LABORATORY DISINFECTION
CODE OF PRACTICE

Introduction
The use of different disinfectants in hospital and medical school laboratories has evolved over a number of years. Some of this use is inappropriate. This document is intended to ensure that an appropriate disinfectant is used for a particular procedure and that it is suitably controlled.

There are many types and formulations of disinfectants on the market. The choice should be limited to those contained within this policy.

Responsibility of heads of departments
Heads of department are responsible for ensuring that a local laboratory disinfection policy is drawn up, based on these recommended disinfectants and the requirements of the individual departments. Suitable instruction and training must also be given.

Recommended disinfectants
The following disinfectants (diluted in water) are recommended for use in laboratories within the Royal Free Hospital and the Medical School:

- Sodium hypochlorite 1000ppm* (0.1%), 2,500ppm (0.25%), 10,000ppm (1%)
- Chlorine releasing tablets and granules
- Alcohol 70%
- Tristel (chlorine dioxide) 280ppm
- Virkon 1%
- Formaldehyde
- Glutaraldehyde 2%(not to be used outside of a safety cabinet)

* ppm: parts per million of available chlorine

NOTE: Clear soluble phenolics (CSPs), of which the prime example is Hycolin, have been withdrawn from sale as a result of the European Biocidal Products Directive and should not be used or stored unless awaiting disposal. See Appendix I for HPA advice on alternatives to hycolin

Properties and usage of recommended disinfectants

Considerations
There must be adequate contact with the disinfectant to enable it to be effective, for example; objects should be fully immersed and air pockets should not be present. Deposits of organic matter should be removed prior to disinfection. Adequate contact time should be allowed for the disinfectant to perform its function. This time will vary according to the type of disinfectant, the presence of inactivating or interfering factors (such as excessive organic material or and the
presence of chemicals) and the microbial load. Disinfectants do not necessarily kill all biological agents and do not usually destroy bacterial spores. Only freshly prepared in-use dilutions should be used since stored dilutions may lose activity. The HSE have recommended that autoclaving of dry discard should replace discard pots with disinfectant wherever possible.

**Hypochlorites**

Hypochlorites are highly effective against vegetative bacteria, viruses and fungi. They have limited activity against bacterial spores and are not very effective against mycobacterium spp. They are compatible with anionic and non-ionic detergents, but are inactivated by organic matter and may corrode metals and damage rubber. Hypochlorites are commonly available as solutions of sodium hypochlorite and as powdered or tableted sodium dichloroisocyanurate (NaDCC) which are also recommended for spillages. Sodium hypochlorite stock solutions will decay with time, light and temperature and should be stored in cool and dark conditions. Working solutions of any hypochlorite need to be changed frequently (at least daily) because of deterioration caused by the addition of organic matter. Commonly used dilutions are:

- **1000 ppm (0.1%)** for general wiping of equipment and benches (not spillages)
- **2500 ppm (0.25%)** for discard containers (if required)
- **10,000 ppm (1%)** for spillages
- **20,000 ppm (2%)** for work involving prions/TSE agents (NADCC not effective)

(for more information on prions see HSE guidelines: [http://www.doh.gov.uk/cjd/tseguidance](http://www.doh.gov.uk/cjd/tseguidance))

Note: hypochlorites should not be mixed with acids as gaseous chlorine is released at low pH, nor with formalin or formaldehyde as a bis chloromethyl 3 ether (a lung carcinogen) is released.

**Peroxygen-based disinfectants**

Virkon is a multi-component peroxygen based oxidising agent. It is effective against bacteria, fungi, and viruses. Activity against bacterial spores and Mycobacterium spp. is variable for peroxygen-based disinfectants. It does cause some corrosion to metals (as hypochlorous acid is formed), but less than hypochlorite solution. Virkon in solution is stable for 7 days and has a pink colour which gradually fades with inactivation; however, good infection control practice indicates that in-use disinfectants should be replaced daily. Virkon (1%) may be suitable in laboratories performing low risk microbiological or Class 1 genetic modification work. This disinfectant should not be used for clinical work.

