

Training and Human Resources Development for Microbiological Containment Laboratories

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Topics to be covered

- The role of line management
- The role of the safety staff
- The safety is a culture, the way we work not an add on
- The importance of risk assessment
- Training requires competency testing
- We learn from laboratory incidents
- Topics covered in training

LABORATORY SAFETY



90%
personnel attitudes
and
individual actions

facilities and
equipment

10%



Management of Safety

- policy of institution
- responsibilities
- written procedures
- safety manual
- safety audits /assessment of work
- incident reporting and investigation

Management of Safety

- OHS committees
- emergency planning
- induction – new staff, visiting scientists, visitors, 'old' staff
- working alone issues
- long hours, deadlines etc

Responsibilities

- **Everyone** has responsibility for OHS
- Line management responsibility
- Director of organisation ultimately responsible
- Safety staff there to provide information, advice, training etc – they are not responsible for it all!

Human Resource Management

Keys to good HR Management

- Clear lines of responsibility
- Open communication between all levels of management
- Promptly reporting things when they go wrong
- Staff selection process
- Annual staff reviews
- Regular discussion between supervisor and staff member
- Training in open communications
- Buddy system (mentoring)

Stages (= OHS)

- Recognition
 - training, lab biosafety manual
- Evaluation
 - Use an appropriate Safety Standard
- Control / Prescription
 - China Standard GB 19489-2004 “Laboratories – General Requirements for Biosafety”
 - “Regulations on Administration of Biosafety in Pathogenic Micro-organism Laboratories Nov 2004” Decree No. 424 of State Council, PRC
 - Testing and surveillance

How to achieve this?

- OHS Risk Assessment of work

Risk assessment of work

Overall procedure

- Identify hazards
- Assess Inherent Risk (without controls)
 - Using Consequence & Likelihood tables
- Determine Management Controls
- Assess Residual Risk (with controls)
- Monitor controls

Procedure

Step 1

- Identify hazards, using checklist (enter on form)

List of hazards

Part A: Section 2a. Workplace Hazards Identification			
Using tick boxes identify all hazards associated with workplace, system of work, equipment and substances used. Each of these hazard categories corresponds to information available on the CSIRO OHSE index http://www.csiro.au/services/humanres/essentials/safely/OHSMSIndex.htm			
Refer to this index for further information and control strategies on each of the hazards listed below.			
Mechanical (Plant)		7. Biological	
1.1 Vehicles, transport	<input type="checkbox"/>	7.1 Biological materials (Refer to Part D)	<input type="checkbox"/>
1.2 Plant, machinery, equipment in motion	<input type="checkbox"/>	7.2 Biological materials (refer to Part D) involves GMO's	<input type="checkbox"/>
1.3 Compression/tension/stored energy	<input type="checkbox"/>	7.3 Allergens / sensitisation	<input type="checkbox"/>
1.4 Noise	<input type="checkbox"/>	7.4 Irritants	<input type="checkbox"/>
1.5 Vibration	<input type="checkbox"/>	7.5 Genotoxins (mutagens, teratogens)	<input type="checkbox"/>
1.6 Firearms	<input type="checkbox"/>	7.6 Zoonoses (refer to Part D)	<input type="checkbox"/>
1.7 Pressure equipment (high/vacuum)	<input type="checkbox"/>	7.7 Handling of small animals	<input type="checkbox"/>
1.8 Tools, sharps, cutting implements	<input type="checkbox"/>	7.8 Handling of large animals	<input type="checkbox"/>
		7.9 Handling of human samples (refer to Part D)	<input type="checkbox"/>
2. Radiation		8. Chemical/Hazardous Substances	
2.1 Ionising (refer to Part C1)	<input type="checkbox"/>	8.1 Carcinogens	<input type="checkbox"/>
2.2 Ultraviolet (refer to Part C2)	<input type="checkbox"/>	8.2 Sensitising agents	<input type="checkbox"/>
2.3 Infrared (refer to Part C2)	<input type="checkbox"/>	8.3 Corrosive/oxidising agents	<input type="checkbox"/>
2.4 Laser (refer to Part C2)	<input type="checkbox"/>	8.4 Irritants	<input type="checkbox"/>
2.5 Radiofrequency (refer to Part C2)	<input type="checkbox"/>	8.5 Genotoxins (mutagens, teratogens)	<input type="checkbox"/>
2.6 Electromagnetic field (refer to Part C2)	<input type="checkbox"/>	8.6 Toxic/harmful substances	<input type="checkbox"/>
2.7 Extremely low frequency (refer Part C2)	<input type="checkbox"/>	8.7 Solvents	<input type="checkbox"/>
		8.8 Generation of dusts, vapours, fumes etc.	<input type="checkbox"/>
		8.9 Asbestos	<input type="checkbox"/>
3. Fire and Explosion		9. Gases	
3.1 Flammable substances	<input type="checkbox"/>	9.1 Flammable	<input type="checkbox"/>
3.2 Explosives	<input type="checkbox"/>	9.2 Asphyxiant inert gas	<input type="checkbox"/>
		9.3 Toxic gas	<input type="checkbox"/>
4. Temperature		9.4 Gas cylinders / tanks	<input type="checkbox"/>
4.1 High temperature materials	<input type="checkbox"/>	9.5 Pressurised lines	<input type="checkbox"/>
4.2 Cryogenic fluids	<input type="checkbox"/>		
5. Hazardous Environments		10. Personal	
5.1 Confined spaces	<input type="checkbox"/>	10.1 Manual handling incl striking & grasping	<input type="checkbox"/>
5.2 Working at heights	<input type="checkbox"/>	10.2 Slips, trips, falls	<input type="checkbox"/>
5.3 Working at sea or in water bodies	<input type="checkbox"/>	10.3 Fixed posture, eg microscopy	<input type="checkbox"/>
5.4 Heat/cold stress	<input type="checkbox"/>	10.4 Repetitive and/or overuse movements, eg keyboarding, pipetting	<input type="checkbox"/>
		10.5 Pressure (diving/altitude)	<input type="checkbox"/>
6. Electrical		10.6 Working alone	<input type="checkbox"/>
6.1 High voltage equipment	<input type="checkbox"/>	10.7 Field work	<input type="checkbox"/>
6.2 Live electrical equipment	<input type="checkbox"/>	10.8 Mental stress	<input type="checkbox"/>
6.3 Static charge	<input type="checkbox"/>	10.9 Overseas travel / work (vaccinations)	<input type="checkbox"/>
		10.10 Engulfment eg in sand	<input type="checkbox"/>
11. Other - Specify:	-----		

