

# Essential oils components

## *Protection and disinfection of the books*

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National Library CR

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2011 - 2015*

# The principal abbreviations

frequently used in the following text

EO = essential oil(s)

EOC = essential oil(s) components

This presentation is the part of the project  
*„Essential oils a tool for saving and increasing of culture  
heritage on paper“*

**The philosophy of the project were**

- Testing of antifungal effectivity of selected **EO vapors** on typical fungi species existing in library depositories
- Selecting the most effective EO and determine their composition
- Selecting volatile and major components of effective EO to test their antifungal effectivity sole and in combinations
- For the most effective EO and EOC find their influence on physical properties of book materials and
- Find the effect of different conditions of their application on fungicidal effectivity
- Developing book disinfection technology
- Find protection books protocol against fungi in depositories with occasional high relative humidity atmosphere

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- Find defence protocol for books against fungi in depositories with occasional high relative humidity atmosphere

**This tasks are splitted between**

**3 research institutions**

# Participants of research

## Academy of Science CR Inst. Analytical Chemistry Brno

- Proposal of the testing instrumentation and cooperation with SZÚ and MBÚ

- **Analyses** of the most effective EO to select principal **components**

- Antifungal activity of **lavandine oil** and its selected **components** at different concentrations

- Determination of absorption and desorption of EO components – different materials

## National Library CR Praha

- Testing **antifungal activity** of saturated vapors of selected **EOS** and their combinations

- Selection of the most effective combination of EOS

- This combination is used to set up quantitative method of determination of the antifungal effectivity of EOS at various conditions of disinfection of paperlike material

- Set up the book disinfection method

- Reality of EOS fungistatic effect in depository with high R.H.

**Let us see the results**

## University Pardubice Pardubice

- Effect of selected EO and EOS on physical and chemical **properties** of the cellulose fibrous materials and PVC

- Theory and mechanism of water and EO vapors **absorption/desorption** in paperlike materials

# Tests with spores in saturated vapors of EO and 75% R.H.

Experiments were done in SZÚ

## Tested EO

1. **LAVENDER** – *Lavandula species*
2. WHITE BIRCH – *Betula album*
3. BERGAMOT – *Citrus Bergamia*
4. CITRONELLA – *Cymbopogon nardus*
5. EUCALYPTUS– *Eucalyptus radiata*
6. **LIME** – *Citrus aurantifolia*
7. JUNIPER BERRY – *Juniperus communis*
8. TEA TREE – *Melaleuca alternifolia*
9. MYRTLE – *Myrtus communis*
10. CLOVE – *Syzigium aromaticum*
11. THYME WHITE – *Thymus vulgare*
12. **CINNAMON** – *Cinnamomum zeylancium*
13. TEXAS CEDARWOOD – *Juniperus asheia*
14. PATCHOULI
15. CORN MINT – *Mentha arvensis*

## Tested fungi sp.

1. *Rhizopus oryzae*
2. *Mucor racemosus*
3. ***Penicillium aurantiogriseum***
4. ***Aspergillus niger (brasiliensis)***
5. *Fusarium oxysporum*
6. *Cladosporium cladosporioides*
7. *Chaetomium globosum*
8. *Alternaria tenuissima*

