University Occupational Health and Safety Guidance Note

UNDERTAKING A RISK ASSESSMENT FOR GENETICALLY MODIFIED MICROORGANISMS

The SACGM Compendium of Guidance Part 2 provides comprehensive guidance on the risk assessment of work activities involving GM microorganisms. This should be used in conjunction the guidance below when completing the GM Risk Assessment Part 1 to inform the classification of the GM project.

Sections (a) to (c) of the GM Risk Assessment should be completed before completing the remaining sections, using the following guidance.

(d) Identify the hazards associated with the GMM

For GMMs that are primarily a potential risk to human health, a detailed risk assessment for human health can be carried out first and a provisional containment level set based upon human health protection.

For GMMs that are primarily an environmental concern, a detailed risk assessment for the environment can be carried out first and a provisional containment level to prevent harm to the environment set. Consider the hazards of the recipient strain, the vector and the inserted material.

Consider:

- Hazards associated with the recipient strain Recipient Strain: Refer to the Advisory Committee on Dangerous Pathogens (ACDP) Approved List of Biological Agents and Specified Animal Pathogens Order (SAPO) Guidance.
- Hazards associated with genetic inserts
    - Hazards arising from the alteration of existing pathogenic/infectivity traits
    - The inserted gene encodes a pathogenicity or virulence determinant
    - The modification affects the infectivity or virulence of the host organism
    - The modification alters susceptibility to the immune system
    - The modification alters tissue tropism or host range.
    - The modification alters the susceptibility of the organism to prophylaxis
- Transfer of harmful sequences between organisms
- Sequence mobilisation in bacteria.
- Recombination between related viruses
- Reassortment between segmented RNA viruses
- The ability of GMM to become established in the host
- The probability that rare events will occur
- Potential mechanisms by which the GMM might pose a hazard to the environment
- If the modification alters stability or survivability.

(e) Are you confident that for all of the GMMs covered by this assessment there are no harmful properties associated with the recipient strain, the vector, or the inserted material? (No or negligible risk)

Select ‘Yes’ only if the GMM poses **NO** or **NEGLIGIBLE** risk to humans or the environment.

(f) Are you confident that none of the final GMMs could be hazardous to humans or the environment? (No or negligible risk)

Select ‘Yes’ only if the GMM poses **NO** or **NEGLIGIBLE** risk to humans or the environment.

(g) Consideration of the nature of the work to be undertaken and a detailed review of the control measures.

When classifying the GMM consideration requires to be given to the parental organism and the particular genetic modification, insert, deletion, disablement or other modification which produces the new GMM. The parental organism will have been assigned an ACDP categorisation based on their ability to cause infection, severity of resultant disease, the risk of the infection spreading to the community and whether any prophylaxis or treatment is available. Therefore dependant on the actual genetic modification the classification of the GMM may alter and the above categorisation criteria must be considered to appropriately give an informed decision on the GMM classification.

(i) Are any of the work procedures likely to generate aerosols?

Consideration should be given to the likelihood of the work activity generating infectious aerosols. Where this is the case work should not be undertaken on the open bench but in an appropriate mechanically ventilated environment such as a Microbiological Safety Cabinet or where animal work may be involved an isolator.

Not all micro-organisms present an inhalation risk however, dependant on the parental organism and the resultant GMM consideration must be given to the potential that infection could arise from exposure to the GMM when working on the open bench. Where this is the case then the work should be carried out in a microbiological safety cabinet or if using the GMM in animal work then an isolator may be required. In addition, consider whether a certain titre would require to be reached before this would become a risk? (See section ix.)

If a microbiological safety cabinet is being used solely for the purpose of material sterility and not user protection, this should be stated.

(ii) Will it be necessary to use sharps?

Does work involve glass Pasteur pipettes, needles? If so, could they be substituted with safer alternatives?

One of the main risks in any laboratory environment is working with sharps eg, hypodermic needles, glass pipettes, graduated and Pasteur pippettes, as well as general laboratory glassware. In the case of glassware the hazard can be eliminated by using plastic alternatives in the majority of cases. Therefore, as part of the risk assessment process you should consider safer alternatives where practical taking into account the risks associated with a sharps injury should one occur and an operator being exposed through a cut or puncture wound to the GMM. If the sharp is not substituted for a safer alternative then appropriate safe systems of work, information, instruction and training play a vital role and you should detail how this will be managed.

(iii) How will waste materials be disposed of?

Include both solid and liquid laboratory waste and waste from experiments with infected animals.

