Safety at CABI BIOSCIENCE UK Centre (Egham)

The UK Health and Safety at Work Act (1974) places responsibility on employers and employees to carry out work in a safe manner. Additionally there are many health and safety regulations that affect the procedures necessary in culture collections. Similar regulations are enforced in countries throughout the world. All, however, are written to ensure that the worker is not placed at undue risk and that adequate measures are taken to reduce all risks in the working environment.

Employers and workers alike become quite concerned by the ever increasing requirements of safety legislation. Common sense must prevail to prevent over-reaction and thus the implementation of unnecessary and excessive procedures that would impede work and generate an antagonistic attitude in employees. Merely to blindly follow legislation does not encompass the intention of the regulations and their recommendations. It is the control of hazards and the generation of a safe and pleasant working environment that is intended. However, employers generally approach safety at work as a matter of compliance with the law and rarely look upon the safety of each individual independently in their employ. Whether it is compliance with the law or duties of a caring employer the basic requirements in order to establish a safe workplace are:

- Adequate assessment of risks.
- Provision of adequate control measures.
- Provision of health and safety information.
- Provision of appropriate training.
- Establishment of record systems to allow safety audits to be carried out.
- Implementation of good working procedures.

Good working practice requires assurance that correct procedures are actually being followed and this requires a sound and accountable safety policy.

The UK Management of Health and Safety at Work (MHSW) Regulations 1992 (Anon, 1992) are all encompassing and general in nature but overlap and lead into many specific pieces of legislation. These regulations require that every employer makes a suitable and sufficient assessment of the risks to health and safety to which any person whether employed by them or not may be exposed to through their work. These assessments must be reviewed regularly in addition to and when changes in procedures or regulations demand, and must be recorded when the employer has more than five employees. Preventive and protective measures may then be required following these risk assessments. The MHSW regulations require the employer to make arrangements for the effective planning, organization, control, monitoring and review of these preventive and protective measures. Health surveillance may be necessary where there is an identifiable disease or adverse effect and a likelihood of its occurring during the work undertaken. There must also be valid techniques available to detect the disease or condition. Competent persons must be appointed to assist the employer to carry out health and safety duties.

Further to this, procedures to follow for serious and imminent danger in danger areas must be established and employees must be made aware of these procedures. Information must be provided to employees on the risks to their health, preventive and protective measures, safe procedures to be followed and the identity of those persons appointed to carry out safety related duties on behalf of the employer. The MHSW regulations go on to cover:

- Cooperation and coordination where two or more employers share a workplace.
- Persons working on host employers' or self-employed persons' undertakings.
- Health and safety capabilities and training of staff and those appointed with special health and safety duties.
- Employees' duties and employers' duties to temporary workers.

The MHSW regulations provide a common sense approach to the major aspects of safety and following the principles outlined within them will enable any culture collection manager to establish safe procedures. The main hazards encountered in culture collections are the organisms held and chemicals used, in media (particularly selective agents), reagents and stains. These are covered by the Control of Substances Hazardous to Health (COSHH) Regulations (1988).

The effect of COSHH on Culture Storage and Supply

Like the 1974 act the COSHH regulations (1988) were compiled and made law in the UK to stimulate and enforce an improvement in Health and Safety in the workplace. All principles embodied in the COSHH regulations are contained in the UK HSW Act 1974. COSHH formalizes, enforces and in some instances extends certain sections of this act. The regulations were an attempt to ensure that employers do not expose their employees to risk as far as reasonably practicable. The latter term is quite commonplace and open to interpretation, so much so that there is a Croner publication (1989) on that phrase. In the employers' case it encompasses the minimum that must be done to meet the law. COSHH requires a suitable and sufficient risk assessment for all work that is liable to expose an employee to any substance that may be hazardous to health.

Health and Safety regulations can do many things other than what is intended of them. Employers may be given a false sense of security as they may feel that as there are safety regulations it must be safe. Others may feel a sense of overkill. This may be a common reaction when far-sweeping regulations come into operation which apply equally from certain typewriter correction fluids to cyanide. The `head in the sand' tactic, hide and it might go away attitude, was I am sure, one reaction to COSHH. Figures supplied at a conference on risk assessment in microbiology in 1990 suggested that 80% of employers had ignored COSHH in the first 18 months. The attitude encountered by many trying to enforce safety procedures is `I've been working like this for 20 years and nothing has happened yet.' On the other hand there tends to be a lot of overreaction for example from suppliers, no doubt to be on the safe side. Where reasonable and sensible people see this they tend to feel less respect for the law. Overkill will lead to boredom, apathy and carelessness. It is therefore essential that common sense is applied to the implementation of procedures to minimize risk.