**Alcohols**

Alcohols are effective against many bacteria including Mycobacterium spp. and fungi. They have variable activity against viruses (less effective against nonenveloped viruses) and have no activity against bacterial spores. Alcohols poorly penetrate organic matter, particularly proteinaceous material therefore cleaning beforehand is essential. Due to their flammability they should not be used near flames or equipment likely to generate sparks. Alcohol sprays must
not be use on electrical equipment whilst connected to the mains. When used to
decontaminate centrifuges, allow time for the alcohol to safely evaporate before
turning the equipment on. A surface wipe is a convenient method of disinfection,
but due to evaporation has a limited effect and therefore should be confined to
surfaces with no visible contamination.

Alcohols should not be used undiluted. The most effective strength for alcohol
disinfection is a 70-80% (v/v) solution of isopropanol or ethanol in water.
IMS (Industrial methylated spirits, which comprises 95% ethanol and 5%
methanol) is also suitable when diluted.

Aldehydes
There are two distinct types with differing uses.
1) Formaldehyde gas is used to fumigate microbiological safety cabinets and
rooms. Safety cabinets should be fumigated at least 6 monthly and before
issuing a permit to work. The procedure of fumigating a safety cabinet requires a
written risk assessment.
2) Glutaraldehyde has long been recognised as a cause of ill health, with
dermatitis and respiratory problems being the most significant effects. It is now
also classified as an asthmagen and respiratory sensitiser and has been
assigned a Maximum Exposure Limit (MEL) of 0.05 ppm.
While glutaraldehyde is an effective disinfectant, its use in the open laboratory is
not recommended. Small amounts may be used for disinfecting equipment inside
safety cabinets, but these cabinets must vent to the exterior and not be of the
recirculating type.

Chlorine dioxide (Tristel)
Tristel (an aqueous solution of chlorine dioxide) has an activity that differs from
hypochlorite as it does not produce free chlorine. It is active against bacteria,
including Mycobacterium spp., viruses, fungi and spores. Activation is required
before use and it is used extensively as an endoscope sterilant. Chlorine dioxide
is not a skin or respiratory sensitiser or irritant, in low concentrations it has a
history of safe use and has been used to sanitize drinking water and swimming
pools since the 1950s. At higher concentrations (280 ppm) it has rapid
bactericidal activity against Mycobacterium spp. and is a more effective
sporocide than hypochlorite. It may effect some metals.

Use of non-listed disinfectants
New disinfectants may only be introduced following written agreement with the
Biological Safety Officer and the Infection Control Officer.
<table>
<thead>
<tr>
<th></th>
<th>Vegetative Bacteria</th>
<th>Tubercle Bacilli</th>
<th>Bacterial Spores</th>
<th>Fungi Lipid (enveloped) viruses</th>
<th>Non-lipid (non-enveloped) viruses</th>
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</thead>
<tbody>
<tr>
<td>Hypochlorites</td>
<td>+</td>
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<tr>
<td>Aldehydes</td>
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<tr>
<td>Virkon</td>
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<td>+</td>
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<tr>
<td>Chlorine dioxide</td>
<td>+</td>
<td>+</td>
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<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

**Recommendations for specific procedures**
- Routine disinfection of benches: sodium hypochlorite or IMS
- Disinfection of safety cabinets: 70% alcohol (ethanol or IMS)
- Disinfection of centrifuges: 70% alcohol (ethanol or IMS)
- Disinfection of tuberculous material: Tristel 280ppm
- Spillages: sodium hypochlorite*
- Discard pots: sodium hypochlorite#
  * Chlorine releasing tablets may be more convenient
  # For the inactivation of Mycobacterium spp. in discard pots, Tristel (Chlorine dioxide) should be used.

