Management of inflorescence and viable seed production of blackgrass (Alopecurus myosuroides) on set-aside in France

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Abstract - Two experiments were carried out, one in a glasshouse and the other in the field, to assess the effect of weeding management (type and date) on blackgrass seed production during set-aside. The results of the field experiment were used to establish relationships between head length and spikelet number per head. Head number per plant was not reduced by one mowing in glasshouse, but it was reduced by double mowing. Early (beginning of anthesis) or intermediate (hal-anthesis) glyphosate reduced head number by 72 or 50 % respectively compared to a control, but later (during seed ripening) application did not. A first mowing reduced length of heads elongated after the mowing by 25 % compared to a control, but less than two successive mowings (by 50 %). Seed viability was greatly reduced by early glyphosate (by 91 %) compared to a control, less by medium date spraying and two mowings (by 17 % and 47 % respectively), and not by one mowing or late spraying. These results were checked at two densities in the field experiment. The results of head length for low density were consistent with those in glasshouse, but head number did not differ between treatments. Head number per plant and head length at the highest density were lower (from 50 to 76 % according to the treatments for head number and from 3 % to 16 % according to the treatments for head length) than for individual plants. These preliminary results give indications, which should be checked in farmers' fields conditions, for better control of blackgrass in set-aside fields.

Key-words: Blackgrass / viable seed production / set-aside

Short title: Head and viable seed production of blackgrass

Introduction

The reforms made to the European Union Common Agricultural Policy in 1992 to regulate crop production imposed set-aside (*i.e.* no crop production on part of the arable land) on European farmers. Rotational set-aside has a variety of not always quantified effects on insect and disease epidemiology (Hancock et al., 1992; Yarham & Symonds, 1992; Dulout et al, 1997), and on weed seed production (Jones & Naylor, 1992; Lechner et al., 1992; Rew et al., 1992, Connolly & Naylor, 1996) and dissemination (Wright & Bonser, 1992). The evolution of flora under and after set-aside has been studied by several authors (for example Aquilina, 1992; Brodie et al., 1992; Clarke & Cooper, 1992; Zwerger et al., 1993; Boberfeld & Jasper, 1994…), and Rew et al. (1992), Lawson et al. (1992) and Jones and Naylor (1992) have assessed the seed production of weeds during set-aside. They observed that seed production could occur, and that the management of the set-aside field greatly affected this seed production. It would therefore be useful to be able to predict the effects of set-aside management on weed seed production, in order to propose set-aside management limiting weed seed production, and an increase in the size of the weed seed bank in the soil.

Alopecurus myosuroides Huds (blackgrass) is a common weed in cereal rotations in Europe (Froud-Williams & Chancellor, 1982; Orson & Harris, 1997; Jouy & Guilbert, 1998), and is frequently found in set-aside fields (Lechner et al., 1992; Shield & Godwin, 1992; Zwerger et al., 1993; Clotuche et al., 1997), where it can be the dominant weed (Chauvel et al., 1995). Blackgrass is one of the most difficult annual weeds for eliminate from fields in a cereal succession. Farmers frequently plan their timing of weed control practices to prevent blackgrass reproduction in set-aside areas. There are several references on the demographic parameters of blackgrass (e. g. Chauvel, 1991; Melander, 1995), among which the studies of Naylor (1970, 1972) and Moss (1979, 1980, 1983, 1985, 1987, 1990) provide the most comprehensive information. Moss (1990) and Chauvel and Gasquez (1993) proposed quantitative models for predicting the development of blackgrass populations. However, they cannot be used to predict blackgrass seed production in set-aside fields because the values were developed using plants in competition with crops. Shield and Godwin (1992) showed that blackgrass occurence is sensitive to cutting frequency and Clarke and Cooper (1992) found that frequent cutting progressively reduced the number of heads regrowing after each cut, but provided no quantitative data. Better knowledge of the effect of weeding on seed production of blackgrass during set-aside is necessary in order to establish appropriate set-aside management. It would also be useful to make weeding in wheat crop more appropriate if it is impossible to completely avoid blackgrass seed production.

