

# Biological safety manual

[Composed by the Biological Safety Officers (BVF's) at the RuG]

## VMT

Safe microbiological techniques



BIOHAZARD

**RuG**

**Arbo en Milieudienst**

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Composed by the Biological Safety Officers  
Co-ordinated by the Arbo- en Milieudienst RUG

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# Work in the Lab

## VMT

(VEILIGE MICROBIOLOGISCHE TECHNIEK = SAFE MICROBIOLOGICAL TECHNIQUES)

Safety measures for activities with Genetically Modified  
Organisms  
(GMO's = GGO's in Dutch)  
at VMT level

Activities with GGO's can only be performed by those  
people that have permission from the Biological Safety  
Officer (BSO = BVF in Dutch)

## VMT

(VEILIGE MICROBIOLOGISCHE TECHNIEK = SAFE MICROBIOLOGICAL TECHNIQUES)

### Goal

**No genetically engineered  
Organism leaves the lab uncontrolled**

<b>Required</b>	Designated lab coats Disinfect bench top daily (70% alcohol) Keep lab clean Autoclave biological waste Keep floor free of bottles, etc. Close door of lab
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<b>Prohibited</b>	Food & drinks and paraphernalia Coats, etc Excess chemicals in the lab Oral pipetting
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<b>Prevent</b>	Aerosol formation (syringes!) Spilling (clean immediately!)
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## VMT

### Lab coats

**LAB COATS HAVE TO BE WORN AT ALL TIMES IN  
A VMT LAB:**

**PLEASE: NO EXCEPTIONS !!!!!**

# Decontamination

**Decontamination basically means:  
Contain and inactivate the hazard!**

**Contain:**

- \* Define workarea (use trays if possible)
- \* Have absorbent material within reach
- \* Collect all contaminated material for sterilization (autoclave)

