Sustainable management of sunflower downy mildew risk

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ABSTRACT

- The sunflower downy mildew caused by *Plasmopara halstedii* is considered as a quarantine pest by the European Union. In France, its control is based on the use of genetic and chemical tools. But the emergence of pathogen strains resistant to chemicals (late 90s) and the occurrence of new pathogen races able to overcome specific resistances (early 2000s), led us to analyze all the aspects involved in the sunflower production to look for a more sustainable management of this disease, focusing on agricultural practices.
- To assess the importance of agronomic factors on the inoculum and primary infection, a pilot programme "Sunflower downy mildew and sustainable management of the risk at the plot level" was carried out in Midi Pyrenees region. A network of 225 field plots was monitored –trough 3 years (2007-2009) with i) a survey of agricultural practices used by farmers and ii) soil bioassays to characterize soil infestation and the pathotypes of *P. halstedii* present.
- Data showed that short rotations and soil type increased significantly the risk of primary contamination. In addition, the study of the genetic resistance profiles of varieties from fifty plots showed that the frequent use of the same resistance profile in the same field was conducive to new races emergence.
- This study carried out at grower scale showed how some agronomic factors were able to reduce the risk of downy mildew.
- This study allowed us to propose a model for sustainable management of downy mildew for farmers and extension services.

Keywords: monitoring network, *Plasmopara halstedii*, soil bioassay.

INTRODUCTION

Sunflower downy mildew is a common disease in many regions where sunflower is grown. The pathogen, *Plasmopara halstedii* (Farlow) Berles & de Toni is an obligate parasite. The disease affects young seedlings when soil moisture is high and the temperature does not exceed 18°C. The systemic infection of seedlings can be caused by both zoospores (asexual spores mainly located on the underside of leaves) and oospores (sexual reproduction) which can survive in the soil for a decade.

P. halstedii shows physiological races able to infect a variable range of sunflower genotypes. The nomenclature of these races is based on the reaction of a set of differential lines (Tourvieille *et al*, 2000). In France, race 100 was first identified in 1965 and was well controlled by two resistance specific genes *Pl*1 and *Pl*2. But in 1988 and 1989, two new races, 710 and 703 appeared (Tourvieille *et al*, 1991). Since then, a monitoring network which includes breeders and extension partners has been conducted by the French Ministry of Agriculture allowing to follow the evolution of the pathogen. Thus, 15 races more could be identified, especially race 304 since 2000 and more recently race 714 (ONPV et al, 2011).

Considered as a regulated pest at the European Union level, *P. halstedii* must be controlled. In France, its control is regulated and mainly based on the use of genetic and chemical tools that provide an efficient control. But the emergence of pathogen strains resistant to chemicals and the occurrence of new races able to overcome specific resistances have led to include cultural practices for a more sustainable management of downy mildew. However recommended cultural practices based on studies carried out on controlled environment should be assessed in field conditions. For that, a 3-year project called "Sunflower downy mildew and its sustainable management at field level" was carried out in Midi-Pyrénées region from 2007 to 2009. The present work provides an overview of the main results of this project.

MATERIALS AND METHODS

Location

The study took place in the South West of France and was carried out with two cooperatives i/ Qualisol located in Tarn-et-Garonne near Beaumont-de-Lomagne and ii/ Gersycoop located in Gers near Fleurance. Over the period 2007-2009 the network included 225 field plots owned by 47 growers. Fields were chosen according to 3 factors: i) downy mildew history : all the fields had already expressed downy mildew during the past 3 years or were located in an area where downy mildew has been usually observed, ii) the crop rotation sequence and iii) the type of soil (calcareous clay, clayey loam and loam).

Survey

For each field data of past cropping history and present cultural practices were collected using a questionnaire designed to assess a priori the downy mildew risk (Thiery, 2007).

Field disease evaluation

In each field, plants that showed typical symptoms of *P. halstedii* infection were observed between the stage "appearance of bud" and the stage "early flowering". The incidence and the severity of downy mildew symptoms were recorded.

When *P. halstedii* sporulation was observed on leaves, these leaves were sampled for the identification of pathotypes according to a host range (Gulya et al, 1998).

Moreover, 90 fields were selected for soil bioassays. Each soil sample was collected in the 2-8 cm seed bed depth. A quantitative analysis was performed on a susceptible line for rating soil infection as DMI (downy mildew incidence) and a qualitative analysis from two sporulating seedlings per bioassay was also performed on a host range to characterize the race (Tourvieille et al, 2008).

