

### PROJECT REPORT No. OS32

DORMANCY AND
PERSISTENCE OF VOLUNTEER
OILSEED RAPE

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# DORMANCY AND PERSISTENCE OF VOLUNTEER OILSEED RAPE

by

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### 1. Executive Summary

When oilseed rape crops are harvested a proportion of the seeds are left in the fields. No harvesting method will collect all the seeds from the mature crop plants. Appreciable quantities of rape seeds can be left behind, providing a source of volunteer plants for subsequent crops. These volunteer plants are important because they compete with following crops, can act as sources of pests and diseases for the next or adjacent rape crops and affect the purity of following rape crops grown in the same field. The current concerns relating to gene escape from genetically modified oilseed rape gives added emphasis to the potential importance of volunteers and feral rape derived from shed or lost rape seeds.

The objectives of the project were as follows:

- 1.) quantify oilseed rape seed losses at harvest,
- 2.) investigate genetic, physiological and environmental factors controlling dormancy,
- 3.) study the influence of agronomic factors on persistence,
- 4.) model the population-dynamics of oilseed rape

### Seed losses

Harvesting losses assessed in 1995 and 1996 were high, confirming the conclusion from earlier work that rape seed losses of several thousand seeds per m<sup>2</sup> are not exceptional. Losses in the region of 5,000 seeds/<sup>2</sup> are common and losses higher than 10,000/m<sup>2</sup> can occur. Most losses occur before harvest, or at the cutter bar and are not the result of poor setting of the harvester. Delayed harvesting dramatically increases seed loss. As the initial seed losses are so high, a very low % survival can still result in an appreciable weed problem.

### • Factors controlling the onset of dormancy

Laboratory studies showed that fresh rape seeds are not dormant (no primary dormancy) but they develop dormancy over the first month after shedding. Petri-dish tests confirmed that water stress and darkness were the major requirements for the development of secondary dormancy. Thus, seeds buried in dry soil have the greatest potential to become dormant. It is also possible that anaerobic conditions and low temperatures can induce dormancy. Preliminary research indicates that this response of rape seeds to light is mediated by the phytochrome system, which controls a number of physiological responses to light in many plant species.

The work also showed that leaving seeds on the soil surface for 2-4 weeks reduced the subsequent induction of dormancy. In extreme cases seeds left on the soil surface for 4 weeks completely failed to become dormant. Also, seeds exposed to alternating temperatures were shown to be less likely to persist than those given a steady temperature. Finally, the work revealed that seeds of different cultivars of rape have different potentials to become dormant. Most rape cultivars have a low dormancy potential but a significant minority, including Apex and a number of other currently popular cultivars, has a greater ability to persist. The differences between cultivars seem to be primarily of genetic origin, as several populations of the same cultivars exhibited broadly similar levels of dormancy. This work has been done in Petri-dishes and although preliminary field work confirmed the general trends further verification in the field is still required.

### • Influence of agronomic factors on seed persistence

The Petri-dish tests identified a number of issues relating to the persistence of volunteer rape that required confirmation in the field. It is clear from the field experiments started in 1995, 96 and 97 that when the soil conditions at harvest are dry, seed persistence is likely to be greatest. We believe that exposure of seeds to darkness in dry soils is the key reason for the number of dormant seeds that are produced. This has been confirmed in field tests. Rape fields should not be cultivated after harvest, as this buries some seeds in darkness, providing conditions for the onset of dormancy. Leaving seeds on the soil surface for several weeks also appears to inhibit the development of dormancy when the seeds are eventually buried. Seed persistence seems to be minimal in wet Augusts.

Optimum post-harvest management strategies are;

- i) keep rape seeds on the soil surface for as long as possible, as this seems to minimise persistence.
- ii) do not plough or cultivate the rape stubble for 2-4 weeks after harvest, particularly if the soil is dry. This incorporates many seeds into the soil providing ideal conditions for the development of a large seed-bank.
- iii) use a herbicide to control germinated seedlings rather than a cultivator or harrow.

### Long-term persistence

Once a soil seed bank has been created, the decline in seed numbers appears to be relatively slow. The data to support this conclusion are rather limited, as few experiments have generated adequate seed banks to follow the decline. There is good evidence that substantial numbers of seeds will last two years, and some indication that perhaps 10% of seeds will survive for 5 years and 1% for 10 years. Persistence is very difficult to predict precisely because of the variation in the percentage initially becoming dormant, the cultivar used and site variables. It must be remembered that because of the high levels of seed losses even a low % persistence can cause serious problems in subsequent crops.

### Modelling the population dynamics of oilseed rape

A model based on the life cycle of volunteer rape is being constructed. This type of model is a valuable tool to explore the key aspects of the biology of this weed that contributes to its long-term survival. What are the main aspects controlling seed numbers in the soil? Unfortunately, long-term data on two parameters that appeared to be important from sensitivity analysis: seed survival in the soil from one year to the next and the proportion of seeds in the seedbank giving rise to seedlings, are based on very limited data. The only information on which to base the derivation of these parameters appear to be our own results from the 1995 field experiments. As we now (as of summer 98) have three year's data on persistence, these are currently being used to improve the prediction model.

This project has generated many answers as to when and why rape seeds persist in the soil and become weeds in subsequent crops. We are still very short of information on some critical aspects and further work is needed, for example, to confirm long-term decline rates and the significance of apparent differences in dormancy between cultivars, in the field.

### 2. General Introduction

When crops are harvested a proportion of the crop seeds remain in the fields. No harvesting method, whether mechanical or by hand, will collect all the seeds from the mature crop plants. Over 20 years ago, Hughes (1974) and Cussans (1978) reviewed the problems arising from 'crops as weeds', concentrating on cereals, potatoes and oilseed rape. Cussans concluded that up to 5% of a cereal crop and 20% of a rape crop could be left in the field after harvest. Thus, volunteer crop weeds, arising from these shed seeds are not new. As far as oilseed rape is concerned, appreciable quantities of seed can be left behind, providing a source of volunteer plants for subsequent crops. These volunteer plants are important because they compete with following crops, can act as sources of pests and diseases for the next or adjacent rape crops and affect the purity of following rape crops grown in the same field. importance, as far as having competitive effects on other crops is concerned, depends on the crop in which they appear. Rape plants can be very vigorous and so when present as weeds, have the potential to be extremely competitive. In crops where a wide range of herbicides are available to control the rape seedlings, their presence is of minor importance but in crops such as linseed, sugar beet and field beans, volunteer rape is more difficult and expensive to control (Lutman, 1993). Control is made more complicated by the propensity of rape seedlings to emerge over a prolonged period of time, making the optimum timing of control difficult.

The importance of volunteers as contaminants of subsequent rape crops, emerged as a serious problem when regulations over glucosinolate levels were changed. Where 'double low' (low erucic and low glucosinolate) varieties were contracted to be grown, the presence of high glucosinolate volunteers had the potential to adversely affect the marketability of the low glucosinolate crop. Similar problems now arise where HEAR and 'conventional' rapes are grown in the same rotation. This problem is likely to increase in the future once the large number of oilseed rape cultivars with specific oil contents, currently under development, reach the market place (Carruthers, 1995). For example, high lauric rape is already being grown on a small scale. A further issue associated with the presence of volunteers relates to the persistence of genetically modified (GM) rape crops. There is much current concern about the potential for genes in GM rape to escape to neighbouring rape crops or hybridise with other related weeds. The presence of GM rape seed volunteers provides a mechanism for gene escape in time. Volunteers containing genes, for example for glyphosate resistance, could appear in subsequent crops, with possible consequences for weed management.

Quantitative data on long-term persistence of rape is limited. Schlink (1995, 1998) demonstrated that a small % of rape seeds buried in undisturbed soils could persist for 5-10 years and persistence up to 10 years is suspected in some arable fields (eg Sauermann, 1993). The surveys of volunteer and feral oilseed rape that have been in progress in Eastern Scotland (Wilkinson et al., 1995) have not definitively identified the longevity of rape populations but data on the cultivars found in recent assessments indicate that those that have not been grown since the early 1980s are still present. Thus, longevity of seeds in agricultural situations remains to be confirmed, but 5-10 years is suspected.

The persistence of rape seeds in the soil is somewhat unexpected, because the seeds when shed from the parent plants appear to have little or no primary dormancy (Schlink, 1994), so have the potential to germinate immediately if adequate moisture is available. This is not surprising because if rape seed had even a modest level of primary dormancy, then seeds collected from winter rape crops and then sown 2-3 weeks later (as the next crop) would not establish properly. It seems that the seeds acquire dormancy in the first few weeks after being shed at harvest. Research in Göttingen indicated that water stress and perhaps oxygen

deficiency associated with darkness were probably the critical environmental factors causing the onset of secondary dormancy (Pekrun, 1994; Pekrun *et al.*, 1997a, b). This work suggested that post-harvest cultivation might play an important role in determining the level of seed persistence.

Thus, volunteer rape is an important weed for a number of reasons and there is a clear need to investigate:

- i) how it occurs,
- ii) why the seeds persist
- iii) how persistence can be minimised.

This project aimed to investigate these three issues, through a combination of laboratory and field research.

### 3. Quantification of harvesting losses

### 3.1 Introduction

There are few-published results on seed losses before and during rape harvest. Estimates that have been published indicate that losses are in the region of 200 to 500 kg/ha, which equates to several thousand seeds/m² (Bowerman 1984; Vera et al. 1987; Meien-Vogeler 1988; Brown et al. 1995; Price et al. 1996). When the project started more data was needed to quantify the level of seed losses, as the input of seeds from a stand of rape determines the initial population of persisting seeds.

### 3.2 Materials and Methods

Harvesting losses were assessed immediately after the rape harvest on fields at Rothamsted in 1992, 1994 and 1995 and on one field of a commercial farm near Harpenden in 1996. In all cases the seeds were collected using an industrial vacuum cleaner, within a few days of harvest. All the material lying on the ground was collected from 20 random quadrats of 0.25 m<sup>2</sup>. Seeds were later extracted from the straw, stones and soil by sieving the material several times and finally floating the seeds in a saturated salt solution. After drying, the seeds were either counted or numbers were estimated from their total weight and their thousand seed weight.

### 3.3 Results and discussion

Seed losses in the 6 fields sampled varied from nearly 5000/m² to nearly 15,000/m² with a mean of 8853/m² (Table 3.1) The variation between the small samples tended to be quite high but we do believe that the overall conclusions are valid. It seems that seed losses during rape harvest can be in excess of 10,000 seeds/m², in weight terms equivalent to 400-450 kg/ha, or more than 10% of an average 3.0 t/ha crop. These figures are not dissimilar to those quoted from other studies, such as those of Bowerman (1984), Vera et al., (1987) and Price et al. (1996) (see above).

Studying some of the data from the 1992 experiment in more detail it appeared that losses immediately behind the combine (in the straw swath) were not appreciably higher than those between the swaths. This suggests that most losses were occurring at, or before, the cutter bar and were not due to poor setting of the combine, so that harvested seeds were lost from the combine with the rape straw. This conclusion concurs with that of Price et al (1996). Their work also showed that approximately 50% of the losses occurred in the standing crop prior to harvest. Thus, it seems that careful adjustment of the harvester will not solve the problem. There is no clear evidence whether swathing results in greater or smaller seed losses. It depends on the weather between swathing and harvest. Bowerman (1984) was unable to detect any clear trends, as the advantages / disadvantages of the two systems varied from site to site and year to year. Price et al (1996) found that when harvesting was done at the optimum time, there was little difference in losses between swathed and direct cut crops, but when harvesting was six days later, losses increased appreciably on the swathed plots but not on the direct cut ones.

**Table 3.1** Seed collected from commercially grown rape fields immediately after harvest. Average of 20 quadrats  $0.25 \text{ m}^2$  in size. Standard error of mean is given in parenthesis

Cultivar	Year	seeds/m <sup>2</sup>	s.e.m.
Libravo	1992	10704	(227.7)
Falcon	1994	6240	(1633.6)
Envol	1994	4769	(326.4)
Apex	1995	8711	(1450.0)
Envol	1995	8192	(1799.4)
Apex	1996	14507	(2384.3)
Mean		8853	

The key issue is to harvest the crop at the optimum time and not to harvest late. A minimum number of seed losses, perhaps 4000 seeds/m<sup>2</sup>, is inevitable until plant breeders develop rape cultivars that have a more robust pod.

