Field Crops Research 160 (2014) 31–40

Contents lists available at ScienceDirect

Field Crops Research

journal homepage: www.elsevier.com/locate/fcr

Black stem by *Phoma macdonaldii* affected ecophysiological components that determine grain yield in sunflower (*Helianthus annuus* L.)

F.J. Quiroz[a], J.P. Edwards Molina[a,b], G.A.A. Dosio[a,b,*]

[a] Sunflower Group, Unidad Integrada (Facultad de Ciencias Agrarias – UNMdP/Estación Experimental Agropecuaria – INTA) Balcarce, Ruta 226 km 73.5, 7620 Balcarce, Argentina

[b] Laboratory of Plant Physiology, Unidad Integrada (Facultad de Ciencias Agrarias – UNMdP/Estación Experimental Agropecuaria – INTA) Balcarce, Ruta 226 km 73.5, 7620 Balcarce, Argentina

ARTICLE INFO

Article history:
Received 28 July 2013
Received in revised form 19 February 2014
Accepted 20 February 2014
Available online 21 March 2014

Keywords:
Leaf area
Intercepted radiation
Radiation use efficiency
Black stem by *Phoma macdonaldii*
Grain yield
Sunflower

ABSTRACT

Black stem (BS) of sunflower (*Helianthus annuus* L.) by *Phoma macdonaldii* has been reported in many production areas of the world associated to losses in grain yield. Their symptoms could be confounded with natural leaf senescence because they appear during the reproductive period. The objective of this work was to evaluate the effect of BS by *P. macdonaldii* on grain yield and the ecophysiological components determining it.

Two field experiments were conducted under natural inoculation of *P. macdonaldii*. Two hybrids were protected or not with a combination of leaf fungicides. Severity of BS, leaf area, leaf senescence, intercepted PAR and dry matter were periodically measured. Nitrogen nutrition index was estimated at flowering. At physiological maturity, BS incidence, grain number and weight, and oil concentration were also measured.

Presence of other diseases was low or null. Incidence of BS ranged from 2.9% to 49% within the protection treatments, hybrids and experiments. The protected treatment diminished BS in both hybrids and experiments in comparison with the unprotected one. Plants from protected treatment showed higher leaf area, leaf area duration, intercepted PAR, radiation use efficiency, dry matter and grain yield and their components, than those from unprotected treatment. Oil concentration was not affected by the disease. Differences in all studied variables were mainly supported for differences in mid and upper leaves.

Black stem was related to premature leaf senescence and thus to reduction of intercepted radiation. An effect not only on the efficiency of radiation interception but also in the efficiency of the radiation conversion was observed.

1. Introduction

The black stem (BS) of sunflower (*Helianthus annuus* L.) by *Phoma macdonaldii* Boerema (teleomorph *Leptosphaeria lindquistii* Frezzi) has been reported in many sunflower production areas of the world (Acimovic, 1984, 1988) including Argentina where it behaves as an endemic disease (Bertero de Romano, 1978). Losses between 10 and 30% in grain yield were associated to BS (Velásquez and Formento, 2003) and premature ripening (Carson, 1991) both caused by *P. macdonaldii*. The use of susceptible hybrids, no-tillage sowing and most intensification practices (high mineral nutrition, high plant density, shortening of row distance) generates predisposing conditions to BS development (Debaeke and Pérès, 2003).

P. macdonaldii is a soil-borne fungus which overwinters as mycelium, pycnidia and pseudothecia in infected stubble residue (Gulya et al., 1997). Black stem symptoms are associated to previous necrosis in veins and/or leaf petiole and to an acropetal progress from bottom to upper leaves. *P. macdonaldii* also attacks roots and collar of the plant producing a stem girdling lesion at the soil level at the beginning of premature ripening (Donald et al., 1987).