The purpose of this research was to determine head and seed production of blackgrass in set-aside as affected by management practice and plant density in glasshouse and field experiments. The viability of the seed produced was also determined. Management practices included simulated mowing or an application of a non selective herbicide (glyphosate), which are the most common weed management practices used in set-aside (Dalbiès-Dulout, 1999).

Materials and Methods

Two experiments were carried out at Grignon (1°58’E, 48°51’N) in the Paris basin (France), in a glasshouse (Experiment 1, 1996) or in loamy soil (Experiment 2, 1995). Blackgrass seed used in both experiments was collected from a population grown on a fallow land near Dijon (Eastern France, 5°02'E, 47°20'N), and stored under dry conditions until used. The growth stages were measured according to the Zadoks scale (Zadocks et al., 1974). A treatment growth stage was a median stage, *i.e*. the stage of half the plants on the recording date. Zadoks growth stages 50 and after were determined on the plant head population: a growth stage was attained when half of the heads had attained this stage. A head is considered to be mature when spikelets at the top of the inflorescence begin to fall. Mowing was simulated by cutting the plants with shears to a height of 100 mm. Glyphosate (Roundup®, 360 g a.i. l^{-1}, 0.75 $l.ha^{-1}$, Monsanto) was sprayed with a hand-sprayer at 270 g a.i. ha^{-1}.

Glasshouse experiment (Experiment 1)

Seeds were kept for two weeks in the dark at 4°C before sowing in March 1996. They were germinated on a potting mix in the glasshouse and when seedlings had reached Zadoks stage 13 transplanted into a 4-liter pot (one plant per pot) filled with a mixture of earth:sand:peat (15:4:1 by volume). Plants were watered as necessary and fertilised with a drip irrigation system with automatic release. Treatments consisted of a non-treated control (T1); a single mowing at Zadoks stage 59 (EM1), 65 (MM1), or 80 (LM1); a single application of glyphosate at Zadoks stage 61 (EG1), 65 (MG1) or 79 (LG1); and an initial mowing at Zadoks stage 67 followed by a second mowing when new heads were at the same stage (2MM1). Treatments were replicated 10 times. Each replicate consisted of an individual plant. Growth stages of individual plants were determined and heads per plant were counted every 3-4 days. The number of heads before mowing or spraying was also recorded. The lengths of 10 randomly chosen heads of each plant from each treatment were measured at the end of the experiment (100 heads per treatment). The seed viability was assessed by performing germination tests at least 2 months after the harvest. Empty (i.e. unfertilised ovules or aborted seeds, E) and full seeds were sorted and counted until 200 full ones were attained. These 200 seeds were germinated on filter paper sprayed with a solution of giberellic acid (100 mg.l^{-1}). The room temperature was about 20°C. The germinating seeds were counted after 10 and 15 days. The number of germinating seeds on the second date (N) was always the same as on the first date. The seed viability ratio (SV) was thus calculated as:

$$SV = N/(E+200) \qquad (1)$$

The seed viability was measured on three replicates (plants) for all treatments, except MM1 (twice) and T1 (four times). Analysis of variance was performed with STATGRAPHICS software for number of heads per plant and head length.

Field experiment (Experiment 2)

The seeds used in experiment 2 were sown directly in the experimental field on March 21, 1995. The experiment was conducted at low and high population densities of blackgrass. Low density consisted of single plants separated by a distance of at least 200 mm from other blackgrass plants, obtained by planting seeds at a depth of 30 mm on a row. Treatments in the low density were a non-treated control (T2), a single mowing at Zadoks stage 69 (MM2), a single application of glyphosate at Zadoks stage 65 (MG2), and an initial mowing at Zadoks stage 69 followed by a second mowing when new heads were at the same stage (2MM2). Each plant within a treatment was considered as a replication. The number of replications per treatment varied due to poor emergence of the blackgrass. The number of replications for T2, MM2, MG2 and 2MM2 were 19, 27, 13 and 20, respectively.