# 4: Identification of genetic, physiological and environmental factors controlling the onset of secondary dormancy.

### 4.1 Introduction

Rape seeds, when freshly shed from pods show little or no primary dormancy (Schlink, 1994; Lutman, 1993) and so have the potential to germinate immediately after shedding, if environmental conditions are appropriate. The many thousands of seeds identified in Section 3 as being left in the field after harvest can germinate and in some years this can be clearly seen by the 'greening up' of rape stubbles with germinating seedlings during August. Yet, rape seeds can persist for many years (see General Introduction). In some way environmental conditions must be causing the onset of secondary dormancy. Preliminary work done in Germany at Göttingen University by Schlink (1994) and Pekrun (1994), over a period of 5 years showed that oilseed rape seeds can develop light sensitivity in the soil and that apparently as a consequence can persist ungerminated but viable for several years. This phenomenon was reproduced in Petri-dishes by subjecting seeds to water stress and darkness, or to an atmosphere low in oxygen (3% O<sub>2</sub>) and darkness. Water stress was simulated by using Polyethylene Glycol (PEG) to create the relevant osmotic potential. Extensive studies in soil in pots showed that it was extremely difficult to create the relevant dry conditions by using lightly moistened soils, because the moisture conditions varied within the pots.

As several aspects of the work that had been done in Göttingen had not been fully resolved, it was concluded that some more detailed laboratory studies were needed at the start of this project, before setting up field experiments on the influence of agronomic practices on the onset of dormancy. These aspects were:

- the effect of light and the combined effect of light and darkness on the onset of dormancy
- the effect of alternating temperatures on dormant seeds
- the implication of genotype for both, onset of dormancy and maintenance of dormancy

Most of this work has been done in Petri-dishes, using PEG to establish the relevant water potentials, but some small scale field work is also included.

### 4.2 General methods - Petri-dish tests

In all Petri-dish experiments four batches of 100 rape seeds were spread out in 9 cm Petri-dishes containing 2 layers of filter paper and 8 ml of a Polyethylene Glycol-solution (PEG 6000/8000), to induce rape seed into secondary dormancy. The concentration of the PEG solution was chosen according to Michel and Kaufmann (1973) to create specific water potentials. After an incubation period of up to 4 weeks all non-germinated, healthy seeds were removed from the Petri-dishes and placed in fresh ones containing 2 layers of filter paper and de-mineralised water. The seeds were kept under these conditions for 2 weeks. At the end of that period, all non-germinated, healthy looking seeds were counted. Occasionally, they were also checked thereafter for viability by stratification at 2 - 4°C for 3 days and subsequent retesting. Seeds tested in this way generally germinated immediately after stratification, as had been regularly observed in previous experiments in Germany. Thus, all those seeds that remained non-germinated at the end of the germination test and that were healthy looking, were defined as dormant.

For statistical analysis, the mean percentage surviving seeds was calculated for each Petri-dish. Where appropriate the untransformed data were used in the analyses but in some data sets transformations were needed. In these cases a logit transformation was used where D = number of seeds treated; n = number responding to treatment. The calculations were as follows:  $(0.5 * \log(P/(1-P)))$  where P = (n + 0.5)/(D+1), to avoid problems with 0 or 100 %.

## 4.3 Effects of light and darkness on the onset of secondary dormancy in six cultivars of rape

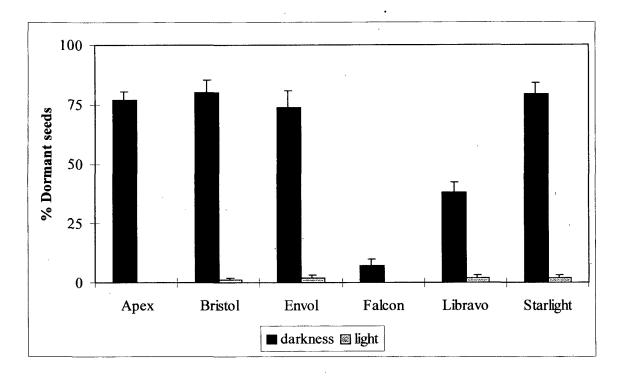
This work was done at the outset of the project to confirm that water stress and darkness were required to induce dormancy. The earlier German work had been done on cultivars that were no longer grown in the UK, so a selection of current cultivars was tested.

#### 4.3.1 Materials and methods

Seeds of six common UK cultivars of oilseed rape were incubated in darkness or in light at 20°C in PEG at a concentration that created an osmotic potential of -15 bar, for four weeks. They were then removed from the PEG and were germinated in darkness in water at 20°C for a further two weeks. Those not germinating were considered to be dormant.

### 4.3.2 Results and discussion

Fig 4.1 Percentage of dormant seeds of six cultivars of oilseed rape following incubation in PEG for 4 weeks in either the light or in darkness. Error bars = one s.e.m



After exposing seeds to either continuous light or continuous darkness during incubation under water stress (- 15 bars), it was evident that seeds could only develop dormancy in darkness and not in light (Fig. 4.1). Five of the six cultivars exhibited an appreciable level of dormancy following incubation in PEG in the dark and germination in the dark. The sixth cultivar, Falcon, showed the same trend but the overall level of dormancy was much lower. After incubation in light almost all the seeds of all six cultivars germinated in darkness and were not dormant. This confirmed earlier observations in Germany on five cultivars (Pekrun *et al.*, 1997b) and suggested that rape seed would probably not become dormant while lying on the soil surface. This experiment also showed that distinct differences existed between cultivars.

## 4.4. Effect of water potential on the onset of secondary dormancy in four cultivars of rape

A critical issue relating to the development of secondary dormancy in rape seeds, is the dryness of the soil. How dry does it have to be to cause the seeds to become dormant? In simple terms the requirements seem to be that the soil should not be so wet that the seeds will immediately germinate but wet enough so that the seeds can become physiologically active. In totally dry soil the seeds will not respond to environmental conditions because the moisture level of the seeds is too low to permit biochemical processes to occur. This would be the same as the seeds being kept in a bag. There is no evidence that dry stored seeds become induced into dormancy if kept in the dark.

### 4.4.1 Materials and methods

Seeds of four cultivars of rape (see Table 4.1) were incubated in darkness for four weeks at 20°C in PEG at concentrations that created a range of water potentials (-2, -5, -10, -15, -20 bar). They were then germinated in water at 20°C in the dark for a further two weeks and the number of dormant (non-germinating) seeds recorded.

### 4.4.2 Results and discussion

**Table 4.1**. Effect of water potential on the induction of dormancy in four rape cultivars. % dormant seeds (logit transformed) with corresponding original values for % dormant seeds, in parentheses

water potential (bars)	Apex		Bristol		Falcon		Starlight	
- 2	-1.734	(3.0)	-1.136	(9.0)	- 2.652	(0)	- 1.544	(4.3)
- 5	-0.934	(13.3)	-0.506	(26.5)	- 2.652	(0)	- 0.701	(19.5)
- 10	-0.265	(37.0)	0.132	(56.5)	- 1.433	(5.8)	- 0.326	(34.3)
- 15	0.533	(74.5)	0.718	(81.0)	- 1.840	(3.8)	0.429	(70.0)
- 20	-0.030	(48.5)	0.777	(81.8)	- 2.448	(0.5)	0.875	(85.3)

During incubation at a high water potential (- 2 bars), few seeds remained (Table 4.1). In contrast, when seeds were subjected to conditions of strong water stress (- 20 bars), the majority of seeds failed to germinate and thus a large proportion of the seeds could become dormant. It should be noted that the low water potential required to generate appreciable levels of secondary dormancy represents very dry soil. Even slightly moist soil has a relatively high water potential. However, the percentage of seeds becoming dormant depended not only on the water potential tested but also on the genotype. Seeds of the cultivars Bristol and Starlight, for example, exhibited a higher level of dormancy than seeds of Falcon, as had occurred in Experiment 4.3 (above).

# 4.5 Effect of duration of darkness during incubation under water stress on the onset of secondary dormancy in two cultivars of rape.

The aim of these two experiments was to establish whether the length of the period in the soil in darkness was important in relation to the onset of dormancy and whether seeds left on the soil surface after shedding lost their ability to become dormant.

#### 4.5.1 Petri-dish test

### 4.5.1.1 Materials and methods

Seeds of cultivars Apex and Bristol were incubated in PEG (-15 bar) at 20°C for four weeks, but during that time they were kept in darkness or light for varying periods of time: ie 0 weeks light / 4 weeks in dark, 1 week light / 3 weeks dark etc (Table 4.2). The seeds were then germinated in the dark for 2 weeks at 20°C and the % dormant seeds recorded.

#### 4.5.1.2 Results and discussion

The results showed (Table 4.2) that the longer the period of darkness and the shorter the period of light, the greater the % of dormant seeds. Two weeks exposure to water stress and darkness seemed to be inadequate, a longer period was needed. Both cultivars showed similar responses. If the onset of secondary dormancy in oilseed rape is associated with changes in phytochrome system, which controls many other light mediated responses in plants, there should have been an increase in the % dormant seeds as the period in the dark increased. The results showed this trend. Further evidence for the involvement of phytochrome in the development of dormancy is presented in Section 5 of this report.

**Table 4.2** Effect of time in light and darkness during a 4 weeks period of water stress on the development of dormancy. % dormancy in darkness as logit transformed means with corresponding % dormancy of original values, in parentheses

weeks in light	0	1	2	3.	4
weeks in darkness	4	3	2	1	0
Apex	- 0.259	- 0.695	- 1.435	- 1.691	- 1.919
•	(37.2)	(24.8)	(7.6)	(3.2)	(1.6)
Bristol	0.006	- 0.227	- 1.092	- 1.777	- 1.866
	(50.0)	(39.2)	(14.0)	(2.8)	(2.0)
s.e.m. 0.218					

### 4.5.2 Field test

### 4.5.2.1 Materials and methods

This experiment was done on Rothamsted's Woburn farm (sandy soil). Gauze bags containing 1200 rape seeds (cv Bristol) were buried 20cm deep on 16 July 96 or were left on the soil

surface for 1, 2, 3 or 4 weeks, before being buried. During this 4 week period the buried seeds were kept in contact with dry soil, by placing a 5 cm layer of air dry soil below and above the seeds, when they were buried and additionally covering the plots with transparent plastic sheeting during the 4 week treatment period. The gauze bags were excavated on 9 October and the number of surviving seeds assessed.

### 4.5.2.2 Results and discussion

Approximately 20% of the seeds buried at the beginning of the experiment were still present in October. The % survival had declined to 10% after being left on the soil surface for one week and then to nearly zero when the period on the soil surface was 2, 3 or 4 weeks (Table 4.3). This effect of pre-treatment agrees to some extent with the Petri-dish test as both experiments showed minimal persistence if the seeds were left on the soil surface for more than one week. This suggests that incorporation of seeds into the soil should be delayed for perhaps two weeks after shedding to minimise the risks of seeds becoming dormant. This effect of the timing of soil cultivation has been explored in other field experiments (see Section 5, below). Other experiments on the long-term persistence of seeds also identified that seeds left on the soil surface for 4 weeks prior to burial failed to develop secondary dormancy (see Section 6, below).

**Table 4.3** Effect of 0 - 4 weeks pre-treatment of rape seeds on the soil surface prior to c. 3 months burial on the % seed survival (s.e.m. = standard error of mean)

	Weeks on the soil surface								
	0	1	2	3	4				
Mean	20.5	10.1	0	0.3	0.4				
s.e.m.	6.01	4.87	na	0.23	0.16				

## 4.6 Effect of the temperature regime during the germination of seeds induced into secondary dormancy.

Seeds induced into dormancy by incorporation into dry soil, would experience considerable diurnal temperature fluctuations if they were near the soil surface, but these variations would be greatly reduced if the seeds were buried at depth, for example by ploughing. It is well known that weed seeds respond to temperature fluctuations, as alternating temperatures are frequently used to break the dormancy of dormant seeds. This experiment was done to test whether temperature fluctuations could affect the germinability of dormant rape seeds.