Procedure

Step 2

- Determine Inherent Risk ie the risk that exists without any controls
 - Determine consequence
 - Determine likelihood
 - Enter into IR table using the numerical values

Consequence

LEVEL	DESCRIPTOR	CONSEQUENCE – DESCRIPTION
1	Insignificant	No injuries, low financial loss
2	Minor	First aid treatment, on site release immediately contained
3	Moderate	Medical treatment required, on site release contained with outside assistance, high financial loss
4	Major	Extensive injuries, loss of production capability, off site release with no detrimental effects, major financial loss
5	Catastrophic	Death, toxic release off site with detrimental effect, huge financial loss

Likelihood

LEVEL	DESCRIPTOR	LIKELIHOOD – DESCRIPTION
1	Rare	May occur only in exceptional circumstances
2	Unlikely	Could occur at some time
3	Possible	Might occur at some time
4	Likely	Will probably occur in most circumstances
5	Almost Certain	Is expected to occur in most circumstances

Risk Matrix

LIKELIHOOD	CONSEQUENCES				
	Insignificant (1)	Minor (2)	Moderate (3)	Major (4)	Catastrophic (5)
(5) Almost Certain	M	M	H	H	H
(4) Likely	M	M	M	H	H
(3) Possible	L	M	M	H	H
(2) Unlikely	L	L	M	M	H
(1) Rare	L	L	M	M	H

Legend

H:	High risk immediate action required
M:	Moderate risk; management responsibility must be specified (significant and moderate combined to be moderate)
L:	Low risk; manage by routine procedures

Procedure

Step 3

- Enter Inherent Risk onto form

Procedure

Step 4

- Determine the Residual Risk ie the risk that remains after implementing measures to reduce it:

Procedure

Step 5

- Enter the existing (management) controls for each hazard onto form
- Determine if existing controls are very good, reasonable or poor

Management Controls

Rating	Management control examples
Very Good	Controls are best practice, involve explicit standards and are followed all of the time. Includes a high emphasise on elimination, substitution or engineering controls.
Reasonable	Controls are in place but not followed all of the time and may not include best practice. Includes a high emphasis on administration and protective equipment.
Poor	There are few or no controls in place. No standards have been identified. Controls do not address Hierarchy of Control principles.

Procedure

- Plot Inherent Risk against the assessed quality of the existing management control of the risk to determine the Residual Risk

Risk Matrix for Residual Risk

<i>I N H E R E N T R I S K</i>	High			
	Mod			
	Low			
		Very Good	Reasonable	Poor
		<i>MANAGEMENT CONTROL</i>		

Legend



Residual risk is **high**
- attend to immediately

Residual risk is **moderate**
- attend to in short term

Residual risk is **low**
- attend to in longer term

Procedure

Step 6

- Take action where necessary!
- Use hierarchy of controls when determining any controls that need to be implemented:

Hierarchy of Controls

- Elimination
- Substitution
- Isolation
- Engineering control
- Administrative control
- PPE (Personal Protective Equipment)

Procedure

Step 7

- Monitor controls

Training

Successful training is not

- Reading the safety manual and signing a form that you have read, understood and will comply with the manual
- Sitting through a series of boring safety lectures
- Being allowed to work unsupervised in the laboratory immediately after training
- Signing a sheet of paper saying you attended training

