

Gene stacking in herbicide tolerant
oilseed rape: lessons from
the North American experience

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Gene stacking in herbicide tolerant oilseed rape: lessons from the North American experience.

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Executive Summary

The aim of the project is to provide English Nature with evidence about the potential rate of formation and spread of 'stacked gene' volunteers of oilseed rape and the management of these plants in order to be better able to advise DETR (now DEFRA) in the risk assessment process. This has involved discussions with UK consultant agronomists and scientists before and after a visit to Manitoba and Saskatchewan in October 2001.

Systems of farming in the Canadian prairies have become very dependent on glyphosate. Despite this, the widespread presence of canola (oilseed rape) volunteers that are tolerant to glyphosate has not compromised these systems.

It is concluded that herbicide tolerant gene-stacked volunteers of oilseed rape would be inevitable in practical agriculture in the UK. It is recommended that more emphasis would have to be placed upon post-harvest cultivations in order to minimise volunteer populations in subsequent crops. However, it is considered that there would be little impact on other agricultural practices if only genetically modified (GM) varieties tolerant to glyphosate (Roundup Ready) or glufosinate (Liberty Link) were introduced into the UK. The main implication for herbicide use would most likely be the increased usage of paraquat +/- diquat pre-drilling of crops. This might have an impact on hares.

Discussions with consultant agronomists suggest that UK farmers may be more likely to try to control feral rape populations on uncropped land if they were to contain herbicide tolerance genes. It is recommended that more emphasis would have to be placed upon preventing their establishment in uncropped land and on methods of control that would have minimal or no impact on biodiversity.

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Background

In 1998, a field of oilseed rape (*Brassica napus*), known as canola in North America, was identified in Alberta as having volunteers with multiple tolerances to glyphosate and/or glufosinate and/or the imidazolinones. The sequential crossing of three herbicide tolerant varieties is the most likely explanation for the observed multiple herbicide tolerance (Hall *et al*, 2000).

In 1999, a further 11 fields in Canada were confirmed as containing multiple herbicide tolerant volunteers. However, weed scientists suspect that the occurrence of volunteers with 'stacked' herbicide tolerant genes is common in the Canadian prairies (Hugh Beckie, Agriculture Canada, personal communication). There is a consortium of Canadian scientists, Suzanne Warwick (Ottawa, Ontario), Ginette Seguin-Swartz (Saskatoon, Saskatchewan), Anne Legere (St. Foy, Quebec) and Hugh Beckie (Saskatoon) who are involved in field research on gene stacking in *B. napus*. In addition, Keith Downey, a world authority on the movement of pollen from canola, works for Agriculture Canada at Saskatoon. Also in Saskatoon is the Saskatchewan Canola Development Commission which funds research *via* a levy on sales. Suzanne Warwick leads Canadian research on gene flow to weed species.

No 'gene stacking' of herbicide tolerance in canola volunteers has been recorded in the United States, although it has occurred in experimental conditions (Brown, University of Idaho, personal communication). The major reason for this is that they only received registration to grow commercially GM herbicide tolerant canola in 2001, and this for two regions only. The key centres for research are the Universities of Idaho and Dakota. Dakota has field conditions and cultivars similar to Canada. The Idaho group has been researching the 'risk' of large-scale copolymerisation of GMO canola for 10 years. It has been looking at gene flow within canola crops (volunteers) and also to related weed species. From field research plots (over two years) there are canola lines that contain three herbicide tolerance genes (glyphosate, glufosinate and the imidazolinones). Also over this time and in similar field trials, canola x weed hybrids that contain two herbicide tolerant trans genes have been identified.

Gene stacking of HT genes in volunteer canola has implications for biodiversity in at least three ways:

- It can provide a source of HT genes to future crops growing in the same or neighbouring fields. This may result in intensification in the use of cultural or chemical control methods in cereals and other subsequent non-canola crops within the rotation in order to minimise the number of canola volunteers.
- There may be changes in the management, both cultural and chemical, of current and subsequent canola crops to try to minimise the number of within crop HT gene-stacked volunteers. This may result in a change of pre-sowing herbicides and also post-harvest cultivations in order to minimise the number of volunteers in the future.
- There may be cultural or chemical methods adopted in non-cropped areas in order to reduce feral canola that may contain HT genes.