To date most of the hard work should have been done and it is only the assessment of new risks, monitoring changes and improving safe procedures that should be occupying managers. However, it can be much simpler dealing with a known chemical than with a named microorganism. The full metabolic and biochemical potential of a microorganism is rarely known and therefore assessing the risk when the hazard is not clearly defined becomes difficult. This is where the COSHH regulations are realistic leaving room for interpretation. The regulations incorporate terms `as far as reasonably practicable', `adequate control', `suitable measures' which enables the employer to set relevant safe procedures that are workable. Microorganisms present different levels and kinds of hazard. It is therefore not possible to treat them all in the same way, but it is impracticable to treat each different microorganism individually. There are 3000-5000 recognized species of bacteria, 72 000 species of fungi and compounded upon this, different strains have different properties.

The culture collection at CABI BIOSCIENCE UK Centre (Egham) presents a good case for treating all microorganisms at one hazard level rather than as individual strains. It holds over 18 000 strains and the full potential of each one is not known. In general CABI BIOSCIENCE UK Centre (Egham) requires all staff to treat microorganisms as potential pathogens in hazard group 2 which requires containment level 2 (unless its potential is known or it is of a higher hazard group) according to the Advisory Committee on Dangerous Pathogens (ACDP; Anon, 1990).

This is done despite only c. 100 strains listed in hazard group 2 or 3. This approach takes into account the production of potentially hazardous mycotoxins. In the last analysis a safe laboratory is the result of applying good techniques, a hallmark of technical excellence.

Containment level 2 is easily achievable and should be standard practice in all microbiological laboratories. Good aseptic techniques applied by well trained personnel will ensure pure and clean cultures and will minimize contact with the microorganism. However, the unexpected, the accident, must also be taken into account when assessing the risk involved in handling microorganisms. The employment of good laboratory practice, good housekeeping, workplace and equipment maintenance and ensuring that staff have the relevant information and training, will minimize the risk of accidents. The establishment of emergency procedures to reduce potential harm is an additional and sensible approach.

Information on Safety

There is a vast array of sources of information necessary to carry out suitable and adequate risk assessments, and it can be somewhat daunting to find the most appropriate. The source documents used by CABI BIOSCIENCE UK Centre (Egham)for safety data on chemicals and other hazardous substances are:

1. HSE Guidance Notes EH 40 Occupational Exposure Limits (revised annually). This provides lists of hazardous substances and their long and short term exposure limits and gives brief information on controlling and monitoring exposure (Anon, 1993c).
2. Manufacturers (mainly BDH, SIGMA) safety data sheets.
3. ACDP Categorization of pathogens according to hazard and categories of containment (Anon, 1990). Not only does this provide lists of organisms under their risk group it also provides details of laboratory facilities and containment levels necessary to handle them.
4. Croner's Substances Hazardous to Health which is updated monthly (Kellard, 1994).
5. IRS Health and Safety Bulletin (Anon, 1994).

Classification of Micoorganisms on the Basis of Hazard

Various classification systems exist which include WHO; USPHS; ACDP; EFB; OECD and EC. Pathogenic organisms are covered by ACDP who list four hazard groups 1-4 with corresponding containment levels. The Advisory Committee on Genetic Manipulation (ACGM) prescribe separate but similar regulations for those organisms that have been genetically modified.

The COSHH regulations work well and can be easily applied in establishments with designed laboratories but may not work so well in the industrial environment where very large volumes and more hazardous techniques may be used. Total containment is rarely applicable.

Assessment of microorganisms

Microorganisms are more difficult to name, less predictable and more difficult to enumerate or measure than chemicals. Virulence and toxicity may vary from strain to strain. In addition to the risk of infection other hazards exist, such as mycotoxin production, allergenicity.

