- 1. Working with biological agents
- 2. Handling of hazardous substances
- 3. Actions in case of accidents in the lab
- 4. Working with lasers
- 5. General behavior in case of emergency

When handling biological agents in the laboratory, special rules of conduct and compliance with certain protective regulations must be observed.



Symbol for biohazard

#### Area of application

cell culture lab (r. 117)
storage room (r. 116 + ante-room)
MRE lab (r. 115)
AFM labs (r. 131 a/b + ante-room)
SpiDi lab (r. 132)
BONG lab (r. 309)
chemistry lab (r. 310)
OS lab (r. 311)

These rooms are labelled as

"BIO II" and

"Gentechnischer Arbeitsbereich S1"

C	BIO II	
Sch	utzstufe 2 nach Bic	StoffV
	und	
GE	NTECHNIS	CHER
AR	BEITSBER	EICH
	S1	
Tür	geschlossen h	alten!

#### **Responsible people**

phone

responsible for genetic and bio labs: Prof. Käs

responsible for biological safety: Dr. Weizenmann

emergency call / technical service:

medical office:

<u>regulatory authority:</u> Saxon State Ministry for Energy, Climate Protection, Environment and Agriculture

0341/9732470

0341/9432592

0341/9734333

0341/9938-00

see operating instructions (page 2)

#### biological agents in general



risk group 1 (e.g. bacteria, yeast)

risk group 2



risk group 3 (e.g. HIV, SARS-CoV-2)

(e.g. salmonella, hepatitis A)

risk group 4 (e.g. variola, Ebola)

https://www.svlfg.de/biologische-arbeitsstoffe

#### genetically modified organisms (GMOs)

## Genetically Modified Organism (GMO)

**Bacteria, Plants, Animals, Viruses** 



organism whose genetic material has been altered in a way that does not occur naturally (crossing or natural recombination) and is capable of replication or of transferring genetic material

https://microbenotes.com/genetically-modified-organism/

#### **Risk classification**

Bio-substances / GMOs are classified into one of the following risk groups according to the risk of infection they pose, based on the state of scientific knowledge:

**Risk group 1:** they are unlikely to cause disease

**Risk group 2:** they can cause a human disease and pose a risk to workers; they are unlikely to spread to the population; effective prevention or treatment is possible

**Risk group 3:** they can cause a serious human disease and pose a serious risk to workers ... effective prevention or treatment possible

**Risk group 4:** they can cause a serious human disease and pose a serious risk to workers ... effective prevention or treatment **not** possible

## 1. Biological agents – Risk classification

## **Biological agents in our group are:**

cell lines and genetically modified organisms (GMOs)

→ risk group 1

- → safety level S1 accordingly Genetic Engineering Safety Regulations (GenTSV)
- commercially acquired cell lines
- already genetically modified cell lines from other laboratories
- cells, transfected in our cell culture lab

primary tissue samples from university hospital

- risk group 3\*\* (pathogens are not transmitted by air)
- → safety level S2 accordingly Regulation on Biological Agents (BioStoffV)
- human tissue samples directly obtained after surgery

## **1.** Biological agents – Risk classification

**Before starting work,** a risk assessment (determination of the hazard potential) must be carried out for the planned work, from which the safety classification of the work results.

In the case of new obtainment of biological agents (cell lines, GMOs, primary tissue etc.), you have to inform the responsible of the lab about the planned work in order to classify the potential risk.

Responsible of the lab: Undine Dietrich, room 315, tel. 32472 (<u>dietrich@physik.uni-leipzig.de</u>)

Likewise the Biosafety Officer (BBS) has to be informed.

BBS of the faculty: Nicole Weizenmann, room 423, tel. 32520 (weizenmann@uni-leipzig.de)

#### Before starting work:

- receive the safety instruction
- get briefing from a trained employee
- inform about potential danger (see also chapter 2 and 3)

Access to laboratories only after safety briefing and

for visitors only in the presence of trained employees.

https://microbiozindia.com/lab-safety-rules-in-pharmaceuticals-all-you-need-to-know/



#### Before starting work:

- inform yourself about
  - operating instruction (Betriebsanweisung)
  - skin protection plan (Hautschutzplan)
  - hygiene plan (Hygieneplan)
  - emergency plan (Notfallplan)