Plots for high population density treatments were established by planting five blackgrass seeds at a depth of 30 mm at 70 x 70 mm intervals in squares measuring 1 x 1 m. Two squares were established for each treatment. Treatments in the high density were a non-treated control (T2h), a single mowing at Zadocks stage 69 (MM2h), and an initial mowing at Zadoks stage 69 followed by a second mowing when new heads were at the same stage (2MM2h). Each plant within a treatment was considered as a replication. The establishment rate was low : plant number ranged 74-112 plants m^{-2} in the 6 squares, with a mean of 87.5 plants m^{-2}.

The final number of heads and head length were measured for each plant of each treatment of the low density. Only the mean head number per plant was measured for the high density treatments, and the head length of all plants. The number of spikelets per head were counted on about 50 heads from each of the three following treatments: 49 for T2h, 46 for MM2h, and 51 for 2MM2h.

Results

Head Length and Spikelet Production

Head length (HL, expressed in mm) and spikelet number per head (SNH) measured on heads of different treatments of high density in experiment 2 were used to establish relationships. The heads obtained from control plants (unmown T2h) were distinguished from those obtained after one mowing (MM2h), and from those obtained after two mowings (2MM2h). Linear fits calculated for each of these three head samples gave the following results (Cf figure 1):

T2h: $SNH = 1.74*HL - 35.97$ (n = 49, R^2 = 0.86) (2)

MM2h: $SNH = 1.61*HL - 20.17$ (n = 46, R^2 = 0.67) (3)

2MM2h: $SNH = 1.40*HL - 4.18$ (n = 51, R^2 = 0.77) (4)

Though comparison of the slopes showed no significant difference at a 5 % level between the three samples, the three different relationships were used in the other experiment to evaluate the spikelet number per head. Equation (2) was used for treatments T1, EG1, MG1 and LG1, equation (3) for treatments EM1, MM1 and LM1, and equation (4) for the treatment 2MM1.

Head Number and Length, Seed Viability

Experiment 1

The results of the management treatments in terms of final head number, head length, seed viability and seed production per head in experiment 1 are shown in Table 1. Variance analysis showed that the number of heads before mowing or spraying was not different (P>0.05) at this stage, whereas the final number of heads per plant and head length were significantly different (P<0.01). A single mowing (EM1, MM1, LM1) always gave the same final number of heads per plant as the control and as LG1. Both glyphosate application timings EG1 and MG1 reduced final head number (less than half the control). EG1 gave a slightly lower final number of heads than the head number before spraying, which means that some developing heads have been killed by the glyphosate after spraying at an early flowering stage (stage 61). The earlier the spraying, the greater the decrease in the final head number compared to the control. Double mowing led to a final head number intermediate between those for early glyphosate spraying and the control or a single mowing.

Differences between treatments were more marked for head length. Double mowing (2MM1) gave the lowest mean value (44 mm), half the control value. One early mowing at the end of inflorescence emergence (EM1), or at half-anthesis (MM1) gave the same result (66 mm), longer than 2MM1, but different from the control, whereas later mowing (LM1) during seed ripening reduced the head length to an intermediate value (48 mm), close to the value of 2MM1. Mowing always resulted in heads that were 50-75 % of the control head length.

Glyphosate spraying (EG1, MG1, LG1) gave head lengths that were always at least equal to the control value. This could be due to the fact that the heads remaining after spraying were the ones that appeared earlier on the plants, which are also probably the longest.

The mean seed viability ratio for the control (T1) was 42.6 %. It was higher for all the treatments with one mowing, and nearly halved by double mowing. SV ranged from 4.0 % for early spraying (EG1) to 61.2 % for late spraying (LG1). A small difference in the date of spraying (EG1 *vs* MG1) gave large differences in the seed viability ratio (4.0 *vs* 35.4 %).

The final estimated number of viable seeds by plant was highly decreased by double mowing and early glyphosate spraying, and at a lower extent by intermediate date of glyphosate spraying and late mowing.

