Handling of Genetically Modified Microorganisms (GMM) at UPSC

We have permission from "Arbetsmiljöverket" to work
for floor 2, 3 and 4 and the "Odlingshus" and SLU for floor 5 and 6, plus the Populus transformation facility on floor 4. These instructions have to be followed; persons that violate the rules will not be allowed to perform work with GMMs. Group leaders are responsible that all persons working for him/her get this information and that the rules are followed.

1. We have a permit to work with these miroorganisms; E.coli, Agrobacterium, Chlamydomonas, Cyanobacteria and Yeast.

2. Each place (freezer, fridge etc.) where GMMs are stored must be labelled with a "GMM-sticker" and given an unique number (there is room for this on the sticker). – 70°C freezers will also get stickers and numbers.

3. Each person working with lab work must keep a UPSC laboratory notebook.

4. Each new GMM has to go through a risk evaluation. What should be evaluated is whether the GMM results in properties that may make the organism harmful to humans, animals, plants or the environment. Wt Agribacterium could be harmful to plants so modifications that does not make it more harmful is not a problem. If you think that there is a possibility that your GMM could be more harmful than the "standard" GMMs in the lab, discuss this with the "GMO/GMM officers".

5. The person working in the lab should make the risk evaluation, and make a note on this in the notebook. Also, it must also be noted where (e g in which GMM-labelled freezer) the GMM is stored. The PI in charge of the research should regularly (e g every week) countersign the notebook to approve the risk evaluations.

6. All solutions containing GMM must be sterilized. This should be done by addition of a sufficient amount of Jodopax, shaking and incubation for at least 30 minutes. The solution can the go into the sink and glassware for normal dishwashing. "Sufficient amount" is addition a 1:100 dilution of 5.2 % Jodopax. You could e g prepare a 20X dilution and add 1/4 volume of that to a bacterial culture.

7. Disposable material that has been in contact with GMM (plates, tips, tubes etc.) should be disposed in "Riskavfallskartonger", clearly labelled with the 2 stickers; UN3245 and Class 9 label. Seal the plastic bags with cable ties, close the lid and seal it with package tape. The boxes should be deposited in a designated room in "Miljörummet" close to "Vaktmästeriet" where they will be picked up for destruction. Remember to sign on the outside of the GMM door.

8. Before you start to work with any other GM organism than mensioned above, you have to consult with the "GMO/GMM officers" Ove Nilsson (SLU) or Stefan Jansson (UmU, GMO) and Thomas Hiltonen (UmU, GMM).

HOW TO PERFORM A GMM RISK EVALUATION

The two UPSC departments have permits to perform research with Genetically Modified Microorganisms (GMMs) as long as the activities are of the "F-class".
"F-activities" are defined as posing "no or negligible risk to harm the health of humans or to harm the environment".
A typical "F-class GMM" has the following properties:

- It is not likely that the receiving organism can cause disease in humans, animals or plants.
- The insert and the vector will not give the GMM such new properties that it is likely that it can cause disease in humans, animals or plants or otherwise harm the environment.
- The resulting GMM will have such properties that it is unlikely to cause disease in humans, animals or plants or otherwise harm the environment.

The effects in question are those that are possible in the environment where the GMM will be in contact in case of an unintentional release.

The two departments have performed a risk evaluation which is "typical" for the kind of F-type GMM activities that are performed. This risk evaluation concerns a disarmed strain of *Agrobacterium tumefaciens* carrying binary vectors with antibiotic selection markers such as hygromycin, kanamycin, carbenicillin or gentamycin and also a plant gene with an eukaryotic-type promoter for subsequent expression in a transgenic plant. All risk evaluations should be compared to this "type evaluation" and if the GMM in question might pose a higher risk to any of the points mentioned above, the GMM safety officers (currently Ove Nilsson, SLU and Thomas Hiltonen, Plantphys) must be consulted before the GMM is constructed.

Points to consider when doing the risk evaluation:

1. First the biological material has to be evaluated for potential harmfulness. Possible harmful effects (see section 3) associated with the donor organism and the receiving organism has to be identified. Also possible harmful effects associated with the vector or the inserted genetic material has to be considered.

2. The potential harmfulness of the GMM which is the result of the genetic modification has to be evaluated. Harmful effects caused by recombination or other changes of the genetic material has to be identified. In the final evaluation one has to consider the potential harmful effects identified in section 1 above (for instance to what extent these potential harmful effects can be expressed in ht e new construction) and the potential new harmful effects that can be caused by the resulting GMM. To see if the risk evaluation is realistic, it should be compared to the "typical" risk evaluation mentioned above. If there is any indication that the resulting GMM might pose more risk than the "typical GMM" described in the "typical risk evaluation" the GMM safety officers should be consulted before construction of the GMM.

3. The following are considered to be potentially harmful effects:

a) Human disease, including infection, effects causing allergies or toxicities and the potential to cause cancer. Also how serious or transmittable these diseases or harmful effects are should be considered.
b) Animal or plant disease.
c) Harmful effects caused the by the fact that a disease is non-treatable.
d) Harmful effects caused by harmful compounds formed by the GMM.
e) Harmful effects caused by establishment or spread to the environment.
f) Harmful effects caused by a natural transmission of the introduced genetic material to other organisms.

After doing the risk evaluation, and if you find that the GMM that you are going to construct clearly falls within the departmental permits, you should check the relevant box at the bottom of the page in your UPSC laboratory notebook and indicate where the GMM is stored.

If you have any questions regarding the risk evaluations, please ask Ove Nilsson (SLU) or Thomas Hiltonen (Plantphys).

How to handle GMMs at UPSC

In the lab where you work with GMM, make sure that you have Jodopax. Veterinary Jodopax is avaliable at Kemiförrådet.

It is convenient to have a special vial on each lab-bench where tips etc. can be disposed. Biohazard labels can be found i the GenomicsLab.

It is practical to have a container with diluted Jodopax (0.05% working concentration) in where you add bacterial contaminated growth medium. An alternative to add Jodopax to each bottle with GMM solution.

GMM contaminated Waste (plates, epptubes, etc) is deposited in “Riskavfallskartonger”. Do not throw other garbage than GMM contaminated stuff. When the box is full, label the box with the 2 stickers; UN3245 and Class 9 label. The stickers can be found in the staircase room on floor 4.

The correctly labeled boxes should be deposited in a special room. To find this, you go into the KBC house on Floor two, and turn left, and pass the “stone collection” in the corridor under the “KBC fik”.

At the end of the corridor turn right to get into the corridor for “Kemiförrådet” and “lastbryggan”.

Turn immediately right to get to the GMM room.

and place your GMM waste in the designated coldroom and sign on the paper on the door.

If any doubt, ask Thomas Hiltonen or Ove Nilsson. In their offices, there are binders with rules, our permits etc.