**General Precautions**
- Employees should carry out a COSHH assessment on the use of all chemical disinfectants to determine the particular precautions for each. In general the following precautions will be needed:
  - Concentrates should be dispensed using closed systems, e.g. pump or syphon
  - Staff should always wear suitable gloves when handling disinfectants.
  - All persons handling concentrated stock solutions of disinfectants should wear suitable safety spectacles, goggles or a full-face visor and a disposable plastic apron if splashing could occur.
  - Some disinfectants give off corrosive and/or toxic vapours and will be subject to Workplace Exposure Limits (WEL). If adequate ventilation is not available during their use or preparation, a respirator which has been assessed as appropriate should be worn. Advice should be sought from the safety office if a respirator is thought necessary.
- Information in this policy is based on the HSE publication ‘Safe working and the prevention of infection in clinical laboratories and similar facilities’ 2003, ISBN 07176 25133
APPENDIX I

HPA ADVICE - Replacement for Hycolin
February 2007

Background
Clear soluble phenolics (CSPs), of which the prime example is Hycolin, have been withdrawn from sale as a result of the European Biocidal Products Directive. (This is not because they are ineffective, but because sales levels do not justify the substantial investment needed for registration). The withdrawal of CSPs from the market was abrupt and has taken users by surprise. CSPs were the agent of choice for mycobacterial disinfection. There are established alternatives to CSPs available for every laboratory requirement other than areas where mycobactericidal activity is required where, up until now, they were the sole disinfectant. There is now an urgent need to find an effective replacement for laboratories handling mycobacteria.

Present predicament and what do we require
Disinfection against TB and other pathogenic mycobacteria is required in routine diagnostic laboratories as well as reference and research facilities. Chemical disinfectants are required in case of spills, for routine environmental decontamination, for equipment decontamination (where corrosion can be problematic) and for disinfection of liquid waste, which may contain a high concentration of organic matter. CSPs have the advantage of low corrosivity and high resistance to inactivation by organic matter.

It may be impossible to find all the attributes of CSPs in a single substitute, however the user’s situation may have to become more complex, with more than one agent required in a laboratory.

There are no standard tests for laboratory disinfectants. European disinfectant tests are usually for environmental disinfection in the absence of gross soiling, in contrast with a laboratory spill, where there may be very high levels of organic matter; similarly with disinfection of liquid waste. The applicability of other European disinfectant tests to laboratory situations is debatable. Many disinfectant manufacturers will claim, most correctly, that their disinfectants will inactivate mycobacteria. However this is only a starting point for consideration of a replacement for CSPs; many other factors can prevent a chemical disinfectant from working effectively. To assess how effectively an agent will perform in practice, experience and judgement by those experienced in laboratory safety and disinfectant properties will have to come into play.

A small HPA group was convened to review options or alternatives for Hycolin. The results of this review and subsequent recommendations are outlined below. These recommendations were derived from peer review published data (see references) with the additional input of HPA users and experts from across the Agency.
**Possible Alternatives to Hycolin**

There was a historical general perception that hypochlorites were unreliable against mycobacteria. More recent work indicates this is not the case.

**Hypochlorites**

These are the main laboratory disinfectants. Their characteristics are: wide microbicidal spectrum, comparatively rapid action, inactivation by organic matter at low concentrations and corrosive to some metals.

Hypochlorites are available from either liquid sodium hypochlorite or as solutions of the solid sodium dichloroisocyanurate (NaDCC). NaDCC has several practical advantages over sodium hypochlorite, mainly the stability of the undiluted disinfectant and the availability of pre-measured tablets of the agent.

Results from a recognised disinfection centre of excellence, the Hospital Infection Research Laboratory (HIRL) at City Hospital in Birmingham, shows hypochlorites from NaDCC to be effective against *Mycobacterium tuberculosis*, a validated substitute for TB, *Mycobacterium terrae*, and atypical mycobacteria such as *Mycobacterium avium-intracellulare* (MAI) though, as with most other disinfectants, activity against atypicals is slower.

At CfI, NaDCC is used at 2500 ppm available Chlorine (ppm av Cl). The work from HIRL showed 1000 ppm to produce a reduction greater than 5 logs in four minutes and 10,000 ppm to produce the same reduction in one minute in dirty conditions (10 percent serum). Similarly, with MAI, those reductions were achieved in 60 and 10 minutes respectively.

From these data we believe that hypochlorites could be used in laboratories handling mycobacteria for disinfection in areas where corrosion is not issue.

Hypochlorites can have their activity checked in use by starch/iodide indicator papers: if sufficient oxidising power is left in a used solution, it will turn the papers blue-black; if there is insufficient oxidising power (showing overloading of the disinfectant), the papers will produce a pallid colour or not change at all. (Care – high concentrations can bleach out the colour change, therefore, if there is no colour change apparent, dilute the test solution to about 1% and retry).