There are two principle methods of treating contaminated material and waste, through chemical inactivation and autoclaving (or both). Any waste consisting of or containing viable GM material must be considered as part of the risk assessment, specifying the type and form of waste(s) generated, their treatment, their final disposal. An indication of the numbers of viable GMOs remaining after treatment (if any) should be provided.

Solid contaminated material or waste may be in the form of:

- Sharps e.g. needles;
- Plastics e.g. tissue culture flasks, pipettes;
- Contaminated equipment e.g. centrifuge buckets used for cultures or contamination from a spillage in e.g. a centrifuge rotor;
- Contaminated material used to absorb a spillage; or
- Possibly animal bedding contaminated with viable GMM where the animal excretes these in urine, faeces or other body fluids.

Disinfection

Dependant on the solid waste it may undergo chemical inactivation, sterilisation via autoclaving or both. The chemical disinfectant used must be validated against the parental micro-organism which is being utilised to create the GMM and through the risk assessment process you must

consider if the genetic modification makes the resultant GMM potentially more resistant to inactivation by the disinfectant.

Disinfectant choice should be determined by:

- The general type or identity of agents for which the disinfectant has demonstrated efficacy;
- The presence of protein or other substances likely to reduce efficacy or be chemically incompatible with the disinfecting agent; and
- The pH and temperature of the waste that are compatible with safe disinfection.

Contaminated items should be completely immersed in liquid disinfectants taking care to prevent air bubbles forming. Intimate contact must be achieved between the disinfectant, and the waste or contaminated surface for a sufficient length of time. Oil and grease residues on surfaces may prevent effective contact with the disinfectant.

Written standard operating procedures for disinfection should be in place and should specify:

- The wastes and contaminated articles that are to be disinfected, eg disposable or reusable articles that are heat sensitive, liquid wastes and effluents other than cultures;
- The disinfectant that is to be used, its use-dilution, final concentration, contact time
- How often the disinfectant should be changed;
- The contact times to ensure inactivation;
- The methods for routine or occasional validation of the disinfection process;
- The safe disposal of used disinfectants and the need for decontamination of containers: and
- The means for the safe removal and disposal of treated waste.

The solid waste or contaminated material may require to be immersed in the disinfectant and if this consists of equipment then you should consider whether the disinfectant may degrade it.

Disinfectants predominately are in liquid form but others are useful in that they are produced in powder form e.g. Virkon. Therefore, the powder can be added to e.g. a tissue culture vessel at a specified weight/volume (w/v) to provide a resultant working ie final concentration. Once this has been left for the specified contact time, for ease this is usually overnight, then the liquid waste can be disposed to drain and the tissue culture vessel can be discarded into waste for disposal via the Clinical Waste Service.

Remember small volumes can hold more viable material than larger volumes therefore consider degree of kill. If use of the disinfectant confers a 10^5 log reduction and there are 10^{10} viable organisms in the original volume then 10^5 viable GMM's still remain in the contaminated material. You would them require to determine if this is as acceptable titre for disposal and would not confer a risk to humans or the environment.

Autoclaving

Autoclaves used for the inactivation of GM waste must be validated annually by 12-point thermocouple. Details of the autoclave cycle should be given (e.g. temperature and hold time).

Written standard operating procedures for autoclaving should be in place and should specify:

- The solid and liquid wastes that are to be autoclaved, eg cultures and media, sharps (if sharps bins are autoclaved prior to disposal through the Clinical Waste Service, they should be able to withstand the process), pipettes, other disposable and reusable articles, gloves and laboratory coats, paper towels and tissues;
- The containers (which allow steam penetration) that are to be used;
- The required sterilising cycle, eg temperature and time settings and cycles;
- Whether biological or chemical indicators are to be used and their location in the load;
- The unloading procedure;
- The checks to be made and recorded by users and others, eg maintenance staff; and
- The emergency procedure in the event of a malfunction or failure.

Following the autoclaving process, the waste should be placed in yellow clinical waste bags and disposed of via the Clinical Waste Service.

(iv) Have any disinfectants been validated under the actual conditions of use?

For example, if disinfectant is being used for the treatment of virus in tissue culture medium, is it known that the disinfectant is effective in the presence of high levels of protein?

Disinfection protocols are required to be in place for both routine use and for use in spills. As well as documenting the disinfection process, the protocol should record that the disinfectant has been assessed for its efficacy under in-use conditions. Efficacy may be determined by:

- Examining the manufacturers' literature;
- By examining the relevant peer-reviewed literature; or
- In-house testing.

This section should indicate the type of disinfectant, in-use concentrations and contact times that are suitable for the biological agents in use.