To meet COSHH requirements a step by step evaluation of a laboratory procedure or an industrial process must be carried out. This is necessary as different organisms provide different hazards and different size inocula are required to cause a problem. The assessment must cover the procedure from the original inoculum or seed culture to the final product or the point where the organism is killed and disposed of. It is not adequate to say that the microorganism is of ACDP hazard group 2 or less and therefore work can be carried out on the laboratory bench apart from those procedures that may create aerosols. Some individuals may respond differently to exposure, being more sensitive than others. It is therefore critical that the full potential of organisms be taken into account and this related to the effect they may have on the particular individual carrying out the work.

Mycotoxins

One of the better known hazards associated with fungi is the ability to produce toxic secondary metabolites. The presence of these in culture media adds to the hazard status of the growing organisms. The toxins produced may be carcinogenic, nephrotoxic, hepatoxic, haemorrhagic, oestrogenic or cause inflammatory effects. The most commonly known is aflatoxin which is considered to be carcinogenic, hepatotoxic and potentially mutagenic and is produced by strains of *Aspergillus flavus* and *A. parasiticus*. The table below lists some mycotoxins that may be present in growth media and present additional problems in both use and disposal.

Mycotoxicoses are poisonings caused by the ingestion of food contaminated (and sometimes rendered carcinogenic) by toxin producing microfungi. Toxins are also produced by many other fungi for example citreoviridin, citrinin, islanditoxin and patulin by species of *Penicillium*, ochratoxin by *Aspergillus* and trichothecenes and zearelenone by species of *Fusarium*, and various other compounds including cochliodinol by *Chaetomium*. It should always be remembered that many fungi have not been studied chemically and because mycotoxins are not reported for a species does not mean it does not produce them. The handling of materials contaminated by these toxins can lead to their ingestion and subsequent poisoning. Inhalation of mycotoxins can also be dangerous. Toxins from *Aspergillus* and *Fusarium* species have caused problems in patients when inhaled. The death of 2 factory workers from liver disease was associated with the inhalation of dust containing aflatoxin.

Some common mycotoxins and examples of fungi producing them

Mycotoxin	**Organisms**
Aflatoxin	*Aspergillus flavus, A. parasiticus*
Aflatrem	*Aspergillus flavus*
Altenuic acid	*Alternaria alternata*
Alternariol	*Alternaria alternata*
Austdiol	*Aspergillus ustus*
Austamide	*Aspergillus ustus*
Austocystin	*Aspergillus ustus*
Bentenolide	*Monographella nivalis*
Brevianamide	*Aspergillus ustus*
Citrinin	*Aspergillus carneus*, *A. terreus, Penicillium citrinum*, *P. hirsutum, P. verrucosum*
Citreoviridin	*Aspergillus terreus, Penicillium citreoviride*
Cochliodinol	*Chaetomium cochliodes*
Crotocin	*Acremonium crotocinigenum*
Cytochalasin E	*Aspergillus clavatus*
Cyclopiazonic acid	*Aspergillus versicolor*
Destruxin B	*Aspergillus ochraceus*
Fumagilin	*Aspergillus fumigatus*
Fusarin	*Fusarium moniliforme*
Gliotoxin	*Aspergillus fumigatus*
Islanditoxin	*Penicillium islandicum*
Malformin	*Aspergillus niger*
Maltoryzine	*Aspergillus* spp.
Moniliformin	*Fusarium moniliforme, F. oxysporum, F. equiseti*
Ochratoxin	*Aspergillus ochraceus, Penicillium viridictum*
Oxalic acid	*Aspergillus niger*
Patulin	*Aspergillus clavatus, Penicillium expansum, P. roquefortii, P. claviforme, P. griseofulvum*
Penicillic acid	*Aspergillus ochraceus*
Penitrem	*Penicillium crustosum*

Roridin	*Myrothecium roridum, M. verrucaria, Dendrodochium* spp., *Cylindrocarpon* spp., *Stachybotrys* spp.
Rubratoxin	*Penicillium rubrum*
Rubroskyrin	*Penicillium* spp.
Rubrosulphin	*Penicillium viridicatum*
Rugulosin	*Penicillium brunneum, P. kloeckeri, P. rugulosum*
Satratoxin	*Stachybotrys chartarum*
Slaframine	*Rhizoctonia leguminicola*
Sterigmatocystin	*Aspergillus flavus, A. nidulans, A. versicolor, Penicillium rugulosum*
Trichodermin	*Trichoderma viride*
Trichothecin	*Trichothecium roseum*
Trichothecenes T2 toxin deoxynivalenol (vomitoxin) nivalenol diacetoxyscirpenol fusarenone 3-acetyldeoynivalenol 15-acetyldexoynivalenol	*Fusarium acuminatum, F. roseum, F. sporotrichioides*
Tryptoquivalene	*Aspergillus clavatus*
Verrucarin	*Myrothecium verrucaria, Dendrodochium* spp.
Verruculogen	*Aspergillus fumigatus*
Viopurpurin	*Trichophyton* spp., *Penicillium viridicatum*
Viomellein	*Aspergillus* spp., *Penicillium aurantiogriseum, P. crustosum, P. viridicatum*
Viriditoxin	*Aspergillus fumigatus*
Xanthocillin	*Eurotium chevalieri*
Zearalenone	*Fusarium culmorum, F. graminearum, F. oxysporum, F. roseum*

