Status: July 2013

DIRECTIVE
ACCORDING TO §12 „GENTECHNIK- SICHERHEITSVERORDNUNG“
FOR LABORATORIES OF SAFETY LEVEL S1

Contents

1. Area of application
2. Reponsible individuals
3. Gene technological procedures
4. Danger potential of GMOs
5. Protective measures, rules of conduct and hygienic measures
- **5.1 Access regulations**
- **5.2 General procedures**
- **5.3 Additional rules**
- **5.4 Hygienic measures**
- **5.5 Prohibitions**
- **5.6 Personal protective equipment**
- **5.7 Special regulations**

6. Behaviour in case of danger
- **6.1 Spilling of biological material**
- **6.2 Fire**

7. First aid
8. Appropriate disposal procedures
9. Links to general rules and standards
10. Special regulations

---

1. Area of application

This directive applies to the gentechnological facilities in the Institute for Physical Chemistry of the University of Muenster, Corrensstraße 28/30. The following rooms are covered:

Laboratories:	1. floor; rooms E119, 120, 122, 124, 133, 134, 135, 136, 138
Autoclave room:	1. Floor, room E133
Storage room:	1. floor, room E122

These rooms are marked by signs „Genlabor S1“ or „Gentechnik-Arbeitsbereich S1“ on the doors.

The social room for breaks is E114.
Bathrooms/changing rooms are in room EK02 („Umkleide“, “Waschraum Damen”) and EK05 (“Umkleide”, “Waschraum Herren”).

2. Reponsible individuals

Project leader:	Prof. Dr. Dagmar Klostermeier Tel. 0251 83 23421	
Biological safety representative:	PD Dr. Joachim Kremerskothen, Tel. 0251 83 25780	
Emergency call	112	
University medical service:	Occupational health service of the University and University hospital	Tel. 83 56081 Tel. 83 55826
	Dr. Saße	Tel. 83 55968
First aid:	Bremann, Daniela	Tel. 83 29166
	Cramer-Kellers, Cornelia	Tel. 83 23412
	Dorstewitz, Julia	Tel. 83 23427
	Feldhues, Hendrik	Tel. 83 29181
	Guddorf, Jessica	Tel. 83 29166
	Steif, Christian	Tel. 83 23422
Hospital:	University hospital Münster	
Safety representative*:*	Herr Euler, Dezernat 4.5	
Fire safety representative:	central**:** Frau Kraus-Brauckmann, Dezernat 4.5,	Tel. 83 30302
	Institute: Frau Dorstewitz	Tel. 83 23427
Fire arms:	112	
University representative:	Herr Wolfgang Mette, Dezernat 4.5 Tel. 83 25795	

3. Gentechnological procedures

The following gentechnological procedures of safety level 1 are performed in the facilities:

Cloning and mutagenesis of genes that code for proteins interacting with nucleic acids for the preparation of recombinant proteins for fluorescence experiments.
Single-molecule FRET experiments ar performed with e.hg. helicases and topoisomerases to investigate their dynamics. PCR-cloning of helicase and toposomerase genes, PCR mutagenesis for the introduction of cysteines for fluorescent labeling.

Gentechnological procedures comprise the genreation, handling, amplification, storage and disposal of genetically modified organisms (GMOs) as well as transport within the facilities.

4. Danger potential by GMOs

The GMOs are classified as risk level 1, meaning that they pose no danger for healthy individuals when handled according to this directive.
A comprehensive risk assessment is part of the documentation according to the „Gentechnik-Aufzeichnungsverordnung“.

5. Protective measures, rules of conduct, and hygenic measures

According to the basic rules of handling microorganisms, and the „Gentechnik-Sicherheitsverordnung“ the following rules apply:

5.1 Access regulations

a) Only individuals who have been instructed about the dangers and the required project-specific safety measures are permitted to work in the lab. Safety instructions according to the directive are required before starting the work, and have to be repreated in annual intervals, and should be related to the specific type of work. This regulation applies to all persons working in the lab, even if they are not directly involved in work with GMOs.

b) Visitors are only allowed to enter the lab in company of a safety-trained individual.

c) Cleaning, maintenance and repair personnel is only allowed to perform work in the lab after authorization by the project leader and instruction about potential dangers.
(Be aware of change in personnel.)
An instruction about the type of work performed in the lab and the main rules of conduct is sufficient.
A contact person hast o be available at short notice at any time.