Successful training for BSL-3 is

- Based on prior experience at BSL-2
- Training in BSL-3 procedures
- Competency testing to ensure that the procedures can be performed
- Participating in a risk assessment of work
- Being supervised in the laboratory until your supervisor is confident that the staff member is competent
- Full training in emergency procedures including spills training
- Annual retraining and competency testing
- Learning to promptly report incidents and near misses

Some examples of training that must be provided

- Organism risk groups
- Using biological safety cabinets
- Sterilisation and disinfection
- Use of centrifuges
- Modes of infection – aerosols
- Respiratory protection
- Handling spills and incidents
- Safety signs

Risk groups for microorganisms

Classification

- Microorganisms are classified into four risk groups, based on several criteria (termed Risk Groups 1 - 4)
- Laboratories are classified into four corresponding Physical Containment levels (termed P1 – P4 laboratories)

Risk group allocation:

- based on the **degree of hazard** to the individual, the community and the environment

Degree of hazard determined by:

- infectivity
- ease of transmissibility
- resultant effect
- host range of agent
- availability of vaccines/effective treatment

TABLE 7 Infectious dose for humans^a

Disease or agent	Dose ^b	Route of inoculation
Coxsackievirus A21	≤18 ^c	Inhalation
<i>Escherichia coli</i>	10 ⁸	Ingestion
<i>Francisella tularensis</i>	10	Inhalation
<i>Giardia lamblia</i>	10-100 cysts ^d	Ingestion
Influenza A2 virus	<790 ^c	Inhalation
Malaria	10	Intravenous
Measles	0.2 ^{c,e}	Inhalation
<i>Mycobacterium tuberculosis</i>	<10 ^f	Inhalation
Poliovirus 1	2 ^{c,e,g}	Ingestion
Q fever	10	Inhalation
<i>Salmonella typhi</i>	10 ⁵	Ingestion
Scrub typhus	3	Intradermal
<i>Shigella flexneri</i>	180	Ingestion
Shigellosis	10 ⁹	Ingestion
<i>Treponema pallidum</i>	57	Intradermal
Venezuelan encephalitis virus	1 ^{c,h}	Subcutaneous
<i>Vibrio cholerae</i>	10 ⁸	Ingestion

^aAdapted from Wedum et al., 1972.

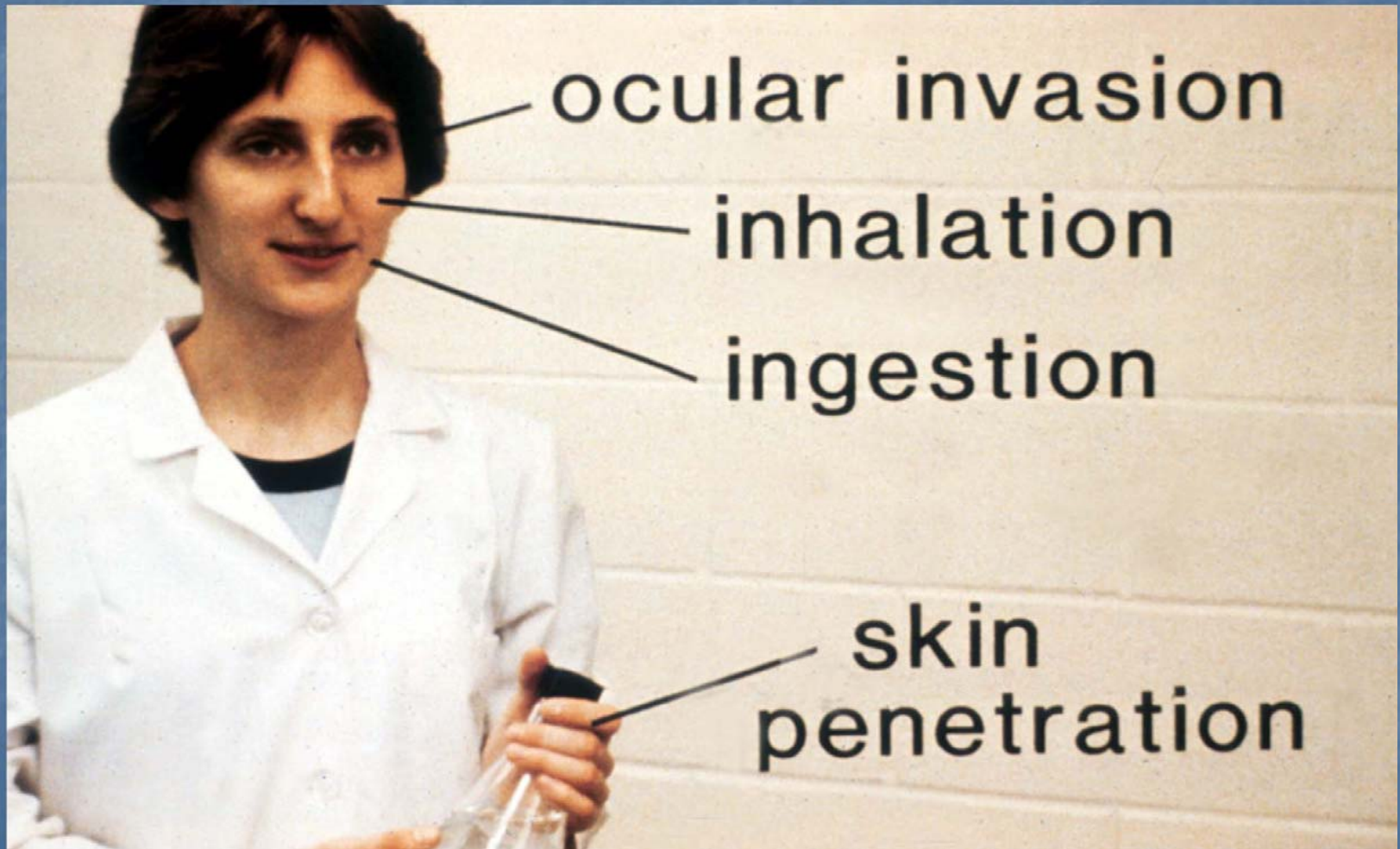
^bDose in number of organisms unless otherwise indicated.