Data from CABI BIOSCIENCE UK Centre (Egham)database and Smith & Moss (1985).

Sensitization

Long periods of exposure to airborne microorganisms can lead to sensitization and occupational asthma. Dermatitis may also occur as a result of exposure to allergenic fungi. Illness can be caused by both viable and non-viable organisms where it is the reaction with cell wall components that causes the condition. Being aware of such possibilities it is relatively easy to prevent exposure. Adopting good laboratory practices and providing microbiological safety cabinets are essential to reduce such risks.

Safety Records

There is now a requirement under UK Health and Safety Legislation to keep records of all significant findings with regard to risk assessments and other details. Records normally kept are:

- Risk assessments.
- Training.
- Document issue.
- Maintenance and service of equipment.
- Personal protective equipment issue and maintenance.
- Minutes of safety meetings.
- Reports on safety inspections; surveys; audits.
- Purchase and stocks of hazardous substances.

Such documentation is also normally complemented by information and literature to assist in risk assessment and analysis. Safety data sheets on all chemicals must be obtained when

they are purchased and issued to users before they are used. It is also essential that safety records and information are made available to employees.

The opportunity for all employees to discuss safety issues and be made aware of the risks they may be exposed to and the preventative and protective measures in place has been given through personal risk assessments. These are carried out, recorded and reviewed during the formulation of forward job plans during annual staff appraisal. The personal risk assessment record lists all duties the member of staff is required to do and the risk assessment for that procedure is noted. At this point the individual will have the opportunity to see and discuss the assessment and its relevance to them. It will also identify any area that may need further assessment or measures introduced to protect the individual. It is during this assessment that the individual's training record will be consulted to ensure they have the relevant training for the tasks they are expected to perform. Areas where the individual feels that they require more information will be identified and thus through this process the real aims of Health and Safety Legislation are addressed.

Storage of safety information

The amount of safety information required under the different pieces of legislation soon leads into storage and access problems. Today the availability of computerized systems makes a solution possible (see Table below). There are packages for handling all kinds of information available from a vast number of software suppliers. Many are suitable and easily adapted to suit individual needs. There are several specifically designed for the purpose, particularly for COSHH. The systems can ensure that all aspects of legislation are covered automatically highlighting specific requirements such as when reviews are due, the requirement for training or the need to introduce emergency procedures. Other systems can be specifically designed either internally or by specialists in the field. Although the latter can be more appropriate this can be much more expensive. However, it is now quite commonplace to have laboratory management systems specifically designed; these should also include the health and safety requirements.

Some available computer storage systems for safety data

Software	**Producer**	**Designed use**
CamHealth	Cam Axys, Harston, Cambridge	COSHH
Accident Management	Cam Axys, Harston, Cambridge	RIDDOR
COSHH-Master	AcQLab, Sheerness, Kent	COSHH
COSHH management/ Hazard	Harley Systems Ltd., Princes Risborough, Bucks, UK	COSHH
COSHH Information Management Systems (CIM)	Seton Ltd, Banbury, Oxon	COSHH
Laboratory Information Management systems (LIM)	Laboratory Microsystems High Wycombe	Laboratory data management including safety data (designed to your requirements)
PC Based COSHH Recording System	Norton Waugh Computing Ltd, Weston under Lizard, Shifriel, Shropshire	COSHH
Safety Information system	ROSPA, Birmingham	COSHH

COSHH use implies that the software not only stores safety data but holds risk assessments and can hold information specifically for your own use. It can also have some form of management system which will highlight needs and automatically produce reports, prompts for training, issue of PPE etc.

