

Guidelines on containment and control measures for the use of GMO plants in Plant Growth Facilities - Greenhouses/Growth rooms and Growth cabinets

Background

Article 35 of the Genetically Modified Organisms (Contained Use) Regulations, S.I. No 73 of 2001, requires GMO users *to apply principles of good greenhouse, growthroom practice ...including such principles (if any) as may be specified or approved by the Agency*. Herewith find guidelines for users who plan to work with GM plants in greenhouses, growthrooms, growth cabinets etc.

Legislative requirements for GM plants under the GMO (Contained Use) Regulations S.I. No 73 of 2001

Part III of the GMO (Contained Use) Regulations S.I. No 73 of 2001 makes reference to Genetically Modified Organisms (GMOs) other than Genetically Modified Micro-organisms (GMMs). Contained use under Part III of S.I. No. 73 of 2001 is defined as *any activity in which organisms other than micro-organisms are genetically modified or in which such organisms are cultured, stored, transported, destroyed, disposed of or used in any other way and for which specific containment and other protective measures are used to limit their contact with the general public and the environment*. GM plants include all reproductive organs including seeds, pollen, bulbs, tubers etc.

In addition, the onus is on the user carrying out an activity involving a contained use, to ensure that all appropriate measures are taken to avoid adverse effects on human health or the environment in accordance with article 5 of the GMO (Contained Use) Regulations S.I. No 73 of 2001

It should be noted that containment measures for the use of Genetically Modified Micro-organisms (GMMs) in plant growth facilities are not covered by this guidance but are regulated in accordance with the Fourth Schedule of S.I. No. 73 of 2001, in particular, Table 1B thereof.

GM plants that are placed on the market under Directive 2001/18/EC on the deliberate release of GMOs into the environment or similar product based Community legislation are not covered under the Contained Use Regulations - S.I. No. 73 of 2001.

General requirements for work with GM plants :

It should be noted that containment and control measures should be determined by the risk assessment carried out in accordance with article 36 and Part II of Seventh schedule of S.I. No. 73 of 2001. The environmental risk assessment (ERA) must be carried out on a case by case basis.

Good Greenhouse Growth room/cabinet Practices, Containment Level 1

We recommend that the following procedures should be adopted as Good Greenhouse/Growth-room Practice when working with GM plants in greenhouses, growth rooms and growth cabinets. These measures should also be adopted during experiments requiring Level 1 Containment.

Level 1 Containment will apply to the following

1. Plants with limited ability to transfer genetic material to indigenous plant species in Ireland. This could occur:

- through a lack of sexually compatible weed species or through artificial means, e.g., removing flower buds or bagging of flowers to minimise pollen transfer
2. Transformed plant cells - using a plant pest (e.g., *Agrobacterium tumefaciens*) where the plant pest is a disarmed strain.

Plant Facility

1. Greenhouses, growth rooms and growth cabinets should be of robust design and construction, to facilitate easy cleaning and withstand normal climatic conditions. Where the level of risk from pollen /seed dispersal is particularly high, polycarbonate glasshouses should be erected in preference to glass. Floors should be impervious.
2. Within the greenhouse/growth room/cabinet GM plants should be labelled with a Biohazard sign in addition to the nature of the genetic modification and the identity of the scientist responsible for the activity
3. A water source and sink should be available in an adjacent preparation area;
4. The glasshouse/growth room doors must be kept closed at all times and locked when unattended;
5. Staff must maintain a high standard of cleanliness and hygiene and suitable protective clothing must be worn by all staff entering the glasshouse growth room/cabinet.
6. Protective clothing should comprise a laboratory coat. Overshoes or dedicated shoes should be worn in the glasshouse growth room/cabinet when it is necessary to minimise or prevent the release of the GM material
7. Eating, drinking, smoking, storing of food for human consumption and applying cosmetics must not take place with the glasshouse growth room/cabinet

