University Occupational Health and Safety Guidance Notes

GM WASTE INACTIVATION

MATERIAL AND WASTE INACTIVATION INCLUDING VALIDATION PRINCIPLES FOR COMPLETION OF THE RISK ASSESSMENT FOR GENETICALLY MODIFIED MICRO-ORGANISMS (GMMs)

1. Purpose

The Genetically Modified Organisms (Contained Use) Regulations 2014 require that GMMs in contaminated material and waste from Class 2 to 4 GM activity are inactivated by validated means. The guidance contained in this document provides information on achieving validated inactivation of waste GM material to achieve compliance with the Regulations. Note that although the regulations now state that inactivation of Class 1 material and waste is only required by validated means where and to the extent the risk assessment shows it to be required, the **University of Strathclyde guidelines are such that ALL contaminated GM material and waste MUST be inactivated by a validated means before disposal.**

2. Waste management

The Regulations define "inactivation" as the "complete or partial destruction of GMMs so as to ensure that any contact between the GMMs and humans or the environment is limited to an extent commensurate with the risks identified in the risk assessment and to provide a high level of protection for humans and the environment". This implies that the degree of inactivation required will vary depending on the nature of the organisms being used. Waste management and contaminated material inactivation which will be applied to the activity must be described in the risk assessment.

The risk assessment must specify the type and form of waste(s) generated, their treatment, ultimate form and fate including an indication of the numbers of viable GMOs remaining after treatment (if any). GM waste is classed as biological material and should be disposed of via the Clinical Waste Service.

There are two principal methods of inactivating waste material; chemically or via steam sterilisation i.e. the autoclave process. The selected treatment option must inactivate or render safe the hazardous component of the waste or contaminated material according to the risk assessment. It should be possible to validate the treatment method to verify that the number of viable organisms in any waste or effluent are within acceptable discharge levels or that the organism is destroyed.

2.1 Chemical Inactivation

Chemical disinfection is widely used for treating liquid wastes and for removing contamination from equipment and other reusable items that may be damaged by steam or heat.

Disinfection is not as effective as steam sterilisation in destroying biological agents nor as easily monitored. Many disinfectants are hazardous to health and may produce toxic or corrosive effects or induce an allergic sensitisation. Details of disinfectants and conditions for their use in the laboratory should be specified in standard operating procedures or laboratory codes of practice. For further information on disinfection see the Information Sheet Selection and use of disinfectants.

Inactivation by chemical disinfection would normally give a 5-log reduction in viability. If any live GMMs are present in the waste to be disposed of this must be justified by the risk assessment.

- Validation data

When using a chemical disinfectant it is acceptable to rely on validation data provided by the manufacturer for the particular micro-organism(s) so long as it is being used at the manufacturers' recommended concentrations and exposure times according to the requirements and conditions of use.

Where more than one species of micro-organism is being utilised in a laboratory it is advantageous to use a broad spectrum chemical agent for disinfection.

It may be necessary to undertake actual measurements to quantify the efficacy of the chemical disinfectant and procedure being used, e.g. When using a chemical disinfectant under non-standard conditions that do not conform with the manufacturer's instructions or non-standard heat treatments etc. Once a particular chemical inactivation procedure has been experimentally validated the results should be recorded along with the exact conditions used during the validation with the particular micro-organism. This will enable the actual percentage kill, log reduction in viability, or similar to be stated. Where it is not possible to determine the percentage kill or log reduction in viability, for example where viability is reduced to below detectable limits, simply state this to be the case and, where possible, indicate the limit of detection. This process then only needs to be repeated if the micro-organism or conditions of use are changed.

In many cases where validation of a chemical disinfectant is required it will not be possible to give the actual degree of kill for the GMM itself at the commencement of a project, as the GMM will not have been constructed at this stage. In such circumstances the statement with regard to waste inactivation in the GM risk assessment should be based upon validation tests using the parental or host micro-organism.

Where there is difficulty in validating chemical inactivation, consideration should be given to the use of autoclaving as an alternative. This may be particularly important if the risk assessment has identified that the GMMs may pose a risk to the environment.