### 4.6.1 Materials and methods

Seeds of the same six cultivars studied in Section 4.3 were exposed to a water potential of -15 bar for 4 weeks at 20°C in the dark. Their germination was then tested in darkness under three contrasting temperature regimes: 15°C constant, alternating 12h periods of 18/12°C and 20/10°C. The number of surviving rape seeds was recorded after two weeks.

### 4.6.2 Results and discussion

The greater the temperature fluctuation in the germination test the lower the % of surviving seeds (Table 4.4). Most seeds survived in the absence of diurnal fluctuations in temperature. Thus, seeds buried deeply by ploughing would be least likely to germinate, whilst those near to the soil surface would be more likely to germinate. As in the previous tests Libravo and Falcon had the fewest dormant seeds.

**Table 4.4**. Effect of three contrasting temperature regimes during a two week germination test on the level of dormancy exhibited by six rape cultivars. % dormancy in darkness as logit transformed means with corresponding % dormancy of original values, in parentheses

Temperature	Apex	Bristol	Envol	Falcon	Libravo	Starlight
15°C	0.533	0.686	0.100	- 1.178	- 0.624	0.757
	(74.8)	(79.6)	(54.8)	(8.0)	(22.4)	(82.4)
18/12°C	0.157	- 0.091	- 0.140	-1.860	- 0.780	0.357
	(57.2)	(50.8)	(43.2)	(1.6)	(16.8)	(32.8)
20/10°C	- 0.390	- 0.254	- 0.628	- 2.308	- 1.270	- 0.793
	(32.4)	(38.0)	(23.2)	(0)	(7.2)	(16.8)
s.e.m. 0.129		. •				

## 4.7 Studies of the variation in the potential of rape cultivars to develop secondary dormancy.

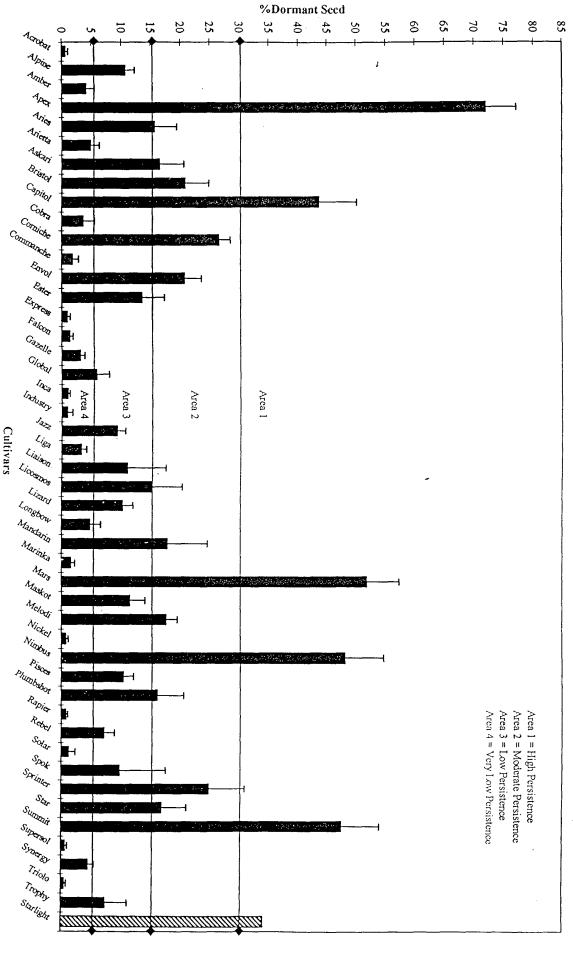
It is clear from the experiments done in Germany (Pekrun et al., 1997b) and in the early work on this project that rape cultivars seem to differ in their potential to develop secondary dormancy. The tests in Section 4.3 and 4.6 clearly showed that the level of dormancy induced by exposure to water stress and darkness in the Petri-dish tests was much higher in Apex, Bristol, Starlight and Envol than it was in Libravo and Falcon. It was not clear how widespread this variation in cultivar response actually was. A series of tests was done with 47 different cultivars included in the NIAB variety trials.

### 4.7.1 Materials and methods

A series of experiments examined the development of secondary dormancy in 26 spring and 21 winter rape cultivars (seeds harvested in 1995). The experiments were repeated four times between summer 1996 and autumn 1997. The seeds were stored in a constant temperature seed store (c. 18°C) prior to testing. As it was not possible to test all 47 cultivars at once, each experiment was split into four to six sub-tests, each including Starlight as a standard. Seeds were soaked in PEG in the dark for 4 weeks, as in the previous experiments, to induce dormancy. The number of dormant seeds present at the end of the two week germination test in darkness, was recorded. The % dormant seeds was analysed using techniques based on the Generalised Linear Model (GLIM), which enabled us to standardise the numbers of dormant seeds in relation to the dormancy level exhibited by the cultivar Starlight in each sub-test. Fuller details are given in the paper by Pekrun, Potter & Lutman (1997)

### 4.7.2 Results and discussion

There were clear differences between cultivars, some showing consistently high potential to become dormant and some showing virtually no potential (Fig. 4.2). More cultivars showed low levels of dormancy than high levels. There was variation between tests, particularly in the overall level of dormancy, but also in the relative positions of a few cultivars. However, although this variation did not affect the general conclusion from the work it would be wrong to use the percentage values as precise estimates of the potential level of dormancy. We do believe that the experiments indicate those cultivars that are more likely to persist and those that are least likely to do so. In Table 4.5 the 47 cultivars are categorised into four groups, high, moderate, low and very low persistence potential. The high persistence cultivars to be avoided if volunteer rape is a potential problem on the farm are: Apex, Starlight, Capitol, Nimbus, Mars, Summit. It is ironic that Apex, the most widely grown cultivar in recent years, was the most dormant in most tests.



rape. Data are the means of four experiments standardised on the basis of the response of the standard cultivar (Starlight) (Vertical bars = s.e.m.) Fig 4.2 Results of Petri-dish tests to establish the percentage of dormant seeds, resulting from exposure to water stress and darkness, in 47 cultivars of

The results of the four tests show a decline in dormancy with age. The last two tests in 1997 showed much less persistence than those in 1996. Using the data from Starlight as an example the mean percentage dormant seeds in the four tests was 53, 61, 14 and 8%. This result has some field relevance in that all the seeds would be very young when shed from the crop at harvest and so would exhibit maximal potential to develop secondary dormancy. However, the tests were only done on one sample of seeds for each cultivar and it is possible that different populations of the same cultivar would not respond in the same way, perhaps because of different environmental conditions experienced during maturation. This aspect of the variability in the potential to develop secondary dormancy is considered in Section 4.8.

Finally, we do not yet know whether the conclusions of the Petri-dish tests will be reflected in differences in persistence in the field. Some preliminary work by Pekrun with the six cultivars studied in Section 4.3, indicated that the same relative behaviour was generated in a field test, but it has not been possible to pursue this aspect of the work in more detail.

**Table 4.5** Categorisation of the levels of secondary dormancy generated in Petri-dish tests in 47 cultivars of oilseed rape

High persistence (>30%)*	Moderate persistence (15-30%)	Low persistence (5-15%)	Very low persistence (<5%)
Apex	Aries	Alpine	Acrobat
Capitol	Askari	Arietta	Amber
Mars	Bristol	Ester	Cobra
Nimbus	Corniche	Global	Commanche
Starlight	Envol	Jazz	Express
Summit	Mandarin	Liaison	Falcon
	Melodi	Licosmos	Gazelle
	Plumbshot	Lizard	Inca
	Sprinter	Longbow	Industry
	Star	Maskot	Liga
		Pisces	Marinka
		Rebel	Nickel
		Spok	Rapier
		Trophy	Solar
			Superol
		•	Synergy
			Triolo

<sup>\*</sup> These are the % dormant seeds from the combined analysis of the four tests (see Fig. 4.2)

### 4.8 Comparison of the development of dormancy in several populations of five rape cultivars

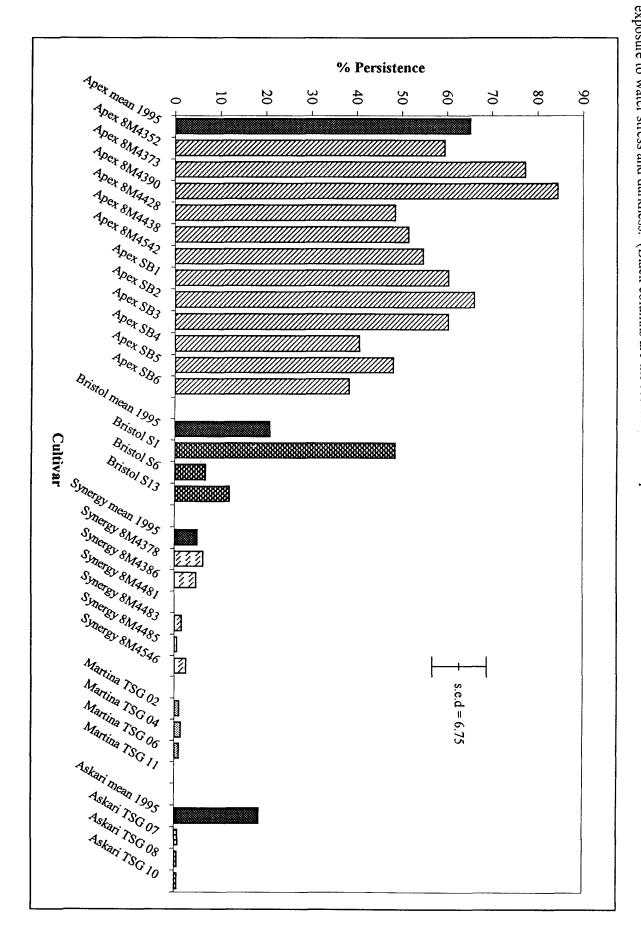
The aim of the initial Petri-dish tests with the 47 cultivars, described above, was to provide some information about the potential to develop secondary dormancy within British oilseed rape cultivars. This work was done with one sample of seeds from each cultivar and so the results obtained could have been due to the genetic background of the cultivar and/or to the environmental conditions that occurred as the seeds were maturing. It is known that the level of dormancy exhibited by weed seeds is influenced by the environmental conditions experienced by the parent plant. For example wild oats (*Avena fatua*) seeds are more dormant when produced in cool summers than in hot ones (Peters, 1982). Consequently, several samples of seeds of oilseed rape cultivars were collected in autumn 97. These were tested for their potential to develop secondary dormancy, as in the previous tests.

### 4.8.1 Materials and methods

A Petri-dish experiment was set up in March 1998. The experiment was conducted using the same Petri-dish method as in the previous tests. The experiment included 5 cultivars and a total of 28 populations as follows: Apex 12, Bristol 3, Askari 3, Martina 4 and Synergy 6. Following incubation in the dark under PEG induced water stress, the percentage of dormant seeds was recorded.

