Advisory Committee on Releases to the Environment

Advice on an application for deliberate release of a GMO for research and development purposes

Applicant: Wild Bioscience Ltd

Application: To release genetically modified wheat lines that investigate altered agronomic performance through the expression of plant regulators of photosynthesis.

Reference: 22/R55/01

Date: February 2023

Advice of the Advisory Committee on Releases to the Environment (ACRE) to the Secretary of State under section 124 of the Environmental Protection Act 1990

ACRE is satisfied that all appropriate measures have been taken to avoid adverse effects to human health and the environment from the proposed release. ACRE sees no reason for the release not to proceed according to the following advice.

To minimise the likelihood that GM wheat from this trial will enter the human food or animal feed chains, the applicant should:

1. Ensure that the 20 m surrounding the trial site is planted with a non-cereal crop and that cereal volunteers are controlled (prior to flowering) in this area during the trial.

2. Plant a wheat pollen barrier, of 3 m width, to flower at the same time as the GM wheat as an additional precautionary measure.

3. Control *Elytrigia repens* (Couch Grass) using a glyphosate herbicide and hand weeding, if necessary, within the trial site and the surrounding 20 m, before flowering and for the duration of the trial.

4. Ensure that any GM or non-GM wheat plant material remaining in the area of release at the end of the trial is disposed of appropriately.

5. Ensure that following harvest, the area of release is lightly tilled twice (once after harvest and again in the following spring) to a depth of 5 cm to stimulate germination of any wheat plant volunteers. The release areas should be left fallow and monitored for wheat plant volunteers for 2 years following harvest.

6. Record the number of wheat plant volunteers that germinate before destroying them with an application of glyphosate herbicide or hand pulling them prior to flowering.

7. Ensure that suitable measures (such as those described in Wild Bioscience's application) are put in place to keep birds (and animals) out of the trial area and that the efficacy of these measures are kept under review.

8. Ensure that machinery used on the site is cleaned thoroughly onsite, including between using it with GM and non-GM material, and that clothing and equipment such as vehicles used by personnel on the site are also cleaned thoroughly before leaving the site.

Comment

ACRE considered the risks to human health and the environment posed by the proposed release of wheat that has been genetically modified to give a phenotype of higher levels of Phytochrome B expression and enhanced photosynthesis and productivity in controlled environments. The aim of this research is to investigate the effect of this over-expression in the field. Furthermore, under controlled environmental conditions the eight identical transgenic lines intended for release were indistinguishable from untransformed controls. No other changes to the plant morphology or development were apparent.

Key characteristics of this field trial with respect to its environmental risk assessment are:

i) It will be on a small scale. The maximum area for the proposed trial in each year will be 500 m^2 including controls and the spacing between plots. The proposed release will be conducted within the GM field trial site at Rothamsted Research, Harpenden, UK. The trial will be planted first in the Spring of 2023 and then sequentially for five years until the final harvest in Autumn of 2027.

ii) The GM wheat and non-GM wheat grown in this trial will not be put into the human food chain or fed to livestock.

The applicant intends to trial 8 genetically modified lines; all contain the same gene construct PCK promoter-PHYB gene T-DNA. The GM lines also contain the antibiotic resistance genes encoding neomycin phosphotransferase I (*npt1*). NPT1 confers resistance to aminoglycoside antibiotics such as kanamycin and neomycin. This gene is used in the development of GM plants to facilitate the selection of plants that have been transformed successfully.

Molecular Characterisation

ACRE noted that the plants for this trial were a genetically modified UK milling wheat variety cv. Cadenza, that can be sown as either a spring or winter type. It was modified using *A. tumefaciens* mediated transformation to incorporate a promoter-gene cassette, as a single T-DNA construct, into a nuclear location. The gene of interest is derived from the wheat photoreceptor Phytochrome B (PHYB) from *Triticum aestivum*. The PHYB gene was placed downstream of the *Zoysia japonica* Phosphoenolpyruvate Carboxykinase (PCK) promoter (Nomura et al., 2005). The inserted T-DNA cassette of the plasmid also contained the *nptII* kanamycin resistance gene downstream of the Sc4 promoter for selection of transformed plants, and the right and left t-DNA borders. Wild Bioscience plan to trial eight separate transgenic lines, each containing the same gene construct outlined above, along with eight segregating wild type (or null) lines as controls.

ACRE made a number of points with respect to the lack of detail within the molecular characterisation presented within the application. These points were also mirrored in several the public representations received on this application. The applicant was therefore asked to address ACRE's concerns with respect to particular sections of the application. The applicant's response is presented here with reference to the application section that it supplements.

