14.0 PATHOGEN AND INSECT USE POLICY

Recommended Guidelines for Pathogen and Insect Use
at the Boyce Thompson Institute
BTI Pathogen Use Committee  4/7/04

1. The Plant Growth Facilities Committee will review all pathogen and insect use in the BTI Plant Growth Facility. A written request for use of new pathogens or insects must be submitted by the Principal Investigator (PI) to the Greenhouse Manager and the Plant Growth Facilities Committee prior to use in the BTI Plant Growth Facility. The PI will be asked to complete a Pathogen Use Form or Insect Use Form (see Appendix A) to be reviewed by the Plant Growth Facilities Committee. If the PI is proposing to work with recombinant pathogens, he/she will be asked to submit a current rDNA MUA and Greenhouse/Growth Chamber Manual approved by Cornell’s Institutional Biosafety Committee (IBC), which will be kept on file in the Greenhouse Office.

2. Each request for use of a new pathogen or insect will be reviewed by the Plant Growth Facilities Committee, and space will be allocated to the laboratory by the Greenhouse Manager. If a pathogen or insect is considered “high risk”¹ by the Plant Growth Facilities Committee, the Greenhouse Manager will attempt to find space within the BTI Plant Growth Facility that can be shared with other labs already using the pathogen or insect successfully under strict containment measures (e.g., Tobacco Mosaic Virus or TMV). Alternatively, the Greenhouse Manager will attempt to locate space at Cornell that is already being used for the pathogen or insect in question and can be shared with BTI scientists.

3. Compliance with containment procedures to prevent pathogen or insect spread to non-target hosts is critical to keep the BTI Plant Growth Facility functioning optimally for all users. All lab members who work with pathogens or insects must understand and follow necessary containment measures for the pathogens or insects they use. Their signature on the Pathogen Use Form or Insect Use Form will indicate that they have read and agree to comply with all necessary containment measures as outlined.

4. Some general guidelines for pathogen and insect containment are as follows:
   - When possible, inoculate plants in the laboratory to keep high concentrations of pathogens or insects away from the BTI Plant Growth Facility.
   - Transport infected plants on designated carts, which may need to be covered prior to moving infected plants. Clearly mark carts that
are to be used only for designated pathogens or insects. After use, clean and decontaminate carts.

- Do not visit “clean” greenhouses or growth chambers after having gone to a pathogen-containing or insect-containing greenhouse or growth chamber the same day.
- The one-glove-rule applies in the BTI Plant Growth Facility as well as other BTI locations.

Pathogen Use Form
Insect Use Form

A. Standard Operating Procedure: Pathogen containment facility (PCF) – Room 216 BTI

I. Introduction

1. Background information on the facility
   a. Room 216 of the Boyce Thompson Institute (BTI) has been designated a plant pathogen containment facility (PCF) and contains four reach-in growth chambers.

2. The PCF is intended for use of plant pathogens whose use in the main greenhouse area is not permitted or undesirable. This includes pathogens for which there is a high risk of contamination of other plants, including (but not limited to), Phytophthora species, Hyaloperonospora parasitica, and tobacco mosaic virus.

3. Use of the PCF is open to BTI labs, subject to availability of space and review of intended use by the greenhouse committee. Application for review should be sent to the greenhouse manager using the BTI pathogen use form available on the BTI website (http://bti.cornell.edu/).

II. Physical Containment Standards

1. Description of facility
a. The PCF is located on the second floor of the BTI which is located on the campus of Cornell University, Ithaca, NY. The building is locked from 5:00 P.M. to 6:00 A.M. on weekdays and locked 24 hours per day on Saturday and Sunday. Access to BTI during “after hours” is controlled by a card reader.

2. Description of safeguards in each compartment of facility
   a. Room 216 is locked for 24 hours per day.

III. Equipment Standard
   1. Solid and liquid waste sterilization
      a. All waste from the PCF will be autoclaved using the Consolidated steam-supplied autoclave in room 214. The autoclave must be set at a minimum of 15 psi, 121 °C for 45 minutes. The proper sterilization of each autoclave load should be confirmed by including indicator or autoclave tape. A change in the tape color or markings will indicate that time, temperature, and the presence of steam have been adequate to ensure sterilization. All autoclaved material should be disposed of for landfill; do not return plastic pots to the greenhouse area; soil and plant material should not be sent for composting. Non-disposable materials should be sterilized as per the individual pathogen SOP.