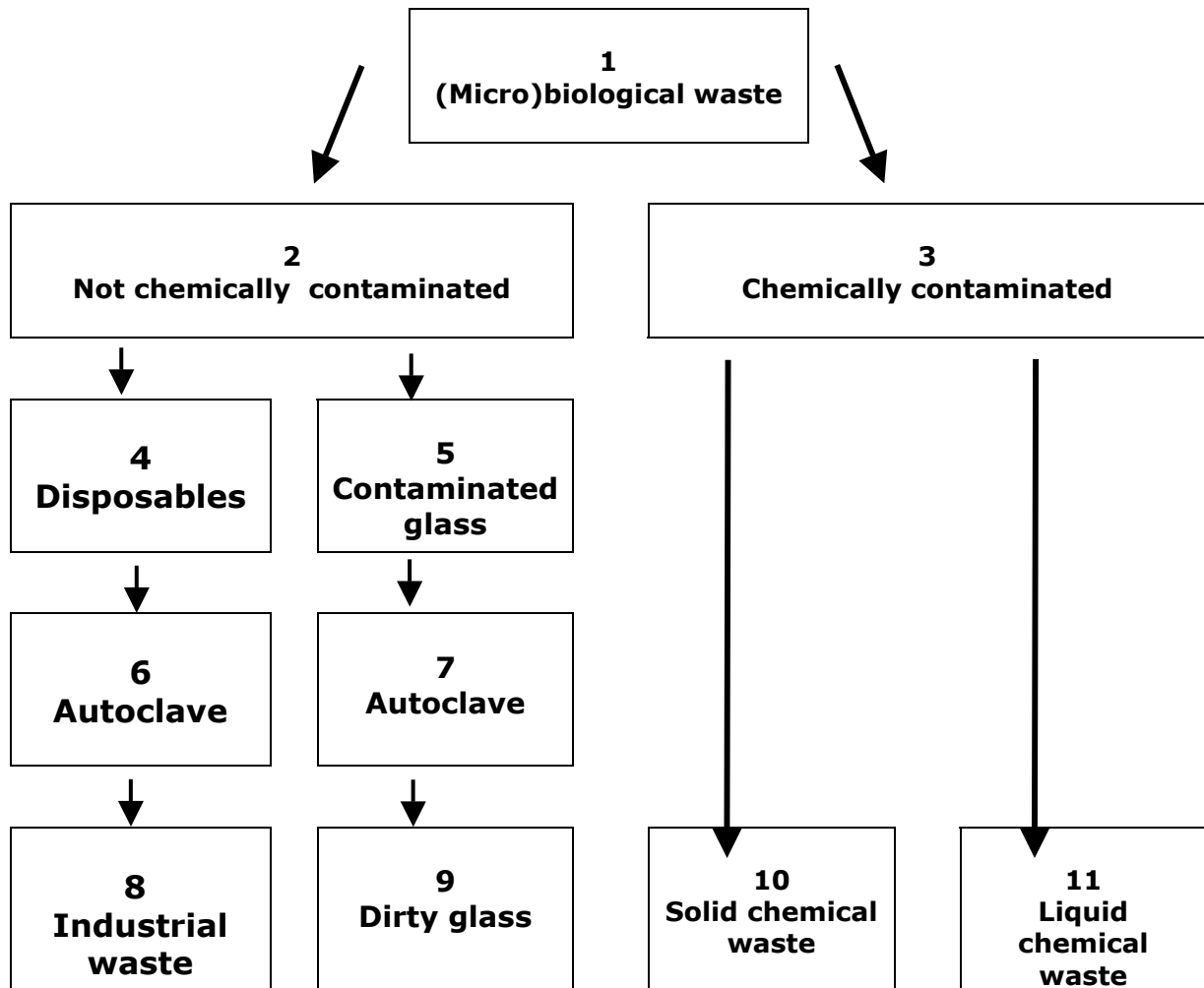
**Inactivate:**

- \* Clean benchtops with 70% EtOH
- \* Use bleach (tablets or liquid, at least 1 hour) to inactivate cultures and waste collecting flasks !!!
- \* Autoclave all solid waste, including (bacterial) agar plates. At least 25 minutes at 120 °C

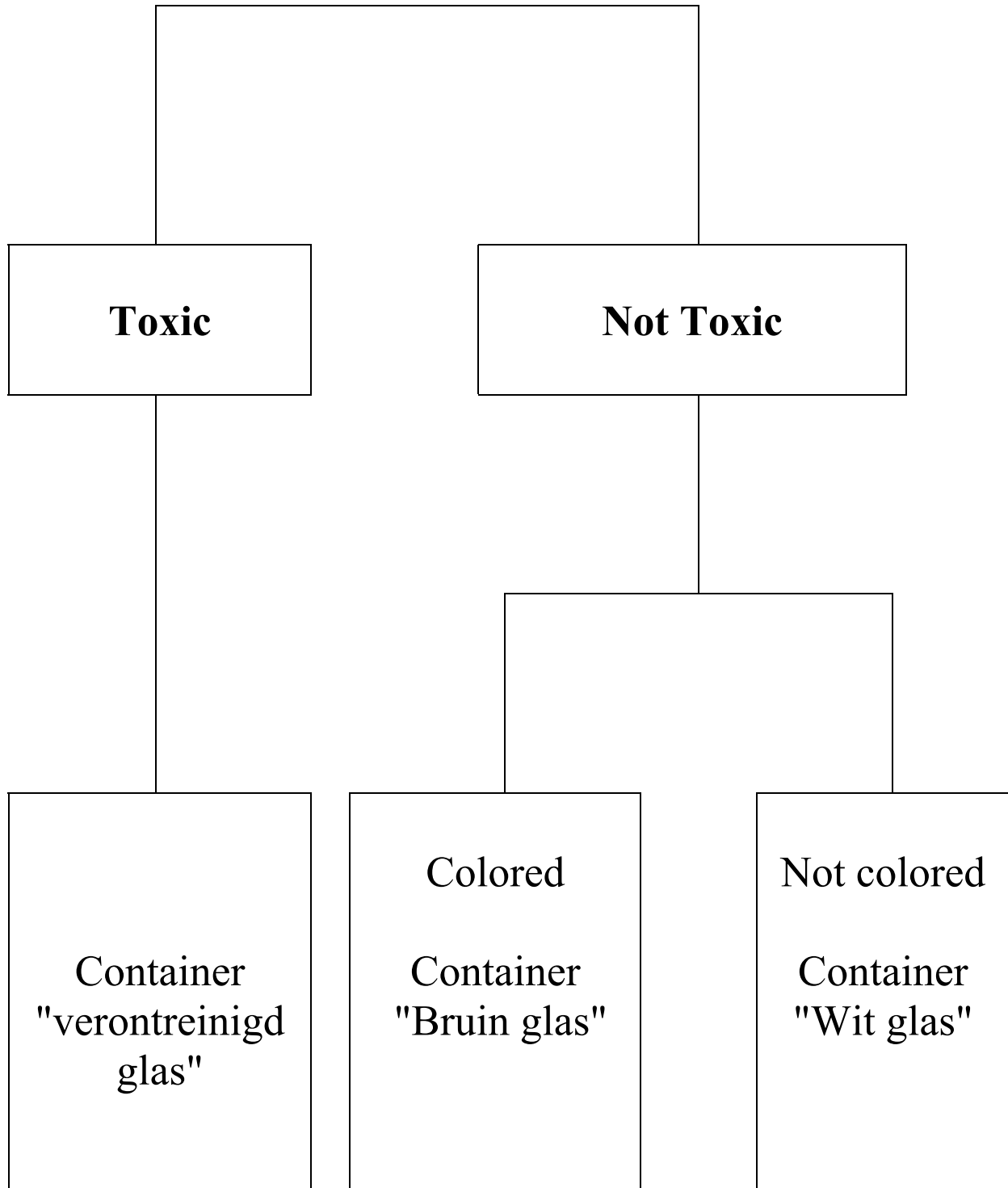
**Wash hands with soap whenever leaving the lab !!!!**

