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## **Effects of wheat volunteers and blackgrass in set-aside following a winter wheat crop on soil infectivity and soil conduciveness to take-all.**

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*Key-Words:* blackgrass, set-aside, soil conduciveness, soil infectivity, take-all, wheat volunteers

### **Abstract**

Two experiments were carried out in France in which disease indices were used to evaluate the effects of wheat volunteers and blackgrass (*Alopecurus myosuroides*) on soil infectivity and soil conduciveness to take-all caused by *Gaeumannomyces graminis* var. *tritici*. Soil infectivity was evaluated by measuring the disease index on susceptible wheat plants grown on soil samples collected from the field. Soil conduciveness to the disease was obtained by measuring disease indices on plants grown on soil samples to which different amounts of take-all fungus inoculum were added. One experiment (expt. 1) was carried out using soils from farmers' fields (two fields in 1994 and two in 1995); soil infectivity and soil conduciveness were evaluated for three experimental situations : bare soil, soil with wheat volunteers and soil with blackgrass plants. In 1994 the soil

infectivity was zero in bare soil, high with the wheat cover, and intermediate with the blackgrass cover. In 1995 the soil infectivity was uniformly low for all three conditions. Soils bearing wheat were less conducive than bare soil, soils bearing blackgrass and bare soils were similarly conducive. A second experiment (expt. 2) carried out in 1995 compared the soil infectivity and soil conduciveness to take-all of soils planted with wheat or blackgrass in set-aside land after periods of wheat monoculture of 0-6 years. The soil infectivity was low for all treatments. The soil was more conducive after blackgrass than after wheat. In both cases, the soil conduciveness was less when the monoculture had continued for more than 4 years. The decline was less after blackgrass than after wheat. Thus, whenever set-aside is set up during the increase phase of the disease in fields with cereal successions, abundant wheat volunteers might hinder the expected positive effect of a break in cereal successions on take-all development. The presence of blackgrass in a set-aside field, with significant soil infectivity and high soil conduciveness, might increase the risks of take-all development in a wheat crop following set-aside.

**Abbreviations:** Ggt = *Gaeumannomyces graminis* (Sacc.) von Arx et Olivier var. *tritici* (Walker)

## Introduction

The Common Agricultural Policy was reformed in 1992 in order to regulate crop production in the EU, and set-aside (*i.e.* no production on part of the arable land) was imposed on European farmers. The set-aside regulations and set-aside rates have changed considerably since 1992. However, farmers in most European countries have been allowed to include set-aside as part of a crop rotation. This means that such rotational set-aside is preceded and followed by a crop. The 1994 data from the French Ministry of Agriculture indicate that rotational set-aside accounted for about half of the set-aside area in France (excluding set-aside for industrial crops); the other half was long-term set-aside.

Rotational set-aside raises questions about its effects on insect and disease epidemiology (Hancock et al., 1992; Yarham and Symonds, 1992). These effects will depend on the nature of the previous and following crops, and on the vegetation in the set-aside field. The vegetation in set-aside fields may vary greatly from one field to another (Fisher et al., 1992; Wilson 1992), since farmers are allowed to sow a range of cover crops, or let natural regeneration take place, which results in a mix of volunteers and weeds. Set-aside fields in which natural regeneration occurs contain large numbers of wheat plants when the preceding crop was wheat, which is common in France. Blackgrass (*Alopecurus myosuroides* Huds.) is also a common weed from cereal successions in France, and is frequently found in set-aside fields, where it may even be the dominant weed (Chauvel et al., 1995).

We have studied the effects of one year set-aside on take-all (*Gaeumannomyces graminis* (Sacc.) von Arx et Olivier var. *tritici* (Walker) = Ggt), whose development is known to be greatly influenced by crop succession (e. g. Colbach et al., 1994; Slope,

1967; Steinbrenner and Höflich, 1984), and which might be expected to be most influenced by set-aside (Yarham and Symonds, 1992). Thus, intensive cereal cropping leads to a greater risk of take-all development, but Gerlagh (1968) and Lemaire and Coppenet (1968) showed the transient effect of wheat monoculture on the increase in the disease, which is followed by a decline. In France, 80 % of the wheat crops are grown as a first wheat, i.e. after a crop other than wheat, mainly to avoid problems due to soil-borne pathogens, such as take-all fungus. The risk of using set-aside covered with wheat volunteers and blackgrass, which is also a host of the fungus (Nilsson, 1969), was assessed by considering the effects on soil conduciveness to take-all and the build-up of pathogenic inoculum in soil cropped with wheat in rotation or in monoculture.