# Results of tests with spores

## 1 week in saturated vapors of EO and 75% R.H.

Experiments were realized in SZÚ

### Effectivity after 1 week

Esence č.	<i>Rhizopus oryzae</i>	<i>Mucor racemosus</i>	<i>Penicillium aurantiogriseum</i>	<i>Aspergillus niger (brasiliensis)</i>	<i>Fusarium oxysporum</i>	<i>Cladosporium cladosporioides</i>	<i>Chaetomium globosum</i>	<i>Alternaria tenuissima</i>
1	N	> 4,2	N	> 5,28	N	> 4,5	> 4	> 5,5
2	> 4,2	2,32	3,82	2,88	> 4,4	> 4,5	> 4	> 5,5
3	> 4,2	2,44	> 5,3	4	N	> 4,5	> 4	> 5,5
4	2,56	3,36	> 6,16	3,04	3,24	2,56	> 4,2	> 5,4
5	3,56	3,2	3,98	1,7	3,56	3,56	> 4,2	> 5,4
6	4,12	4,08	> 6,16	> 5,16	N	4,12	> 4,2	> 5,4
7	3,74	3,52	3,9	3,88	N	3,74	> 4	> 5,2
8	3,03	2,92	2	2,12	2,22	3,03	> 4	> 5,2
9	> 5,14	> 4,3	4,12	3,96	3,2	> 5,14	> 4	> 5,2
10	3,7	3,62	> 5,42	3,52	N	3,7	> 4,16	> 5,5
11	3,6	2,36	2,22	2,52	N	3,6	> 4,16	> 5,5
12	> 5	> 4,4	> 5,42	> 5,32	N	> 5	> 4,16	> 5,5
13	2,76	1,84	0,4	0,08	0	2,76	> 4,2	> 5,3
14	3,88	1,22	1,52	0,1	3,86	3,88	> 4,2	> 5,3
15	> 5	> 4,3	5,92	3,66	N	> 5	> 4,2	> 5,3

# Selections for detailed experiments

## Selected EOS

from most efficient EO

- Alfa-pinene
- Citral
- Linalyl acetate
- D-limonene
- Linalool
- Menthol
- Menthon
- Ocimene

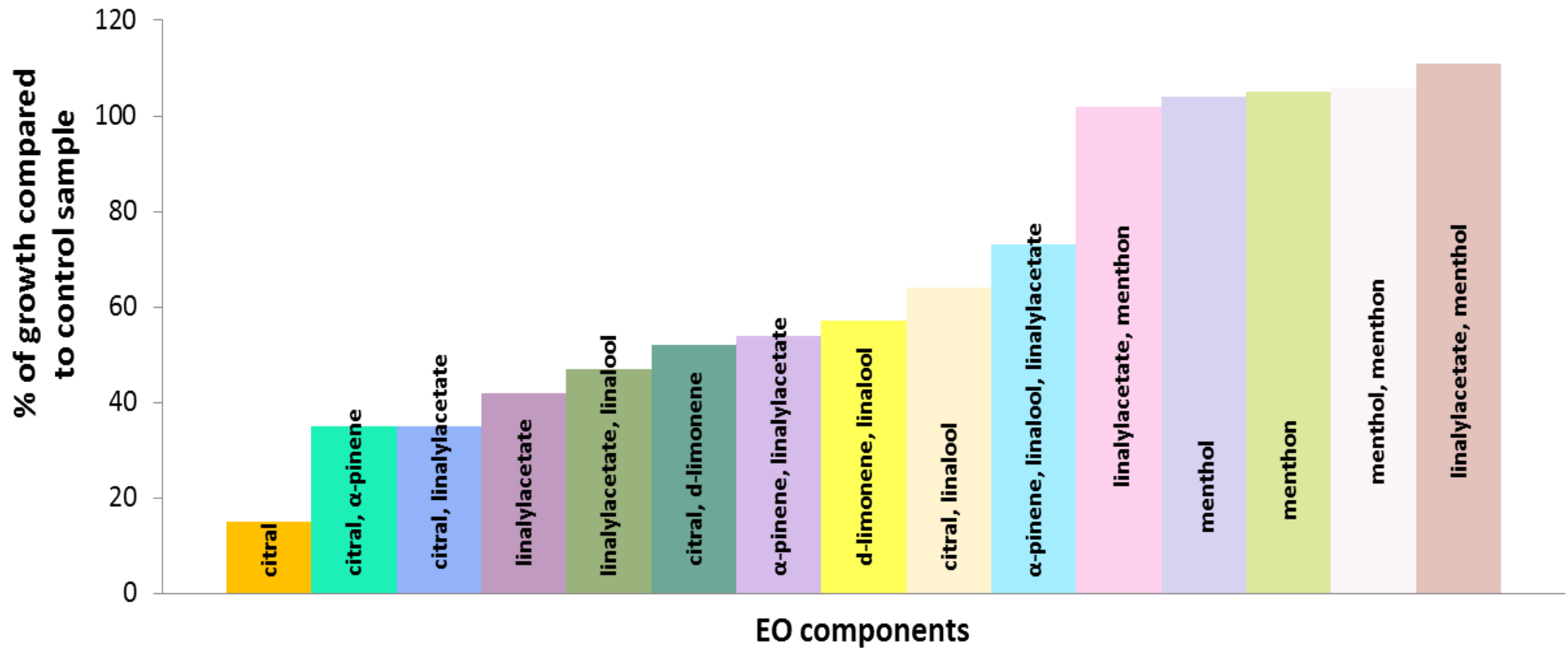
## Selected fungi sp.