Experiment 2

The mean number of heads per plant and the head length for both densities are shown in Table 2. The number of heads per plant at low density was not significantly different between treatments at the 5 % level. Differences between mean values, and the mean values themselves, were far lower than in experiment 1, and standard variations were greater. At low density, treatments had a significant (P<0.001) effect on head length. The control had the longest heads, slightly less than the control in experiment 1. As in experiment 1, one mowing at the end of flowering (MM2) reduced head length, and double mowing was yet more effective. Spraying with glyphosate at half anthesis resulted in head length intermediate between those after one and two mowings. The final estimated number of seeds by plant (viable and non viable together) was more decreased by double mowing than by intermediate date of glyphosate spraying and by single mowing, as was the final estimated number of viable seeds by plant in the experiment 1.

The number of heads per plant for the high density treatments were not subjected to variance analysis, as only mean values were recorded. This mean final head number was much lower, about 25-50 % of those recorded for treatments with low density. The head lengths of control, mowing and double mowing treatments at low and high density were compared, by performing three variance analysis using plants as replicates. Head length was always significantly ($P<0.01$) lower in high density treatments than in the similar low density treatments. Differences were nevertheless slight, from 15 % (double mowing) to 3 % (single mowing). The final estimated number of seeds by plant (viable and non viable together) was very low in all the treatments for the high density, compared to the low density in the same experiment.

Discussion

The relationships between head length and spikelet number are close to that given by Chauvel and Gasquez (1993). A medium head length of 70 mm resulted in about 90 spikelets per head, which is close to the 100 value given by Moss (1990), lower than the average of 120 in Naylor (1972), and far less than the 150 value found by Stryckers and Delputte (1965). In our experiment the relationship did not differ a lot whether the heads were from plants mown or not. This seems to mean that the inflorescence structure is not very affected by plant management, and that it is mainly the head length which supports the reported (Menck & Börner, 1971) differences obtained when plants are in different growth conditions.

The differences for head number and head length between experiment 1 and 2 in both absolute and relative values might well be due to differences in genetics or growth conditions, but as the seeds were all from the same batch, genetic differences must be lowered. Nevertheless the predicted spikelet number of 8 plants from a single UK field which had a significant relationship of spikelet number and head length (Naylor, 1973) ranged from 104 to 140. The plant density in experiment 2 (low density treatment) was very low, and there was no competition for light. The blackgrass plants in the second experiment in the field were given water, but no mineral fertilizer was added, as was the case in the glasshouse (experiment 1). The difference in head length of the controls between the two experiments might also be due to differences in the plant growth conditions in the two experiments. The results from experiment 1 (increased head length due to glyphosate spraying during flowering) were not confirmed in the field, maybe due to greater between-plant differences within each treatment in the field.

Our results showed that cutting the blackgrass plants decreased the number of seeds more by decreasing the head length that the number of heads per plant, though Lechner *et al*. (1992) showed differences in head number also. These differences between our results and those of Lechner *et al*. (1992) might be due to the differences in growth conditions in experiments: new growth after cutting is probably far more difficult in highly competitive conditions with other plants (e.g. within a crop, or a set-aside field with a lot of weed species) than in individual plant conditions (Dulout et al., 1998). Nevertheless the effect of cutting was more marked after two mowings. But large differences in head length between mowing treatments were observed in both experiments. These differences probably reflect the differences in growth conditions during inflorescence development, and the effect of mowing on the reduction of the green leaf area, which resulted in a large decrease in the number of seeds per plant, especially after late mowing or double mowing. This preliminary result on the effect of mowing on head length should be checked in farmers' field conditions.