### 4.8.2 Results and discussion

The % dormant seed of Apex was high in all populations, varying from 40% - 85% (Fig 4.3). The overall mean was similar to that recorded in the earlier experiments with the seed collected in 1995 (using means unadjusted for Starlight - see section 4.7.1). The three Bristol populations were rather variable with one relatively dormant population and two with a much lower dormancy. Because of the variability in the responses of the three populations tested it is not possible be sure that they reflected the likely responses of this cultivar. All populations of Synergy, Martina and Askari demonstrated little dormancy. The 1997 populations of Askari were less dormant than the one tested in the earlier work. Synergy responded similarly in both tests. Martina, which seemed to be a low persistence cultivar, had not been tested in the earlier work. The best comparisons of the relative importance of the genetic and environmental components of secondary dormancy are between the 12 Apex and 6 Synergy populations. The sample number for Askari, Bristol and Martina was inadequate for a clear indication of the variation in dormancy between populations. It is clear that with Apex that there is variation between populations but the over-riding effect is due to the cultivar, as all populations showed relatively high levels of dormancy. The same conclusion can be drawn from the Synergy data. Thus, both environment and genotype play a role in determining the persistence of rape seeds, but the genetic constituent is the more important.



exposure to water stress and darkness. (Black columns are the results from the previous tests with seeds harvested in 1995, see Fig 4.2) Fig 4.3 Results of Petri-dish tests to establish the percentage of persisting seeds in a range of populations of five cultivars of oilseed rape, resulting from

### 4.9 Importance of phytochrome in the dormancy responses in oilseed rape

NB This work was done as an associated piece of research by a visiting post-doctoral researcher from Spain (Dr Francisca Lopez-Granados)

It is common that the mechanisms controlling the development of dormancy and subsequent germination of weed seeds in response to light is mediated by phytochrome, a pigment that exists in cells in two forms that respond to different wavelengths of light (red and far-red) (Bewley & Black, 1994). The presence of the far-red absorbing form of phytochrome (Pfr) is required to induce light sensitivity in seeds of many species. Prolonged exposure to far-red light (FR), as can occur under dense crop canopies, induces a secondary light requirement in some weeds due, it is believed, to the reversion of Pfr to Pr (red absorbing form of phytochrome). As Pfr also reverts to Pr in darkness non-dormant, but buried, rape seeds could acquire dormancy because of this phytochrome reaction. Furthermore, the proportion of Pr to Pr + Pfr influences seed germinability. Information is available on the response to light quality of seeds of many species of crops and weeds, including members of the Cruciferae (eg. Thlaspi arvense L. (Hazebroek & Metzger, 1990)). Despite the absence of primary dormancy, seeds of oilseed rape also appears to respond to light.

Experiments were done to see whether non-dormant rape seed could be induced into dormancy by exposure to far-red light as well as to darkness.

#### 4.9.1 Materials and methods

Seeds of rape cultivars Libravo and Falcon were used in these experiments which were all done in Petri-dishes, using techniques similar to those described in the previous sections. Full details of the methodology are given in Lopez-Granados & Lutman (1998). The induction of secondary dormancy was studied by imbibing seeds at 6, 12 and 20°C, in the presence and absence of water stress (-15 bar using PEG), for various periods of time from 5 mins to 28 days, under white light, far red light or darkness. After imbibition the seeds were tested in the dark for their ability to germinate at 12°C.

#### 4.9.2 Results and discussion

The treatments at 12°C, in darkness or white light, in absence of water stress, failed to induce secondary dormancy for all incubation times. If incubation was for less than 5 days, even under water stress and in darkness or under far-red light, the seeds also failed to become dormant. Rape seeds do not respond to far-red light very quickly, unlike some other seeds where exposure for less than 60secs can induce dormancy (Bewley & Black, 1994). Imbibition under water stress for 28 days in darkness and under far red light resulted in an appreciable number of dormant seeds (Table 4.6). When incubation time was only 14 or 5 days, fewer seeds became dormant. Libravo tended to produce more dormant seeds than Falcon, as had occurred in the experiments in Section 4.3. There was some evidence that far-red light produced more dormant seeds than darkness. This was particularly evident in the absence of water stress, when 5 and 14 days incubation under far red light resulted in some dormant seeds. When incubation was for a longer period, without water stress, the seeds tended to germinate during the incubation period.

Table 4.6 Effects of incubation time and temperature on the development of dormancy in seeds of cultivars Libravo and Falcon placed in darkness or under far-red (FR) light. Values = % dormant seeds (statistical analysis of data given in Lopez-Granados & Lutman, 1998)

Days Cultivar incubation			Incubation at 12°C				Incubation at 6°C			
		No water stress		Water stress (-15 bar)		No water stress		Water stress (-15 bar)		
		FR	Dark	FR	Dark	FR	Dark	FR	Dark	
5	Libravo	11.9	0+	4.7	2.2	19.4	0+	14.7	7.2	
	Falcon	1.9	0+	1.1	0.3	27.2	0+	5.6	0.3	
14	Libravo	12.8	0+	10.8	6.9					
	Falcon	15.3	0+	3.3	0.8	,				
28	Libravo	0*	0+	26.7	20.3					
	Falcon	0*	0+	13.1	5.0					

<sup>\*</sup> All seeds germinated during incubation, prior to germination test

When the temperatures were kept low, previously ineffective short periods of incubation were enough to induce dormancy, even in the absence of water stress, provided the seeds were under far red light.

There appears to be a balance between the mechanisms involved in seed germination and those concerned with the development of dormancy. If germination is inhibited by water stress or by low temperatures this provides time for the seeds to become dormant. If temperatures are high, water stress is needed to reduce germination growth and to give time for the phytochrome mechanism to induce dormancy. The evidence suggests that the positive shift in phytochrome's status caused by far-red light, induced more dormant seeds than the slower reversion in phytochrome induced by darkness. Thus, the evidence from this work indicates that phytochrome is involved in the onset of secondary dormancy. Some more work to confirm this, in the light of more recent studies covering other aspects of seed biology in rape, is required to confirm this conclusion.

<sup>+</sup> All seeds germinated during germination test

# 5. Field studies on the factors influencing the persistence of rape seeds

### 5.1 Introduction

The conclusions from the early work in Petri-dishes and from research done previously in Germany (Schlink, 1994; Pekrun, 1994), indicated that the most likely reason for the development of dormancy in rape seeds was the exposure of seeds to a combination of darkness and water stress. Rape seeds left in the field after rape harvest would be exposed to contrasting environmental conditions, depending on the weather conditions and the post-harvest management, particularly cultivations, employed by the farmer. Consequently, field experiments were set up, initially in autumn 1995, to examine whether it was possible to confirm this theory in the field, by comparing different post-harvest cultivation systems.

### 5.2 Influence of Agronomic Factors on Rape Seed Persistence - 1995 Experiments

The aim of these experiments was to simulate seeds shed at rape harvest by creating known populations of rape seeds on a stubble, and then to explore the effects of different cultivations on the survival of the seeds. It was thought that this type of approach would be better than using natural, but variable, populations of shed rape seeds.

### 5.2.1 Materials and methods

Early in August 1995 two experiments were started, one at Rothamsted (flinty silty clay loam) and the other at Woburn (loamy sand), to study the effects of post-harvest cultivation on the survival of rape seeds in the soil. Five cultivation treatments and three cultivars were tested in a split-plot design with 4 replicates, with the cultivation treatments being allocated to the main plots and the cultivars to the  $3 \times 6 \text{ m}^2$  sub-plots. Both fields were fallowed in the previous year. To ensure dry soil conditions, the soil was cultivated with a power harrow several times before the start of the experiment. Seeds, derived from crops harvested at the end of July, were broadcast by hand on each plot to simulate seed losses of 500 kg/ha from the previous rape crop, which resulted in approximately 10,000 seeds/m² (cultivars Apex, Bristol and Envol = 9484, 12109 and 10915 seeds/m²).

The cultivation treatments were as follows:

- immediate ploughing (20 cm) after seed distribution: "Plough"
- 3 times shallow stubble tillage with a spring tine (5 cm) at weekly intervals followed by ploughing (20 cm) in week 4: "3 St. + pl."
- 4 weeks delay and then spring tine (5 cm) plus plough (20 cm): "delay pl."
- 4 weeks delay and then non-inversion tillage with flexi tine plus discs (10 cm): "delay NI"
- no tillage: "Zero-till."

To avoid germination of the seeds directly after broadcasting as a result of rainfall, the plots were covered with plastic sheets during the first 4 weeks of the experiment (at Rothamsted only two of the four replicates). On 18 September (Rothamsted) and 20 September (Woburn) emergence was assessed in 10 quadrats/plot (size depending on plant density: 1/40 - 1/4 m<sup>2</sup>). The following spring (mid. Feb.- mid. April 1996) the soil seedbank was assessed by taking 20 soil cores (2.5 cm-diameter, 30 cm deep) from every sub-plot. The samples were washed through 2 mm-sieves and seeds were extracted by floating them in a saturated salt solution.

Samples of seeds were checked for viability in a germination test using gibberellic acid or stratification to break their dormancy. For statistical analysis of the percentage seeds found in the field a log transformation was used ( $\log_n (\% + 0.5)$ ). Percentages were calculated using the number of seeds broadcast for each variety. Seedling counts are shown on an area basis.

In spring 96 the two sites were re-cultivated (harrowed and ploughed) in May to simulate preparation for a late spring crop. Rape seedlings were counted, at Rothamsted on 21 June 1996 and at Woburn on 2 July. In July 1996 the seedbank of the Woburn-experiment was assessed a second time but this time taking only 10 cores per plot. During the following winter the sites were ploughed in preparation for sowing crops in spring 97. Further soil cores (10 /plot) were taken from the Woburn site (Bristol plots only) in November 96.

Spring wheat was sown at Rothamsted and spring beans at Woburn in spring 97. In June rape seedlings were counted in ten 1m<sup>2</sup> quadrats on all plots on both experiments. During the autumn both sites were ploughed in preparation for sowing linseed at Woburn and maize at Rothamsted, in spring 98. In November 97 soil cores (20/plot) were again taken from the Woburn site and seed numbers recorded. Seedling numbers were assessed on both sites in May/June 98, whole plot counts at Woburn and 10 x 1m<sup>2</sup> quadrats at Rothamsted

### 5.2.2 Results and discussion

### 5.2.2.1 Autumn 95 - Spring 96

The soil was very dry prior to the start of the experiments and virtually no rain fell during the first 4 weeks of the experiment, so covering the plots with plastic sheets appeared to have been unnecessary (see Pekrun, Hewitt & Lutman, 1998 for details). Greater numbers of seedlings emerged in September at Rothamsted than at Woburn (Table 5.1). Maximum emergence occurred at Rothamsted on the non-cultivated plots ("Zero-till.") in which ca. 60 % of the broadcast seeds produced seedlings. At Woburn emergence was greatest on plots that had been disced and spring tined 4 weeks after the start of the experiment ("delay NI" = delayed non-inversion tillage). Few seedlings emerged at both sites on the plots that received the other three treatments which included ploughing.

Persistence assessed in the following winter/spring (Table 5.2) revealed almost a mirror image of the preceding data. More seeds were found on the sandy soil at Woburn than on the heavier soil at Rothamsted. Persistence at both sites was greatest in plots that had been ploughed immediately after the seeds had been broadcast ("Plough"). It was somewhat lower in plots where a shallow stubble tillage was done several times before ploughing ("3 St. + pl."). Persistence was very low in plots, in which post-harvest cultivation had been delayed for 4 weeks prior to ploughing ("delay pl.") and was even lower following delayed non inversion tillage ("delay NI"). In plots where no tillage was done at all, almost no seeds were found in spring. No consistent differences were found between the three cultivars. The only marked difference was the extremely high value for Envol in the ploughed plots at Woburn. Almost all retrieved seeds were found to be viable.