CABI BIOSCIENCE have taken the decision to have a system designed around a personal risk assessment. Individual records for staff are linked relationally to computer stored files on hazard risk assessments, training, document issue, personal protective equipment (PPE)

issue, PPE assessments and exposure records to hazardous substances. This system highlights the need for training, document or information issue, new assessments where necessary, issue of PPE, where required, when a member of staff is requested to perform a new procedure or duty.

No computer system can carry out the necessary actions for COSHH. These computer systems that help store the necessary information, aid retrieval, collation and its management are considered useful. It is important that it is remembered that it is not the act of recording assessments that is important but it is the establishment of mechanisms that ensure a safe environment that is required.

Handling Microorganisms at CABI BIOSCIENCE UK

Filamentous fungi, yeasts and bacteria of hazard group 2 or below (ACDP; Anon, 1990) are commonly handled at CABI BIOSCIENCE. They are received for different purposes, including taxonomic or physiological study, molecular biology, identification or inclusion in the dead dried herbarium collection or for deposit in the living collection. However, as the CABI BIOSCIENCE UK Centre (Egham)runs an identificaton service and receives unnamed organisms from all over the world (approximately 4-5000 cultures or specimens annually) the low hazard rating cannot be guaranteed. CABI BIOSCIENCE UK Centre (Egham)therefore operates parcel reception procedures in a laboratory equipped to contain risk group 3 organisms. When organisms of this group are suspected the material is either destroyed or passed to the Mycological Reference Laboratory of the Public Health Laboratory Service (PHLS), Bristol. The screening ensures that the organisms handled outside of the parcel reception facility are of low hazard group. The fungi of hazard group 2 are extremely unlikely to infect healthy individuals and usually only sufferers of autoimmune disease, those suffering severe ill health or those on immunosuppressant drugs can be infected.

CABI BIOSCIENCE sends cultures to laboratories outside the CAB INTERNATIONAL organization as culture sales, exchanges or as part of collaborative studies or contracts. The despatch of such includes safety information. This covers the fungus hazard group and is quite general. It includes information on the hazard status including the potential for toxin or allergen production, information on containment levels required, how to handle, transport, deal with spillages and dispose of the culture when the work is complete. Instructions on the package instructs the recipient to open the parcel in a laboratory. Customers are informed on ordering, before supply, if they have ordered a hazard group 2 organism and they are asked if they have the facilities for handling them. Where data is known on the toxic products of the strains this information is provided to the customer. However, it is rare that the full potential of a strain is known.

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a. To accompany organisms not assigned to hazard groups 2-4

Date of issue:
Revised 1995

Information on supplied Cultures of Microfungi as required under COSHH regulations and HSW Acts 6(4)(c)

Fungus names:

The fungi supplied (as listed above) are not categorised as Risk Group 2, 3 or 4 under EU Directive 90/679/EEC Classification of Biological Agents, and implemented in the UK through The Advisory Committee on Dangerous Pathogens, and therefore fall into Risk Group 1, i.e. a biological agent that is most unlikely to cause human disease. However, the fungi should be handled with care.

- Avoid all contact with the organism, growth media or materials on which they have grown. Many fungi produce extracellular metabolites which may be toxic.

- Avoid inhalation of spores as they may cause allergic reactions. Such allergies can be caused by the spores of many fungi including *Alternaria, Aspergillus, Cladosporium, Doratomyces, Sporobolomyces*.

To avoid these possible hazards and reduce the risk in handling, normal aseptic microbiological techniques should be employed.

All parcels containing fungi should be opened in a laboratory with Containment Level 1 as described by the Advisory Committee on Dangerous Pathogens (*Categorisation of pathogens according to hazard and categories of containment*, 4 edition London HMSO) and summarised below.

CONTAINMENT LEVEL 1

Containment level 1 is suitable for work with organisms of hazard group 1. Laboratory personnel must have received instruction in the procedures conducted in the laboratory.