^cMedian infectious tissue culture dose.

^dBlacklow et al., 1972.

^eIn children.

Routes of Exposure



Risk group 1

- low individual and community risk
- many microorganisms
- secondary schools, undergrad teaching

Risk group 2

- moderate individual risk
- limited community risk
- viruses, parasites, fungi, bacteria
 - Legionella, Shigella, Hepatitis B
- teaching, primary health labs, most research labs

Risk group 3

- high individual risk
- Limited to moderate community risk
- viruses, bacteria, fungi
 - JEV, SARS-CoV rabies virus, *B. anthracis*, *Brucella spp.*, *Y. pestis*, MDR *Mycobacterium tuberculosis*
- clinical pathology labs

Risk group 4

- high individual and community risk
- viruses
 - Ebola, Hendra, Nipah

Note: some require additional precautions at each level

Risk group = Physical Containment level

Each risk group must be worked at
corresponding laboratory containment
level

eg Risk Group 2 organisms --> P2 labs
(BSL-2)

The biological safety cabinet (BSC)

Structure and Function of a BSC

Principle:

- prevents escape and spread of aerosols hence forming primary barrier

Types of cabinets

- Class I - operator and environment protection
- Class II - operator, environment and product protection
- Class III - 'total' protection



Class III Biological Safety Cabinet Line





Note:

'Clean workstation', 'clean bench cabinet', or 'laminar flow cabinet' provide NO operator protection (protects product only)

Points to remember when using Class II BSC

- plan work
- minimise materials in cabinet
- minimise arm movements in and out of cabinet
- no bunsen burners
- ergonomics (OOS): short pipettes and pipettors

Quote found on side of a BSC

Cabinets have the ability to enhance the level of sterility created by the operator, but not to produce it independently of the user and the surroundings.

It should however be understood that prevention of cross-contamination is perhaps ensured more by good aseptic technique than by any miraculous action of the cabinet itself. The actions of the operator must always complement the operation of the cabinet.