Application part A1, question 14:

(a) the size and structure of the insert and methods used for its characterisation, including information on any parts of the vector introduced into the genetically modified plant or any carrier or foreign DNA remaining in the genetically modified plant.

The total size of the DNA insert is 9601 base pairs (bp) and contains two coding sequences, one of which encodes a generic selectable marker, and the other the specific wheat gene of interest (PHYB). The insert contains foreign DNA components that are routinely used for plant transformation: *Agrobacterium tumefasciens*-derived t-DNA borders flank the insert, as is typical for agrobacterium transformation (each 23 bp long).

The selectable marker module contains an *E.coli*-derived aminoglycoside phosphotransferase (nptII) gene (813 bp coding sequence), in which a plant intron has been introduced from Arabidopsis thaliana FAD2 (1135bp), and expression of this gene is driven by the Sc4 promoter sequence which is derived from subterranean clover stunt virus (532 bp). Each of the two coding sequences comprising the insert contain *Agrobacterium tumefasciens*- derived nopaline synthase (Nos) terminator sequences (253 bp).

The other part of the DNA insert is the reason for the application itself: to specifically explore the impact of increasing the activity of a gene that is already encoded in the wheat genome; this part of the insert comprises the *Zoysia japonica* derived PCK promoter region (1656 bp) and the coding sequence derived from *Triticum aestivum* PHY B (3501 bp).

[subsection **(b) the size and function of the deleted region or regions,** is not applicable in this case]

(c) the copy number of the insert,

Molecular characterisation was carried out on the inserted sequences as follows. Primer pairs specific to the *nptII* gene were used to quantify copy number in T0 regenerating transgenic plants by digital PCR. These dPCR assays were repeated in the next (T1) generation and carried out for every T1 plant grown for each of the eight lines. This analysis confirmed that six lines contained a single copy of the insert, and the remaining two lines had two copies of the insert. All of these lines followed normal rules of Mendellian inheritance in the T1 generation consistent with the segregation of a single locus, from

which a population of plants that were heterozygous for this transgenic locus, homozygous, or segregating azygous were identified. The latter two populations were bulked for field trials.

(d) the location or locations of the insert or inserts in the plant cells (whether it is integrated in the chromosome, chloroplasts, mitochondria, or maintained in a non-integrated form) and the methods for its determination.

[Similarly, more detail was required in answer to question 15, **(b) Conclusions on the molecular characterisation of the genetically modified plant.**]

The molecular characterisation in answer to Question 14(c) above, described how copy number was quantified for each line. This was complemented with additional PCR testing to analyse insert integrity and length. For each line, every T1 plant was further analysed by PCRs using primers specific to the gene of interest; one primer pair amplified a small DNA sequence starting upstream of the PCK promoter region and reading into the 5' end of the promoter. Another primer pair was designed to specifically amplify the DNA sequence spanning the 3' end of the gene of interest coding sequence and start of the terminator. The results of these PCRs were checked for consistency with the dPCR data collected for the *nptII* gene and used to confirm that a full-length promoter and coding sequence had been integrated into each transgenic plant and was absent from segregating azygous plants and wild type controls. Based on consistent PCR findings across different primers, detection methods, and plant generations, Wild Bioscience concluded that the insert was stably integrated and segregating as a single locus.

Some of the public representations criticised Wild Bioscience's molecular characterisation of the GM lines for not including information on unintended effects on the genome, including the role this may play in altered phenotype of the resultant plants. These data are not required in applications for small trial releases of GM plants unless they are needed to inform the risk assessment. ACRE considered whether this information would provide useful data on the biological and agronomic characteristics of these plants compared to gathering data from the field. As part of this, ACRE were reminded of their previous discussions on what intrinsic characteristics of wheat these (or other) alterations would need to change in order for them to confer an environmental risk e.g. to make wheat a problem weed[1].

It is inevitable that there will be differences between plant lines. This is the case for conventional plant breeding as much as it is for GM. Attempting to interpret these differences is challenging and not constructive unless there is an indication of what hazard to look for. Under controlled conditions, the GM plants are indistinguishable from untransformed plants. An objective of the trial is to determine whether this is the case

[1] Chepil W.S. (1946) Germination of Weed Seeds I. Longevity, Periodicity of Germination, and Vitality of Seeds in Cultivated Soil. Scientific Agriculture **26**: 307-346.
Anderson, R. L. and G. Soper. 2003. Review of volunteer wheat (Triticum aestivum) seedling emergence and seed longevity in soil. Weed Technology **17**: 620–626.

under field conditions. Monitoring of GM plants is a standard requirement in any consent that is issued for a GM field trial.