1. The laboratory should be easy to clean. Bench surfaces should be impervious to water and resistant to acids, alkalis, solvents and disinfectants.
2. If the laboratory is mechanically ventilated, it is preferable to maintain an inward airflow into the laboratory by extracting room air to the atmosphere.
3. The laboratory must contain a wash-basin or sink that can be used for hand washing.
4. The laboratory door should be closed when work is in progress.
5. Laboratory coats or gowns should be worn in the laboratory and removed when leaving the laboratory suite.
6. Eating, chewing, drinking, smoking, storing of food and applying cosmetics must not take place in the laboratory.
7. Mouth pipetting must not take place.
8. Hands must be disinfected or washed immediately when contamination is suspected, after handling viable materials, and also before leaving the laboratory.
9. All procedures must be performed so as to minimise the production of aerosols.
10. Effective disinfectants must be available for immediate use in the event of spillage.
11. Bench tops should be cleaned regularly after use.
12. Used laboratory glassware and other materials awaiting sterilisation must be stored in a safe manner. Pipettes if placed in disinfectant, must be totally immersed.
13. All waste material which is not to be incinerated should be rendered non-infective before disposal.

14. Materials for disposal must be transported in robust containers without spillage.
15. All accidents and incidents must be reported.

Opening cultures and ampoules All parcels containing microorganisms must be opened in a laboratory by trained personnel and, ideally, in a cabinet that will prevent inhalation of aerosols.

The fungus is supplied either growing on a suitable agar slope or as a freeze-dried preparation. Both may be stored in the laboratory prior to use, the first in a 20-25°C microbiological incubator for 2 - 6 weeks and the latter in a refrigerator for microbiological use for 5 - 30 years.

Details of suitable media, incubation temperatures for the growth of the strains and any known special hazard are given with the strain(s) supplied.

The freeze-dried preparation is opened and transferred to growth media as follows:

Care should be taken in opening freeze-dried or lyophilised cultures. The ampoule should be marked with a glass cutter/file near the middle of the cotton wool plug and a hot glass rod used to crack the glass. Time is allowed for air to filter through the plug into the ampoule and the pointed tip should be snapped off.

Failure to allow air to gently enter will result in a rush of air generating an aerosol and releasing the microorganism. The plug may be impregnated with dried culture and should not be handled. This plug should be replaced with a fresh sterile plug. The addition of water/medium should be a gradual process avoiding squirting the liquid onto the dried material which will generate an aerosol. Transfer of the now liquid culture after a period for absorption of water (at least 30 min) should be carried out carefully.

Transport

If the materials are to be transported to another laboratory they should be packaged with enough absorptive material to absorb all contents of the containers in case of breakage. They should be placed in containers that will prevent breakage. If they are to be sent abroad they must be placed in a tin within another tin clearly labelled as containing biological specimens and all postal regulations of the recipient country must be followed. Fungi should not be forwarded to third parties outside your company.

Disposal

All fungi, media and containers should be sterilised by autoclaving at 121°C for 15 min before disposal by suitable means such as incineration.

Procedures in case of spillage

If the fungus is spilt or its container broken, thoroughly wet with a disinfectant, such as 4% sodium hypochlorite, and allow 30 min before swabbing up and transferring into a container for autoclaving.

To accompany cultures assigned to hazard group 2 (CAB Bioscience UK Centre (Egham) does not supply organisms of hazard groups 3 or 4.

Date of Issue:
Revised 1995

Information on Supplied Cultures of Microfungi as required under COSHH regulations and HSW Act s.6(4)(c)

Fungus names:

The fungus (or fungi) supplied (as listed above) are Risk Group 2 organisms under EU Directive 90/679/EEC Classification of Biological Agents to be adopted by the Advisory Committee on Dangerous Pathogens (ACDP) *Categorisation of biological agents*, 4 edition. These are biological agents that may cause human disease and which might be a hazard to laboratory workers but are unlikely to spread to the community. Laboratory exposure rarely produces infection and effective prophylaxis or effective treatment is available.

- Avoid all contact with the organism, growth media or materials on which they have grown. Many fungi produce extracellular metabolites which may be toxic.

- Avoid inhalation of spores as they may cause allergic reaction. Such allergies are caused by the spores of fungi including *Alternaria, Aspergillus, Cladosporium, Doratomyces, Sporobolomyces* and many more.

To avoid these possible hazards, and reduce the risk in handling, normal aseptic microbiological techniques should be employed. All parcels containing fungi should be opened in a laboratory with containment level 2 as described by The Advisory Committee on dangerous pathogens 1990 (*Categorisation of pathogens according to hazard and categories of containment*. London: HMSO). Any work that may result in the creation of an aerosol containing the organism must be carried out in an appropriate microbiological safety cabinet.