Sterilisation and decontamination

Methods of Decontamination

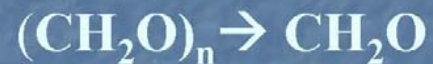
- Autoclaves
- Incineration



- Rendering
- Alkaline Digestion

Methods of Decontamination

- Decontaminating Instruments
- Lab Waste
- Digesting
- Other

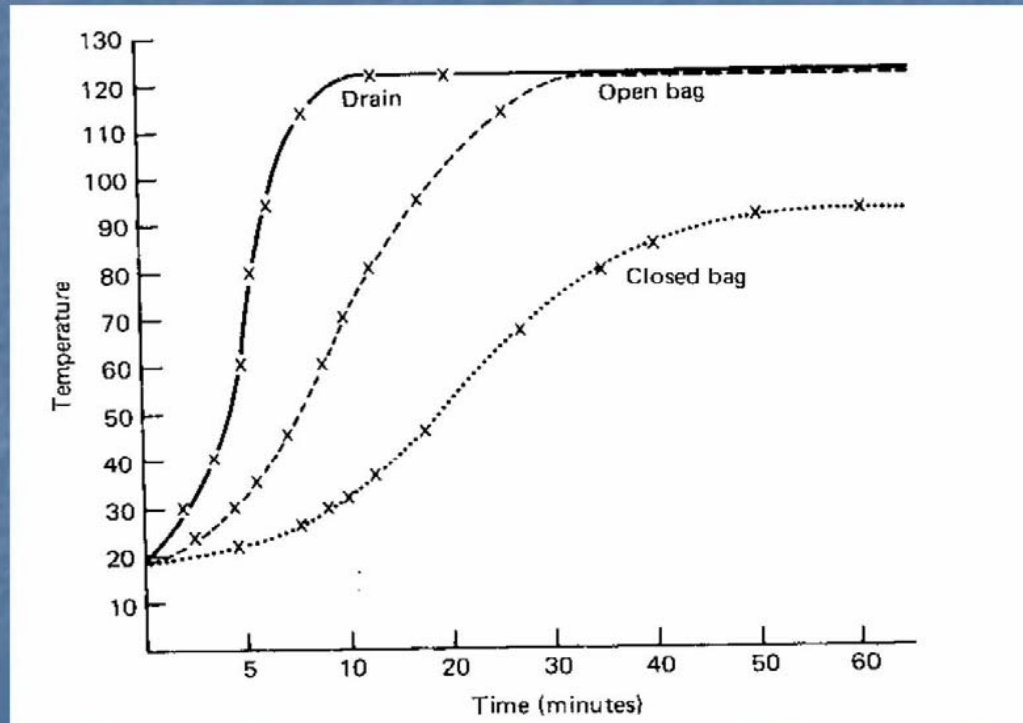


Autoclaves

- open up autoclave bags/containers
- put biohazard bags on tray
- select cycle parameters for load and allow penetration time
- hypochlorite solutions should not be autoclaved



Load penetration time (Collins CH)



Monitoring of autoclave cycles

- autoclave tape
- Browne's tubes
- biological indicators – spores
- Bowie - Dick packs
- thermocouples

Laboratory Decontamination Methods

- Formaldehyde
- Hydrogen peroxide (vapourised)
 - 30% liquid hydrogen peroxide is vapourised; vapour degrades to water vapour and oxygen
- Chlorine dioxide
 - Can be used down to 20mg/L; optimum RH 50%

Safety considerations

- Use two persons at all times
- At end of decontamination, sample air before entry (use two methods eg formaldemeter, sampling tubes specific for formaldehyde)
- Wear PPE to allow for higher levels of HCHO eg HCHO filter in a full-face mask (protects eyes also), not one for nuisance levels organic vapours



Potential problems

- Deposits (repolymerization) of paraformaldehyde on walls if temperature is too low or RH too low
- Gas absorbed by many materials eg sponge
- Off-gassing afterwards
- Pockets of gas can remain

Disinfectants

- refer Appendix E of AS/NZS 2243.3
- choose appropriate disinfectant
- make up fresh if necessary
- allow contact time

Types of disinfectants

- Halogens eg chlorine and iodine
- Aldehydes eg formaldehyde, glutaraldehyde
- Oxidising agents eg peracetic acid, hydrogen peroxide
- Alcohols eg ethanol
- Phenolics
- Quaternary ammonium compounds
- Chlorhexidine
- Acids and alkalis

Centrifuges

General and microbiological safety

- Training course
- Refresher training
- Balance tubes
- Correct placement of swing-out rotors
- Load tubes, bottles in BSC
- O-rings – in good order and greased (seal)
- Use biocontainment rotor (sealed bucket and sealed rotor)

Biocontainment buckets, tubes

Buckets with biocovers



One-touch sealing tubes



Liners for pellets

Biocontainment rotors

Dual-locking lid



Double seal with filtered caps, plugs



Hermetic seal with O-ring



Fluid containment annulus

Failed Centrifuge Rotor



Laboratory acquired infections and aerosols

Laboratory Acquired Infections

Historically:

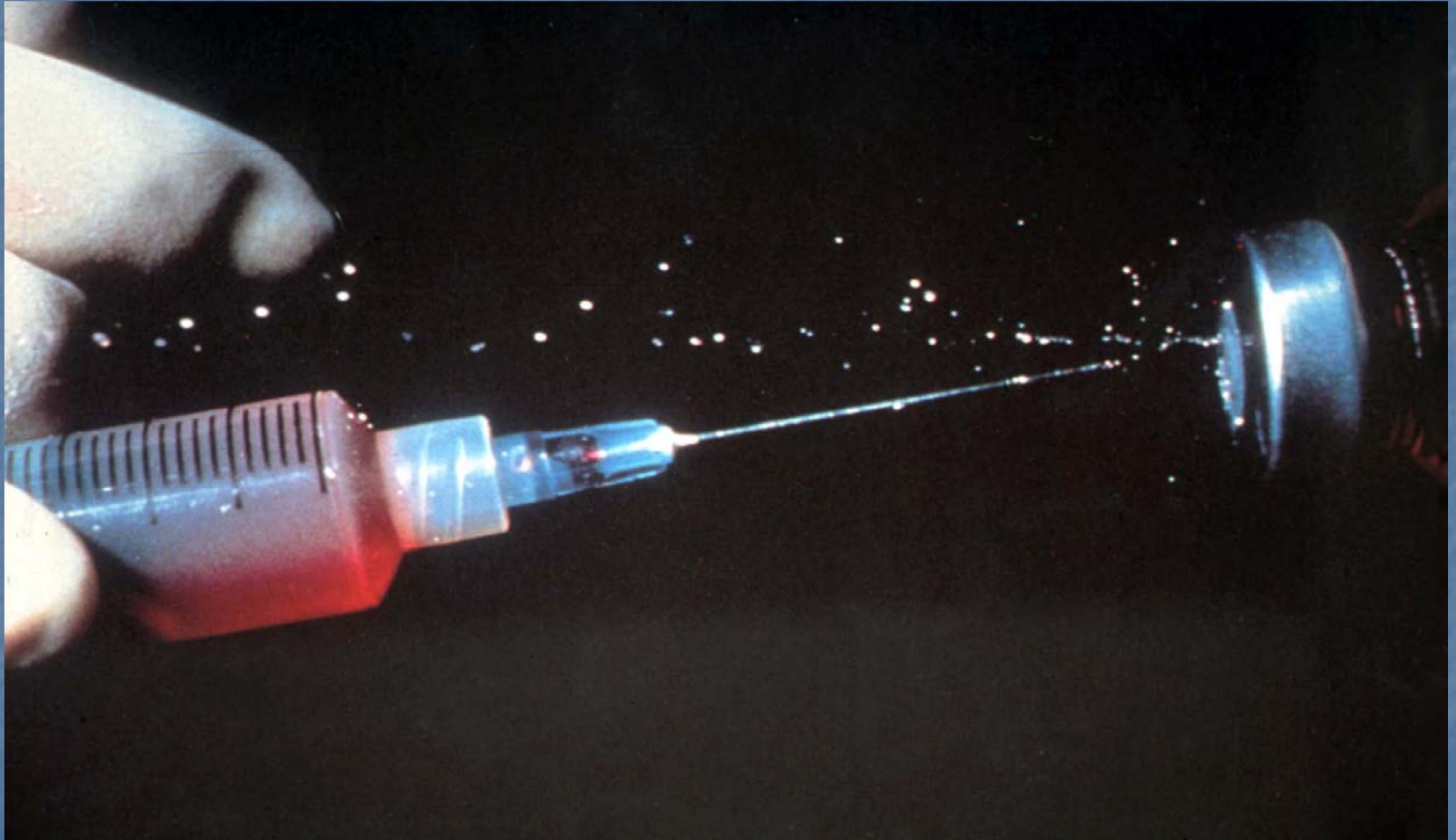
- only 20% from recognised accidents
- 80% unknown, ie no recognised accident or knowledge of how transmission occurred

Remaining 80% of infections due to aerosols

- Vortexing
- Sonication
- Homogenisation
- Dropping cultures of high titre/spills
- Blowing out drops in pipettes
- Removing needles from syringes/rubber seals



Withdrawing Syringe from Vaccine- Stoppered Bottle



Aerosol production: the invisible hazard



Breaking the surface of any liquid creates a cloud of aerosol droplets that can spread pathogens