# VMT

## Disposal of biological waste



# Disposal of glass waste



# **10 COMMANDMENTS for VMT**

## **(safe microbiological techniques)**

- 1. All VMT related work can only be performed by those people that have permission from the Biological Safety Officer (BVF). Work according to the rules, even if you believe there is no apparent risk.**
- 2. During VMT related work all doors and windows have to be closed. Verify that insects and other pests are not present in the lab.**
- 3. Wear a closed laboratory coat. Do not take this labcoat outside the VMT area unless it is absolutely necessary for the experiment. In case of a contamination of the labcoat, sterilize the labcoat first, with bleach or by autoclaving, before washing.**
- 4. Clean and sterilize spills immediately. Report serious contamination immediately to the BVF.**
- 5. It is absolutely prohibited to eat, drink or smoke, or to have cups, plates, mugs or silverware in the VMT area.**
- 6. Pipetting by mouth is prohibited. Used pipettes are collected in a disinfecting solution.**
- 7. Prevent aerosols. These may be created by -splashing drops; -pouring of liquids; - discharging pipettes; -opening of moist plugs; - using inoculating loops that are too hot. Use needles only if there is absolutely no alternative.**
- 8. Glassware and instruments that have been in contact with GMO's (Genetically Modified Organisms, GGO's in Dutch) have to be sterilized or disinfected before being washed, reused or discarded. Biological waste has to be collected in autoclavable plastic bags, which are autoclaved before discarding (use indicator tape to demonstrate that the bag was autoclaved).**
- 9. Wash hands with soap and water after work and before leaving the room. Bench surface areas have to be cleaned and disinfected daily. Keep area clean and organized.**
- 10. Record the general nature of the work clearly in a lab journal.**

# **10 GEBODEN voor VMT**

(veilige microbiologische techniek)

- 1. De werkzaamheden mogen uitsluitend worden verricht door personen die daartoe toestemming hebben van de BVF. Werk volgens de regels, ook als u denkt dat er geen risico is.**
- 2. Tijdens het werk dienen deuren en ramen gesloten te zijn. Controleer de werkruimte op afwezigheid van insecten en ander ongedierte.**
- 3. Draag gesloten laboratoriumkleding. Het dragen van deze labjas buiten de werkruimte is alleen toegestaan waar gedeelten van het experiment worden uitgevoerd. Indien de labjas door morsen of ongeval besmet raakt, wordt deze vòòr het wassen ontsmet of gesteriliseerd.**
- 4. In geval van besmetting direct ontsmetten; ernstige besmettingen dienen direct gerapporteerd te worden aan de BVF.**
- 5. Eten, drinken en roken, de aanwezigheid van eet- of drinkgerei in de werkruimte is verboden.**
- 6. Pipetteren met de mond is verboden; deponeer gebruikte pipetten in een desinfecterende vloeistof.**
- 7. Vermijd aerosolvorming. Handelingen waarbij aerosolen ontstaan zijn: - vallende druppels; -gieten van vloeistoffen; - uitblazen van pipetten; - openen van vochtige stoppen; - centrifugeren van open buizen; - te hete entnaalden (verkeerd gebruik). Gebruik van injectienaalden mag alleen indien daarvoor geen alternatief is.**
- 8. Glaswerk en instrumenten die in aanraking zijn geweest met GGO=s worden gesteriliseerd of ontsmet voordat zij worden gewassen, hergebruikt of afgevoerd. Biologisch afval wordt gedeponneerd in een steriliseerbare afvalzak voorzien van indicatortape en gesteriliseerd.**
- 9. Was de handen met zeep na afloop van de werkzaamheden. Werkoppervlakken dienen aan het einde van iedere werkdag ontsmet te worden. Werkruimte schoon en netjes houden.**
- 10. De aard van de werkzaamheden dient overzichtelijk in het labjournaal geregistreerd te worden.**

## Paper work

(to the disposal of the BVF, the Biological Safety Officer)

<b>Secretariat BVF</b>	<b>Units</b>	<b>Everybody</b>
Permits	List of:	Lab journal
List and records of VMT workers	-Strains	
Reports	-Vectors	
	-Constructs	
	-GMO's (GGO's in Dutch)	

Labjournals should be understandable by others (note dates, sources, etc). Identify labjournals (give it a number or code) and use page numbers.