   Problem associated with the Consolidated autoclave should be reported immediately to the Boyce Thompson Institute mechanic shop (Phone: 607-254-1201).

IV. Operation Standards
   1. Containment
      a. Responsibility for overseeing containment in the PCF rests with the following containment directors:

         Primary Investigator: Dan Klessig  
         Home ph: 607-756-8206  
         Office ph: 607-254-4560  
         Email: dfk8@cornell.edu

   2. Responsibilities of Containment Director
      a. The containment directors are responsible for training lab personnel in the proper handling of pathogens according to the guidelines of the SOP.

   3. Emergency Contacts
a. In case of an emergency (fire, flood, tornado, etc.) in which containment may be breached, contact Cornell University Environmental Health and Safety (ph. 911) or Cornell University Police (ph. 911) and inform Containment director(s) listed above as well as BTI building director Larry Russell (ph. 607-254-1359 [BTI], 607-220-4357 [cell], 607-589-4281 [home]) and David MacDonald (ph. 607-254-1201 [BTI], 607-220-4358 [cell], 607-589-6884 [home]).

4. Sanitation and Containment Procedures
   a. Individual pathogens should be handled according to specific SOPs. The greenhouse committee must be apprised of all new pathogen usage, to be accompanied with appropriate SOPs.
   b. All work should be performed within Cornell Institutional Biosafety Committee (IBC) guidelines for working with recombinant DNA and genetically modified organisms.
   c. No greenhouse equipment including carts should be used in the PCF.
   d. Plants may be transported to the hallway outside the PCF, and transferred to a cart specifically designated for the PCF facility.
   e. When working in the facility, personnel will be required to wear lab coat and disposable gloves. When work is completed, the gloves should be disposed of and autoclaved. Wash hands thoroughly with soap. Change gloves often. Do not touch door handles, control panels of chambers and autoclave, non-PCF carts, autoclaved waste, notebooks, lab equipment etc with gloved hands.
   f. Personnel should have a PCF-designated lab coat, to be stored in room 216 and not used outside 216 except to transport waste or to perform experiments in the laboratory. Contaminated lab coat should be sterilized in the Consolidated autoclave in 214 before they are given to the laundry service through the BTI stockroom.
   g. Personnel working in the PCF are not allowed to enter other plant growth facilities on the same day, and must bathe and launder clothing before returning to work the following day.
   h. Pathogen infection of plants should be performed inside of the designated growth chambers in room 216 whenever possible, such as for infection with TMV. For other pathogens infections, infections should be performed as per the specific SOP. Do not touch outside of the chamber and other surface of room 216 with gloved hands that have been used for infection. The chambers will be labeled with the pathogen(s) in use.
   i. All plant material, plastic pots and soil used in the designated chambers will be sealed in autoclavable plastic bags and sterilized in the Consolidated autoclave in room 214. The PCF cart should be used to transport waste to the autoclave in room 214 and immediately
be returned to room 216. Do not use the same cart to subsequently transport autoclaved waste.

j. The chamber should be cleaned and disinfected between experiments to avoid pest build-up and prevent contamination of future experiments. This includes cleaning and wiping down all surfaces as in (5. a).

k. Control pests in the growth chambers to avoid potential transmission of pathogens. In the event of discovery of pests, contact the Greenhouse manager for direction. Be aware that pesticides cannot be used in room 216 and the best strategy for control is preventative practices including frequent chamber clean-out.

5. Decontamination
   a. In the event that maintenance and repairs of equipment and structures within the containment area is required, all plant materials and tools will be removed from the area and secured in another area of the facility. The affected area will be decontaminated with a solution of either 40% household bleach solution, or 0.1N NaOH, or by irradiation with an ultraviolet lamp (2 W/m²) for at least 30 minutes or as prescribed by the SOP for the pathogen in use. Wear proper protection gears to handle bleach, NaOH, and ultraviolet.

   b. In the event of a spill of contaminated material either inside or outside the PCF, the affected area will be decontaminated as in (a).