Programme of visit to Canada

Monday, 8 October 2001:

Visit farms near Winnipeg with Martin Entz, University of Manitoba who has been quoted in the Canadian press as having some concerns over gene stacking of HT genes in canola

Tuesday:

University of Manitoba, Winnipeg to discuss the issues with Lyle Friesen (expert in gene stacking) and also canola breeders, agronomists and extension workers

Wednesday:

a.m. - Hugh Beckie, AgCanada Saskatoon (Canadian canola gene stacking project)

p.m. - Dr. Keith Downey, AgCanada and Mr. Roy Button, Executive Director, Saskatchewan Canola Development Commission

Thursday

a.m. - Mr Eric Johnson (Scott Research Farm) weed biologist and former extension person

p.m. - Dr. Alan McHughen, University of Saskatchewan, transgenic oilseed breeder

Friday:

a.m. – Regina, to visit with Dr. Bill Greuel, provincial transgenic oilseed specialist

p.m. - visit two farmers who were co-operators in a major *B. napus* gene stacking study in 1999-2000

Cultivation of both GM and non-GM herbicide tolerant (HT) canola in Canada

Development of current cropping systems in the Prairie Provinces of Canada

The cropping systems on the Canadian prairies have changed dramatically over the last few decades (Table 1). The changes have met the objectives of increasing and diversifying output, reducing labour and machinery costs and conserving soil and moisture through improved tillage techniques and weed control. The availability and price of glyphosate has been a major driver of these changes over the last ten years. This has allowed chemical based rather than cultivation based fallowing and direct seeding. It is now estimated that 30% of the crop area is direct seeded without any cultivation and 60% of the area is tilled to 5-10cm, mainly in the spring.

Despite the reliance on glyphosate, the introduction of canola cultivars, genetically modified to tolerate this herbicide (Roundup Ready), has not appeared to compromise these systems. Indeed, herbicide tolerant canola is now providing an additional 'weed-cleaning crop' to wheat and fallow. This, in turn, is allowing the increased adoption of 'weed-dirty crops' such as peas and lentils into the rotation, at the expense of summer fallow. In addition, herbicide tolerance has also enabled canola to be grown on very weedy fields and also allowed the control of weeds that had become resistant to conventional selective herbicides used in this crop.

Table 1 Idealised changes in cropping sequences on the Canadian prairies (all crops are spring sown)

| | Year 1 | Year 2 | Year 3 | Year 4 | Year 5 | Year 6 |
|-------------|-----------------|--------|-----------------|-----------------|-----------------|--------|
| 1971 | summer fallow | wheat | wheat | - | - | - |
| 1976 | summer fallow | canola | wheat | - | - | - |
| 1981 | summer fallow | canola | wheat | Peas or lentils | cereal | - |
| Early 1990s | chemical fallow | canola | wheat | cereal | peas or lentils | cereal |
| 2001 | HT canola | wheat | peas or lentils | wheat | - | - |

Herbicide tolerant canola crops in Canada

There are two plant species of canola grown in Canada, *Brassica napus* (the same species as oilseed rape in Europe) and *Brassica rapa*. Only *Brassica napus*, which constitutes 90% of the canola area of western Canada and all the canola area of eastern Canada, has been modified to be tolerant of herbicides. There are currently four types of herbicide tolerant (HT) *Brassica napus* crops commercially available in Canada (Table 2). Three have been genetically modified to be tolerant to a specific herbicide, whilst the cultivars that are HT to the imidazolinones have been produced by conventional breeding techniques. Roundup Ready canola cultivars are the most commonly grown, despite the dependence on this herbicide in other parts of the rotation (Figure 1). The reason for their popularity is the effectiveness and cost of glyphosate, despite farmers having to pay a \$15Canadian/acre technology fee to Monsanto and having to sign an unpopular Technology Use Agreement with Monsanto. Surveys on why farmers adopt HT crops in general suggest that the ease of and better weed control (particularly herbicide resistant and perennial weeds) is the major reason. However, higher yields are also obtained because herbicide tolerant crops can often be sown earlier in the spring when it is still too cold for conventional herbicides to work effectively.

A survey of canola growers in western Canada carried out in 2000, but reviewing crop management practices from 1997 to 2000, recorded 2.13 herbicide applications in Roundup Ready and Liberty Link (i.e. transgenic) crops, compared to 1.78 applications in conventional crops. Both these numbers include pre-plant herbicides such as glyphosate. Conventional crop growers used more pre-plant soil incorporated herbicides, resulting in more cultivation of the soil. However, there does not appear to be a trend towards more or fewer applications of herbicides in herbicide tolerant crops. Recent research (in press) suggests that one well-timed post-emergence application of glyphosate is usually sufficient to retain yield because subsequent applications are controlling later germinating weeds that do not compete with the crop.