Used in consecutive experiments

- Penicillium aurantiogriseum*  
(frequent occurrence in library)
- Aspergillus niger (brasiliensis)*  
(high resistance)



# EOS saturated vapors effectivity against *Aspergillus brasiliensis* spores



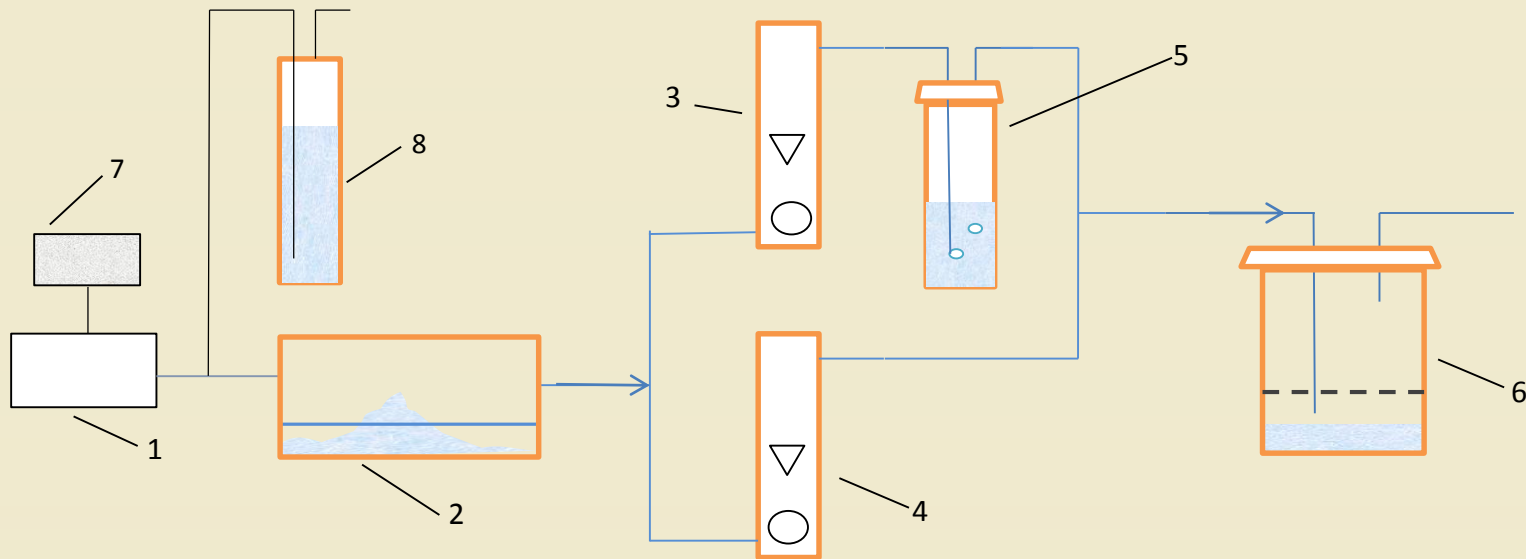
*Growth of molds after exposition of spores  
to saturated vapors of selected EO components*