**Table 5.1** Effect of the five cultivation treatments on the number of rape seedlings/m<sup>2</sup> counted on 18/09/95 at Rothamsted and on 20/09/95 at Woburn. (NB c. 10,000 seeds/m<sup>2</sup> present at the start of the experiment)

		(a) Roth	namsted		
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.
Apex	234	283	310	2265	5359
Bristol	354	280	433	3776	6289
Envol	411	375	274	3271	6249
SE of means	327.2 (compariso	ons within the sam	e level of cultiv	ation 243.8) (d.f	: 30)
		(b) W	oburn		
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.
Apex	3	37	31	2680	1380
Bristol	1	24	14	2959	2410
Envol	4	21	18	3300	1648
SE of means	188.6 (compariso	ons within the sam	e level of cultiv	ation 168.6) (d.f	: 30)

**Table 5.2** Effect of the five cultivation treatments on the seed numbers in the soil expressed as a % of seeds sown ( $log_n$ (% seeds + 0.5)), assessed February - April 1996 at Rothamsted and Woburn. Means of original values, in parentheses

	(a) Rothamsted								
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.				
Apex	2.70	2.66	1.57	0.14	- 0.69				
	(18.3)	(18.0)	(4.6)	(1.1)	(0)				
Bristol	3.05	2.48	1.54	0.90	-0.45				
	(21.2)	(14.3)	(4.8)	(2.1)	(0.2)				
Envol	2.47	2.29	0.51	-0.43	-0.69				
	(12.4)	(10.0)	(1.6)	(0.2)	(0)				
SE of me	ans 0.299 (com	parisons within th	ne same level of	f cultivation 0.283	3) (d.f. 30)				
		(b) W	oburn o						
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.				
Apex	3.74	3.29	1.58	0.30	-0.69				
	(55.0)	(36.8)	(5.4)	(1.1)	(0)				
Bristol	3.80	3.38	2.06	-0.32	-0.69				
	(62.4)	(33.6)	(7.4)	<b>(0.4)</b>	(0)				
Envol	4.20	3.36	1.55	-0.43	-0.17				
	(90.3)	(29.6)	(4.7)	(0.2)	(0.5)				
SE of me	ans 0.338 (com	parisons within th	ne same level of	f cultivation 0.28′	7) (d.f. 30)				

### 5.2.2.2 Summer 96

Seedlings/m² were counted on 21 June (Rothamsted) and 2 July (Woburn). Few seedlings were present at both sites, except on the plots that had been immediately ploughed at Woburn, where there were up to 35 plants/m². (Table 5.3). The numbers present did, however, reflect the seedbank, as no or virtually no seedlings were found on the plots with delayed non-inversion tillage or no cultivation. There were no clear differences between cultivars, although more seedlings of Apex appeared to be present on the 'ploughed' plots. However, in relation to the seedbank assessed in spring the number of seedlings was very low: < 1 % of the seedbank.

In July 1996 soil sampling was repeated at Woburn (Table 5.4). This showed that the number of persisting seeds had declined appreciably compared to the spring assessment, particularly on the Bristol and Envol plots. The fate of the seeds that did not persist and did not emerge is unknown. They may have either died, or germinated and then died due to failed emergence, predation or fungal or microbial attack. When testing the seeds' viability, seeds always showed 100 %, or nearly 100 % viability, which would suggest that the majority of seeds did not die as seeds but after germination. Most seeds were present on the 'immediately ploughed' plots

**Table 5:3.** Effect of five cultivation treatments on the number of rape seedlings/ $m^2$  on 21/06/96 at Rothamsted and on 2/07/96 at Woburn. Mean of 4 replicates

(a) Rothamsted						
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.	
Apex	0.53	0.28	0.32	0.03	0	
Bristol	0.47	0.21	0.19	0.04	0	
Envol	0.35	0.39	0.12	0.04	0	

(b) Woburn							
	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.		
Apex	34.7	4.5	0.7	0	0.01		
Bristol	24.6	3.6	0.8	0	0		
Envol	26.1	3.6	0.8	0	0		
SE of means	3.68 (comparison	ns within same lev	vel of cultivation	n 3.21) (d.f. 30)			

**Table 5.4** Seedbank, as % sown ( $log_n$ (% seeds + 0.5)), assessed at Woburn in July 1996.. Means of original values, in parentheses

	Plough	3 St. + pl.	delay pl.	delay NI	Zero-till.
Apex	3.61	2.48	0.14	-0.27	-0.69
	(48.9)	(21.5)	(1.1)	(0.5)	(0)
Bristol	2.67	2.31	0.91	0.19	0.33
	(22.3)	(14.7)	(3.4)	(1.3)	(1.7)
Envol	3.10	2.75	0.62	-0.16	0.10
	(23.8)	(21.0)	(1.9)	(0.9)	(2.8)

SE of means 0.548 (comparisons within the same level of cultivation 0.478) (d.f. 30)

**Table 5.5** Seedbank, as % sown ( $log_n$ (% seeds + 0.5)), assessed at Woburn in November 1996, for Bristol only. Means of original values, in parentheses

	Plough	3 St. + pl.	delay pl.	delay NI
Bristol	2.92	1.81	0.19	-0.69
	(20.6)	(7.6)	(1.3)	(0)
	:	SE of means 0.390 (	d.f. 9)	

Table 5.6 Rape seedlings/m<sup>2</sup> counted on 8/06/97 at Rothamsted and on 13/06/97 at Woburn.

(a) Rothamsted							
	Plough	3 St. + pl	Delay pl.	Delay NI	Zero-till		
Apex	1.2	0.9	0.3	0.0	0.0		
Bristol	1.4	0.8	0.5	0.0	0.0		
Envol	1.2	0.9	0.3	0.0	0.0		
SE of means (	0.30 (comparisons	s within the same	level of cultiva	tion 0.16) (d.f.	30)		
· · · · · · · · · · · · · · · · · · ·		(b) Wo	burn		<del></del>		
	Plough	3 St. + pl	Delay pl.	Delay NI	Zero-till		
Apex	11.0	4.7	1.6	0.6	2.4		
Bristol	37.9	11.1	1.8	0.7	3.1		
Envol	16.4	7.7	1.8	0.8	0.4		
SE of means 3	3.09 (comparisons	s within the same	level of cultiva	tion 2.35) (d.f.	30)		

### 5.2.2.5 Autumn 97

In November 97 soil cores were again taken from this experiment. Seed numbers/m² had declined slightly between the two assessments. Very few seeds were found on the plots where cultivation had been delayed, when the project was started in August 95 (Table 5.7). Similarly there were few seeds on the direct drilled plots. Slightly more than 10% of the seeds sown in August 95 were still present on the 'ploughed' plots and 5-8% of the tined cultivated and ploughed plots. Differences between cultivars were small.

**Table 5.7** Seedbank, as % sown ( $log_n$ % seeds + 0.5)), assessed at Woburn in November 1997. Means of original values, in parentheses

Woburn						
	Plough	3 St. + pl	Delay pl.	Delay NI	Zero-til	
Apex	2.39	1.08	-0.69	-0.69	0.08	
Арех	(11.2)	(5.4)	(0)	(0)	(1.0)	
Bristol	2.47	1.91	0.54	-0.14	0.12	
Distoi	(12.8)	(7.7)	(1.8)	(0.5)	(1.0)	
Envol	2.5	1.95	-0.14	-0.29	-0.69	
Liivoi	(12.8)	(7.7)	(0.5)	(0.5)	(0)	

#### 5.2.2.8 Summer 1998

Very, very few rape seedlings were found in the linseed crop sown in 1998 at Woburn when assessed at the end of May. Even on the immediately ploughed plots where the seedbank was still in excess of 1000 seeds/m<sup>2</sup> (Table 5.7) there were fewer than 1 seedling/4m<sup>2</sup>. Only occasional plants were present on all the other plots. Further assessments in June and early July failed to identify any more seedlings.

Table 5.7 Rape seedlings/m<sup>2</sup> counted on 25/06/98 at Rothamsted

(a) Rothamsted						
	Plough	3 St. + pl	Delay pl.	Delay NI	Zero-till	
Apex	22.0	8.9	5.1	1.0	0.1	
Bristol	20.6	6.7	6.9	1.2	0.2	
Envol	11.7	9.8	6.4	0.8	0.1	
SE of means 3	3.19 (comparisons	s within the same	level of cultiva	tion 3.54)*		

<sup>\*</sup> the statistical analysis applies only to the 'Plough, 3St + pl and Delay pl.' treatments as the others were effectively zero.

Conversely, on the Rothamsted site where few seedlings had been recorded in 1996 and 1997 (Tables 5.3, 5.6), appreciable numbers emerged, mainly in the plots that had been immediately ploughed in August 1995. There were over 10 plants/m² on all the 'ploughed' plots (mean = 18.0), somewhat fewer on the repeatedly tined cultivated plots and on the delayed ploughed plots and virtually none on the other cultivation treatments. This correlates very well with the seed samples taken in winter 96 (Table 5.2) and the relationships of the three cultivars on the 'ploughed' plots are similar, with most seedlings and seeds on the Apex and Bristol plots and least on the Envol. Clearly, seeds still remain on these plots.

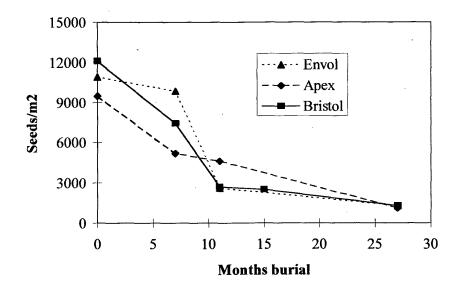
### 5.2.3 General discussion

One of these experiments (Woburn - loamy sand) had a considerable number of seeds which became dormant and persisted in the soil. These seeds have provided an ideal resource for the monitoring of long term seed decline. The site has been ploughed once each year and sown with spring crops in 1997 and 1998. Rape seedling numbers and soil seed bank levels have been measured each year. Of particular interest are the data on seed persistence. The last sample was taken in November 1997. The results show an exponential decline rate with a rapid early loss of seeds (Fig. 5.1a). Approximately 10% of those originally present were still present two years later. Transforming seed numbers to log<sub>10</sub> seed numbers/m<sup>2</sup> permitted the calculation of decline rates (Fig. 5.1b). This analysis suggests a 50% decline rate of about 9 months, indicating that it would take about 3 years to lose 90% and 9 years to lose all the seeds. Further soil cores will be taken in Autumn 1998, to provide more data on the decline rate. The number of seeds present in the soil at Rothamsted site were lower making definitive conclusions on long-term decline rates more difficult to draw. However, the appearance of appreciable numbers of rape seedlings on the 'ploughed' plots in 1998 would suggest that further soil coring to investigate the remaining seed bank on these plots would be worthwhile. In previous years the absence of seedlings at Rothamsted suggested that there were few seeds remaining.

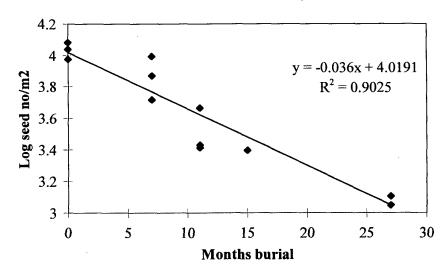
Comparing the seed bank levels at Woburn with the seedling numbers present in the relevant years would suggest that 1% or less of the seedbank emerges as seedlings. It is a very low proportion of the total. This figure equates to some data collected with 'normal' broadleaved weeds such as chickweed, red dead nettle and cleavers, where again seedlings only represented a very small percentage of the seedbank (Miller *et al.*, 1998).

Fig. 5.1 Decline in the numbers of rape seeds persisting in the soil from August 1995 to
November 1997 (Expt Woburn - Aug 1995)

a) seeds/m² for the three cultivars



b) decline rate for the mean of the three cultivars



### 5.3 Influence of Agronomic Factors on Rape Seed Persistence - 1996 Experiment

Following the 'success' of some of the cultivation treatments in the experiments started in 1995, in creating dormant rape seed populations in the soil, it was decided that the work at Woburn should be repeated in 1996, to establish whether the main conclusions were valid under the anticipated different weather conditions in 1996.

### 5.3.1 Materials and methods

The experiment was basically the same as those done in 1995. Seeds of three cultivars of rape were broadcast onto the soil surface on 14 August (10,000/m²) and were then cultivated (ploughed or non-inversion cultivation) at various dates after spreading. Seedling emergence was monitored on 28 August 96 and soil cores (20 cores, 2.5cm diameter/plot) were taken in Feb-March 97 to estimate the rape seed bank. The same soil sampling techniques were used as in the 1995 experiments. Rape seedlings were counted on the plots on 6 June 97.

The experiment was a split-plot design with four replicates, five cultivation treatments and three cultivars (Apex, Envol and Bristol). The cultivation treatments were :

- immediate ploughing "Plough"
- immediate non-inversion tillage "NIT"
- non-inversion cultivation delayed for 4wks "NI4"
- ploughing delayed for 4wks "P4"
- stubble cultivation once a week for three weeks, followed by ploughing in week 4
   "S3P"

### 5.3.2 Results and discussion

Soil conditions were much wetter in 1996. The week before the experiment started there was 26mm of rain. Consequently, the soil was wet when the seeds were broadcast. It remained dry from 14 - 21 August but then 14mm of rain fell on 22 August and the weather remained wet for the next week. As a result of all the rain many rape seeds germinated in August and seedling counts on 28 August were more than 2000/m² on virtually all plots, except those ploughed on 14 August (Table 5.8). As a consequence of the high germination few seeds persisted and so few were found in the soil cores extracted in Feb-March 97. There were less than 1% of the sown seeds on all treatments except the immediately ploughed plots where 2.6% were present (Table 5.9). In all cases this is a very low proportion of those sown. Similarly, few seedlings emerged in summer 97, a maximum mean of 2.2 seedlings/m² on the immediately ploughed plots and less than 1/m² on all the other cultivation treatments (Table 5.10). In the assessments, because of the low numbers of surviving seeds and the variability in the data, it was not possible to detect differences between cultivars.