CONTAINMENT LEVEL 2

Containment level 2 is suitable for use with agents of in Hazard Group 2. Laboratory personnel must receive instruction and training in handling pathogens and an appropriate standard of supervision of the work must be maintained.

1. The laboratory should be easy to clean. Bench surfaces should be impervious to water and resistant to acids, alkalis, solvents and disinfectants.
2. Access to the laboratory should be limited to laboratory personnel and other specified persons.
3. There should be adequate space ($24m^3$) in the laboratory for each worker.
4. If the laboratory is mechanically ventilated, an inward airflow into the laboratory must be maintained by extracting room air to atmosphere.
5. The laboratory must contain a wash hand basin which should be located near the laboratory exit. Taps should be of a type which can be operated without being touched by hand.
6. An autoclave for the sterilisation of waste materials must be readily accessible, normally in the same building as the laboratory.
7. The laboratory door should be closed when work is in progress.
8. Laboratory coats or gowns, which should be side or back fastening, must be worn in the laboratory and removed when leaving the laboratory suite. Separate storage (pegs) must be provided in the laboratory suite for this clothing.
9. Eating, drinking, smoking, storing of food and applying cosmetics must not take place in the laboratory.
10. Mouth pipetting must not take place.
11. Hands must be disinfected or washed immediately when contamination is suspected, after handling infective materials, and also before leaving the laboratory.

12. In general, work may be conducted on the open bench, but care must be taken to minimise the production of aerosols. For manipulations such as vigorous shaking or mixing and ultrasonic disruption etc., a microbiological safety cabinet (Class 1 or Class II) or equipment which is designed to contain the aerosol must be used. The cabinet must exhaust to the outside air or to the laboratory air extract system (see para 34, 14(b) of ACDP Categorisation of pathogens and their containment levels).
13. Effective disinfectants must be available for routine disinfection and immediate use in the event of spillage.
14. Bench tops should be disinfected after use.
15. Used laboratory glassware and other materials awaiting sterilisation must be stored in a safe manner. Pipettes if placed in disinfectant must be totally immersed.
16. Material for autoclaving must be transported to the autoclave in robust containers, without spillage.
17. All waste materials must be made safe before disposal or removal to the incinerator.
18. All accidents and incidents must be immediately reported to, and recorded by, the person responsible for the work.

Opening cultures and ampoules All parcels containing microorganisms must be opened in a laboratory by trained personnel and, ideally, in a cabinet that will prevent inhalation of aerosols.

The fungus is supplied either growing on a suitable agar slope or as a freeze-dried preparation. Both may be stored in the laboratory prior to use, the first in a 20-25°C microbiological incubator for 2-6 weeks and the latter in a refrigerator for microbiological use for 5-30 years.

Details of suitable media, incubation temperatures for the growth of the strains, and any known special hazards, are supplied with the strains.

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Transport If the materials are to be transported to another laboratory they should be packaged with enough absorptive material to absorb all contents of the containers in case of breakage. They should be placed in containers that will prevent breakage. If they are to be sent abroad they must be placed in a tin within another tin clearly labelled as containing biological specimens and all postal regulations of the recipient country must be followed. Fungi should not be forwarded to third parties outside your company.

Disposal All fungi, media and containers should be sterilised by autoclaving at 121°C for 15 min before disposal by suitable means such as incineration.

Procedures in case of spillage If the fungus is spilt or its container broken, thoroughly wet with a disinfectant, such as 4% sodium hypochlorite, and allow 30 min before swabbing up and transferring into a container for autoclaving.

- **Guidelines for the establishment and operation of culture collections**

If you are considering setting up a collection of your own or enhancing the one you have why re-invent the wheel or make the same old mistakes. There are lots of sources of information, look through the UKNCC web pages for advice or contact a collection. The UKNCC follow a quality management system which will provide relevant information. Read the UKNCC Quality Manual for further information.

The World Federation for Culture Collections have published guidelines which they are currently updating. The old edition (1990) are available from the WFCC secretariat (see organisations) and on the WFCC web site.

WDCM (World Data Center for Microorganisms)
Center for Information Biology, National Institute of Genetics, 1111 Yata, Mishima, Shizuoka 411, Japan. Tel: +81-559-81-6895; Fax: +81-559-81-6896. URL: http://wdcm.nig.ac.jp/

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Some further reading that may help:

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