## **Materials and Methods**

### *Soils and sampling*

Experiment 1 : soil samples were collected from farmers' set-aside fields in the Brie area, in the central part of the Paris basin (France). The soil was loamy with 14-24% clay. The crop preceding the study had been winter wheat in all the fields. Set-aside with natural regeneration of vegetation following the wheat crop led to a great diversity of flora within each plot under study. Each plot included areas of bare soil (a few m<sup>2</sup> without any growing plant), soil with wheat volunteers, and soil with blackgrass. Samples were taken from two fields in 1994 (fields A and B) and from two others in 1995 (fields C and D) in April before the canopy destruction (seven months after the harvest of the previous winter wheat crop). The plants were removed from the non-bare soil, and 20 kg samples of soil were taken from the blackgrass, wheat and bare areas.

Experiment 2 : soil samples were collected from plots involved in a long term rotation experiment at Grignon (1°58'E, 48°51'N) in the Paris basin (France). This long-term experiment compared the effects of different durations of cereal monoculture (Colbach and Huet, 1995). A total of seven durations of wheat monoculture were selected from this long-term experiment in autumn 1995 and used for experiment 2. The durations ranged from 0 (potato in 1994-95) to 6 years of continuous wheat crop (Table 1). Each crop succession was present in three plots (10.5 m<sup>2</sup> per individual plot) in a block design. Plots were divided into two subplots (3.5 m<sup>2</sup>) where the set-aside sown with blackgrass or with wheat were compared. Soil samples (20 kg) were taken from each subplot after 7 months of wheat and blackgrass growth.

All experimental soil samples (soil from 30 sampling points per plot sampled with a small shovel and mixed) were collected at a depth of 0-15 cm. They were air-dried and ground to give particles of 5 mm or smaller.

#### *Soil conduciveness measurement*

The method described by Lucas et al. (1989) was used. The ability of a soil to allow expression of pathogenicity in a population of susceptible host plants was assessed by introducing increasing amounts of inoculum. Ggt inocula were grown on barley seeds that had been soaked in water (W/V = 1) and autoclaved (1h, 120°C) twice within 24 hours. After a 3-week incubation at 20°C, the colonized seeds were air-dried, ground, and sieved to obtain propagules (infectious particles) of size 1-1.6 mm. The propagules were mixed into soils at concentrations of 0, 150, 500 or 1500 units kg<sup>-1</sup>. For each concentration, four pots were filled with 500 g soil; each pot was considered to be a replicate. Pots were then seeded with 5 caryopses of wheat cv. Talent and kept for 5

weeks at 15°C day, 10°C night, 14 h photoperiod, 80-90% relative humidity and 150  $\mu\text{mol.m}^{-2}.\text{s}^{-1}$  radiation. The plants were harvested, the roots were gently washed under running water, and root necrosis was recorded. Each plant was assigned to one of five disease severity classes (0, 1, 2, 3, 4) corresponding to zero, 1-10, 11-30, 31-60, 61-100% of the root system showing take-all lesions. A disease index (DI) was calculated for each inoculum level using the following formula:

$$DI = \sum_0^4 (n_i * i) * (\sum_0^4 n_i)^{-1}$$

where

$n_i$  = number of plants assigned to the  $i$  class

$i$  = severity class

The conduciveness of soil to the disease is illustrated by the disease indexes produced in response to the range of infestations (Lucas et al., 1989).

#### *Soil infectivity measurement*

The disease index measured in non-artificially-infested soils (rate 0) was considered to be a measurement of the infectivity of these soils (i.e. the expression of the resident Ggt inoculum).