- Compatibility

The chemical agent selected should be compatible with other substances or materials that may be present in the waste load so that its efficiency is not reduced and to ensure that toxic or hazardous products are not thereby formed nor released.

Any chemical agents used to inactivate waste must be active against the species of micro-organism being used and this information can be gained from the manufacturer. Some disinfectants have a broad range of activity, whilst others may be of more limited efficacy and the conditions of use should not reduce the activity of the chemical agent to an unacceptable level. For example, many disinfectants are sensitive to the chemical environment in which they are used and in some cases their activity can be greatly reduced by the presence of large amounts of organic, protein or particulate matter and the nature of the surfaces, items or equipment which will be exposed to the chemical disinfectant. Therefore, it is important when completing a risk assessment that as well as naming the chemical agent being used the contact time and final concentration are also incorporated.

- Inactivation of waste prior to autoclave

The GM risk assessment may indicate that liquid waste should be autoclaved rather than use chemical disinfection as a means of primary waste treatment. If liquid waste is autoclaved it should be noted that it is bad practice for material to be left untreated for any length of time prior to autoclaving as it could lead to an increased chance of a breakage or spillage during a prolonged storage period. Any bulk liquid should be chemically disinfected whilst it is awaiting autoclaving. The use of the disinfectant would be a precautionary measure rather than the primary means of waste treatment and as such it would not necessarily need to be validated. Consideration must be given to the choice of disinfectant and the amount of disinfectant treated waste that will be passing through the autoclave. Autoclaving some disinfectants can result in risk to health, and / or damage to the equipment from the vapours produced. It may be advisable to consult the disinfectant manufacturer or supplier prior to autoclaving disinfectant treated waste.

2.2 Autoclaving

Where autoclaving is used, it is well established that a cycle of 121^{o}C for 15 minutes, with full steam penetration to the centre of the load, is sufficient to render most materials sterile (waste containing prions requires special consideration). The sterilisation times and temperatures vary depending on the type of load ie solid, liquid, porous, equipment decontamination and will require the appropriate validation of each of these cycles where required.

Standard operating procedures for autoclaving should specify:

- The solid and liquid contaminated 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 lab manager; and,
- The emergency procedure in the event of a malfunction or failure.

Effective sterilisation by autoclaving depends on:

- Installation and commissioning using test loads to validate load temperatures and other operating conditions;
- Effective removal of air from the vessel and all parts of the load including the use of containers that allow steam penetration;
- Achieving and maintaining suitable load temperatures and holding times and the ability to validate these under operating conditions by independent thermocouple tests rather than by the use of biological and chemical indicators; and
- Regular examination and testing by a competent person under a written scheme of examination including the checking of safety valves, steam pressure indicating valves and in the case of bench top autoclaves water level indicators.

For further information on autoclaves see Guidance Notes on the Safe Use of Autoclaves.

There should be appropriate monitoring or indicating devices to warn the user to shut down the autoclave safely if critical operating conditions are not achieved. Emergency procedures should be established to deal with an unsterilised or partly sterilised load so that the waste can be repackaged for example, transferred to another autoclave. The standard operating procedure should specify the conditions, eg the use of robust, leak-proof and sealed inner and outer container, under which the removal of waste to an autoclave outside the laboratory or an alternate mechanism for treatment for example, through the Clinical Waste Service, but this should only be required in extreme cases and must be discussed with the University Biological Safety Adviser. Where materials cannot be autoclaved then standard operating procedures should specify the disinfectants and disinfection methods that are to be used.

Validation requires annual testing of the autoclave to demonstrate that the correct pressure has been reached for the required time. This involves 12 independent thermocouples being placed throughout the autoclave chamber in order to ensure that all waste, no matter where it is placed in the chamber reaches the required temperature for the specified time. Principal Investigators/Academic Supervisors must ensure that the autoclaves used within their department, to inactivate waste, have been appropriately validated.

On subsequent runs verification that the correct conditions were reached can be obtained through the use of, for example, chart recordings/printouts or appropriate indicators such as Browne's colour indicator tubes or spore strips autoclaved with the load. A record should be retained of all monitoring, maintenance and performance tests carried out on the autoclave together with a logbook or similar record of all routine disposals including the temperature charts and details of the load.