Selection pressure on pathotypes of *P. halstedii*

Data of 10-year succession of sunflower cultivars are available for 47 fields. The cultivars were classified into two groups describing two types of resistance patterns i/ varieties with *Pl*6 or *Pl*7 gene which are resistant to races 100, 710 and 703 but susceptible to race 304 and ii/ all other varieties which are either susceptible to races 703 and 710 or resistant to races 100, 703,710 and 304. A repetition index (RI) was calculated and took into account the number of varieties with the same resistance pattern and the delay of cropping the same resistance pattern. RI was considered as a qualitative variable. More RI was greater more the same resistance pattern was used in a short cropping system (Dordolo M., 2008).

Data analysis

Three explanatory variables qualify the inoculum: 1) the intensity of downy mildew (qualitative variable), the soil infection rate (quantitative variable) and the emergence of new races (qualitative variable).

Four explanatory variables were selected as the most related to the inoculum: soil type, tillage, crop rotation and soil drying capacity.

The independence of explanatory variables was checked using a χ^2 test. A MCA was performed to identify trends between agricultural practices and inoculum. The qualitative variables were tested by χ^2 test and the quantitative variables by ANOVA. Given the over-dispersion data, GML model had to be used.

RESULTS

Field surveys

It was difficult to obtain a balanced data set despite the fact to rebalance the sample from year to year. Over the three years, the most prevalent soil type was clayey calcareous. The crop rotation was essentially short, alternating sunflower with wheat. Plough was used in most of fields (Fig 1).

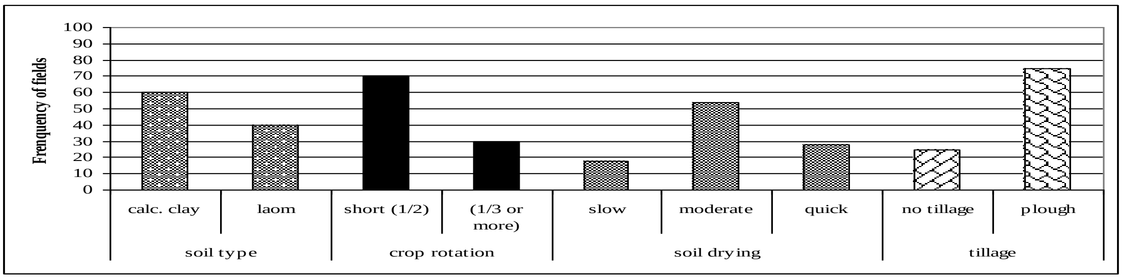

Figure 1: Distribution of the 225 fields according to 4 agronomical factors

Monitoring of downy mildew

From all the 225 monitored fields 54% of fields had no symptom of downy mildew while some infected plants were observed in 46% of fields. In general the intensity of downy mildew was low: only 12% of the fields showed significant attacks (> 3% infected plants), two fields presented foci with more than 30% of disease incidence and one field showed an overall attack rate above 10% with some foci with 30% of infected plants.

Looking at the results of soil bioassays of 90 field plots, the soil infection was much higher than the disease incidence expressed in the field. So, 68% of tests showed downy mildew symptoms with 22% of field plots presenting DMI from 10 to 30%, 10% of field plots where DMI was above 30%. One field provided the highest DMI that reached 67%. Thus even in the absence of symptoms in the fields, downy mildew was still present in the soil and could initiate an epidemics when favourable conditions were met.

Seven races were detected over 61 analyzed field plots: race 703, the most common in the region, was also the race mostly detected in these soil samples (31 plots). Detected in 27 plots, race 304 was also well-established. Race 704 and the previous races 100, 700 and 300 were sporadically present. A new race 707 was present alone or in combination in 13 plots. Twenty plots showed a combination of 2 races (Table 1).

Races	**703**	**304**	**707**	**704 - 100 - 300 - 700**
No of plots	31	27	13	1

Table 1: Distribution of downy mildew races in 61 field plots

Influence of agronomic factors on downy mildew intensity

The MCA performed with the variables “soil type” and “crop rotation” (both other variables depended on them) showed that infected plots were more frequently associated with clayey calcareous soil and short crop rotations. In the contrast the absence of DM would be more related to loamy soil type and long crop rotations (Fig 2).

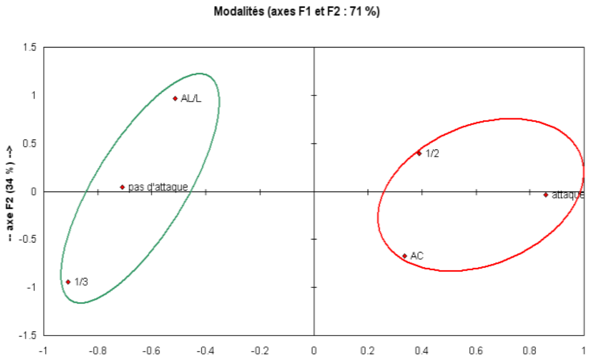

Figure 2: Influence of agronomic factors on the intensity of DM in fields (MCA).