#### *Statistical analysis*

The influences of blackgrass and wheat volunteers and of bare soil on the conduciveness and infectivity of each soil sample were analyzed by an analysis of variance on the

disease index for each dose of introduced Ggt and each canopy (GLM procedure, SAS software, SAS Institute, 1989). Mean values for each rate were then compared using the Student Newman Keuls test.

## **Results**

### *Experiment 1*

The results are shown in Figure 1. The soil infectivity for the bare-soil treatment was zero or very low in the four fields. Wheat and blackgrass treatments gave different results in 1994 and 1995. For the two fields studied in 1994, the soil infectivity was greater for the wheat treatment than for bare soil; blackgrass treatment gave an intermediate value. For the two fields studied in 1995, infectivity was zero or very low for all three soils.

The disease index of the bare soil increased with the increase in introduced Ggt for all four fields. The values for the highest concentration were 2.9-3.4, depending on the field considered. A similar trend was found for the blackgrass treatments, and the highest values obtained for the highest amount of Ggt were 3.4-4.0. The difference in disease indices between 0 and 1500 propagules  $\text{kg}^{-1}$  were very similar for bare soil and blackgrass treatments in the four fields (Table 2). Wheat treatment yielded different results : the disease index did not increase continuously in field A in 1994 with the amount of Ggt introduced. The difference in the disease indices between 0 and 1500 propagules  $\text{kg}^{-1}$  was far smaller for this treatment than for bare soil and blackgrass treatments (Table 2). Values for the highest rate in wheat treatment were lower than for the two other treatments. Though less pronounced, similar results were obtained for

wheat treatment on field B. The disease index for wheat treatment increased with the Ggt concentration in 1995, but it was often lower than for bare soil and blackgrass treatments. Soil conduciveness was lower for wheat treatment than for bare soil or blackgrass treatment in 1994 and 1995. The higher the soil infectivity, the lower was soil conduciveness in wheat treatments.

### *Experiment 2*

The disease indices for 0 and 1500 propagules  $\text{kg}^{-1}$  are shown in Figure 2 for each of the seven plots and for both wheat and blackgrass treatments. The soil infectivity (Figure 2a) was very low for all the plots that had undergone 0-6 years wheat monoculture before set-aside, but became greater than zero for both wheat and blackgrass treatments when the duration of monoculture had exceeded 2 years. There was no decline in the disease index with the duration of the monoculture for either treatment.

The disease indices for wheat and blackgrass treatments for different durations of monoculture yielded the same pattern at concentrations of 1500 (Figure 2b) 150 and 500 propagules  $\text{kg}^{-1}$  (not shown). For wheat treatment, the disease index increased slightly when the wheat monoculture had continued for 0 to 4 years, and declined with longer periods. The disease index for blackgrass treatment was higher than for wheat treatment, especially for durations of monoculture longer than 4 years.

## Discussion

The zero soil infectivity for bare soils in experiment 1 is consistent with the absence of susceptible plants needed for the survival or development of the fungus. The presence of roots that are potential hosts for the fungus (Nilsson, 1969) in set-aside flora made up of wheat or blackgrass, explains why the disease index was higher than zero without introduced Ggt. The expression of the Ggt inoculum resident in the soil was different in 1994 and 1995. These differences in soil infectivity measured with the same susceptible cultivar may be due to factors such as inoculum density, inoculum energy, inoculum virulence and biotic environment, components of the inoculum potential as defined by Lockwood (1988). In contrast, the patterns of soil conduciveness to the disease in 1994 and 1995 were similar, which suggests that the biotic and abiotic environments do not really account for the differences observed. Inoculum energy and inoculum density are difficult to evaluate and inoculum virulence was not measured. Nevertheless, the most important factor determining year-to-year differences in infectivity was probably the inoculum density. The density of Ggt populations at the time of sampling is the result of an increase in soil contamination due to the growth of the previous susceptible wheat crop and to the presence of susceptible wheat volunteers or blackgrass after harvest of the wheat crop. The climatic conditions during the experiment probably accounted for most of the year-to-year difference.