The effect of mowing on the viability of seeds from new heads had never been studied, and we found no clear effect. The seed viability ratio we observed for the control in experiment 1 (42.6 %) was low, but consistent with the 49 % obtained by Naylor (1972), the 43-76 % reported by Moss (1983), and the 49-87 % reported by Chauvel (1996). One mowing did not affect the seed viability ratio; two mowings decreased it. This could be due to a poorer pollen flow and a subsequent lower fertilisation ratio, to worse seed growth conditions for later flowering and ripening, or one more time to reduced assimilates available for inflorescence growth due to the decrease of green leaf area due to mowing. The increase in density from low to high greatly decreased the number of heads per plant, and was more than three times as severe as the effect reported by Moss

(1990) for a plant density 0-100 per m². This could be due to the fact that Moss (1990) reported results for blackgrass in competition with a wheat crop, where the intra-species competition effect was probably biased by the inter-species effect at low densities. Our results confirm that blackgrass is sensitive to competition and seem to indicate that it adapts its head production to the available growth factors. But the differences in head length between high and low density treatments were far smaller, although significant. This suggests that the number of heads per plant is the major variable that is influenced by growth conditions for the blackgrass plant.

Glyphosate treatment gave highly date-dependant results. Early spraying resulted in a very low viable seed production, whereas late spraying did not reduce the seed below that of the control. The number of heads per plant was decreased after an early spraying (EG1), but it was mainly the seed viability ratio which led to the differences from the control and from the spraying six days later (MG1). Shuma et al. (1995) had already shown a similar importance of the spraying date on seed viability for *Avena fatua*. If confirmed in field trials, this is an important practical result for deciding when to treat blackgrass in set-aside fields. Farmers often use the blackgrass head appearance as a signal for deciding the date of treatment in set-aside (Dalbiès-Dulout, 1999), and our results show that this decision rule provides high risk of seed production if the treatment is delayed even by a short time. More accurate studies about the mechanisms (herbicide translocation in the plants) which are responsible for the date of application effect are needed.

Finally, our results show that the demographic parameters of blackgrass we have studied are very different in set-aside compared to measurements in competition in winter cereals crops reported by different authors. Our results also show that single mean values for demographic parameters cannot be used for modelling blackgrass dynamics in set-aside, because head number, head length and seed viability all seem to depend on the type of weeding used and application date. The current results could be used to adapt the existing demographic models to the case of set-aside.

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Table 1. Number of heads per plant before management, final number per plant, head length, seed viability ratio and calculated number of viable seeds per plant for the treatments in experiment 1.

Treatment	Head number before treatment	Final head number per plant	Head length (mm)	Seed viability Ratio (%)	Estimated viable seed number per plant
T1	18.5 a	53.5 d	88 d	42.6	2670
EM1	21.9 a	58.3 d	66 c	66.5	3338
MM1	18.4 a	47.8 cd	66 c	63.7	2621
LM1	19.7 a	57.5 d	48 b	66.6	2187
2MM1	18.5 a	35.8 bc	44 a	22.7	467
EG1	17.4 a	15.2 a	100 e	4.0	84
MG1	20.6 a	26.9 ab	106 f	35.4	1414
LG1	18.6 a	47.7 cd	89 d	61.2	3471

Means values within columns followed by the same letter are not significantly different at the 0.05 level of probability as determined by the Newman-Keuls test.

Table 2. Final number of heads per plant, head length, calculated seed number (viable and non viable together) per plant for the treatments in experiment 2.

Density	Treatment	Final head number per plant	Head length (mm)	Calculated seed number per plant
Low	T2	26.4 a	77 a, α	2587
	MM2	21.9 a	65 b, α	1850
	MG2	21.5 a	62 c	1546
	2MM2	17.9 a	49 d, α	1150
High	T2h	6.3	69 a, β	528
	MM2h	11.0	63 b, β	894
	2MM2h	7.2	41 c, β	383

Roman superscript letters (a, b, c, d) indicate significant differences within a plant density (Newman-Keuls test; $P<0.05$). Significant differences (Newman-Keuls test; $P<0.05$) between high and low density are indicated by Greek superscripts (α,β) for homologous treatments.

Fig. 1. Relationships between number of spikelets per head and head length (*a*: without mowing; *b*: one mowing; *c*: double mowing).