There is little or no information on changes in weed spectra occurring as a result of the use of non-selective herbicides in HT canola in Canada. Weed scientists expect that changes may occur in time.

Table 2 Herbicide tolerant crops currently registered in Canada in canola (*Brassica napus*)

| Herbicide tolerance | Common generic description | Variety registration | Food safety approval | Breeding system of canola | Weediness of canola | | |
|---------------------|----------------------------|----------------------|----------------------|---------------------------|---------------------|-----------------|---------------|
| | | | | | Crop land | Disturbed areas | Natural areas |
| Glyphosate | Roundup Ready | ✓ | ✓ | 20-30% outcrosser | ✓ | ✗ | ✗ |
| Glufosinate | Liberty Link | ✓ | ✓ | “ | ✓ | ✗ | ✗ |
| Imidazolinone | Clearfield | ✓ | ✓ | “ | ✓ | ✗ | ✗ |
| Bromoxynil | Navigator | ✓ | ✓ | “ | ✓ | ✗ | ✗ |

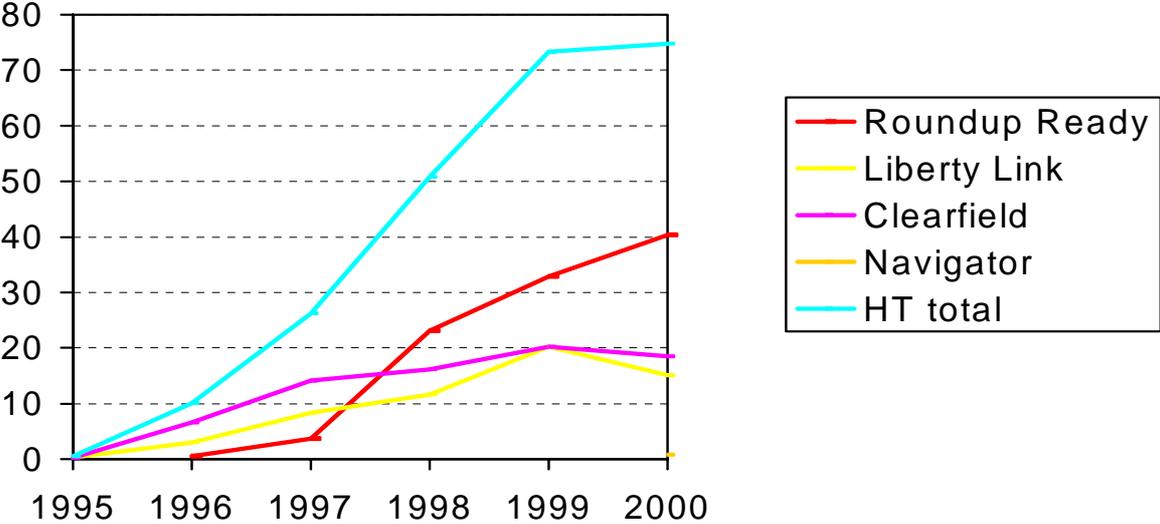


Figure 1 Herbicide tolerant (HT) canola, as a percentage of the total area grown in Western Canada. 4.8 million ha grown in 2000 (R K Downey, personal communication)

Existing regulations and guidelines for the cultivation of HT canola in Canada

There is little or no guidance to growers on the cultivation of HT canola in Canada. There is a recollection that a distance of 175 metres between HT crops and crops with other types of HT or conventional cultivars has been suggested but there was no supporting written material available (Beckie *et al*, 2001). This lack of guidance has resulted in HT crops being commonly grown next to other HT crops or conventional varieties and it is now considered that gene-stacked volunteers are not uncommon in Canada. A spray contractor near Winnipeg says that he assumes volunteers contain at least the Roundup Ready gene unless he has very good reason to suspect otherwise.

Commercial seed in Canada should contain less than 0.25% off-types, including individual seeds that contain unintended HT genes. Surveys show that HT genes regularly occur in conventional varieties but usually below the 0.25% level by having isolation distances of at least 200 metres from commercial crops for non-hybrid seed production. Breeders now have to take particular care to ensure their basic or foundation stocks are clear of unintended HT genes. The seed standard for foundation stock is less than 0.1% off-types. Breeders are accepting that because of gene flow from commercial crops, it may be impossible to produce hybrid varieties in Canada (because the process of which involves the use of male sterile plants) that meet their seed standards. There is also an acceptance that varietal associations, which again include male sterile plants, may not meet a standard of having HT genes in less than 1% of the seed from a commercial crop.

