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Analysis of the influence of a sunflower canopy on *Phomopsis helianthi* epidemics as a function of cropping practices

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ABSTRACT

Phomopsis stem canker (*Diaporthe helianthi* Munt.-Cvet.) can cause drastic reductions in sunflower (*Helianthus annuus* L.) yield and oil content in the main areas of production. The influence of crop management on the incidence and severity of phomopsis stem canker have already been studied in field experiments. However, a more thorough study was required to analyse the effects of soil, crop status (including canopy development) and microclimate on the epidemics of this airborne pathogen, under the influence of cropping practices throughout a growing season. In a 2-year study (2010–2011), carried out in Toulouse (France), the effects of cropping practices (plant density, N fertilisation, and irrigation) and genotypic tolerance (susceptible to tolerant cultivars) on the epidemics of *D. helianthi* were monitored under conditions of reinforced inoculum. Data on inoculum, plant injury, microclimate, crop N status and canopy development were collected. Development of disease injury was broken down into three stages: leaf infection, leaf-to-stem passage and girdling spots on stem. This enabled us to analyse the effects of cropping practices on each of these stages in order to acquire knowledge for the prediction of phomopsis stem canker severity on sunflower. We concluded that the: (i) number of leaf symptoms was determined by the canopy microclimate, especially during vegetative stages, but that after flowering, green leaf tissues potentially available for leaf infection could be an additional limiting variable; (ii) leaf-to-stem passage was determined by leaf length and senescence rate, either natural or induced by phoma black stem; (iii) proportion of girdling symptoms on stems was related to stem diameter. This knowledge will help the design of crop management systems, reducing the risk of crop losses caused by phomopsis stem canker on sunflower.

1. Introduction

Phomopsis stem canker is caused by the fungus *Diaporthe helianthi* (anamorph: *Phomopsis helianthi*; Muntanola-Cvetkovic et al., 1981). It appeared in south-western France in 1984 and then spread over the entire French sunflower cropping area until 1992–1993 (Delos and Moinard, 1995). This fungus has also been reported in many regions of sunflower production worldwide, such as Argentina, Australia, China, Eastern Europe and USA. Ascospores are actively released from perithecia maturing on the sunflower debris and are rain splashed and wind dispersed throughout the growing season. Leaves are infected at the margin via guttation drops (Gulya et al., 1997). The pathogen reduces yield by up to 1.5 t ha^{-1} and oil content by up to 25% due to premature leaf senescence, plant wilting and lodging (Masirevic and Gulya, 1992).

In France, disease control is currently based on the use of disease-tolerant genotypes and to a lesser extent on fungicides. An erosion of tolerance in the case of heavy infections was observed in regions where the disease has been endemic for 30 years, such as Serbia (Masirevic and Forgic, 2000). Moreover, with the European incentives for the reduction of pesticides in agriculture, cultural control is required to limit yield loss. Debaeke et al. (2003) reported that high N fertilisation and high plant density generally increase the proportion of stems with necrotic lesions, although magnitude of response depends on inoculum pressure and timing of spore release, both governed by weather conditions. To our knowledge, the effect of crop management system on epidemics of an airborne pathogen through modifications of soil and plant status, including canopy development and microclimate, has not been extensively explored (Caubel et al., 2012). However, this information is required to represent biotic factors in crop simulation models.

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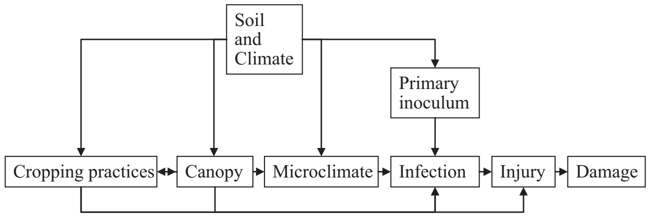

Fig. 1. Relationships between cropping practices, environment, sunflower crop and phomopsis at the field scale (even if primary inoculum is produced at the landscape level).

The severity of phomopsis stem canker is often scored by the final number of girdling spots per plant (Pinochet, 1995). The value of this indicator is obtained by combining three variables: (i) the number of leaf spots per plant, (ii) the proportion of spots that produce a stem lesion and (iii) the proportion of stem lesions turning into girdling lesions. These three variables depend on environmental and crop variables. Among climatic variables, relative humidity (and precipitation) are known to influence ascospore germination and leaf infection (Delos and Moinard, 1997). A relative humidity of 90% must be maintained for at least 10–12 h for leaf infection to be successful (Gulya et al., 1997). The effect of crop management on disease infection could be better represented by taking into account crop microclimate (Debaeke and Moinard, 2010). Relative humidity within the sunflower canopy is governed by the development of leaf area, which is a function of plant density, nitrogen rate and genotype (Stanojevic, 1985; Connor and Hall, 1997). LAI (Leaf Area Index) and $fPAR_i$ (fraction of photosynthetically active radiation intercepted) are frequently used to characterise sunflower canopy development in epidemiological studies (Debaeke and Estragnat, 2003).