PPE – Respiratory protection

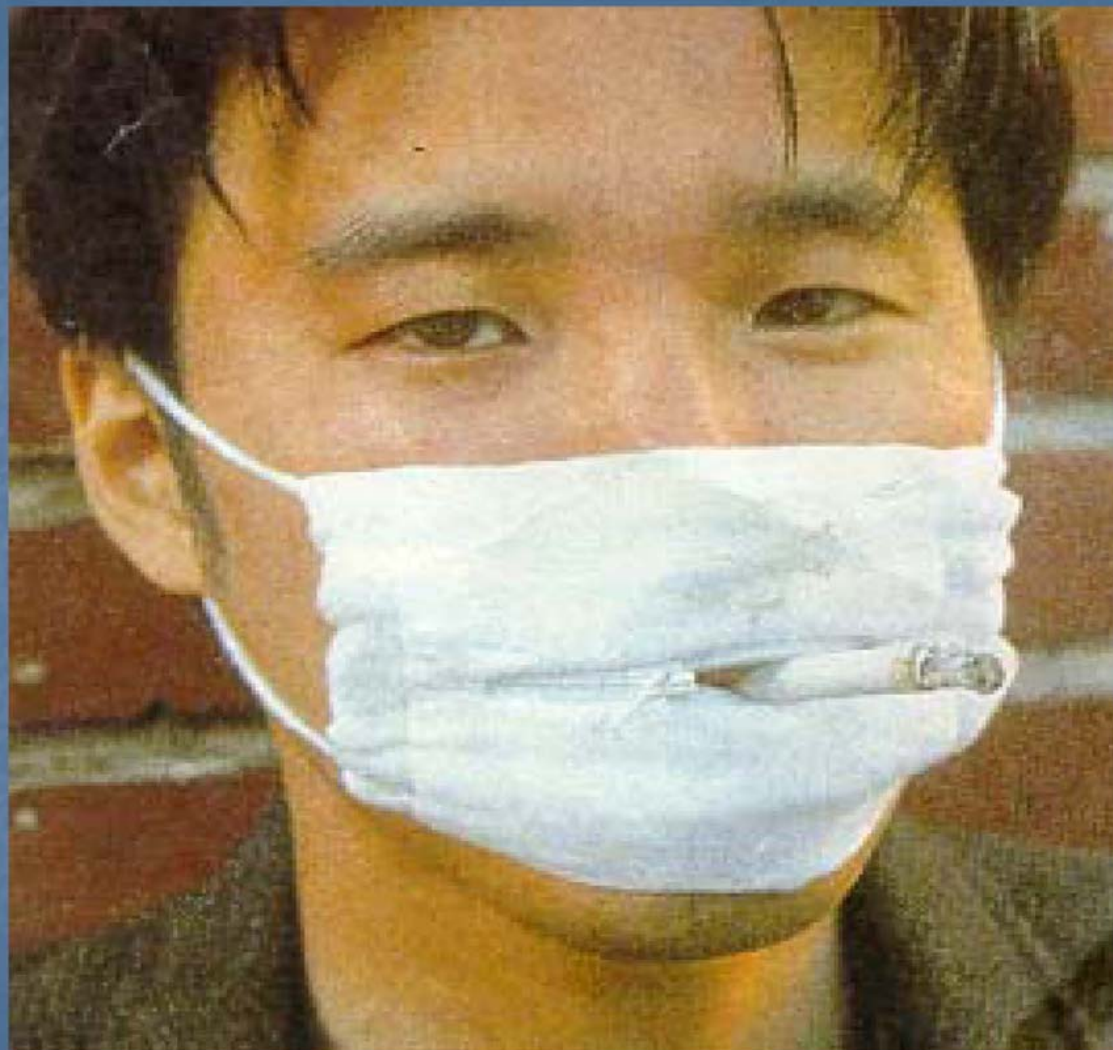
Qualitative or Quantitative Fit Testing



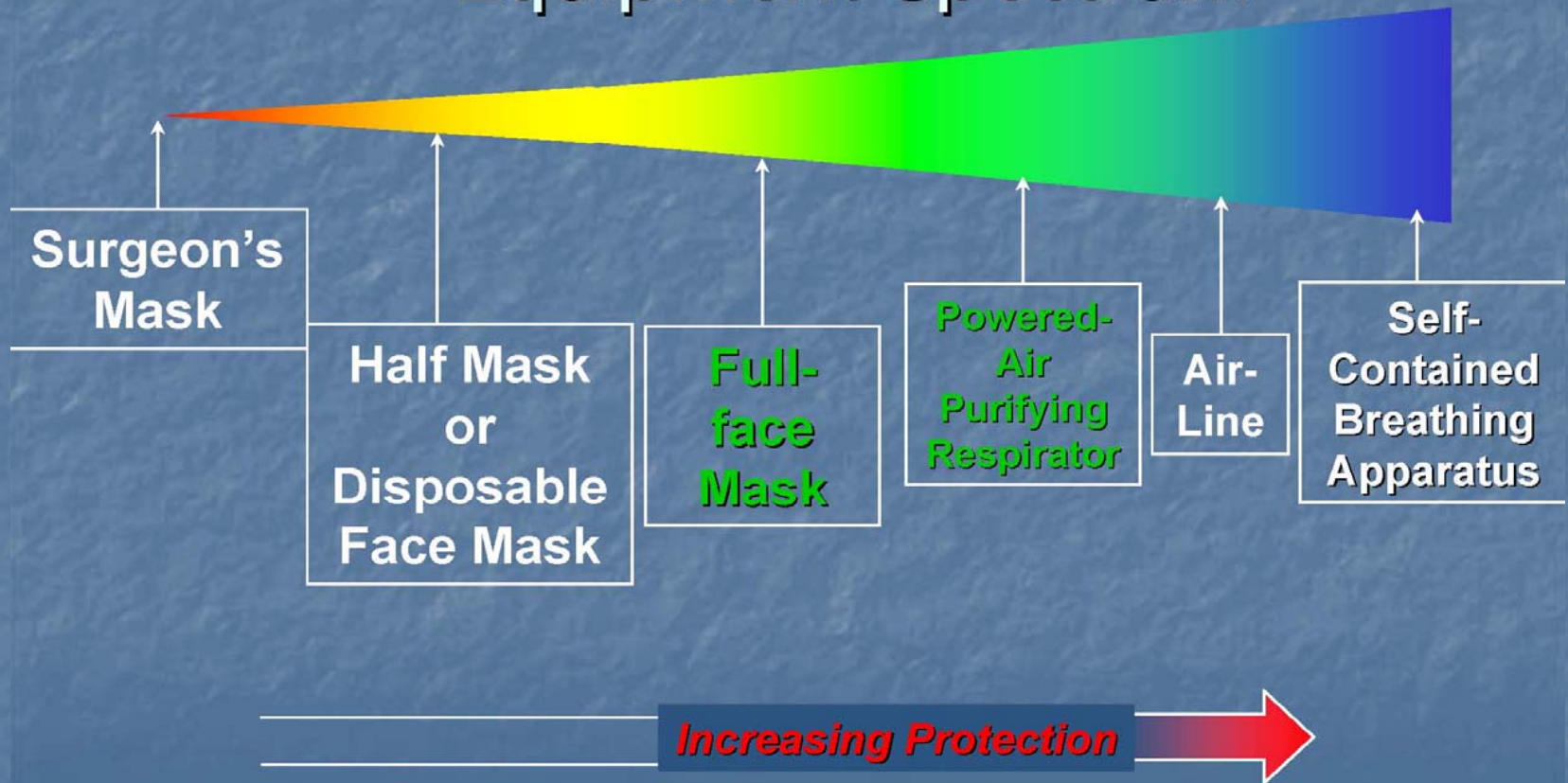
Methods to determine that an individual can get a face fit when using a certain model respirator