The incidence and spread of canola volunteers containing two or more stacked HT genes and changes in herbicide use associated with the management of gene-stacked volunteers

There is no monitoring of the extent of gene-stacked volunteers in Canada. Eleven instances were documented in 1999 and it is accepted that gene stacking of the three most commonly grown HT crops (Roundup Ready, Liberty Link, Clearfield) can readily occur in practice. An Agriculture Canada project recorded stacking of genes in volunteers in all 11 locations where they measured the implications of growing Roundup Ready and Liberty Link crops in adjoining fields in 1999 (Beckie, 2001). The level of gene flow was similar between the two crops and matched data obtained in the UK by (Ingram, 2000). Pollen flow to the 11 Roundup Ready fields is provided in Figure 2 and shows that it amounted to just over 1% on the field edge but varied between 0.1-0.2% from 50 to 400 metres within the crop. However, gene flow was detected to the maximum distance recorded of 800 metres. The measurements may have been compromised, at least in some instances, by the presence of gene stacking in the drilled seed.

Therefore, it is safe to assume that gene stacking between HT crops is almost impossible to prevent unless the crops are very widely dispersed.

Gene-stacked volunteers are generally managed in Canada by the addition of a low rate of 2,4-D to the pre-seeding application of glyphosate. Those emerging with the crop are controlled by post-emergence herbicides. 2,4-D also has to be used post-emergence in cereals where volunteers contain the Clearfield tolerance gene that results in resistance to ALS inhibitor herbicides, such as the sulfonylureas, which are commonly used in this crop. There are one or two instances where there are concerns. Pre-seeding applications of 2,4-D cause unacceptable crop damage to sunflowers on the heavy clay soils near Winnipeg. Post-emergence herbicides are unable to remove volunteers, gene-stacked or conventional, that have emerged before the crop. Here, alternative pre-seeding herbicides to be mixed with glyphosate are being evaluated, notably aminotriazole (no longer available in the UK). Some farmers are now considering Liberty Link as the preferred HT option, rather than Roundup Ready or Clearfield, because of the low usage of glufosinate in non-HT crops and hence the ease of controlling volunteers. In addition, the cost of Liberty has been reduced recently to ensure that total costs are similar to that of the Roundup Ready system.

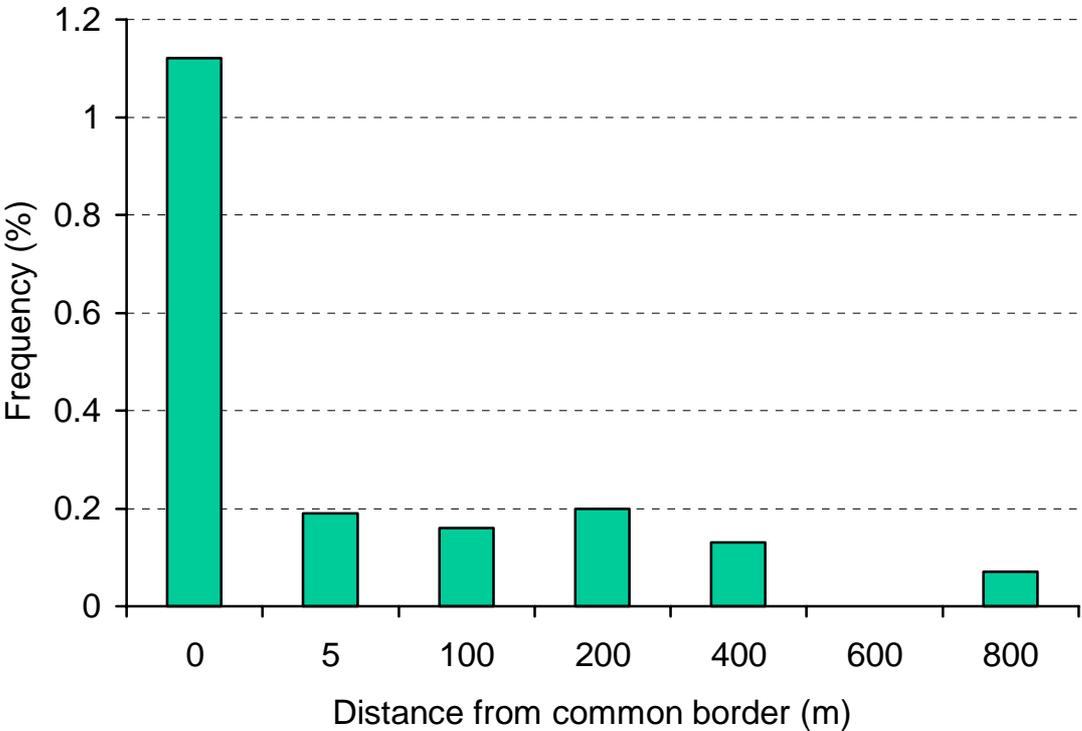


Figure 2 Frequency of gene stacking (% plants) due to pollen flow from 11 Liberty Link fields to adjoining Roundup Ready fields 1999 (Beckie, 2001)