(instructions in every lab and on our PWM website  $\rightarrow \rightarrow \rightarrow$  internal – lab safety)

take the necessary protective measures

# Work with bio-substances of risk group 3\*\* may only be carried out in laboratories of safety level S2.



These laboratories comply with safety level S2:

- cell culture lab (r. 117)
- storage room (r. 116 + ante-room)
- MRE lab (r. 115)
- AFM labs (r. 131 a/b + ante-room)
- SpiDi lab (r. 132)
- > OS lab (r. 311)

## There is an obligation to keep records for work with GMOs and bio-substances.



These laboratories comply with safety level S1:

BONG lab (r. 309)

chemistry lab (r. 310)

## There is an obligation to keep records for work with GMOs and bio-substances.

University of Leipzig, Faculty of Physics and Earth Science, Dept. of Soft Matter Physics and Dept. of Moleculare Biophysics

#### OPERATING INSTRUCTIONS

ACCORDING TO

§12 GENTECHNIK-SICHERHEITSVERORDNUNG FÜR GENTECHNISCHE LABORBEREICHE DER SICHERHEITSSTUFE \$1

There is an obligation of the compliance of the operating instruction. The instruction according to Ordinance on Biological Agents (BioStoffV) correspond to those for Genetic Engineering Safety Regulations (GenTSV)

**Detailed operating instruction** are displayed in every laboratory, as well as a **summary**...

... and on our **PWM website**. → → → internal – lab safety

#### Wear protective equipment:

closed lab coat



- disposable protective gloves
- sturdy and closed footwear
- protective eyewear if necessary (liquid nitrogen)

Remove protective clothing before leaving the laboratory. Do not wear protective gloves outside the laboratory!



#### further generel rules:

- doors and windows must be closed during work
- keep laboratories clean and tidy
- keep order and cleanliness at the workplace
- wash and disinfect hands after finishing work or before leaving the laboratory (see skin protection and hygiene plan)
- > eating, drinking, smoking and storing food is prohibited
- knowledge of operating instructions, hygiene and skin protection plan

further generel rules:

no "storage" of packaging material or other empty containers in the laboratories, each employee has to ensure <u>immediately</u> disposal:

- polystyrol boxes in the yellow container for plastic waste in the courtyard
- > empty ethanol containers in the yellow container as well
- > cardbord boxes in the **blue container** for paper in the courtyard
- empty glass bottles or glass breakage in the container for glass in the courtyard - no glass breakage in normal waste

#### further generel rules:

- use syringes, cannulas, scalpels only if absolutely necessary, disposal in puncture resistant, autoclavable container
- do not replace the protective cap after using cannulas, syringes, scalpels – risk of injury - but dispose immediately in container







#### further generel rules:

- avoid formation of aerosols (e.g. during decanting, stirring, shaking, pipetting, centrifuging, working with ultrasound)
- for internal transport of GMOs or bio material, use tightly closed, break-proof, liquid-tight and autoclavable containers





 storage of GMOs or primary samples in liquid nitrogen (r. 116)

## **1. Biological agents – Proper disposal**

**Solid waste** that contains GMOs or bio material must be inactivated before disposal and then discard as residual waste.

#### autoclav (r. 116, ante-room) 121 °C, 20 min, program P3







- > get briefing from a trained employee
- every using of the autoclave must be documented in the equipment log book
- autoclaving should take place during the working day if possible

Liquid wastes containing GMOs or bio material are inactivated with 1% Na-hypochlorite solution and collected in appropriately labelled containers.



Other hazardous substances in accordance with the **M**aterial **S**afety **D**ata **S**heet (MSDS) of the respective manufacturer (see also chapter 2. Handling of hazardous substances).

## 1. Biological agents – Hygienic measures

#### After finishing work:

- clean work surfaces, equipment and instrument (ethanol 70%, Bacillol AF)
- switch off all devices
   (in the hood, microscope, cell counter etc.)