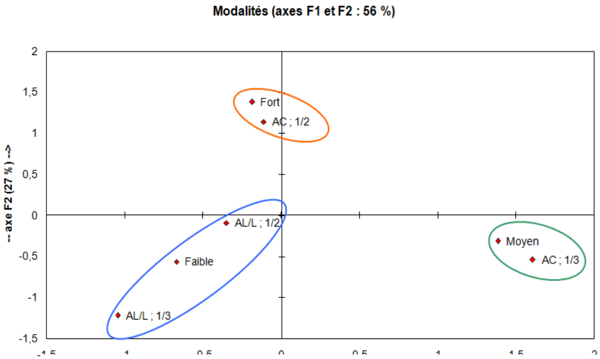

Figure 3: Influence of agronomic factors on DMI from soil bioassays

Influence of agronomic factors on DMI in soil bioassays
The results of GLM analysis indicated that the soil type and the length of rotation were the main two variables that influenced significantly the DMI. By MCA, 56% of the variability could be explained by these two variables (Fig 3). The results suggest that clayey calcareous soils and short crop rotation are associated with high levels of DMI while loamy soils with short or long crop rotation are grouped with mild DMI. Thus, clayey calcareous soils would be more conducive to the survival of the inoculum than loamy soil.

Relationships between resistance patterns of sunflower and emergence of new races
The emergence of new races depends significantly on the frequency of sunflower in the cropping system (χ^2 test: p=0.03) and the succession of a same resistance pattern (χ^2 test: p=0.0008). Results show that fields where hybrids with *Pl*6 or *Pl*7 gene were sown over the past 10 years have a greater probability to see the emergence of a new race than fields where *Pl*6 or *Pl*7 hybrids alternate with another resistance pattern. So, sunflower hybrids resistant to previous races such as race 703 seem favourable to the emergence of race 304, due to a selective pressure on the soil inoculum (Fig 4).

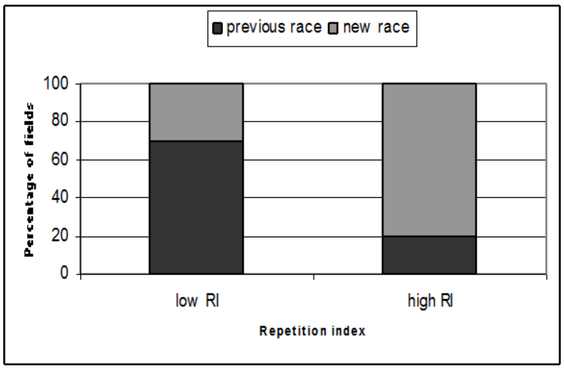

Figure 4: Evolution of races according to the repetition index RI

DISCUSSION
For the 3-year project the incidence of downy mildew was quite low. The lack of damage was mainly explained by the use of resistant varieties. Most of cultivated varieties are resistant to races 100, 703 and 710 and many of them are also resistant to race 304. So such varieties did not allow the expression of prevalent races in the production area. Nevertheless the soil bioassays allowed to overcome the filter of resistant varieties and to study the influence of various agronomic factors on inoculum.
Among agronomic factors, short rotations (4 or 5 sunflower crops per decade) could favour more the survival of inoculum in the soil than long rotations (3 or less sunflower crops per decade). However the inoculum was always present since it can survive 6 to 10 years into the soil (Cohen and Sackston, 1974).
The calcareous clayey soils seem to be more favourable to the conservation of the inoculum compared to the loamy soils. Because loamy soils dry more slowly, the presence of open water during a long time after

rainy periods could be increase the germination of the oospores and thus reduce the amount of inoculum in the soil. In contrast, filter calcareous clayey soils dry quickly, so the duration of open water in the soil is shorter and the exhaustion of the inoculum could be slower.

A relationship between the frequency of the same DM resistance pattern in a given field and the emergence of new races was suggested by our analysis but this result has to be confirmed with a larger number of fields. Nevertheless it is consistent with a study carried out under controlled conditions from 2000 to 2004 (Tourvieille et al, 2004). This study demonstrated the emergence of new races (704, 714) after only four years of single crop farming of sunflower hybrids with the same DM resistance pattern (genes *Pl*6 or *Pl*7). Moreover compared to the single crop farming of one kind of resistance pattern, the strategy of alternating resistance genes can significantly limit the long-term attacks and the emergence of new races.

The impact of agronomic factors on downy mildew led us to propose a sheet decision system based on the soil type of the field combining with the crop rotation, the succession of varieties (resistance pattern). The results of all these practices provide a coloured scale of risk indicator.

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