# The most perspective combination of EOS

Citral + Linalyl acetate (1 : 1)

is used in all following experiments

# Testing line



## Testing line

1. Outlet compressor or compressed gas
2. The closed container with saturated aqueous salt for setting the desired relative humidity
3. Rotameter with adjustable air flow to the EOS saturation impinger
4. Rotameter with adjustable air flow to dilute the EOS saturated vapor
5. Impinger for saturation of the air flow with EOS vapors
6. Testing container for samples inoculated with fungi spores
7. Input filter
8. Manostat

# Testing line



# Results from testing line

Effect of:

- relative humidity
- concentration of EOS
- balast paperlike material
- nitrogen atmosphere

*on the fungicidal activity of EOS*

# Testing line

## Effect of relative humidity

### **RH lower than 50%**

- action of EO is not effective

### **RH higher than 70%**

- effective EO action

# Testing line

Effect of EOS concentration in % of saturation

Concentration	Treatment time	<u>Viability after 7 days of cultivation</u>	
		Aspergillus	Penicillinum
10%	3 weeks	100%	100%
10%	4 weeks	100%	75%
17%	3 weeks	0%	0%
50%	1 week	50%	0%
50%	2 weeks	0%	0%

# Testing line

## effect of balast paperlike material

From absorption tests we know that after 10 days in atmosphere of 50% saturation with EOC vapor the paper board contains 0,3% of EOS.

The flow rate through 8 liter testing vessel is 80 ml/min and so the movement of atmosphere over the sample surface is laminar and very slow.

So EOC concentration at the surface of the board is to be very low at the beginning of the experiment with balast paper board.

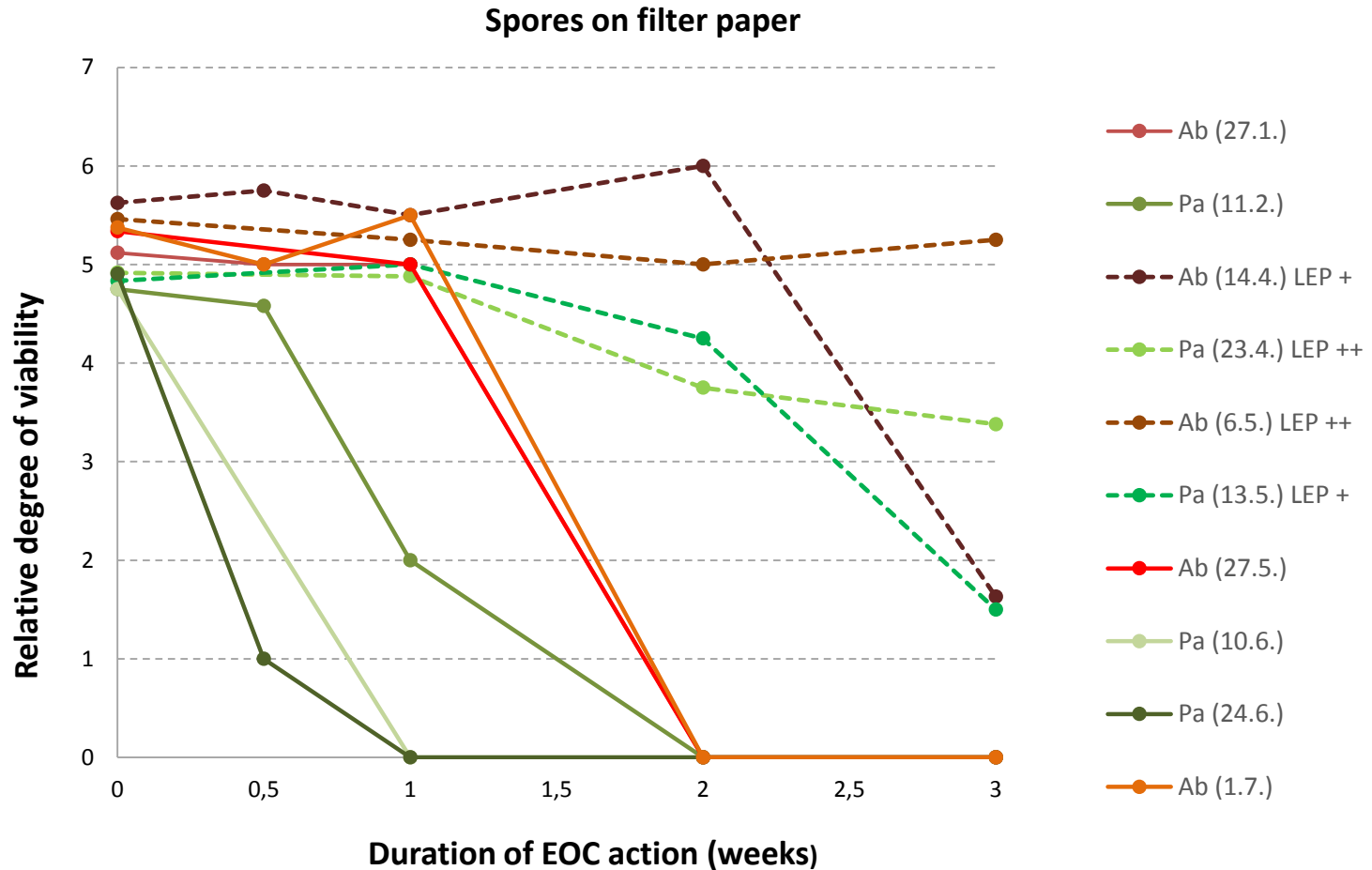
Theoretical mass of EOC transferred through the vessel at the flow rate 80 ml/min during 10 days is 0,35g.