ACRE considered further the implications of the inserted gene cassette including that of the presence of a selectable marker gene, in the environmental risk assessment section below. The structure of the genes and their associated promoters in the T-DNA cassette was standard for experiments of this type and therefore ACRE did not conclude that there were any further molecular aspects to be concerned about. Members noted that the insertion site was not characterised in molecular detail, but given the phenotypic analysis, and that this will be a small-scale trial where plants won't enter into the human food chain, ACRE concluded that in the case of this particular trial, additional data on molecular characterisation would not be helpful in addressing risk-based questions.

The Environmental Risk Assessment

ACRE concluded that its advice was broadly in agreement with Wild Bioscience and that it was highly unlikely that intended or unintended effects of the genetic modification would result in major changes to invasiveness or persistence. This was based on the phenotype reportedly seen by Wild Bioscience when these GM plants were grown under controlled environmental conditions. That is enhanced leaf-level photosynthetic carbon assimilation, increased vegetative biomass, along with an increase in number and total mass of seeds per plant. If Wild Bioscience were to apply for wide scale cultivation of these GM plants in the future, data from small-scale field trials on the comparative agronomic and phenotypic characteristics are likely to be required.

Wheat is naturally self-pollinating but under experimental conditions can be crossed with various wild grasses. The application discusses sexual compatibility with wild relatives present at the trial site. *Elytrigia repens* (common couch) is the only one of these commonly found on the Rothamsted estate and it is proposed to control it and other grasses and weeds in and around the larger GM trial site either by applying herbicides or hand pulling. No cereals or grass species will be allowed to grow within 20 m of the trial area itself. It should be noted that the applicant reports that no spontaneous hybrids between wheat x *Elytrigia* have been found. Public representations alluded to hybridisation occurring between cultivated and wild species when oilseed rape trials as part of the farm scale evaluation work carried out in the early 2000s. ACRE noted that these results illustrated the importance of greater isolation distances being specified for crops like OSR that can more readily outcross with wild relatives, than is probable with elite wheat cultivars such as Cadenza used here.

There is within the application an assessment of the likelihood of horizontal transfer of the gene cassette and specifically of the antibiotic resistance gene, along with consideration given to recombination with soil bacteria. Furthermore, a large proportion of public representations reflected concern that growing plants containing antibiotic resistant marker genes would compromise the use of associated antibiotics in human and veterinary medicine. ACRE has discussed the use of resistance marker genes in GM plants on a number of occasions and taken into consideration the statement from the European

Medicines Agency (EMA) on the importance of preserving the therapeutic relevance of the antibiotics.

ACRE emphasised that the *nptII* gene is present at high frequency in agricultural soils[2]. Antibiotic resistant bacteria occur naturally in the environment, but many are a result of contamination with human and animal excreta in sewage, slurry and manure. Antibiotic resistance in humans and other animals has resulted from the strong selective pressure associated with the substantial use of industrially made antibiotics in human and veterinary medicine and as food supplements for farm animals.

Public representations expressed concern about increasing antibiotic resistance. This is an increasingly well-studied area, with an increasing awareness that it is driven by amongst other things micro-organisms in waste effluents, and there is also evidence that this is more important than crop plant-driven sources. For example, Fonti *et al.* (2021)[3] analysed mobile genetic elements conferring antibiotic resistance in post-treated waste-water outflow into the Mediterranean Sea and found the associated presence of faecal indicator and pathogenic microorganisms. This research was conducted to inform risk assessments and hence improve the treatment regime carried out on wastewater. The authors also considered what the presence of these mobile genetic elements alongside a diverse set of microorganisms meant for risk assessments related to increased antibiotic resistance in the latter, for example when later isolated from the marine environment.

By contrast there are well developed arguments in relation to the consequences arising in, for example, the issue of the possible risks of transfer and subsequent expression of antibiotic resistance genes used as selective markers from GM plants into bacteria. This relies on extensive experimental data and has firm theoretical underpinnings from which it can be concluded that transfer of antibiotic resistance, and other genes, does not exist to any significant level with plants or animals where gene flow is controlled by strict rules on crop separation and by biological barriers to crossbreeding.