As yet, there are no reports of sunflower genotypes resistant to BS. However, several tolerant genotypes with resistance based mostly on additive genes effect were characterized (i.e. Darvishzadeh et al., 2007). The existence of tissue-specific

* Corresponding author at: Unidad Integrada (Facultad de Ciencias Agrarias – UNMdP/Estación Experimental Agropecuaria – INTA) Balcarce, Ruta 226 km 73.5, 7620 Balcarce, Argentina. Tel.: +54 2266 439100x504; fax: +54 2266 439101.
E-mail address: gdosio@mdp.edu.ar (G.A.A. Dosio).

http://dx.doi.org/10.1016/j.fcr.2014.02.011
0378-4290/

resistance genes for *P. macdonaldii* has been suggested (Abou Al Fadil et al., 2007).

The effect of foliar diseases as BS can be evaluated through the study of structural variables (leaf area, chlorophyll, interception of radiation) and/or functional variables (photosynthesis) (Sadras et al., 2000a). A characterization and evaluation of the effects of crop management (irrigation, nitrogen and plant density) via the crop leaf area index or the fraction of PAR on frequency and severity of BS was reported for sunflower (Debaeke and Pérès, 2003). Research in alfalfa (*Medicago sativa*) demonstrated that leaf spot injury caused by *Phoma medicaginis*, *Phoma sclerotioides*, *Leptosphaerulina trifolii* and *Stemphylium botryosum* reduced photosynthetic efficiency (Hwang et al., 2006). In other studies, *Phoma* spp. and *Phyllosticta* spp. decreased leaf area in trees of *Cecropia schreberiana* (Myster, 2002).

Leaf functionality is associated to incident light and therefore to its position on the stem. Bottom leaves receive an intensity and a quality of light (red/far red ratio) poorer than upper ones which make them senesce before (Rousseaux et al., 1996). Leaf age is also a factor which modulates photosynthesis. As was reported in sunflower, photosynthesis capacity in old leaves is lower than in younger ones (English et al., 1979). Consequently, the effect of BS can be more or less significant depending on these factors.

The objective of this work was to evaluate the effect of BS by *P. macdonaldii* on grain yield and ecophysiological related components in sunflower. To accomplish this objective, yield and ecophysiological components were measured in plants with different level of disease manifestation, achieved by (i) applying or not a fungicide protection against BS infection; (ii) selecting two hybrids with different susceptibility to BS and (iii) different environmental conditions as a consequence of experiments performed in two growing seasons.

2. Materials and methods

2.1. Cultural details

Two experiments, hereafter referred to as Exp. 1 and Exp. 2, were conducted at the INTA Balcarce Experimental Station, Argentina (37°45′ S, 58°18′ W). The soil in both experiments was a Typic Argiudol (USDA taxonomy).

Hybrids Payé (KWS, S.A., Balcarce, Buenos Aires, Argentina) and Paraíso 30 (Nidera S.A., Junín, Buenos Aires, Argentina) were sown the 22 Nov. 2002 and the 28 Oct. 2004, in Exp. 1 and Exp. 2, respectively. Hybrid Payé was reported as more susceptible to BS by *P. macdonaldii* and Verticillium wilt than Paraíso 30 in the cultivar evaluation trial network of INTA, year 2001, where severity was evaluated at physiological maturity. Potential number of leaves in experiments with good nutritional and water conditions was 33 in hybrid Payé and 40 in Paraíso 30, while oil potential content was 53.7% in hybrid Payé and 53.1% in Paraíso 30. Emergence occurred 7 and 16 days after sowing, in Exp. 1 and Exp. 2, respectively. Plant density was 7.2 and 5.6 plants m^{-2}, in Exp. 1 and Exp. 2, respectively. The experiments were conducted under conditions of natural inoculation of *P. macdonaldii* in plots infected with the pathogen (verified in previous experiments). Good nutritional conditions were assured by fertilization before sowing with 75 kg ha^{-1} of diammonium phosphate (46% of P_2O_5 and 18% of ammoniacal nitrogen) and at developmental stage V7 (Schneiter and Miller, 1981) with 60 kg ha^{-1} of urea (46% N). Soil water content was monitored every 5–7 days by neutron probe (Troxler 4300, Troxler Electronic Laboratories, Inc., Research Triangle Park, NC). Soil water was maintained by furrow irrigation above 40% of maximum available water in the firsts 0.60 m of soil during the entire growing season. The amount of applied water was 150 mm and 250 mm in Exp. 1 and Exp. 2, respectively. Weeds and insects were controlled adequately through cultural and chemical techniques.