Per unit, a list of strains, vectors and constructs that are used for longer than a few weeks should be maintained (e.g., use first labmeeting of the month for entering records!!).

Please check with BVF if permits are still in order.

## Example forms

Following are some example lists for keeping records. Master copies are available from BVF.

### Strains

<b>date</b>	<b>ID</b>	<b>Short description</b>	<b>Lab journal reference</b>	<b>Store</b>
<i>XX-XX</i>	<i>XL1-blue</i>	<i>E.coli strain from Stratagene</i>	<i>catalogue Stratagene</i>	<i>-80 glycerol</i>

### Vectors

<b>date</b>	<b>ID</b>	<b>Short description</b>	<b>Lab journal reference</b>	<b>Store</b>
<i>XX-XX</i>	<i>pBR322</i>	<i>Amp resistant plasmid for cloning in Ecoli</i>	<i>page 001 A01 John Doe</i>	<i>-80 DNA</i>

## Constructs

<b>date</b>	<b>ID</b>	<b>Short description</b>	<b>Lab journal reference</b>	<b>Store</b>
<i>XX-XX</i>	<i>pML001</i>	<i>Phosphatase gene in pBR322</i>	<i>page 001, A01 John Doe</i>	<i>-80 DNA</i>

## GGO's (Genetically modified organisms)

<b>date</b>	<b>ID</b>	<b>Short description</b>	<b>Lab journal reference</b>	<b>Store</b>
<i>XX-XX</i>	<i>ML001 in XL1-blue</i>	<i>Phosphatase gene in amp resist. plasmid in E.coli strain</i>	<i>page 001 A01 John Doe</i>	<i>-80 glycerol</i>

# CONTACTS AND EMERGENCIES

## BVF's (Biological Safety Officers) at the RuG:

		Room nr.	Tel.nr. 050-
Nijenborgh 4 (Groningen)	Dr. M.H.K. Linskens	5114.0132	3637290
	Prof. dr. D.B. Janssen	5115.0103	3634008
A.Deusinglaan 1 A.Deusinglaan 50 (Groningen)	Dr. A. J. Scheffer	AZG 31.2.09	3633515
	Drs. W. Hofstra	3122.N.106	3633032 *81041
Biologisch Centrum (Haren)	Dr. O.M.H. de Vries	6521.0144B	3632328
	Dr. J.S. Lolkema	6514.0236B	3632155
	Dr. W.T. Stam	6515.0050A	3632252

### E-mail addresses of BVF's

[W.Hofstra@med.rug.nl](mailto:W.Hofstra@med.rug.nl)

[D.B.Janssen@chem.rug.nl](mailto:D.B.Janssen@chem.rug.nl)

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## EMERGENCY telephone numbers

First aid: **8050**

Fire: **8050**

## VMT Emergency procedures

- \* Contain spills
- \* Clear area
- \* In case of personal injury: call first aid **8050**
- \* In case of fire: call **8050**

*In all cases:*

- \* *Report incidents involving GGO's to BVF*

## SUMMARY OF RESPONSIBILITIES

- \* BVF is responsible for infrastructure, check-ups and provides information
- \* VM applies for permit or gives notification, and insures record keeping and good lab practices in the group
- \* VMT worker (e.g. YOU) remains responsible for performing VMT work according to applicable procedures and reports problems to VM or BVF

### Literature and useful links:

- Regeling Genetisch Gemodificeerde Organismen en Richtlijnen van de COGEM bij deze Regeling (juni 1998; Commissie Genetische Modificatie)
- Administratieve organisatie Biologische Veiligheid RUG (april 2000)

### Useful links:

- <http://www.minvrom.nl/minvrom/>
- <http://www.rivm.nl/csr/bggo.html>
- <http://www.icgeb.trieste.it/biosafety/bsfdata1.htm>  
Biosafety: This database contains literature on biosafety and Biotechnological risk assessment.