To add to the above, ACRE gave the following advice on plant to bacterial gene transfer in a previous field trial application:

Even though the scientific consensus is that selection pressure on bacteria containing antibiotic resistance genes is the driver of antibiotic resistance gene frequency in the environment, ACRE discussed the potential for bacteria in the environment to be transformed with antibiotic resistance genes from the gene edited wheat plants. Studies of horizontal gene transfer from plants to bacteria suggest that this phenomenon is extremely

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[2] Walsh F, Duffy B (2013) The Culturable Soil Antibiotic Resistome: A Community of Multi-Drug Resistant Bacteria. PLoS ONE **8**: e65567.

[3] Fonti, V.; Di Cesare, A.; Šangulin, J.; Del Negro, P.; Celussi, M. Antibiotic Resistance Genes and Potentially Pathogenic Bacteria in the Central Adriatic Sea: Are They Connected to Urban Wastewater Inputs? (2021) *Water*, **13**, 3335. https://doi.org/10.3390/w13233335

rare (Please refer to a review by Keese, 2008[4]). ACRE noted that even if a recombination event were to occur between DNA from a plant and a bacterial genome, in order for the gene to be expressed, it would need to be combined as a fully functional transcription unit in the bacterium, which is unlikely. If it were to occur, it would most likely result from a homologous recombination event at a site in the bacterial genome where a version of antibiotic resistance gene already exists.

A number of public representations referred to a paper about the relatively high levels of a synthetic antibiotic resistance genes detected in Chinese rivers, which the authors (Chen *et al.* 2012[5]) attributed to improper disposal of laboratory waste. By way of contrast, LaPara *et al.* (2015)[6] did not detect any of these genes in wastewater effluent or river water samples from the upper Mississippi River in the USA. The authors attribute this to stringent regulations on destroying laboratory waste containing recombinant DNA being followed. The UK's Genetically Modified Organisms (Contained Use) Regulations 2014, apply to the use of plasmids with antibiotic resistance genes under laboratory conditions and address the management of waste.

ACRE were also concerned about the repeated use of the following phraseology in **Part A4** under the Environmental risk assessment. Beginning on page 1 of that section the application states:-"If out-crossing to plants outside the trial area where to somehow occur, selection pressure to maintain the genes in the environment would exist only where kanamycin-based herbicides were used." ACRE commented that this phraseology was of concern, because there is no such product as a kanamycin-based herbicide, meaning that this is not a realistic comment. The same comment is made later in the risk assessment Table in steps 3, 4 and 5 under the headings of **Increased invasiveness in natural habitats or persistence in agricultural habitats due toinserted trait** and **Selective advantage or disadvantage conferred to wheat or other sexually compatible plant species.** In Step 5 of the same response, it is also stated that "No antibiotics will be applied to the soil to provide additional selection pressure for the gene to persist in the environment. "This same comment is made in the next response on page 8:- "No antibiotics will be applied to the soil to give selective advantage."

Public representations, as mentioned above, for this application concerned themselves with the presence of antibiotic resistance genes in these GM plants under consideration for release in field trials. Therefore, ACRE wished that the application be revised such that it more appropriately and clearly highlighted that under no circumstances would anyone

[4] Keese P. (2008). Risks from GMOs due to horizontal gene transfer. Env Biosafety Research. **7**(3): 123 – 149

[5] Chen, J.; Jin, M.; Qiu, Z.-G.; Guo, C.; Chen, Z.-L.; Shen, Z.-Q. Wang, X.-W.; Li, J.-W. A survey of drug resistance bla genes originating from synthetic plasmid vectors in six Chinese rivers. Environ. Sci. Technol. 46: 13448−13454.

[6] LaPara, T.M., Madson, M., Borchardt,S., Lang, K. S and Johnson T. J (2015). Multiple Discharges of Treated Municipal Wastewater Have a Small Effect on the Quantities of Numerous Antibiotic Resistance Determinants in the Upper Mississippi River. Environ. Sci. Technol. 49: 11509−11515.

seriously consider applying antibiotics under such conditions, so there is effectively no selection pressure. Furthermore, it would suffice to cover off only once in the text the fact that antibiotic-containing herbicides are yet to be formulated as they are unlikely to be efficacious under field conditions (and possibly also prohibitively expensive).

Wild Bioscience provided the following correction to replace the phraseology mentioned above:

“Kanamycin is not used in herbicide formulations and as such there are no realistic situations in which kanamycin antibiotics would be applied in field conditions. Practically speaking this means that there would be no selection pressure applied to maintain these genes in the environment.”