This experiment confirmed that in wet conditions after the rape harvest most rape seeds will germinate and few will persist. It must be remembered, however, that even 1% of a conservative 'normal' level of seed shedding of 5000 seeds/m², will result in 50 seeds/m² remaining, enough to cause perhaps the emergence of 1 seedling/2m² which could cause a problem in some cropping systems.

**Table 5.8** Effect of cultivation and cultivar on the number of rape seedlings/m<sup>2</sup> counted on 28 Aug 1996. Mean of 4 replicates (s.e.m values in parentheses) \* for details of cultivation treatments see 5.3.1

	Plough*	NIT	NI4.	P4	S3P
Apex	2.3	2874	1899	2400	1885
	(0.75)	(390.2)	(84.5)	(116.1)	(184.1)
Bristol	1.9	3589	2239	2378	2572
	(0.64)	(760.9)	(119.2)	(77.5)	(410.4)
Envol	3.3	3689	2457	2506	1987
	$(0.34)^{-1}$	(363.7)	(375.7)	(254.5)	(132.5)
Mean of cultivars	2.5	3384	2198	2428	2148

**Table 5.9** Effect of cultivation and cultivar on the number of surviving seeds in February 1997, expressed, as % sown seeds. Mean of 4 replicates. \* for details of cultivation treatments see 5.3.1

	Plough*	NIT	NI4.	P4	S3P
Apex	0	0.18	0.18	0	0.17
Bristol	0.35	0	0.35	0.88	0
Envol	7.43	1.77	0.53	0.17	0.17
Mean of cultivar	2.59	0.65	0.35	0.35	0.11

**Table 5.10** Effect of cultivation and cultivar on the number of seedlings/m<sup>2</sup> counted in June 1997. Mean of 4 replicates. \* for details of cultivation treatments see 5.3.1

	Plough*	NIT	NI4.	P4	S3P
Apex	3.8	0.1	0.1	0.3	0.1
Bristol	0.1	0.2	0	1.6	0.4
Envol	2.6	0.6	0	0.5	0.2
Mean of cultivar	2.2	0.3	0	0.8	0.2

### 5.4 Influence of Agronomic Factors on Rape Seed Persistence - 1997 Experiments

The work on the cultural factors affecting the development of persistence soil seedbanks of oilseed rape seeds in 1995 and 1996 had been restricted to the two Rothamsted farms (Woburn and Rothamsted). It was felt that the work would benefit from a wider range of experiences, so a larger number of simpler experiments were planned on different soil types and in different parts of the country. It was hoped that this work would provide confirmation of the main factors that control the onset of dormancy and also provide further data on the length of time that stubbles should be left uncultivated to minimise the risk of persistence. The work in 1995 and 1996 had compared immediate cultivation with cultivations delayed for 4 weeks. The laboratory work and the small scale field test (Section 4.5) suggested that 2 weeks might be adequate. This period was more likely to be acceptable to farmers, who may be reluctant to leave rape stubbles uncultivated for 4 weeks, because of the perceived risks of disease and pest spread to adjacent newly planted rape crops. Consequently, the 1997 experiments were designed to compare immediate cultivation, with cultivation delayed for 2 or 4 weeks.

### 5.4.1 Materials and methods

Six experiments were set up in July/August 97 at the following sites and dates (Table 5.11). We are grateful to the management and staff at IACR Long-Ashton, Arable Research Centres (ARC) and ADAS-Boxworth for agreeing to prepare the plots and make the cultivation treatments. The two experiments at Rothamsted and Woburn were slightly more complex than those at the four other sites.

<b>Table 5.11</b> <i>I</i>	Details of	the six	experiments	started in	1997
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Site	Date Started	Soil Type	Cultivar
IACR-Rothamsted	6/08/97	Clay loam with flints	Apex
IACR-Woburn	4/08/97	Fine Sandy loam	Apex
ADAS-Boxworth	23/07/97	Hanslope clay	Apex
IACR-Long Ashton	31/07/97	Fine Sandy loam	Apex
ARC-Shuttleworth	28/08/97	Sandy loam	Alpine
ARC-Cirencester	12/08/97	Brashy calcareous fine loam	Apex

On four of the experiments 10,000 rape seeds/m<sup>2</sup> were broadcast by hand onto the plots. At IACR Long Ashton and ADAS-Boxworth the rape seeds were naturally occurring from the previous rape crop. After seed distribution the following cultivation treatments were done:

- 1. immediate non-inversion tillage "NI1"
- 2. immediate ploughing. "P0"
- 3. plough 2 wks later "P2"
- 4. plough 4 wks later "P4"
- 5. non-inversion tillage after 4 weeks "NI4"

Treatments 2, 3 and 4 were applied to all experiments. Treatments 1 and 5 applied only to Rothamsted and Woburn. Additionally, on these two experiments, plots at 2 and 4 weeks were split and half was left uncovered and the other covered with a plastic sheeting between sowing and cultivation. This was to prevent the seeds becoming wet and to keep birds off the plots. Each experiment had four replicates.

Twenty soil cores were taken from each plot on 28 October 97 (Rothamsted) and 11 November 97 (Woburn). Forty soil cores were taken from the other experiments on 12 December 97 (ADAS-Boxworth), 2 December 97 (Long Ashton), 8 January 98 (ARC-Shuttleworth) and on 15 January 98 (ARC-Cirencester). Most of the samples were frozen and then washed through a 2mm sieve. The seeds were extracted by wet sieving and flotation in a saturated salt solution, as in the previous experiments. Further assessments were done on the Woburn experiment on 28 May 98 and at Rothamsted on 1 July to record the number of rape seedlings.

#### 5.4.2 Results and discussion

Seed survival was not great at any site (Table 5.12). No seeds were found on any plots at Shuttleworth. This lack of persisting seeds can be mainly attributed to the rather wet weather that occurred during August 1997. Soils were wet/damp at the start of the work and the rape seeds were exposed to rain at irregular intervals once present on the soil surface. Thus, many seeds germinated in the wet soil, or on the surface prior to cultivation. However, there were Seed survival was relatively greater on the some exceptions to this general picture. immediately ploughed and tine cultivated plots at Woburn and on the plots ploughed after two weeks at Woburn and Rothamsted. At Woburn this only applied to the covered plots, as pigeon grazing removed most of the seeds on the uncovered plots, within two days of being broadcast. Pigeons were much less of a problem at Rothamsted and the data from the covered and uncovered plots were quite similar. The higher survival on these treatments appeared to be associated with the soil moisture levels prior to and at the time of cultivation. Rothamsted, although the soil was damp prior to broadcasting the seeds, little rain fell in the two weeks between the first and second cultivations and so the seeds ploughed in after two weeks were buried into drier soils than those buried at the start of the experiment. The higher levels of persistence of the immediately cultivated plots at Woburn can be attributed to the ability of this very light sandy soil to dry out. Although a lot of rain fell within the four days prior to the start of the experiment, this mostly fell on the first two of the four, giving the sandy soil two days to dry out. This may have been adequate. Most seeds were found on the immediately ploughed plots at Cirencester. Again this site had no rain for the four days prior to the seeds being broadcast, but rainfall was appreciable before the two week plough treatment resulting in the germination of most of the surviving seeds.

The assessments done on the Woburn and Rothamsted sites in summer 98 reflected the seed bank assessments and the type of cultivations. Few seedlings were present on the ploughed plots (> 3/m²), because either there were few surviving seeds or the seeds were buried too deep to emerge. Most seedlings (7.2 seedlings/m² at Woburn and 4.6/m² at Rothamsted) were found on the plots that had been tine cultivated. At Woburn there were clearly more seedlings on the plots tine cultivated immediately after the seeds were broadcast, than on the plots where tine cultivation was delayed, but this was less obvious at Rothamsted. These responses reflect the differences in the seed bank of the tined plots, the previous autumn.

**Table 5.12** Number of surviving rape seeds in the soil (seeds/m²) at 6 sites in autumn 1997 following cultivations immediately after seed shedding, two weeks and four weeks later.

Site	Cultivation Treatments					
	Immediate	Ploughing	Ploughing	Immediate	Non-inv. Cult.	
	plough	after 2 weeks	after 4 weeks	Non-inv. Cult.	After 4 weeks	
Rothamsted	217	854	140	102	217	
Woburn	612	892+	77+	650	178+	
LARS*	153	0	77			
Boxworth*	26	128	102			
Shuttleworth	0	0	0			
Cirencester	1173	51	26			

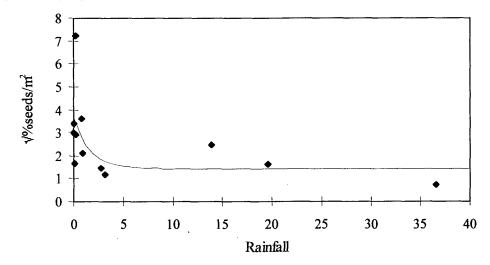
<sup>\*</sup> Starting seed population not known as seeds were shed from a rape crop; other sites all had 10,000 seeds/m² spread prior to cultivation

Further analysis of the data from the four of these six experiments with known starting populations, together with some from the earlier experiments at Woburn and Rothamsted has attempted to compare the numbers of surviving seeds to the amount of rainfall in the four days preceding the cultivation treatment. This is not ideal, as it does not measure the wetness of the soil, but it does give an indication of soil moisture, in the absence of soil samples to assess actual moisture levels. In the sites and treatments where persistence was highest (e.g. Woburn and Rothamsted two week ploughing, Cirencester immediate ploughing), rainfall was low. In contrast where rainfall was higher few seeds were found in the soil. Figure 5.2 shows the relationship between prior rainfall and seed persistence. It appears that if rainfall exceeds c. 4mm persistence will be minimal. This is not very much rain, so one must conclude that soils need to be very dry to maximise persistence. This conclusion agrees with earlier laboratory studies which showed that a low water potential (less than -10bar) was needed to generate dormant seeds.

The absence of persisting seeds at Shuttleworth may be due to the high rainfall prior to the start of the experiment (26 mm) but may also relate to the cultivar used. All the other sites in 1997 used Apex, which is known from the laboratory studies (Section 4.7) to be particularly persistent but the Shuttleworth site used Alpine, a low persistence cultivar. It may have been a combination of both these two factors that resulted in the total absence of surviving seeds.

<sup>+</sup> data from covered plots only

Fig. 5.2 Relationship between rainfall in the four days prior to cultivation and the % of persisting rape seeds



# 6. Persistence of buried rape seeds

#### 6.1 Introduction

The work described in Section 5 was primarily concerned with factors controlling the development of secondary dormancy and how to minimise the risks of it happening. However, under certain circumstances, such as dry soil conditions, it is clear that seeds can become dormant and persist. Having created a seedbank of rape seeds, we have made use of this opportunity to monitor the decline over several years (see Section 5.2). This work is particularly useful as the amount of published data on the persistence of rape seeds is very limited. It is clear that unless the initial soil conditions are appropriate, experiments designed to study persistence will fail because few seeds will become dormant. This is clearly what happened in the burial experiments at Rothamsted and Long Ashton reported by Miller et al. (1998) where stored rape seed buried into wet soil in October failed to persist. The same may well be true of the work on feral rape persistence reported by Hails et al (1997), where bags of rape seeds were buried in a range of habitats and few seeds survived at most sites. As details of the time of year and soil conditions at burial are not given in the paper it is impossible to be sure. Our own early attempts at establishing persistent rape seed populations in soils failed probably for similar reasons: the soils were too wet when the seeds were buried. The work of Schlink (1994, 1998) is an exception, as her burial experiment generated persistent seeds, 0.5% of those buried in 1987 were still present in 1997. However, this work has the drawback that it investigated the persistence of undisturbed seeds, rather than those that receive at least annual disturbance, as would occur from cultivations in arable fields. Despite this limitation several experiments have also been done at Rothamsted to investigate the persistence of undisturbed seeds, to provide information on what could be considered the maximum potential persistence.