**Peracetic acid**

Peracetic acid (PA) is a wide spectrum, rapid disinfectant that is more expensive than hypochlorite, but without its corrosivity and degree of inactivation by organic matter. PA is currently used as a disinfectant for flexible fibre optic endoscopes, where corrosion is highly undesirable.
HIRL results show 0.35% PA to produce a 5 log reduction in TB in one minute under dirty conditions. With MAI, the same reduction was achieved in four minutes in dirty conditions. Similar, though less quantitative, work from University College Hospital showed a different PA product (0.2%) to be of similar activity.

From these data we believe that PA could be used in laboratories handling mycobacteria for disinfection in areas where corrosion must be avoided.

**Chlorine dioxide**
Chlorine dioxide is a wide spectrum, rapid disinfectant that is more expensive than hypochlorite, but without its corrosivity and degree of inactivation by organic matter. Chlorine dioxide is currently used as a disinfectant for flexible fibre optic endoscopes, where corrosion is highly undesirable.

HIRL results show that chlorine dioxide at 1100 ppm produces a 5 log reduction in one minute under dirty conditions. With MAI the same reduction was also achieved in one minute in dirty conditions.

From these data we believe that chlorine dioxide could be used in laboratories handling mycobacteria for disinfection in areas where corrosion must be avoided.

**Recommendations**
Therefore, based on evaluation of peer reviewed papers, we recommend that the following disinfectants are considered for use in laboratories working with mycobacteria. However, their use must be subject to local assessment of suitability and risk assessments. The recommendations relate only to replacement of Hycolin and are not intended to suggest replacement of other disinfectants currently in use.

- There is a range of hypochlorite products that can be used for the disinfection of liquid waste such as sputum digest supernatant, if steam sterilisation is not feasible. Hypochlorite has the advantage that a highly concentrated initial solution can be diluted by adding liquid waste such that the final volume contains the desired available chlorine concentration. Activity can be checked by starch-iodide paper (see above). Hypochlorites can also be used for spills. Hypochlorite presentations such as absorbent granules are particularly suited to spill clearance. Occasional use of high hypochlorite concentrations should not cause corrosion on most laboratory surfaces. Hypochlorite however, should not be used on laboratory equipment such as centrifuges.

- Peracetic acid and chlorine dioxide products – either or both of these solutions could be used in situations where corrosion must be avoided. Although these are used on endoscopes without corrosion, this does not guarantee they will not have corrosion, including where this might have a significant safety element such as centrifuges. We recommend that users make attempts to ascertain compatibility with equipment
manufacturers. We realise this may not always be possible and where it is not, every effort must be made to ensure that centrifuges and components are inspected and maintained in accordance with HPA Centrifuge Guidance. The references describe possible products which may be suitable within this category.

- Chemical disinfectants are an inherently poor quality assurance method of decontamination. Dry discard followed by steam sterilisation has superior quality assurance and should be used wherever feasible. Liquid discards may need to be immobilised with a gelling agent. Safe methods of transport between point of use and point of disposal will also have to be considered.

### Summary

<table>
<thead>
<tr>
<th>Method</th>
<th>Spills in cabinet</th>
<th>Equipment</th>
<th>Routine surface decontamination</th>
<th>Liquid waste</th>
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</thead>
<tbody>
<tr>
<td>Steam sterilisation</td>
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<td>x</td>
<td>NA</td>
<td>√</td>
</tr>
<tr>
<td>Hypochlorite</td>
<td>√</td>
<td>x</td>
<td>√*</td>
<td>√</td>
</tr>
<tr>
<td>Chlorine dioxide</td>
<td>√</td>
<td>√</td>
<td>√</td>
<td>NA</td>
</tr>
<tr>
<td>Peracetic acid</td>
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<td>√</td>
<td>√</td>
<td>NA</td>
</tr>
</tbody>
</table>

* only if surface will withstand repeated hypochlorite use.

### References


J. Holton, P. Nye and V. McDonald. (1994) Efficacy of selected disinfectants against Mycobacteria and Cryptosporidia. Journal of Hospital Infection 27; 105-I 1.5