Naturally, there could be a problem in controlling gene-stacked volunteers post-emergence of a subsequent canola crop but this is no worse than the situation that was faced before the introduction of HT crops. However, canola is grown no more than once in four years and surveys show that the numbers surviving the previous crop are less than half of one plant per square metre (Legere *et al.* 2001). In this situation, a similar number of gene-stacked volunteers could be introduced from the seed, despite meeting the seed standards of 0.25% off-types.

There appeared to be no instances where additional cultivations or where crop margin management is changed in order to control gene-stacked volunteers and feral rape was not considered an issue. Hence, no control measures were attempted in non-cropped areas.

General conclusions regarding the likelihood that gene stacking could become a problem in the UK and the predicted speeds/densities at which this could occur, based on the North American experience

Experience in Canada suggests that the cheap and effective weed control made possible by HT oilseed rape is attractive to farmers because of increased margins and ease of crop management. In addition, particularly in spring oilseed rape in the UK, the flexibility of timing of weed control in HT crops may result in more crops not being treated with herbicides at all. Currently, the cheapest and most effective selective broad-leaved herbicides are applied either pre-emergence or immediately post-emergence in the UK.

However, it should be noted that weed surveys in Canada suggest to the author of this report that competition from broad-leaved weeds is more severe in canola in Canada than in oilseed rape in the UK. In addition, HT crops will not enable earlier sowing in the UK.

Many UK farmers may be particularly anxious to grow HT oilseed rape because of the widespread occurrence of herbicide resistance in black-grass (*Alopecurus myosuroides*). These crops offer cheap and effective control of this weed with a mode of action that is not associated with herbicide resistance in the UK. Hence, this might also reduce the selection for herbicide resistance.

The UK organisation SCIMAC (Supply Chain Initiative on Modified Agricultural Crops) have issued guidelines that address the specific on-farm issues raised in relation to HT crops. These include guidance on volunteers in subsequent crops, cross-pollination with other crops and wild or weed relatives and gene stacking.

There is little doubt that gene-stacked volunteers would occur as soon as differing HT crops were grown in proximity. The SCIMAC guidelines include separating a HT crop from other oilseed rape crops by at least 50 metres. Both UK gene flow data and Canadian experience suggest that this would be effective in reducing significantly the occurrence of gene flow to other canola crops that are not varietal associations or possible partially restored hybrids, and perhaps both the practicalities and the data suggest that there is little benefit in aiming for a greater separation distance.

In Canada, it is believed that the risk of gene stacking between HT cultivars that are not varietal associations and perhaps not HT partially restored hybrids, is based on simple mathematics. This simple relationship is not totally accepted by UK scientists. However, if proven to be correct: not only is the separation distance between two HT cultivars important but so too is the percentage of off-types or volunteers containing another HT gene in the recipient conventional crop.

The inevitability of gene stacking suggests that the issue is not whether gene-stacked volunteers occur but whether their existence creates additional problems to say volunteers from Roundup Ready rape alone. It is possible in the first instance that Liberty Link and Roundup Ready oilseed rape may be introduced into the UK. BASF have stated that the UK weather is too cool for the reliable activity of the imidazolinones. There is still the issue of achieving registration to use glufosinate in the winter months and this may significantly reduce the likely adoption of Liberty Link winter oilseed rape, which dominates the area grown. However, there could still be adoption of Liberty Link cultivars, but treated with conventional herbicides, if they offered agronomic advantage.

Glufosinate is little used in the UK. This is because it works most effectively at warm temperatures and also should be sprayed whilst there are two hours of sunlight remaining. Much of the spraying in the UK is done after the wind drops in the evenings.

In practice, there is little or no additional significance in controlling gene-stacked volunteers containing the Roundup Ready and Liberty Link HT genes as there is in controlling Roundup Ready volunteers. Agronomists do not perceive glufosinate as an alternative to glyphosate where Roundup Ready volunteers occur.