Experiments in the field, started in 1990 and 1991, where seeds were buried in known positions and then excavated subsequently, failed to demonstrate high levels of persistence (Lutman, 1993; Lutman & Lopez-Granados 1998). This was probably for the reasons outlined in the previous paragraph, but there were indications that once the seeds had become dormant the % decline in the seedbank was relatively slow. For example, although only 3.8% of seeds buried in September 91 in Warren field were still present four months later, 4.0% were still there after 27 months (Lutman & Lopez-Granados, 1998). Similar, conclusions were reached with the pot experiment started in 1992, when seeds were buried in 25cm pots and then excavated after increasing periods of time. In this experiment 2.2% of seeds persisted for 6 months and 1.6% for 30 months (Lutman & Lopez-Granados 1998). Two further burial experiments were started in 1993 and 1994.

#### 6.2 Materials and methods

In late summer/autumn 1993 and 1994 rape seeds were buried c. 12cm deep in pots (25cm diameter) filled with a loam soil mixed with a small amount of grit. These pots were placed outside, buried up to within 2cm of the top, in a sand bed. Pots were emptied at intervals and surviving rape seeds were extracted from the soil by washing and sieving. Samples of the surviving seeds were tested for viability, as in the field experiments.

#### **6.2.1** 1993 Experiment

Seeds of cultivars Libravo and Falcon were collected from crops at the end of July. These were kept in a seed store (18-20°C) until the start of the experiment on 15 September. One thousand seeds of each cultivar were buried in 11 pots. These were excavated individually at frequent intervals from 6 - 14 months after burial.

#### **6.2.2** 1994 Experiment

As in the other experiments, seeds were collected from rape crops at harvest. Again cultivars Falcon and Libravo were used in this study. One thousand seeds were buried in each of the 12 pots for each cultivar immediately after harvest on 3 August. Two further samples of seeds were retained for pre-treatment. One sample was left outside in aluminium seed trays until 9 September. However, because of the rainfall during August almost 100% of the seeds germinated. The second sample was also kept outside but a sheet of glass was placed c. 20cm above the trays, protecting them from rain. Eight hundred of these seeds were buried in 9 pots for each cultivar. The pots were excavated after 6, 10, 15, 20 and 26 months, either two or three pots/cultivar on each occasion. As there were not enough pots of pre-treated seeds to be excavated after 26 months, the last sample was processed after 20 months.

#### 6.3 Results

## **6.3.1** 1993 Experiment

Quite a high proportion of the seeds of both cultivars survived on this experiment. After 6 months about 25% of the seeds were still present (Table 6.1). The % survival declined over the following 8 months so that by the end of the experiment only 3-4% remained. In most samples there were more seeds present in the pots containing Falcon than in those containing Libravo. There was considerable variation in the number of surviving seeds between individual pots, so these values must be considered to represent trends rather than absolute values Almost all surviving seeds were viable.

#### **6.3.2** 1994 Experiment

Even more of the seeds buried immediately after harvest survived in this experiment than in the previous one. After 6 months burial 45-66% of the seeds were still present (Table 6.1). More Libravo seeds survived than Falcon. The % survival declined down to 18% for Falcon over the next 20 months but there was little evidence of a decline for Libravo, as 68% of the seeds were still present at the end of the experiment. Where the seeds had been pre-treated outside under a glass plate, prior to burial, only one seed was found in all 18 pots. Surviving seeds were nearly all able to germinate, given appropriate conditions.

**Table 6.1** Percentage of buried rape seeds (cvs Libravo & Falcon) surviving in pot experiments started in 1993 and 1994

Length of	1993 Expt		1994 Expt	
burial (months)	Libravo	Falcon	Libravo	Falcon
6	25	28	66	45
7	9	19		
9	16	17		
10	7	13	47	34
14	4	3		
15			60	36
20			65	41
21				
26			68	18

## 6.4 Discussion of persistence

In 1994 the decline rate of the surviving seeds, particularly for Libravo was very slow. In the 1993 experiments and in Falcon in 1994 the decline was quicker. Overall, one would conclude from this work and from that done in 1990 - 92, that undisturbed seeds will certainly persist for two years. It is also highly likely that a proportion of the seeds will survive much longer, as for example with the Libravo seeds in the 1994 experiment. The number that persist seem to depend mainly on the number that initially become dormant, as in most of the experiments the differences in the % persistence between the first and last samples were much smaller than the decline between the start of the experiment and the first sample. It is difficult to identify the causes for the differences in persistence between these two experiments and those that were done earlier, but they are probably related to the moisture levels experienced by the seeds in the first few weeks after burial. Comparisons of the 1992, 93 and 94 data from pot experiments, with rainfall in the first four weeks after burial, showed that most persistence occurred in the driest year and least in the wettest year.

A further issue relating to persistence data, is the variability between pots / plots. The number of persisting seeds can vary greatly between replicates of the same treatment. The reasons for this are unclear, but as variability is also a feature of the Petri-dish test described in Section 4, and in the earlier work of Schlink (1994) and Pekrun (1994). It appears to be an intrinsic feature in the behaviour of seeds in dormancy studies with rape. It may related to small differences in the environment of the seeds that have a critical effect on the onset of dormancy. More work is needed to explore this issue.

The absence of surviving seeds in the treatments where seeds were after-ripened on the soil surface prior to burial, as in the 1994 experiment, is interesting. It supports earlier work by Schlink (1998), where after-ripening reduced but did not prevent the production of persistent seeds. However, the after-ripening conditions used by Schlink were different from those we used, and this may account for the differences in responses. She also noted that the

after-ripening response differed between cultivars, a conclusion that would concur with our work on the genetic variation in the development of secondary dormancy (Section 4.7). This work on the failure of after-ripened seeds to become dormant adds further weight to the conclusions from Section 5 that leaving seeds on the soil surface after shedding, is likely to minimise seed persistence in both wet and dry conditions.

It seems from the data of the pot and field experiments that once the seeds are in the soil and in a dormant state, they will remain for some years. The field experiment at Woburn started in 1995 should be continued to provide further data on the long-term decline rates, which are a crucial issue in the long-term management of this weed, and for which we have little data.

# 7. Modelling the population dynamics of volunteer rape seeds

#### 7.1 Introduction

As can be seen from the previous Sections there are few data available on long-term changes in seedbanks of oilseed rape. To our knowledge the only published work is that of Schlink (1998), the persistence experiments reported by Miller et al., 1998 and our own data presented in Sections 5 and 6. There is, however, more information available that provides data on each step of the life-cycle of volunteer rape, which could be used to estimate longer-term changes in populations. Consequently, construction of a model incorporating this information could give further insight into the problem. We have therefore combined all data available on volunteer rape into a simple deterministic model, based on the life cycle given in Fig 7.1. This work is still in progress and so this report only presents the structure of the model and some preliminary conclusions. The model concentrates on the agronomic factors that influence the population dynamics of the seeds, rather than on the factors affecting plant growth and seed production, which are key features of the model developed at the Scottish Crop Research Institute (Squire et al., 1997a).

The main benefit of this approach is that the model will identify the steps in the population dynamics cycle (Fig 7.1) that have the main influence on the persistence of volunteer rape through the rotation.

### 7.2 Description of model

The model is based on a series of steps starting from seed losses from a rape crop and calculating seed survival, seedling emergence and seed production in subsequent crops. The key elements are as follows:

seed losses at harvest, seed movement due to soil cultivation, dormancy induction in relation to water potential, germination from the seedbank as a function of burial depth, emergence in relation to burial depth, plant survival as influenced by control measures and seed production in subsequent crops as affected by competition.

The basic model is as follows.

$$\mathbf{N}_{t+1} = \mathbf{D}_{j} \{ \mathbf{C}_{j} \{ \mathbf{I} - \mathbf{G} \} \mathbf{N}^{t} + \mathbf{d}(\mathbf{M}_{i}) \mathbf{C}_{j} \{ \mathbf{1}_{i} + \{ p \mathbf{s}_{i} \mathbf{e} \ \mathbf{G} \mathbf{N}^{t} \} / \{ \mathbf{s}_{i} \mathbf{e} \ \mathbf{G} \mathbf{x}^{t} + \mathbf{d}_{i} \} | \ 0 | \ 0 | \ 0 \} \ \}$$

 $N^{t}$  is the current seedbank,  $N_{t+1}$  is the seedbank in the following year,

 $C_j$  and  $D_j$  are matrices derived from Cousens and Moss (1990) that represent the proportions of seed moved to each soil layer by first cultivation after seed shedding  $(C_j)$  and all following cultivations  $(D_j)$ ,

'j' indexes the cultivation implement; current options: no cultivation, 5 cm spring tining, 10 cm flexi tining, and 20 cm ploughing,

I is the identity matrix, which represents the distribution of freshly shed seed after the first cultivation,

G is a diagonal matrix whose values represent the proportion of persisting seeds that germinate at each depth per year. These values are mainly based on burial experiments by Schlink (1994). It is assumed that the difference in the number of seeds persisting in year t and seeds persisting in year t + 1 is a result of germination in that layer. As Schlink's (1994)

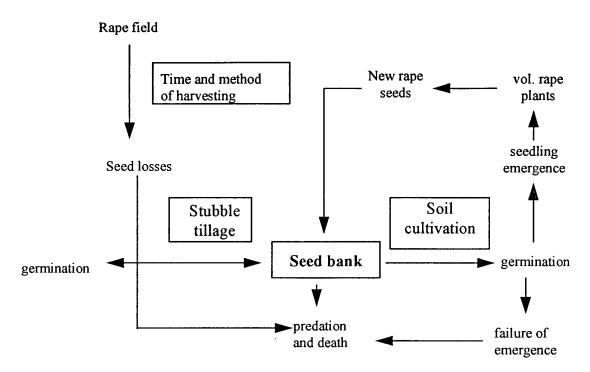
- experiments were done in uncultivated soil, further data has been included from an arable field experiment set up at Rothamsted to monitor the effects of cultivation on weed seed persistence (see Miller et al., 1998)
- M is a diagonal matrix whose values represent the water potential at each depth in the soil after crop 'i',
- 'd' gives the proportion of seeds becoming dormant at each water potential. The values are currently fixed and were derived from the experiment presented in Table 4.1.
- $'l_i'$  represents the amount of seed shed by crop 'i',
- 'p' represents the production of seeds/m by an average rape stand. Currently the value chosen is 60,000/m, which represents the number of seeds produced by an average rape crop. Data based on Leach *et al.* (1994).
- 's<sub>i</sub>' represents the proportion of volunteers surviving in crop 'i'. It is assumed that survival is largely determined by the weed control measures applied to a certain crop. 's' can be varied between 0 and 0.1. The latter value (0.1) represents volunteer survival in the absence of direct control measures, as for example volunteer survival in an oilseed rape crop.
- e is a vector determining the proportion of germinated seeds reaching the soil surface. This value was derived from an experiment by Lutman (1993), which was designed to test the effect of burial depth on emergence of non-dormant rape seed.

#### 7.3 Running the model

Sensitivity analysis on all parameters of the model has been carried out. Unfortunately, this showed that two parameters that appear to be particularly important for the output of the model have a very weak basis of data. These are the longevity of rape seeds in soil and the proportion of seedlings emerging from the seedbank each year. The only data that were available for these parameters were from the work of Schlink 1994 and Miller et al., (1998). However, two field experiments started in 1995 at Rothamsted and Woburn (see Sections 5 & 6) have now begun to generate relevant data and this information will be used in future updates of the model. It is our intention to continue the development of the model this autumn, incorporating the new data. The indications from the work, to data, are that the critical issues are the numbers of seeds shed by the rape crop at harvest, seed survival in the soil and the relationship between the seeds in the soil and the number of volunteer plants. The model does support the conclusion that number of seeds shed at harvest is very important, a conclusion also reached by the Squire model (Squire et al., 1997a). This model also concluded that the efficacy of herbicide use in the crops following rape and density dependence of seed production were also important.