Containment

8. The potential for transfer of genetic material should be minimised in accordance with the Risk assessment. Screens should be erected over doors and windows in order to prevent entry of pests, rodents, birds, insects, etc.
9. Personnel should receive instruction on the procedures to be observed in the glasshouse/growth rooms/cabinets in order to minimise release of the genetically modified material and the development of disease;
10. Floor drains should be filtered to minimise seed dispersal or to prevent invertebrate/vertebrate ingress. Filters should be inactivated by validated means prior to disposal.
11. Pest and disease control measures must be adopted and maintained;
12. Standard Operative Procedures (SOPs) must be drawn up for the following :
 - Training of new staff
 - Treatment and clean-up of spillages
 - Cleaning of equipment
 - Transport of GMOs
 - (a) within the greenhouse/growth room/growth cabinet/building,
 - (b) from the laboratory to the greenhouse/growth room/growth cabinet
 - Operation, testing and maintenance of containment equipment
 - Entry /exit procedures to Glasshouse/ growth room/cabinet and measures for limiting access to the GMO facility
 - Disposal of GMO waste

13. The risk assessment procedure should consider any possible effects on non-modified plants, (e.g., cross-pollination) grown in the same facility at the same time. Segregation of plant material may be required.

Waste

14. An autoclave must be available on site for the treatment of greenhouse/growth room waste material. All pots, trays and glasshouse mix (compost) must be sterilised prior to cleaning or disposal. Soil and growth media must be sterilised on site by validated means before disposal to kill any residual organisms;
15. If autoclave is situated at a site remote from the Greenhouse/Glasshouse the waste material must be transported from the Glasshouse Growth room/cabinet in a labelled, non-breakable, durable, sealed container

Containment Level 2

Level 2 Containment will apply to the following

1. Genetically modified plants that can transfer genetic material to indigenous species in Ireland;
2. Plant species that are potential pests.

The requirements of Good Greenhouse/Growth room/Cabinet Practices should be followed in all GMO premises requiring containment level 1. The Agency recommends that the following **additional requirements** be implemented for containment level 2:

1. Access to the Glasshouse/growth room/cabinet must be restricted to named personnel. A list of those designated personnel must be posted at the entrance to the facility. Entry should be via a key pad/electronic system.
2. Protective clothing, pots, tools, equipment etc. must be decontaminated by a validated means after use and must be bagged within the facility and removed therefrom in labelled, closed, durable, non-breakable containers.
3. A record must be kept of all experimental plants and micro-organisms present in the greenhouse/growth room/cabinet comprising the nature of the modification, the responsible scientist, commencement/termination dates and how disposal of any residual material was achieved
4. The transfer of material containing genetically modified organisms from the laboratory to the greenhouse/growth room/cabinet and/or from the greenhouse/growth room/cabinet to the autoclave must be achieved in labelled closed durable non-breakable containers in order to prevent the dissemination of viable GM material
5. Where pollen and seed borne dispersal presents a potential hazard, pollination and seed formation must be prevented, however, where yield data is required or is an object of the study, pollination/seed dispersal would need to be considered on a case by case basis.
6. Where waterborne and/or soil borne dispersal presents a potential hazard all effluent must be treated by chemical inactivation or other validated means
7. The application of differential pressure between the Glasshouse, growth room/cabinet and adjacent areas (such that there is an inward airflow) and the filtration of exhaust air must be employed where airborne dispersal presents a

potential hazard. The filter must be inactivated by validated means prior to disposal.

8. The use of floor drains should be avoided. Where they are deemed necessary they should be fitted with a disinfection trap that will render any discharged material non-viable.
9. Disinfectants must be available for disinfection;
10. The growing of host or related plant species in the vicinity should be restricted. The area outside the greenhouse/growth room should be monitored for escapes etc;

Pollination and Seed Borne Dispersal

11. Where it is necessary for pollination and seed formation to take place the following control measures should be adopted in addition to those referred to under Containment Levels 1 and 2
 - exclusion of pollination vectors
 - bagging of plants to prevent uncontrolled pollen dispersal
 - use of male sterility
 - Transformation by chloroplast genome to prevent transfer by pollen
 - Alteration in time of flowering so that no genetically compatible plants are simultaneously flowering in the environment
12. In addition to wearing overshoes or dedicated shoes sticky mats to trap adherent material should be employed

Pollination and Seed Borne Dispersal via invertebrate vectors

13. Where unintended dissemination of pollen by an invertebrate vector presents a potential risk the following containment measures should be employed in addition to those referred to under containment Level 1 and 2
 - Doors, air inlets and outlets must be covered with one or more mesh and have brushes around the edges, especially the bottom. Differential pressure must be employed and management procedures must be strictly observed.
 - A pesticidal regime must be implemented
 - If feasible plants should be grown when the vector is not present in the environment