The lower soil infectivity in the areas of blackgrass than in the wheat-bearing plots in 1994 is consistent with the lower susceptibility of blackgrass to take-all (Nilsson, 1969). Soil bearing wheat was more suppressive than bare soil. This is consistent with the development of a microflora that is antagonistic to Ggt in the infected wheat rhizosphere and which is partially responsible for the soil conduciveness

(Cook and Rovira, 1976). The variations in disease index were probably due to the actions of both pathogenic and antagonistic microfloras. The antagonistic microflora populations increase at and close to the sites of necrosis due to take-all (Sarniguet and Lucas, 1992; Sarniguet et al., 1992), leading to reduction of the disease. Our results for wheat treatment in experiment 1 obtained under farmers' field conditions thus corroborate previous reports on the build-up of antagonistic microflora. The conduciveness of the soil bearing blackgrass was the same as bare soil, and greater than that of wheat-bearing soil, suggesting that the antagonistic activity of the microflora did not increase, despite an increase in take-all inoculum infectivity. This might indicate that the association between susceptible plants and pathogenic inocula does not always lead to the development of a microflora antagonistic to Ggt. Thus, though blackgrass plants in set-aside fields resulted in less soil infectivity than wheat volunteers, they did not reduce the soil conduciveness to the disease. Set-aside that allows natural regeneration may therefore have deleterious effects if blackgrass develops. Bare soil yielded low soil infectivity, whereas wheat volunteers, despite an increased soil infectivity, maintained or enhanced natural soil suppressiveness to disease. This is important whenever wheat monoculture is continued for long enough to observe a take-all decline phase. But there is still need for more information on the soil infectivity and conduciveness of soil to take-all immediately after a previous wheat crop has been harvested, and prior to colonization by wheat volunteers, blackgrass, or in the absence of colonization by plants. The uneven distribution of wheat or blackgrass volunteers in set-aside fields might be due to the heterogeneity of the previous wheat crop. Take-all could have developed in irregular patches during the growth of the wheat crop, as it usually does, resulting in the local production of smaller wheat seeds that fall onto the soil during harvest, and abundant weed development in the same places due to the poor

growth of the wheat crop, as also often happens. Thus, patches of take-all in the previous wheat crop might account for the irregular patches of wheat volunteers and blackgrass in a set-aside following a wheat crop. But the effects of blackgrass, wheat volunteers and bare soil on the disease remain the same, although they should be linked not only to a set-aside effect but also to the previous cropping history.

The blackgrass and wheat volunteers did not result from natural regeneration in experiment 2, but were sown according to a standard procedure on randomly selected plots, after the harvest of the previous wheat or potato crop. The soil characteristics of wheat and blackgrass treatment plots were thus assumed to be similar. The low soil infectivity for both treatments and for any of the wheat monoculture durations (Fig 2a) may be explained in the same way as the 1995 results in experiment 1, by a year effect, influenced by climate. The disease index, which decreased after 4 years of continuous wheat cropping according to our bioassay of soil conduciveness, corroborates the phenomenon of take-all decline in wheat monocultures (Fig 2b). The greater disease index in the blackgrass soils is consistent with the results of experiment 1, and confirms that blackgrass resulted in less suppression of take-all than did wheat. Nevertheless, there was still some reduction in the disease in soils studied after 4-6 years of monoculture, showing that the mechanism leading to the decline of take-all after prolonged wheat monoculture is only partly influenced by 7 months of blackgrass growth.

The data on the infectivity and conduciveness to take-all of soils bearing wheat volunteers in set-aside fields with natural regeneration are in keeping with those obtained by Yarham and Symonds (1992). They measured take-all contamination in wheat growing after a period of set-aside, and showed that a large number of wheat volunteers in set-aside fields reduced the effect of set-aside as a break in cereal

successions. Our results indicate that wheat volunteers in a set-aside field maintain the decline in take-all that results from prolonged wheat monoculture. Set-aside might even be included in the management of wheat monoculture, when it is a feature of farming practice, by replacing the wheat crop whenever yield losses are at a maximum because of high levels of disease before a decline. This can only be of value if wheat volunteers are dominant in a set-aside canopy. We have shown that the effects of blackgrass in set-aside fields are related to the ability of the weed to develop a significant level of infectivity and to make soils more conducive to Ggt. Its presence in a field could thus increase the risk of take-all developing on the next wheat crop. In conclusion, the impact of set-aside flora on the risk of take-all should be measured taking into account both aspects of disease development (soil infectivity, soil conduciveness) together with the cropping history of the field.