In Canada, 2,4-D is added to glyphosate prior to seeding and selective herbicides are used post-emergence of the crop to control Roundup Ready volunteers or for the control of volunteers that contain the stacked genes from Roundup Ready, Liberty Link and Clearfield. The only implication to post-emergence herbicide selection is where the Clearfield gene is present: this results in resistance to all ALS inhibitor herbicides, which include the sulfonylureas that dominate post-emergence broad-leaved weed control in cereals both in North America and Europe. In this instance, on occasions, an additional dose of a phenoxy herbicide (2,4-D in North America, mecoprop in Europe) may have to be added to sulfonylurea herbicides. However, should Clearfield be introduced into the UK, the control of the transgene alone or with other HT transgenes in volunteers, should have little significance because mecoprop is often added to sulfonylurea herbicides in cereals in the UK to control rape volunteers. The only other crops where HT Clearfield volunteers could cause a change of post-emergence herbicide policy in the UK is in potatoes and sugar beet where sulfonylureas are used but alternative herbicide policies could be adopted to overcome the problem. However, this may result in additional cost and management time.

However, in the UK 2,4-D (or mecoprop or MCPA) is not approved for use prior to seeding a crop and the crop safety of this use has not been thoroughly tested. Hence, the likely registered approach in the UK would be to revert to paraquat + diquat for pre-seeding control of gene-stacked volunteers or Roundup Ready volunteers. However, some consultants say that they may consider the use of glufosinate where it is known that only the Roundup Ready gene is present. UK consultants are aware that, due to gene flow, HT volunteers could appear after a conventional crop but feel unable to say how they would tackle this issue. Monsanto have employed staff to physically destroy Roundup Ready volunteers where this situation has occurred in Canada.

In addition, diquat rather than glyphosate will be used to desiccate peas and beans that may still contain HT Roundup Ready volunteers. Also, there will be a direct reduction in the area desiccated prior to harvest with glyphosate in Roundup Ready crops and glufosinate in Liberty Link crops of oilseed rape.

Table 3 shows the current usage of glyphosate in the major crops and indicates the farmer's current preference for glyphosate rather than paraquat +/- diquat for pre-seeding weed control. However, it is not known how many hectares are sprayed to kill volunteer oilseed rape as one of the target weeds. Estimates by consultants in the Eastern Counties suggest that this varies between 10-25% of the pre-seeding use of glyphosate. Glyphosate is preferred both on cost grounds and operator safety issues. In addition, the Environmental Information Sheet on paraquat suggests that its use could impact on hare populations.

Disease surveys carried out by ADAS/CSL suggest that another broad-leaved crop is commonly grown two years after an oilseed rape crop. There could be significant numbers of volunteers emerging pre-seeding or post-emergence in such a broad-leaved crop, particularly if the cultivations after the oilseed rape do not follow SCIMAC advice of delaying any cultivation after harvest (Pekrun *et al.*, 1998). Any volunteers that survive seedbed cultivations may be difficult to control selectively post-emergence in these broad-leaved crops, particularly in broad and field beans and peas. However, the numbers may not hinder cultivations and provided that the plants are small, they may be controlled by the seedbed cultivations. This is confirmed by the fact that, at most, 30% of peas or beans receive a pre-seeding application of a non-selective herbicide but the control of volunteer oilseed rape may not be required in many of these cases. It is also interesting to note that at least 7% of the pea crop was treated with glyphosate pre-harvest. There could be a reduction of this use in favour of diquat if Roundup Ready oilseed rape volunteers survive the selective herbicides used in peas.

Table 3 Sprayed area of paraquat +/- diquat and glyphosate in arable crops (ha and % of crop area) in Great Britain, 1998

