region. The sunflower infestation with *O. cumana* should be considered urgently with a severe quarantine and control measurements in order to reduce the risk of expansion of infested areas. Furthermore, research studies and breeding programs should be undertaken urgently to limit *O. cumana* spread.

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