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Figure 1. Soil infectivity (at rate 0) and soil conduciveness (disease index at other rates) for bare soil ( $\sigma$ ), wheat ( $\bullet$ ) and blackgrass ( $\blacksquare$ ) treatments, for fields A and B in 1994 (*a* and *b*), and C and D in 1995 (*c* and *d*). Letters within a rate give the groups by Student Newman-Keuls analysis at 0.05.

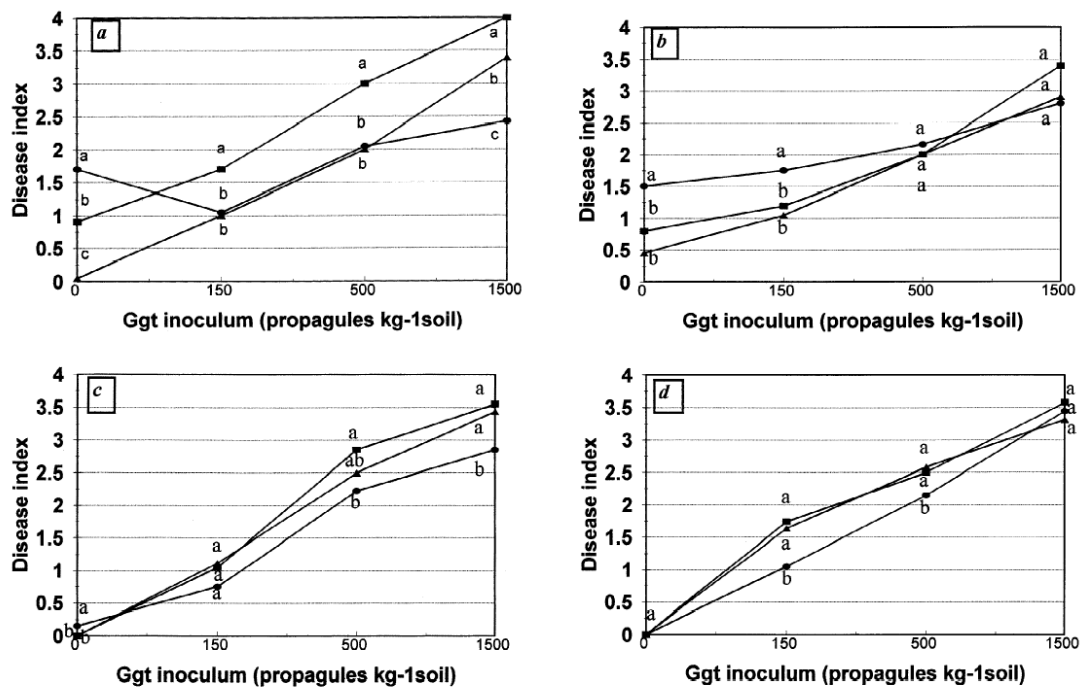
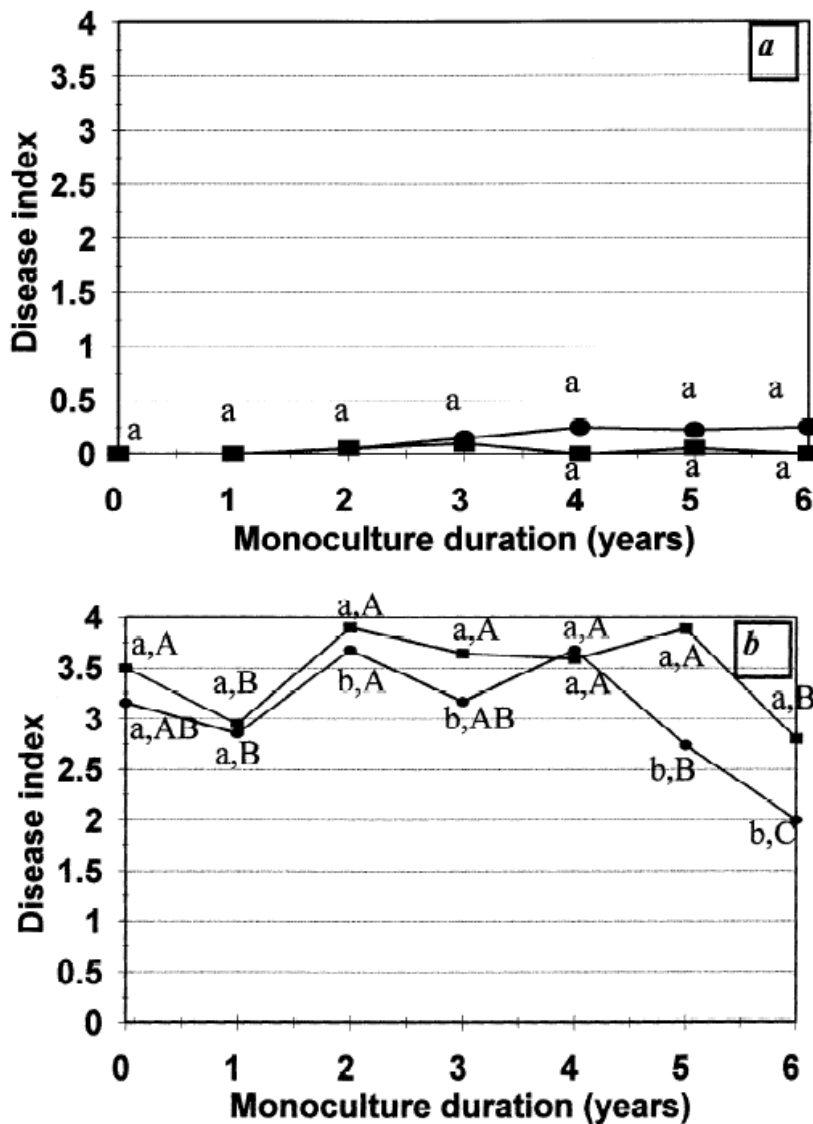