As typical infection starts at the leaf margins then spreads along the main vein of the leaf, lamina length could be a factor influencing the probability that a leaf spot turns into a stem lesion. Similarly, stem diameter could modify the time taken for a necrotic lesion to girdle the whole stem, hence affecting the final proportion of girdling lesions. These variables both depend on some crop management options (plant density, N fertilisation) and especially the genotype (Degener et al., 1999; Seassau et al., 2012). The rate of fungal growth in plant tissues may depend on genotype susceptibility (Vear et al., 1997), plant N content and leaf temperature (Agrios, 2005). Leaf length and speed of fungal growth were assumed to determine the time needed to produce a stem lesion from initial infection. However, before reaching the stem, fungal growth may be stopped by leaf or petiole physiological senescence, but also by *Phoma macdonaldii* which develops on leaf axils and provokes premature leaf senescence (Debaeke and Peres, 2003). Therefore, the respective influence of phoma black stem and the natural senescence rate should be assessed by a diagnostic approach.

A detailed study was conducted to analyse *P. helianthi* infection processes in contrasting sunflower canopies differing in genotypic susceptibility, plant density, nitrogen status, and water availability following the conceptual scheme of Fig. 1. Data on inoculum, plant injury, microclimate and canopy development were assessed together and related (Table 1). The objective was to determine step-by-step by experimentation and simulation: (i) how weather conditions influence primary inoculum production (measurement and assessment of the predictive quality of a model), (ii) how cropping practices influence crop N status, plant architecture and canopy development, (iii) how weather conditions and canopy development both affect crop microclimate (measurement and modelling) and (iv) how these environmental and agronomic conditions can affect phomopsis leaf infection and its fate from leaf to stem.

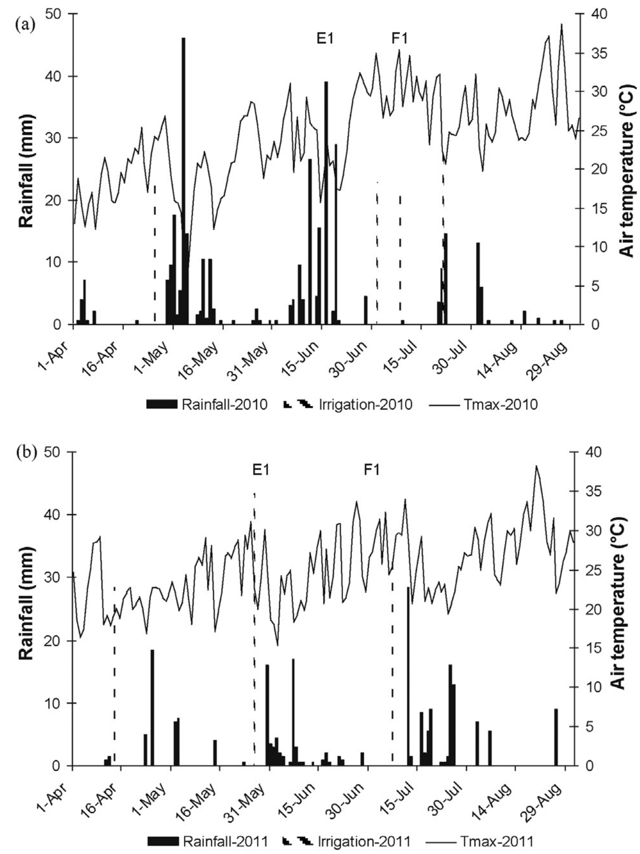

Fig. 2. Weather data: rainfall (with irrigation) and maximum temperature. Growth stages are indicated according to the CETIOM scale (E1: star bud stage, F1: flowering; Merrien, 1992). (a) 2010 and (b) 2011.

2. Materials and methods

2.1. Experimental design and crop management systems

In 2010 and 2011, a sunflower crop was grown under semi-natural infection at INRA experimental station in Auzeville, near Toulouse (Haute-Garonne, SW France, 43°36 N, 1°26 E) on a deep silty-clay soil (24–34% clay in the 0–30 cm layer).

In the semi-natural protocol of infection, fragments of sunflower stalks from infected fields were placed between the crop rows according to the method described by Viguié et al. (2000) in order to reinforce the natural inoculum. In 2010, two stalks per crop row were laid down on 16 June. In 2011, two stalk fragments per row were introduced on 20 May within each of the central crop rows.

In 2010, the experiment was set up as a three-factor split-split-plot design with three replicates in blocks of 90 m^2 each, with water regime (irrigated or not) applied to the main plots, which were split into two sub-plots with two levels of nitrogen fertilisation, which in turn were split into two levels of plant density. Cultivar Melody (moderately susceptible to *P. helianthi*) was sown on 19 April and thinned to two plant densities (D1 = 3.5 plants m^{-2}, D2 = 7 plants m^{-2}) with nitrogen fertilisation (N150 = 150 kg N ha^{-1}: 75 N at sowing + 75 N on 21 June) or without (N0 = 0 kg N ha^{-1}). Two irrigation regimes were applied (IRR0, no irrigation except to promote plant growth and leaf infection; IRR1, 21 mm on 8 July + 28 mm on 21 July; Fig. 2a).

In 2011, the number of modalities was reduced. The field experiment was set up as a three-factor split-split-plot design with three replicates, with irrigation or not as the main plots, each split into two plots with confounded treatments (D1-N150: 150 kg N ha^{-1}, 3.5 plants m^{-2}; D2-N0: no N-fertilisation, 7 plants m^{-2}).