Flowering date of a plant was considered as the day on which all florets from the outer ring of the head showed their stamens (R5.1, Schneiter and Miller, 1981). Flowering date of a plot was registered as the day on which 95% of the plants had flowered and occurred 55 days after emergence in both hybrids in Exp. 1, and 59 and 63 days after emergence in Exp. 2, in hybrids Payé and Paraíso 30, respectively. Time was expressed on a thermal time basis by daily integration of air temperature with a threshold temperature of 6 °C (Kiniry et al., 1992) and with flowering as thermal time origin.

The following treatments were applied from 30 to 104 (743 Cd after first flowering, R5.1, Schneiter and Miller, 1981) and 105 (635 Cd after first flowering, average of the two hybrids) days after emergence in Exp. 1 and Exp. 2, respectively:

(1) A combination of two fungicides in 150 l ha^{-1} spray volume:
   (a) a systemic, preventive, and curative fungicide whose active principle is carbendazim + epoxiconazole (Duett®, Basf Co.) applied every 15 days at 0.750 l ha^{-1} dose, and
   (b) a systemic and contact fungicide with a preventive, curative and antisporulant action, with azoxistrobin as the active principle (Amistar®, Syngenta Co.) applied every 30 days at 0.5 l ha^{-1} dose.

   This treatment will be referred hereafter as F+.
(2) A sole application of water every 15 days, named hereafter as F−.

Hybrids and protection treatments were combined factorially in a randomized split-plot design with three replicates. The hybrids were assigned to the main plots and the protection treatments to the subplots. Each subplot consisted of four rows of 6 m long, spaced at 0.7 m. Physiological maturity in Exp. 1 was taken as the date in which individual grain weight did not increased compared to the previous sample and in Exp. 2 it was estimated visually from the hard yellow color of the head back face and the brown color of the bracts (R9, Schneiter and Miller, 1981) using a correlation between physiological and visual techniques calculated in Exp. 1 (data not shown). Physiological maturity in Exp. 1 occurred 95 (610 Cd after first flowering) and 100 (697 Cd after first flowering) days after emergence in control and protected treatments, respectively, in hybrid Payé, and 105 (752 Cd after first flowering) days after emergence in both treatments in hybrid Paraíso 30 while in Exp. 2, 109 (703 Cd after first flowering) and 111 (731 Cd after first flowering) days after emergence in control and protected treatments, respectively, in hybrid Payé, and 113 (710 Cd after first flowering) days after emergence in both treatments in hybrid Paraíso 30.

2.2. Measurements

2.2.1. Global radiation, temperature and rainfall

Daily global incident radiation, daily mean air temperature and rainfall were measured in a weather station located 400 m from the experiments. Radiation and temperature were averaged weekly after emergence.

2.2.2. Number of leaves and leaf area

The number of total, green and senescent leaves was measured in three plants *per* plot ($n = 9$) at flowering. A leaf was considered as green or senescent when at least 50% of its surface was green or senescent, respectively.

The green and senescent area of each leaf of the plant was periodically measured in three plants *per* plot ($n = 9$) with an area meter (LI 3100 Area meter Li Cor, Lincoln, Nebraska USA). Leaf area *per* plant was calculated as the sum of the area of all leaves. Leaf area index was estimated at first flowering and at physiological