Further work is needed to explore fully the validity of the model and its ability to highlight the critical factors implicated in the survival of the seed and the emergence of volunteer plants.

Fig 7.1 Life cycle of volunteer oilseed rape



# 8. General discussion and conclusions

A number of issues arising from the research described in the previous sections merit further discussion. Volunteer rape is a serious weed, particularly in broad-leaved crops and the current concerns about gene escape from transgenic crops, especially herbicide resistant types, gives added emphasis to the importance of this weed. It must be noted that herbicide resistance has also been bred into rape by conventional means. Conventionally bred rape cultivars resistant to triazine and imidazolinone herbicides are already been grown in Canada as well as the genetically modified rape cultivars resistant to glyphosate and glufosinate. Volunteer herbicide resistant rape may be more difficult to control in subsequent crops and so any way of minimising the persistence of the seeds will be welcome. For example, post-harvest stubble applications of glyphosate to control volunteer seedlings will be ineffective on glyphosate resistant volunteers, or triflusulfuron used to control weeds in sugar beet may fail to control imidazolinone resistant rape volunteers.

### 8.1 Seed shedding at harvest

Volunteer rape would not occur if harvesting removed all the rape seeds from the crop and transferred them to the combine tank. In reality, up to 10% of the crop can be left in the field depending on harvest conditions. It is clear that delayed harvesting is the main generator of high numbers of seeds (see Price et al., 1996). This is controlled by the weather and is not really in the farmer's control. It is self-evident that careful setting of the combine and timely harvesting will help to minimise losses, but even with optimum harvesting, it is still likely that c. 5,000 seeds/m² will be left behind, far more than are needed to create a new crop. Research funders are aware of the need to reduce harvest losses and an EU funded project is looking at ways of reducing pod shatter (Child et al., 1995). Unfortunately, this is not a very easy task as there is little variation in resistance to shatter in current rape cultivars.

Thus, we must conclude that for the foreseeable future all rape crops will leave some seeds in the field at harvest, to form a potential nucleus of seeds for future infestations of volunteer rape.

### 8.2 Dormancy in rape seeds and the effect of post-harvest soil cultivation

Previous research in Germany and the work carried out in this project all concludes that rape seeds have virtually no primary dormancy and that persistence arises from the generation of secondary dormancy. It is interesting that Schlink's work (1994) showed that green seeds tested 8 weeks after flowering possessed some primary dormancy (only c. 80% of seeds germinated in germination tests) but once the seeds are mature they seem to have no primary dormancy. Consequently, if the seeds fall on wet ground at, or just prior to, harvest they will germinate swiftly, as is seen in wet Augusts. We can find no evidence for the prevention of germination in these fresh seeds, if the soil is wet enough to cause germination. It is clear that the primary mechanism that causes the seeds to develop secondary dormancy is dry conditions and darkness. Thus, seeds buried in dry soil will become dormant. This was confirmed by both our laboratory and field tests.

The questions that need to be answered are; how wet does the soil have to be to permit germination, how long does it take for the seeds to become dormant, are their differences between cultivars and what happens if the seeds are left on the soil surface (after-ripening)? Our research has generated many of the answers. Firstly, the soil does not have to be very wet

to cause the seeds to germinate. In Petri-dish tests the osmotic potential of the solution had to be less than 10 bar to induce dormancy, which in soil terms is very dry. It is not possible to say how many ml of water / g of soil this equates to, as it depends on the soil type. However, it seems from the 1997 field tests that 4mm of rain prior to seed shedding was enough to prevent the onset of dormancy, and cause the seeds to germinate, except on the sand soil, where more rainfall was needed. It must be emphasised that in order for the seeds to become dormant they must have access to sufficient water to let them become physiologically active but not so much that they start to germinate. This must be the case, as seeds kept in bags in seed stores do not become dormant. Indeed our Petri-dish test described in Section 4 indicated that the potential to become dormant declined in seeds stored dry in constant conditions. However, this requirement for darkness and small amounts of water to initiate the development of secondary dormancy may explain why rape crops drilled into dry soil at the end of August fail to establish properly when the rain eventually arrives. To conclude, in general, seeds will only become dormant if they are in the dark and under conditions of moisture stress. It is possible that other environmental conditions that allow seeds in the soil to be physiologically active but not able to germinate, such as low soil temperature (below 6°C). The effect of low temperatures on seed germination has been explored by Squire et al.. Anaerobic conditions may also play a role in increasing the persistence of rape (1997b). seeds. However, such conditions are likely to occur only in late autumn and are unlikely in August and September, after rape harvest.

Secondly, how long do the seeds have to be in the dark for them to become dormant? The evidence from our work seems to be that they have to be in these conditions for at least two weeks (Section 4), but we do not have very strong evidence for this. We can say that if seeds are left on the soil surface for more than two weeks, prior to burial they are less likely to become dormant. It seems from both the laboratory studies, pot experiments and the field work, that leaving seeds on the soil surface prevents them becoming dormant when they are eventually buried. This conclusion supports that of earlier work by Schlink (1994, 1998) who had found lower persistence in buried after-ripened seeds. We are sure that four weeks on the soil surface will minimise the subsequent development of dormancy, but have less evidence that two weeks is adequate. The field studies in 1997, were aimed at answering this question, but the wet August in 1997 made it difficult to collect the relevant data. In practical terms rape stubbles should be left uncultivated for at least two weeks after the harvest, particularly if the soil is dry and there is little or no rainfall. If it is wet, then cultivation will not generate dormant seeds. In dry conditions, even shallow stubble cultivations generated more dormant seeds than leaving the stubble uncultivated (see Section 5). Intuitively one might believe that mixing the seeds shallowly into the soil would be a good idea, as it would encourage germination. In reality it has the opposite effect, reducing soil moisture levels and putting some seeds in the dark where they become dormant.

Our work shows that there are differences between cultivars in their potential to persist. Most cultivars have a low potential to persist (see Table 4.5) but a minority, including several of the more widely-grown cultivars including Apex, have a higher potential. The environmental conditions experienced by the seeds as they mature, may influence their ability to become dormant, but the overriding influence is of the cultivar. This conclusion is primarily based on Petri-dish tests and so requires further confirmation in the field. It has not been possible to cover this aspect in the current project, and it remains to be done.

Finally, we have demonstrated that deeply buried seeds, as occurs after ploughing, are more likely to persist than shallowly incorporated ones. This is because the lower diurnal temperature variation in the soil at plough depth, increases the percentage of dormant seeds.

Conversely, variation between day and night temperatures seems to decrease the generation of dormant seeds.

#### 8.3 Persistence of seeds

Having generated a soil seed bank of rape seeds, how long will they survive? This is a key question and it is clear from anecdotal evidence that rape seeds can survive for a number of years (eg Sauermann, 1993). Prior to the initiation of this project the only research on the long-term survival of rape seeds was that started by Schlink and Bauemer in 1987 (Schlink, 1998). Schlink found 0.5% of seeds still present after 10 years. However, her work was in an undisturbed situation and does not really reflect what might happen in an arable field. A few other projects have looked at the persistence of rape seeds but in most instances conditions at burial have not been appropriate to generate adequate numbers of dormant seeds to estimate long-term survival. (Hails et al., 1997; Miller et al., 1998). In this project we have conducted several pot experiments and started two field experiments in 1995, one of which (Woburn) generated large numbers of dormant seeds. The overall conclusion of these experiments, is that the first critical factor in the long-term survival of rape seeds, is the percentage that become dormant in the first autumn (see Section 8.2). Once seeds have become dormant they will persist for some years. In the pot experiments, decline rates varied from high seed loss to almost no loss over 1-2 years. In the two field experiments seeds are still present two years after burial. The experiments are continuing. In the Woburn experiment the decline rate has been closely monitored and this predicts 90% seed loss in c. 3yrs but complete loss only in 9 years (Fig 5.1) but we need more data to confirm this.

The overall, if slightly speculative, conclusion from our work both in the field and in pots, with both undisturbed and disturbed (cultivated) seeds, and that of Schlink (1998), is that once dormant seeds have been created, persistence for 2-3 years is certain, persistence of perhaps 10% of seeds could be for 5 years and persistence of about 1% could be for 10 years. Persistence is very difficult to predict precisely because of the variation in the percentage initially becoming dormant, the cultivar used and site variables. To reiterate, the critical issue is the percentage that becomes dormant initially, once the seeds are dormant in the soil, decline is relatively slow.

It must also be emphasised that as initial seed densities can be higher than 10,000 seeds/m<sup>2</sup>, only a very low % survival can still result in an appreciable weed problem. For example only 1% survival would leave 100 seeds/m<sup>2</sup>, which according to other data would result in 1 plant/m<sup>2</sup>, enough to cause yield losses in poorly competitive crops such as onions.

#### 8.4 Control of volunteer rape

Although how to control volunteer rape seedlings was not part of this project, it is obviously of significance. If the problem is to be minimised, seed production by volunteers must be minimised, otherwise the rape seedbank will increase. The primary method of control is with herbicides, but rape seedlings are not very easy to control, especially in broad-leaved crops, partly because large seedlings (bigger than 4 leaves) are more resistant and partly because the period of emergence of the seedlings is very long, making timing of control rather difficult.

Control in cereals is relatively straightforward, as many of the broad-leaved herbicides used in wheat and barley are effective (Table 8.1). The choice in broad-leaved crops is more limited, the costs can be higher and the timing of control is more difficult. For example, in field beans production costs would not justify more than one treatment of bentazone, but it is

extremely difficult to time this single application; as the early emerging rape may be too large, the late ones may not be emerged and the temperatures may not be high enough to achieve good activity. The need for control is greatest in the less competitive crops such as spring field beans, sugar beet and linseed. It is even more acute in uncompetitive vegetable crops such as onions, where several applications may be needed solely to control this weed. The difficulty of controlling volunteer rape has been one of the main reasons why many sugar beet growers have not grown rape in the same rotation as beet. The recent arrival of triflusulfuron has improved control and the anticipated development of herbicide resistant sugar beet will also simplify volunteer rape control (provided the rape is not also resistant to the same herbicide), encouraging beet growers to include rape in their rotations.

Table 8.1 Herbicides effective on volunteer rape seedlings

Crops	Herbicides	
Cereals	eg. diflufenican, mecoprop, metsulfuron	
Field beans	bentazone	
Peas	fomesafen, pendimethalin + prometryne, isoxaben + terbuthylazine +	
	bentazone	
Potatoes	metribuzin, rimsulfuron	
Sugar beet	triflusulfuron, sequences of phenmedipham +/- metamitron +/- lenacil	
Linseed	metsulfuron, bentazone, bromoxynil + clopyralid	

## 8.4 Practical advice to minimise the problems arising from volunteer rape

The following are some key issues that farmers should consider, in approximate order of importance, to minimise the effects of volunteer rape.

- 1) Leave rape stubbles uncultivated for at least two weeks, preferably longer, after harvest if the soil is dry.
- 2) Do not be tempted to tine or disc the rape stubble in the first two weeks after harvest as this may encourage persistence, by burying seeds.
- 3) If some volunteers have germinated, use a herbicide rather than a cultivator to kill them.
- 4) Try to harvest as efficiently as possible, so that seed shedding is minimised.
- 5) Preferably follow the rape with a cereal crop, as this crop offers the best opportunity to control volunteers.
- 6) If you are following rape with a cereal, avoid ploughing, if possible, as this will maximise volunteer rape seedlings in the cereal where control is less difficult and minimise numbers in the second following crop.
- 7) If the rape stubble is ploughed be aware that the second crop after rape is likely to have more volunteers than the first one (as a result of reploughing the seeds back to the surface), so do not choose a second crop where control is particularly difficult, if high volunteer numbers are expected.
- 8) If volunteer rape is likely to pose serious problems in other crops in the rotation, avoid growing cultivars that seem to have a high potential to persist.

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# Appendix 2 - Acknowledgements

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