Contact person is Prof. Dr. Dagmar Klostermeier, Tel. 83 23421

5.2 General procedures

a) Individuals worling in the lab have to make sure they know the location and use of desinfectants, body and eye showers, first-aid equipment, fire extinguishing equipment, as well as escape routes

b) All laboratories within the S1 area have to kept organized and clean. Material on the benchtop should be limited to instruments and material absoluteley needed at any given time. Stocks have to be stored in the designated storage areas.
During handling of dangerous substances (chemical reagents, flammable liquids, pressurized gases, etc.) the legally required protective measures have to be applied ("Gefahrstoffverordnung", "Technische Regeln für Gefahrstoffe" etc.).

c) Use of desk areas within the lab hast o be limited to protocoling. They are neither to be used for gentechnical procedures nor for office tasks beyond protocoling. The presence of catalogs, books ec. has to be limited to the absolutely necessary.

d) Doors to the laboratoriers should be kept close during gentechnological work. Windows may be opened for aeration as long as it does not adversely affect the functionality of hoods.

e) Pipetting has to be done with pipetting helpers.

f) The use of syringes, needles, blades etc. should be minimized. Sharps have to be collected and disposed of in impenetrable, autoclavable containers.
Needles must not be kinked or reinserted into the protective haft. Containers have to be supplied at the required bech spaces before the experiments are started.

g) In the microscopy compartments of laboratories 122 and 124, only optical experiments with GMOs are permitted.

h) During all experimental work, aerosole formation has to be avoided. Relevant proceses where aerosole formation is possible include stirring, high pressure applications, inoculation, shaking, pipetting, centrifugation and experiments involving ultrasound.

Possible measures to avoid aerosole formation:
-use closed flasks,
-wait long enough for aerosoles to settle before opening,
-avoid formation of bubbles,
-maintain low heights of fall when pouring or pipetting liquids
-don´t blow out pipettes or expunge syringe contents into the air

i) When working with GMOs of risk group 1 with sensitizing or toxic effects, meaures minimizing exposure of employees have to be taken (e.g. safety bench, respiratory protection or avoidance of spore forming phases of fungi, etc.))

j) The identity of organisms used hast o be confirmed at regular intervals

k) User manuals for centrifuges and autoclaves (short versions fom the manufacturer) have to be followed.

l) For internal transport of GMOs, closed, unbreakable and labeled containers have to be used.

m) The GMOs are stored in a – 80°C freezer in laboratory E122.

5.3 Additional rules

a) Directive Dangerous Chemicals according to §20 GefahrstoffV/TRGS 555
b) Directive Centrifuges according to UVV "Zentrifugen"
c) Directive Autoclaves
d) „Strahlenschutzanweisung" according to „Strahlenschutzverordnung"
e) Directive "Storage with and handling of luquid nitrogen/CO_2"

5.4 Hygienic measures

a) Bench surfaces have to be cleaned after finishing the experimental work. (Terralin Protect Ready-to-use solution, 15 min, 2%).
b) After finishing the laboratory work, hands have to be desinfected prophylactically, and then washed. (Sterilium 30 s-1 min).
c) Instruments have to be cleaned prophylactically at regular intervals (Kohrsolin solution, 3%, 4h).
d) To initiate eradication measures, the project leader hast o be informed in case of vermin appearance.

5.5 Prohibitions

a) Food and cosmetics must not be stored within laboratories. Sorage of these items has to be confined to social room E114.
b) Smoking, eating or drinking are prohibited in the laboratories. The social room E114 is available for breaks. The social room must not be entered with lab coats.
c) Mouth pipetting is prohibited.
d) Suction devices (e.g. water-jet vacuum pumps) must not be used for liquids that may contain GMOs, unless an upstream sterile filter prevents leakage of GMOs.

5.6 Personal protective equipment

a) In the S1 laboratories, lab coats are required that have to be washed regularly.
   Lab coats and gloves have to be taken off within the S1 area before leaving.
b) Suitable gloves have to be chosen according to their resistance to the respective chemicals in use. Single-use gloves have to be disposed of after using.
c) To avoid contaminaton, regular clothing and lab coats have tob e stored separately:
   Lab coats: within the lab
   Regular clothing: lockers outside the lab

6. Behaviour in case of danger

- Stay calm, avoid hasty, unreflected reactions.

- Warn endangered persons, ask them to leave the room if required.

- Stop dangerous experiments, switch off gas, water, electricity, if required. Cooling water has to be left on.

- The project leader hast o be notified of any emergency situation.