- > after cleaning, switch on UV lamp of the safety work benches
- clean your hands before leaving the laboratory (Sterillium, Baktolin, Baktolan ... see skin protection and hygiene plan)





by the way: the weekly cleaning of the floor is done with Microbac forte, please follow the dosage of **20 ml to 1 l water** 

## What are hazardous substances?

Hazardous substances are substances and preparations which are very toxic, toxic, low toxic, corrosive, irritant, explosive, oxidising, highly flammable, highly flammable, carcinogenic, fruit-damaging, mutagenic and/or biohazardous,

or

which, when used, give rise to dangerous or explosive hazards substances or preparations are formed or released.

#### Resulting from this: ...

Inform about potential dangers **before** starting work and take the necessary protective measures.

First information are on the packages / bottles in form of pictograms:



https://chemicals.ie/msds/

Safety Instruction, PWM 2023





harmful

toxic / very toxic

corrosive

highly flammable / inflammable

fire accelerating

endangering the environment

For detailed information see the corresponding Material Safety Data Sheet available at the internet site of the manufacturer.

#### **MSDS** contains information about:

- hazards identification
- composition
- first aid measures
- firefighting measures
- accidental release measures
- handling and storage
- exposure controls/personal protection
- physical and chemical properties
- stabiltiy and reactivity
- toxicological information
- ecological information
- disposal considerations

https://triadmachinery.com/blog/october-safety-corner-safety-data-sheets-requirements/



#### **General rules of conduct:**

transport of glass bottles in appropriate transport containers (risk of breaking) - e.g. bucket or basket



... and never carry bottles on the neck

- mark vessels: name of the substance, name of the user, corresponding hazard symbols
- storage only in containers which are suitable for the hazardous substance (e.g. no plastic bottles for solvents)

#### **General rules of conduct:**

- keep very toxic, toxic, carcinogenic, fruit-damaging and mutagenic substances under lock and key
- when openly handling gaseous, dust-like or hazardous substances that have a high vapour pressure work always in a fume cupboard

when the sash is pulled down, the effect of the fume cupboard is at its best

average air velocity in the sash gap (m/s)



sash gap (cm)

proper disposal (s. MSDS)

Liquid nitrogen (LN2):

- has a boiling point of -196°C
- > is heavier than air and accumulates on the ground

#### Liquid nitrogen hazards:

- > skin contact with liquid nitrogen can lead to cold burns and frostbite
- LN2 has a suffocating effect in high concentrations without noticeable signs, since oxygen in the breathing air is displaced by an accumulation of nitrogen
- depending on the duration of inhalation and the remaining oxygen concentration, drowsiness, malaise, blood pressure rise and shortness of breath occur
- plastic containers can become brittle in LN2, danger of splittering when material bursts

#### **Protective measures and rules of conduct:**

- always wear suitable protective gloves and eyewear
- clothing should cover all parts of the body (closed footwear!)
- > when filling the bio-rack: do not open the shut-off valve jerkily
- do not leave container unattended during filling process
- do not use elevator together with LN2 container



#### Behaviour in case of danger:

Leakage of the bio-rack - the LED display of the level monitor lights up and a continuous signal sounds

- leave the room immediately or do not enter it
- leave the door open and come for a second person
- then enter the room only one at a time and ensure oxygen supply, open the window

#### First aid:

- skin contact: treat as burns or frostbite, do not rub, cover with sterile dressing
- > eye contact: immediately rinse with water for at least 15 minutes
- ➤ inhalation: high concentrations may cause asphyxiation → fresh air supply, keep warm and calm, seek medical advice, if breathing stops artificial respiration

## 3. Actions in case of accidents in the lab

## In order to avoid panic in case of emergency, everyone has to inform himself before starting the work about:





location of eye showers in room 310 permanently installed, otherwise <u>flushing bottles</u>







location of body shower (in room 310)



first aid facilities in room 310 and 116 (anteroom)  $\rightarrow$  label at the lab door

## 3. Actions in case of accidents in the lab

In order to avoid panic in case of emergency, everyone has to inform himself before starting the work about:



function of disinfectants (Bacillol, Sterillium)

fire-extinguishing equipment

escape and rescue routes

## 3. Actions in case of accidents in the lab

#### Leakage or spillage of hazardous substances / GMOs / bio-substances

Put on gloves, secure affected area – take decontamination measures:

- surfaces / devices: collect material, dispose it properly or in case of biological agents take up material with paper towels or other autoclavable material and autoclave, then wipe disinfection
- > skin: rinse off contaminated skin areas with plenty of water / disinfect
- mucous membranes / eyes: rinse off with plenty of water or take an eye shower
- clothing: take off and autoclave
- inform laboratory manager / project manager, consult doctor if necessary



Lasers are classified based on their potential for causing injury — especially eye damage, since the eye is most susceptible to excess laser light.