Managing the trial site

ACRE has considered the potential risks of this trial to human health and the environment in the context of it being a small-scale trial from which no material will enter the food or feed chains. The committee considered, in detail, management plans to minimise the persistence of GM material at the trial site and the dispersal of GM material from the site. ACRE recognised that the proposed trial was similar in size to that a previous wheat trial in terms of area, and therefore a 3 m pollen barrier + 20 m isolation distance was adequate to minimise the probability of out-crossing to an acceptable degree.

The relatively small scale of the trial is reflected by the fact that GM plants are being planted by hand as seedlings into the release site and that harvesting will also be undertaken by hand. A wheat pollen barrier will be in place that is designed to flower at approximately the same time as the GMO crop, ACRE acknowledge that this is not an exact science. However, the trial is further contained by a surrounding 20 m isolation distance and the probability of crossing with wild species particularly *Elymus* and *Elytrigia* is very low. The GM plants are susceptible to a wide range of herbicides and therefore, if necessary, it will be straightforward to kill off the GM plants. ACRE considered that the post-harvest processing protocol was robust, and that the described trial management procedures reflected the level of experience that the Rothamsted farm staff have in handling GM trials.

Gene flow

Wheat is a self-pollinating crop with very low rates of cross-pollination with other wheat plants. This is because fertilization often occurs before the florets open, which makes out-crossing unlikely; in addition, wheat pollen is relatively heavy and tends to travel shorter distances than pollen from other grass species that are wind-pollinated. Studies have detected cross-pollination rates of 1–2% between wheat plants in close proximity, but this rapidly decreases with the distance between plants. There are several relevant studies

involving GM wheat field trials, most recently those of Foetzki *et al*. (2012)[7] and Miroshnichenko *et al.* (2016)[8].

The maximum area for the proposed trial each year will be 500 m^2 total area including controls and intra plot spacing. (c.100 m^2 of this will be GM plants comprising 32 plots each of 3m x 1m). There will be 0.5 m separation between plots and a wheat pollen barrier of 3 m width entirely surrounding the trial plots, with a further 20 m surrounding that, in which no cereals or grass species will be left to grow.

ACRE noted that the separation distance required to prevent hybridisation between different wheat varieties when certified seed is produced for marketing purposes is 2 metres. The application proposes to sow a 3-metre-wide wheat pollen barrier (comprising the same variety as the GM wheat) around the trial. ACRE recommended a 2-metre-wide pollen barrier in its advice on previous GM wheat trials as this is an additional precautionary measure to the 20-metre separation distance. But, as this trial is the first such GM release applied for by Wild Bioscience, ACRE is content with the greater depth of pollen barrier in this case to further reduce the probability of any unacceptable gene flow. In order to maintain the separation distance, ACRE advises that the 20 m surrounding the trial site is planted with a non-cereal crop and that cereal volunteers are controlled (prior to flowering) in this area during the trial and for two years afterwards.

ACRE members considered that in terms of the pollen barrier, the key was timing to make sure both the experimental crops and the pollen barrier crop were at the same stage of development. That can be difficult if one is looking at experimental seed that does not have all the characteristics and stability of a commercial variety. The committee concluded that, in their view, if synchronisation proves difficult, then the 20 m separation distance would be an acceptable risk mitigation.

The applicant does not describe the need to move the specific site around within the bounds of the Rothamsted GM field trial site to allow post trial monitoring and to avoid the effects of take-all disease on the plants. However, this is likely to be required and therefore, the location of the specific growing site will require careful consideration to ensure that the 20 m isolation distance remains within the bounds of the Rothamsted site as a whole.

Wheat plant volunteers

The trial will receive standard farm practice as regard to herbicides, fungicides, nitrogen,

[7] Foetzki A., Diaz Quijano C., Moullet O., Fammartino A., Kneubuehler Y. and Mascher F. (2012). Surveying of pollen-mediated crop-to-crop gene flow from a wheat field trial as a biosafety measure. GM Crops and Food: Biotechnology in Agriculture and the Food Chain **3**(2), 115–122.

[8] Miroshnichenko D., Pushin A and Dolgov S (2016). Assessment of the pollen-mediated transgene flow from the plants of herbicide resistant wheat to conventional wheat (*Triticum aestivum* L.). Euphytica **209**:71–84.

sulphur and other fertilisers.