(v) If the work involves the experimental infection of animals is it known whether the animal will shed the GMM?

This section is only applicable if infecting an animal with the GMM. If this is the case then consideration must be given to whether the animal will excrete or shed viable GMM. For example, will the animal bedding be contaminated with viable GMM where the animal excretes these in urine, faeces or other body fluids?

(vi) Does the organisms multiplication involve a complex life-cycle where the work involves the propagation of organisms that are in stages in that life-cycle that are particularly hazardous?

Examples include the propagation of the infective stages of parasites or the release of spores from fungi. Consideration should be given to all potential routes of transmission including those that might not be used naturally.

(vii) Occupational Health

Does the nature of this work preclude it being undertaken by any workers who have a serious skin condition (e.g. eczema) or other health problems that might make them more susceptible to infection (e.g. some kind of immunological defect)? Will workers require to receive any vaccinations or health surveillance?

Intact skin under most circumstances acts as a natural barrier to prevent the entry of a micro-organism into the body however, where a worker has a skin condition, such as eczema, then their skin may not protect them from exposure to the GMM. Consideration then must be given to the whether this worker is at risk from infection when working with this organism. Good laboratory practice should ensure that any cuts are covered prior to entering the laboratory.

Personal protective equipment is the last line of defence in the hierarchy of control measures and may not always be the solution depending on the location of the condition, for example the face, and the potential for exposure in an accident or spillage.

In addition, is there a risk of entry through the mucous membranes such as the eyes?

Will pregnant workers be at an additional risk from the GMM therefore should they be precluded from working with the organism or entering the environment in which it is used?

The Occupational Health Service can be contacted for further advice.

(viii) What personal protective equipment (PPE) is required?

PPE is equipment including clothing, which is intended to be worn or is held at work and protects the person against the risks to their health and safety. In the laboratory this will range from laboratory coat, gloves, eye and face protection to respiratory protective equipment. Consider the risks from the GMM, the tasks being undertaken, and the equipment used to determine PPE required.

It must be remembered that PPE in the hierarchy of controls is the last line of defence and other considerations such as elimination, substitution or engineering controls, such as the use of a microbiological safety cabinet may also be required to prevent exposure to the GMM.

PPE is used as a minimum requirement in some laboratories, it is important to state whether PPE being used is for user protection in relation to the GM work being undertaken.

(ix) Are there contingency arrangements should a spillage occur? Please specify what procedures are in place.

Detail how a liquid spill would be decontaminated, and any affected surfaces cleaned. Include information on procedures should a user be affected by a spillage.

When drawing up contingency plans a number of different factors/scenarios will need to be considered to determine the most appropriate course of action:

- Type of agent - the classification of the GMM, route of transmission, infectious dose (if known), stability in the environment.
- Type of accident - instantaneous or delayed - for example, a dropped flask as compared to a broken centrifuge tube which may be undiscovered until the centrifuge is opened.
- Severity of accident - amount and concentration of material that could potentially be released and form, for example, is aerosol formation likely?
- Numbers of staff potentially exposed - this may depend on location of accident.
- Location within the laboratory - an accident in the open laboratory may require evacuation, as compared to a more 'contained' accident in a microbiological safety cabinet.
- Are there any sharps involved eg. broken glassware which requires to be dealt with? This hazard should be attended to first to ensure this risk is removed and disposed of to a suitable container e.g. broken culture vessel in a rotary incubator.
- How would a spillage in equipment within a centrifuge or rotary incubator be dealt with? Would the disinfectant degrade the surface of the equipment?
- Consider the size of the spillage and how this would be attended to?
- A minor spillage involving little splashing and which is limited to a small area should be handled by applying disinfectant to the spillage and leaving for an appropriate period. E.g. the use of a powder disinfectant such as Virkon. If using a liquid disinfectant then place paper towels over the affected area and then apply the disinfectant. This will prevent the generation of aerosols. The spillage, disinfectant and disposable paper towels should then be discarded as clinical waste and then clean the area again with the liquid (form) disinfectant.
- For larger liquid spills, it may be appropriate to contain the spill using absorbent material such as vermiculite or absorbent matting, this can be removed and later decontaminated through autoclaving and applying disinfectant to the remaining contaminated area.
- A major spillage may involve considerable splashing and/or aerosol production therefore appropriate procedures must be in place to handle these events also. This has been described previously in the main guidance section of the document.

For additional information please refer to The Genetically Modified Organisms (Contained Use) Regulations 2014, and the SACGM Compendium of Guidance.