Respiratory Protective Equipment Spectrum



Respirator Types

Negative pressure

Positive Pressure

Half facemasks - Reusable & Maintenance Free



Supplied Air Systems

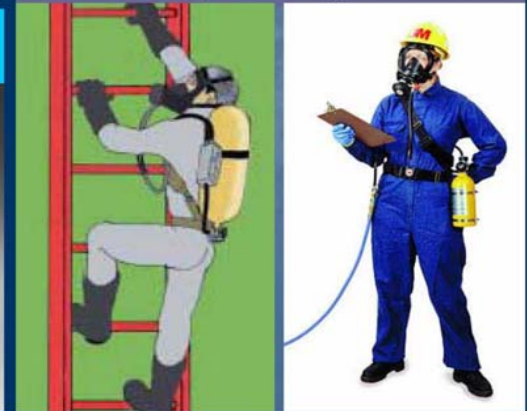


Powered Air Purifying Respirators (PAPR's)



Self Contained Breathing Apparatus (SCBA)

Full facemasks





Dealing with microbiological spills

Planning for spills

- Develop and implement spills procedure
- Train staff
- Spills clean-up team for large and/or high risk spills
- Spills clean-up kits
- Emergency clothing packs
- First-aid considerations

High risk spills in P3 labs

Note: procedure may vary with design

- Avoid breathing aerosols
- Warn others and notify external staff via emergency system
- Remove any contaminated PPE and all leave facility immediately after washing hands
- If necessary and there is an emergency shower in the airlock, take a shower on exiting (or a regular shower after exiting)

High risk spills in P3 labs

- Close outer airlock door and place biohazard warning sign with 'Do not enter' on it
- Stay out of area for at least 30 mins
- Assemble spills clean-up team (must be familiar with BSL-3 procedures) – discuss situation
- Put on appropriate PPE before entering
- Place absorbent material wetted with suitable disinfectant over spill, lay down gently from outside in

High risk spills in P3 labs

- Allow at least 10 mins
- Remove sharp objects eg glass with forceps to sharps bin
- Starting from outside, wipe towards centre of spill
- Using disinfectant, wipe over any other areas likely to have been contaminated
- Treat and dispose of waste appropriately, including PPE
- Wash hands, shower if necessary

Incidents

Process

- Immediately report incident to designated authority
 - Assumes that there is an incident reporting process in place and staff have practised it
- Incident needs to be contained and staff safety/care issues addressed (including medical monitoring, if appropriate)
- Incidents need to be investigated
- Preventative actions need to be put in place to prevent incident reoccurring
- Sign off by Safety Committee and senior management

Investigation of incident

- Gather information
- Take photos
- No blame
- Prepare a report

Reasons

- To prevent incident happening again
- To learn – other institutions also
- Report near-misses/hits
- In the event of later illness

Safety Signs

Two types of general signs

- For potentially harmful situations:
 - Warning signs: black and yellow; use of exclamation mark inside triangle plus words if required



Two types of general signs

- For potentially fatal situations:
 - Danger signs: red, black and white signs with the word 'DANGER' and appropriate words if required



Specialised warning signs

- For microbiological hazards:



Signs for PC laboratories



General sign
for
microbiological
laboratories

Hazards in this Laboratory



Protective Measures Required



Laboratory Supervisor:

Emergency: Ring CMS 5005

Healthy Safe Clean Science

Other signs



Topics covered

- The role of line management
- The role of the safety staff
- The safety is a culture, the way we work not an add on
- The importance of risk assessment
- Training requires competency testing
- We learn from laboratory incidents
- Topics covered in training