Figure 2. Soil infectivity (at rate 0) (a) and disease index measured in the soil conduciveness test for infection with 1500 *Gaeumannomyces graminis* var. *tritici* propagules kg<sup>-1</sup> (b) for soils with wheat (●) and blackgrass (■), after different durations of wheat monoculture prior to set-aside. Capital letters within a monoculture duration (year) and lower-case letters within a plant treatment (canopy) give the groups by Student Newman-Keuls analysis at 0.05.



*Table 1.* Crop successions in experiment 2. The number of the treatment indicates the duration of wheat monoculture (in years) prior to the experiment.

Treatment	1988	1989	1990	1991	1992	1993	1994	1995
6	<b>S. Beet</b>	Wheat	Wheat	Wheat	Wheat	Wheat	Wheat	- Wheat - Blackgrass
5	Wheat	<b>S. Beet</b>	Wheat	Wheat	Wheat	Wheat	Wheat	- Wheat - Blackgrass
4	Wheat	Wheat	<b>Potato</b>	Wheat	Wheat	Wheat	Wheat	- Wheat - Blackgrass
3	Wheat	Wheat	Wheat	<b>Potato</b>	Wheat	Wheat	Wheat	- Wheat - Blackgrass
2	Wheat	Wheat	Wheat	Wheat	<b>Potato</b>	Wheat	Wheat	- Wheat - Blackgrass
1	Wheat	Wheat	Wheat	Wheat	Wheat	<b>Potato</b>	Wheat	- Wheat - Blackgrass
0	Wheat	Wheat	Wheat	Wheat	Wheat	Wheat	<b>Potato</b>	- Wheat - Blackgrass

*Table 2.* Differences in the disease index between the concentrations 0 and 1500 propagules of introduced Ggt inoculum per kg.

	A	B	Field	C	D
Wheat	0.73	1.30		2.70	3.45
Blackgrass	3.09	2.60		3.60	3.58
Bare soil	3.35	2.45		3.56	3.31