The site will be monitored regularly (at least weekly) both during and for two years after the trial. For the Post- trial monitoring period, the trial area will remain in stubble to enable monitoring of volunteers. The applicant does mention shallow cultivation being carried out to encourage volunteers but does not specify when this will be done. ACRE advice on previous trials has been lightly till twice (once after harvest and again in the following spring) to a depth of 5cm to stimulate germination of any wheat plant volunteers. The persistence of such volunteers from winter wheat in cultivated soil has been studied for a long time and is well-characterised.[9,10]

In common with previous GM wheat field trial applications, public representations have raised the issue of volunteers found by USDA APHIS in three states some years after trials had been carried out. There had been more than 100 large-scale trials conducted across 16 states, and although USDA's investigations were inconclusive, it was as previously discussed by ACRE, most probably due to the persistence of volunteers that GM seed had mixed with other seed and in this way become spread to other fields. Interestingly, the presence of volunteers was preceded by field trials for which the USDA asked for notification of; but did not set regulatory conditions. The finding of the afore-mentioned volunteers prompted the US authorities to review this process and put in place monitoring requirements that are comparable to those adopted here in the UK and in the EU for such trials. No further volunteers have come to light since this change was put in place, that is from 2014 onwards.

There are several relevant publications, of which the most detailed are two specifically designed to consider longevity of spring wheat in the seed bank in the context of GM (Kristi *et al*. 2007[11] and Ryan *et al.* 2009[12]). These studies conclude that survival of buried seed beyond the next spring is extremely rare and longer-term persistence in a field is most likely to occur from seed produced from volunteers that escape detection in the following season and then set seed. This conclusion is supported by the more recent study by Kalinina *et al*. in 2015.[13]

This trial proposed by Wild Bioscience is on a very small scale and has a number of

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[9] Chepil W.S. (1946) Germination of Weed Seeds I. Longevity, Periodicity of Germination, and Vitality of Seeds in Cultivated Soil. Scientific Agriculture **26**: 307–346.

[10] Anderson, R. L. and G. Soper. 2003. Review of volunteer wheat (Triticum aestivum) seedling emergence and seed longevity in soil. Weed Technol **17**:620–626.

[11] Kristi A. De Corby, Rene C. Van Acker, Anita L. Brûlé-Babel, and Lyle F. Friesen (2007). Emergence Timing and Recruitment of Volunteer Spring Wheat**.** Weed Science **55**(1): 60–69.

[12] Ryan L. Nielson, Marc A. McPherson, John T. O'Donovan, K Neil Harker, Rong-Cai Yang, and Linda M. Hall (2009). Seed-Mediated Gene Flow in Wheat: Seed Bank Longevity in Western Canada. Weed Science **57**(1): 124–132.

[13] Olena Kalinina, Simon L. Zeller, Bernhard Schmid (2015). Persistence of seeds, seedlings and plants, performance of transgenic wheat in weed communities in the field and effects on fallow weed diversity. Perspectives in Plant Ecology, Evolution and Systematics **17**: 421–433.

measures, including post trial monitoring, to ensure that any volunteers are detected and removed.

Seed movement

ACRE were content with the applicant's outline of how the release will be monitored regularly during all stages of development and harvested at maturity. Some seeds from the GM and control plots will be conditioned, threshed and stored in appropriate GM seed stores. A sample of plants may be hand-harvested, conditioned and threshed to supply seeds for research purposes. All such small samples removed from the trial site will be stored in containment prior to use and will eventually be autoclaved before disposal. The remainder of the site will be harvested by the plot combine.

Grain that is not required for analysis or to provide seed for future trials and all other material, including that from the pollen barrier rows, will be disposed of by incineration, autoclaving, or deep burial at a local authority-approved landfill site using an approved contractor, while any material remaining after analysis will be autoclaved before disposal. Transportation of waste materials will be in secure containers. All straw will be chopped and left on site. The combine will be cleaned prior to leaving the site so that all traces of plant material from the trial will remain in the trial area. All transport of material will be logged.

Other items arising from public representations

Some 80 public representations were received, where these covered areas within the remit of ACRE they were addressed within its assessment, as summarised above. Many of the representations concerned areas beyond the remit of ACRE, and/or these topics are not relevant to the environmental risk assessment when considering a small-scale field trial. Some of these comments would be of relevance if the application had been for commercial-scale cultivation and/or food and feed use. Others were more political in nature and are outside of ACRE's remit; for example, they questioned whether GM techniques should be used in the development of plant varieties and more specifically whether it is necessary to develop wheat with enhanced photosynthesis and productivity using this technology.