| | Paraquat +/- diquat | | Paraquat +/- diquat | | Glyphosate | | Glyphosate | | Glyphosate | |
|---------------|------------------------|-------|------------------------|-------|------------------|-------|------------------|-------|----------------|----------------|
| | Pre-em/preseeding | Other | Pre-em/preseeding | Other | Preseeding/preem | Other | Preseeding/preem | Other | Before harvest | Before harvest |
| | ha | % | ha | % | ha | % | ha | % | ha | % |
| Wheat | 9,654 | 0.5 | 2,208 | 0.1 | 99,209 | 4.9 | 91,947 | 4.5 | 57,983 | 2.8 |
| W. Barley | 4,563 | 0.6 | 720 | 0.1 | 40,106 | 5.3 | 43,466 | 5.7 | 8,439 | 1.1 |
| Spring barley | 823 | 0.2 | 0 | 0 | 16,901 | 3.7 | 26,823 | 5.9 | 7,767 | 1.7 |
| Oats | 896 | 0.9 | 290 | 0.3 | 1,126 | 1.2 | 4,346 | 4.6 | 825 | 0.9 |
| Winter OSR | 9,425 | 2.1 | 1,325 | 0.3 | 19,201 | 4.3 | 48,065 | 10.8 | 40,369 | 9.1 |
| Spring OSR | 99 | 0.2 | 625 | 1 | 8,058 | 13.0 | 13,378 | 21.6 | 3,131 | 5.1 |
| Beans | 149 | 0.1 | 733 | 0.7 | 16,914 | 15.3 | 8,168 | 7.4 | 692 | 0.6 |
| Peas | 3,127 | 3.1 | 2,551 | 2.5 | 19,065 | 18.7 | 10,263 | 10.1 | 7,270 | 7.1 |
| Linseed | 320 | 0.3 | 470 | 0.5 | 13,556 | 13.6 | 21,541 | 21.6 | 16,571 | 16.6 |
| Sugar beet | 6,180 | 3.3 | 3,945 | 2.1 | 43,725 | 23.2 | 10,079 | 5.4 | 1,711 | 0.9 |
| Potatoes seed | 4,558 | 28 | 10,665 | 65.6 | 105 | 0.6 | 190 | 1.2 | 0 | 0.0 |
| Potatoes ware | 16,760 | 11.8 | 85,436 | 60.1 | 13,437 | 9.5 | 5,845 | 4.1 | 1,452 | 1.0 |
| Other crops | 422 | 3.2 | 470 | 0.5 | 7,982 | 21.0 | 9,269 | 32.4 | 7,134 | 24.0 |
| Set aside | 716 | 0.2 | 1,097 | 0.4 | 8,251 | 2.7 | 118,599 | 38.2 | 5,367 | 1.7 |
| | 57,692 | | 110,535 | | 307,636 | | 411,979 | | 158,711 | |

On the other hand, in cereals, provided that the number of volunteers do not hinder sowing, large Roundup Ready volunteers that survive seedbed cultivations may be controlled post-emergence of the crop. However, many farmers are anxious to control volunteer oilseed rape pre-seeding of cereals in order to reduce the risk of slug damage.

Current discussions with crop consultants confirm that farmers appear to have zero tolerance of volunteer oilseed rape in their crops. This is because of their concern over populations of this plant in future crops, particularly in succeeding oilseed rape crops. Rape is grown more intensively on the heavy soils in the UK where it has a higher gross margin than other broad-leaved crops. It is grown every other year on a few farms but is normally grown every four to six years on heavy land farms. The current downturn in farming has resulted in farmers being less certain of their future cropping, which can only increase the pressure to control volunteer oilseed rape.

In the future, farmers may receive encouragement to leave some weeds in crops in order to encourage biodiversity. One of the weeds identified in a recent report for DEFRA as being important to farmland birds is charlock (*Sinapis arvensis*), which like oilseed rape is from the Cruciferae. Unfortunately, the herbicides that selectively control volunteer oilseed rape will also control charlock. On the other hand, the ease and low cost of broad-leaved weed control offered by HT crops in broad-leaved crops may reduce the current over-zealousness to control other 'desirable' annual broad-leaved weeds in the intervening cereal crops.

Gene stacking in rape volunteers does have implications when they need to be controlled in other HT crops, such as maize and sugar beet. It is possible that HT crops could be rotated so that, for instance, Roundup Ready rape volunteers could be controlled in Liberty Link maize or sugar beet. However, gene-stacked volunteers may result in it being necessary to add a selective herbicide to a post-emergence treatment in HT crops. This would appear to have little or no significance for biodiversity.

Non-cropped land

Feral rape is often noted on roadsides and sometimes in the vegetation of non-cropped areas in the UK. The Scottish Crop Research Institute (SCRI) in 1998 found genes in feral rape from a variety that had not been grown since 1986. This suggests that herbicide tolerant genes may persist in feral rape for a considerable time. However, the same researchers found that individual plants could survive for no more than two winters, even when flowering was prevented and there was no competition from other plants.

Discussions with UK crop consultants confirm that the desire to control feral rape plants may be increased if they contain HT genes. However, it is impossible to predict UK farmer actions in this respect and in Canada, feral rape rarely occurs.

There could be issues relating to the control of feral rape. Currently glyphosate is used on a large scale for weed control on hard surfaces (such as footpaths and the edge of roads). The presence of Roundup Ready rape volunteers would suggest that an additional herbicide would have to be used for their control but the obvious candidates may not be suitable for use because of rapid movement to water from these areas.