The effect of balast material depends on its absorption capacity



# Testing line

## effect of balast paperlike material



# The method of quantitative determination of fungicidal effect

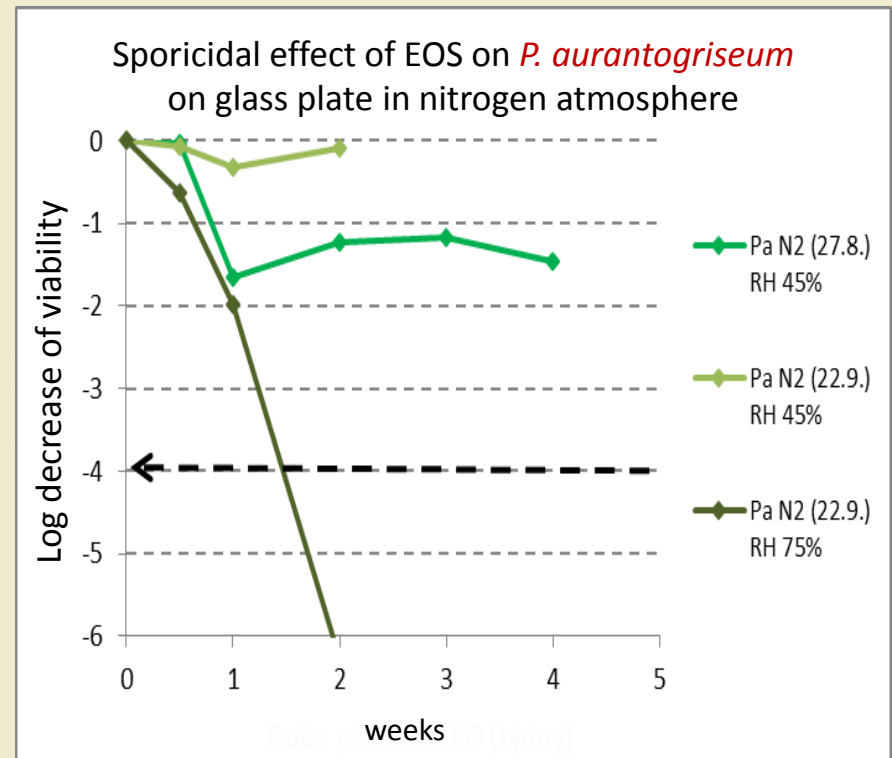
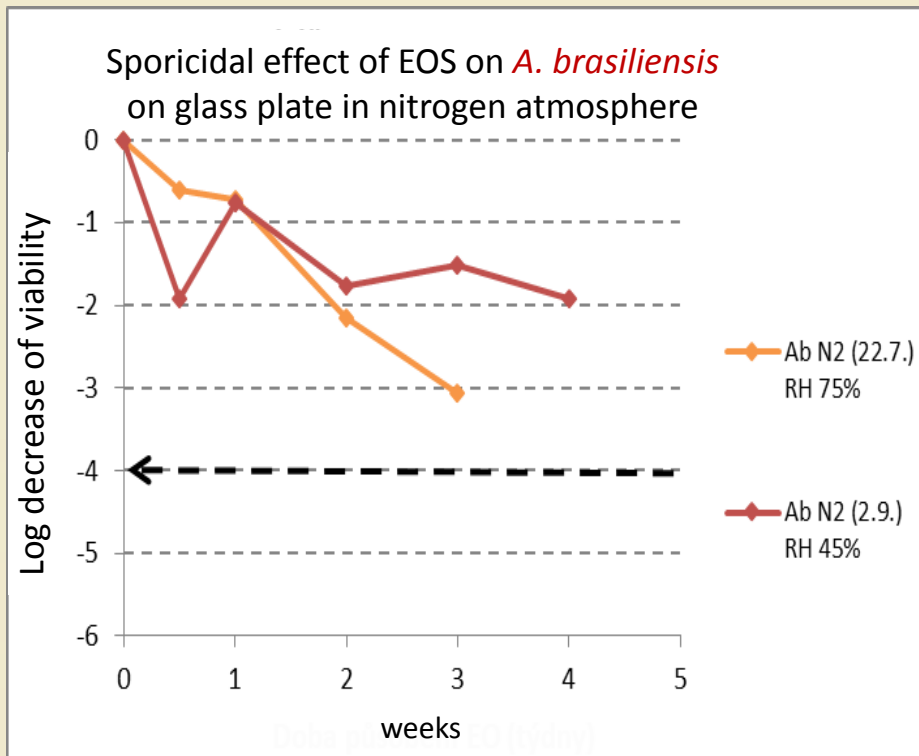
The standard decimal dilution method was adapted for use with porous paperlike materials.