(Calculations are for visible light, a 1 milliradian beam, and a 1/4 second Maximum Permissible Exposure limit.)

There are four main classes for visible-beam lasers: Class 2, Class 3R, Class 3B and Class 4. The first two are relatively safe for eye exposure; the last two are hazardous. The chart shows that the eye injury hazard increases as the laser's power increases.

https://www.laserpointersafety.com/laserclasses.html

#### Hazard potential by laser:

- deposition of thermal energy in tissue
- tissue reactions or damage, dangerous for eye and skin (deep skin burn at IR wavelengths)
- Iocal temperature rise, fire and explosion hazard

#### Damage can be caused by:



In order to assess the risk potential, we have to distinguish between commercially purchased and own experimental setups.

In **purchased devices**, the lasers are encapsulated and existing safety devices prevent the unintentional escape of the laser beam. If used properly, there is no potential danger from laser beam.

The devices concerned are:

- ✓ all AFMs from JPK (r. 131 / 1 and 2 and 309)
- ✓ SpiDi from Zeiss (r. 132)
- ✓ OS from RS Zelltechnik

In our labs with self-designed experimental setups the following rules of conduct apply:

#### 1. Protection against laser radiation





- 1. warning light in front of the entrance door
- 2. protective curtain separates input from the laser area
- **3.** laser protective eyewears at the entrance range

#### applies to room 309 - optical tweezer and room 311 - optical stretcher

#### 2. Rules of conduct for persons who work on the laser

- switch on warning light interlock
- > warn all present persons before switching on lasers
- work on the laser or with the laser beam without arm jewellery such as watch, ring or bracelet (reflected, scattered beam possible)
- restrict laser beams as good as possible on the experimental area
- > put on suitable laser protective eyewear!
- each setup and any modification of a setup must be approved by the person responsible for laser safety

responsible for laser safety in the lab: Bernd Kohlstrunk (phone: 32485)

#### 3. Rules of conduct for persons who don't work on the laser

- if the warning light is switched on: before entering the laser lab, ask for sources of danger!
- wear suitable laser protective eyewear (available in the entrance area)
- do not bring head to beam height, i.e. be careful when bending down
- no unauthorized handling of optics or lasers!

#### Laser protective eyewear



- must comply with DIN EN 207 and be clearly marked
- > are always specially matched to a laser (wavelength)
- eyewears has to be completely intact

#### Laser alignment eyewear

- may only be used for adjustment work on laser devices of classes 3B and 4 which emit in the visible spectral range between 400nm and 700nm
- > must comply with DIN EN 208 and be clearly marked

## **5. General behavior in case of emergency**

- give the alarm immediately
- phone dispatcher of the university (34333) or in case of fire not indicated by alarm system (acoustic signal) additionally the fire brigade 0-112
- keep calm human lives before firefighting
- close windows and doors, do not lock doors
- > switch off electrical appliances, except for room and staircase lighting
- leave the building immediately and go to the meeting place (Friedenspark)
- don't use elevators

#### 5. General behavior in case of danger

#### meeting place in case of fire alarm - Friedenspark



- ... and at the end an important, general note:
- please avoid to work outside the opening hours of the faculty
- if you are in the building after 19 o'clock, sign in the attendance book so that in case of emergency it is known that you are in the house (the book is in the main entrance beside the monitor)

- if you work alone, please inform an other person and keep in contact via phone
- in case of emergency call 34333 (Dispatcher)



#### **Responsible persons:**

project leader / group leader: Prof. Josef Käs (32471), jkaes@physik.uni-leipzig.de

responsible for lab: Dr. Undine Dietrich (32472), <u>dietrich@physik.uni-leipzig.de</u>

appointee for biological safety: Dr. Nicole Weizenmann (32592), <u>weizenmann@uni-leipzig.de</u>

appointee for laser safety and responsible for electrical safety in the labs: Dipl.-Phys. Bernd Kohlstrunk (32485), <u>bkohl@physik.uni-leipzig.de</u>