If the presence of HT rape volunteers results in the treatment of non-crop vegetation with herbicides, then other broad-leaved plants will be affected. This is because such treatments are likely to be based on mecoprop, which affects a range of plants. The areas where HT feral oilseed rape volunteers may establish, but may not persist between oilseed rape crops, are where plant competition has been reduced by herbicide drift from the application of Roundup or Liberty to the HT crop or drought or in such areas as conservation stewardship within Arable Stewardship. DEFRA suggests that feral rape should be controlled by cutting the one or two metres of conservation headland closest to the cultivated land throughout the season. In addition, not sowing the outer cultivated metre of the field with HT oilseed rape would radically reduce seed shed into uncropped land such as hedge side vegetation or grass field margins.

Arable Stewardship crop prescriptions include severe limitations on herbicide choice for the control of annual broad-leaved weeds. However, the herbicide that can be used in the low-input cereal option for the control of annual broad-leaved weeds is amidosulfuron (Eagle). This will control volunteer oilseed rape up to the flower bud stage of growth.

Gene flow from HT crops to related plants (including weeds) in North America

There is a major project in Canada that aims to identify gene flow from HT canola to related plants. None has been recorded so far but weed scientists expect to record gene flow to *Brassica rapa* crops or volunteers. In addition, there is a low probability that gene flow to *Raphanus raphanistrum* (wild radish) and *Erucastrum gallicum* (Dog mustard) could occur. *E. gallicum* does not occur in the UK.

Conclusions

- Gene stacking in volunteers is inevitable but the SCIMAC guidelines of a separation distance of 50m between HT cultivars that are not varietal associations and perhaps not HT partially restored hybrids will reduce its occurrence. Increasing separation distances between non-hybrid crops will only have a small impact on its occurrence unless the isolation distances are increased to 400m or more.
- The number of HT volunteers in subsequent rape crops and the proportion of seed that contains HT genes will have a significant influence on the occurrence of gene-stacked volunteers.
- The risk of outcrossing increases very significantly within varietal associations because they contain a significant proportion of male sterile plants.
- Based on current information, it is likely that only Roundup Ready (glyphosate tolerant) and Liberty Link (glufosinate tolerant) oilseed rape will be released in the UK, subject to registration. Due to the very limited usage of glufosinate in the UK, stacking of glufosinate and glyphosate tolerance genes in rape volunteers will have little or no additional practical significance over and above volunteers that contain only the glyphosate tolerance gene. The greatest practical impact of gene stacking may be when HT rape volunteers need to be controlled in other HT crops such as maize or sugar beet. Here, the additional use of selective herbicides may be required. This is unlikely to have a

significant effect on biodiversity because the selective herbicides are being used to restore the level of weed control to that normally achieved with either glyphosate or glufosinate alone.

- The presence of Roundup Ready volunteers may result in a significant increase in the pre-seeding usage of paraquat +/- diquat, unless 2,4-D or mecoprop receives approval for pre-seeding application. The Environmental Information Sheet on paraquat produced by the manufacturer suggests that it may have an impact on hare numbers although such a warning may be more relevant where grassland is being destroyed rather than the pre-seeding usage in arable rotations.
- Currently, farmers appear to have zero tolerance of volunteer oilseed rape in arable crops but in the future, they may receive encouragement to leave some weeds in crops in order to encourage biodiversity. One of the weeds identified in a recent report for DEFRA as being of potential importance to farmland birds is charlock, which like oilseed rape is from the Cruciferae. Unfortunately, the herbicides that selectively control volunteer oilseed rape will also control charlock. On the other hand, the ease and low cost of broad-leaved weed control offered by HT broad-leaved crops may reduce the current over-zealousness to control 'other desirable' annual broad-leaved weeds in the intervening cereal crops.
- The presence of HT rape volunteers may possibly result in the treatment of non-crop vegetation with herbicides and so other broad-leaved plants will be affected. This is because such treatments are likely to be based on mecoprop, which affects a range of plants. The areas where HT oilseed rape volunteers may establish, but may not persist between oilseed rape crops, are where plant competition has been reduced by herbicide drift from the application of Roundup or Liberty to the HT crop or drought or in such areas as Countryside Stewardship headlands.
- It is recommended by the author that more emphasis should be placed upon:
 - cultivations post-harvest of HT oilseed rape to reduce future populations of volunteer oilseed rape;
 - the possible cost and practical implications of herbicide tolerant volunteers. Canadian farmers openly admit that initially they did not give this aspect sufficient consideration;
 - implications of HT feral rape;
 - methods of reducing populations of feral rape;
 - the responsible control of feral rape.

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