6.1 Spilling of biological material

If biological material is spilt, the affected area hast o be cordoned off. Spilt material that may contain GMOs has to be inactivated immediately.

Decontamination measures:

- Surfaces: Wear protective gloves. Take up spilt material with autoclavable tissues and autoclave. Desinfect the contaminated area with Kohrsolin solution (3%, 4h).
- Instruments: In case of glass breakage, shards have tob e desinfected, and to be disposed off using protective gloves and suitable tools.
  Wear protective gloves. Take up spilt material with autoclavable tissues and autoclave. Desinfect the contaminated instrument with Kohrsolin solution (3%, 4h).
- Clothing: Take of contaminated clothes. First autoclave, then wash clothes.

- Skin: Desinfect affected skin with Sterilium, rinse off with (plenty of) water after sufficient residence time (30 s- 1 min).

- Eyes: Rinse eyes with plenty of water (eye shower), consult optomoterist if necessary.

-Mucosa: Rinse mucosa with plenty of water, consult doctor if necessary.

6.2 Fire

Small fires can be extnguished with fire extinguishers present in every room. In other cases, the emergency plans have to be followed.

7. First aid

- injuries

- Wounds should be desinfected and bandaged immediately as part of the primary care.
- The project leader has to be notified immediately.
- Upon exposure or the possibility of exposure to hazardous substances, a doctor has to be consulted.
- Injuries occuring during work with GMOs have to be documented. The documentation has to be stored for at least 10 years.

- Inhalation of swallowing of GMOs

The project leader hast o be notified immediately. A doctor hast o be consulted to decice whether treatment is required. The project leader and the doctor have to be notified of the identity and amount of organisms that have been taken up.

8. Appropriate disposal procedures

Before starting the experimental work, a disposal container with an autoclavable bag has to be provided on the benchtop.
Solid and liquid waste containing GMOs hast o be inactivated before it is disposed of. Inactivation is achieved e.g. with autoclaving at 121°C for 20 min. Autoclaves are located in room 133. Collected waste will be autoclaved at regular intervals.

9. Links to general rules and standards

Laws/Orders/Technical rules

- „Gentechnikgesetz“ (GenTG)
  - „Gentechnik-Sicherheitsverordnung“ (GenTSV)
  - „Gentechnik-Aufzeichnungsverordnung“ (GenTAufzV)

- „Arbeitsschutzgesetz“

- „Biostoffverordnung“ (BioStoffV)
  - „Technische Regeln für Biologische Arbeitsstoffe“ (TRBA)

- „Gefahrstoffverordnung“ (GefStoffV)
  - „Technische Regeln für Gefahrstoffe“ (TRG)

- „Mutterschutzgesetz“ (MuSchG)
  - „Mutterschutzrichtlinienverordnung“ (MuSchRiV)

Rules from occupational cooperatives

- General rules
- Occupational medicine-prophylaxis
- Biologal material
- Laboratories
- Centrifuges

These and other rules related to occupational health can be found at http://www.uni-muenster.de/Rektorat/Sicherheit/gvv/gvv_inx.htm

Publications of the Robert Koch Institute and the DGHM

- List of donor- and recipient organisms that have been assesssed with respect to their risk for gentechnological work
- List of desinfectants and desinfection procedures tested and recognized by the Robert Koch Institute
- List of desinfectants by the German Society for Hygiene and Microbiology

10. Special regulations

- Duty of Notification

The project leader hast o be notified of any event that does not correspond tot he expected cours of gentechnological work.

- Safety training

Emloyees have to be trained according to this directive before starting, and at regular intervals during the work (at least once a year). The training should be related tot he work performed. Content and time have to be documented. Employees have to confirm their participation by signature.

- Duty of documentation

Only S1 work is alllowed in the gentechnological facilities. All experiments hav eto be documented. Donor and recipient organism, GMO, vector and transferred Gene are essential poarts of risk assessment, and this information has to be part of the documentation. Documentation has to be kept at least 10 years after finishing gentechnological work.

- Penalties

In case of violation of the rules of the „Gentechnikrechts“, penalties of up to 50.000,- € 5 years of imprisonment can apply. Damage compensation up to 85.000.000,- € may due.

11. Updates

The content of this directive corresponds to the German version from 23.4.2013
Version: *10.7.2013* was edited. New version:

12.

Münster, 10.7.2013

Prof. Dr. Dagmar Klostermeier .....

....

....

(Name/s und signature/s project leader/s)