The result of testing is number we get as **difference log of viable spore number before and after disinfection.**

The higher is this number the more effective is the disinfection.  
If this number is **greater than 4** the disinfection is taken as successful.

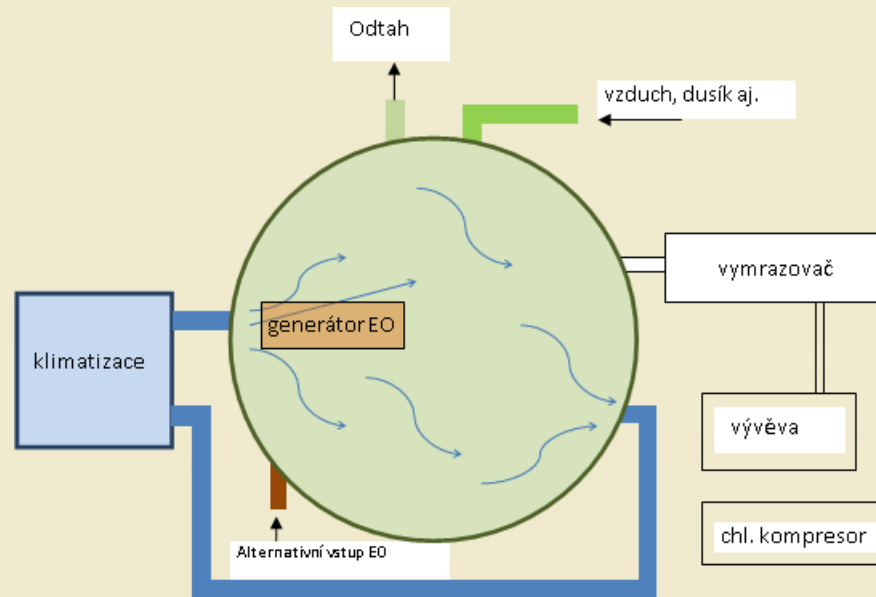
# Testing line

effect of nitrogen atmosphere at different humidity

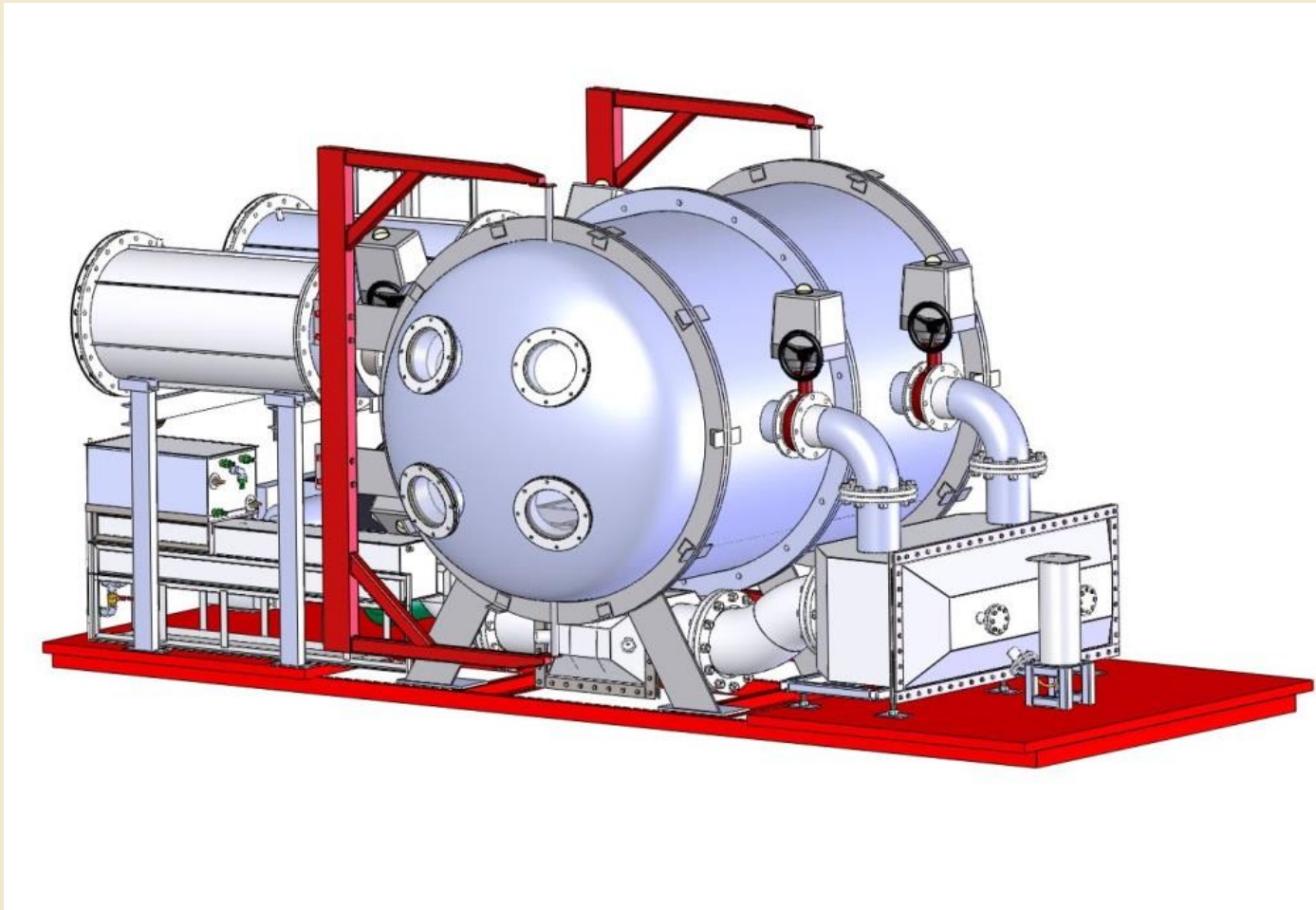


Nitrogen prolongs the necessary time for disinfection in testing line

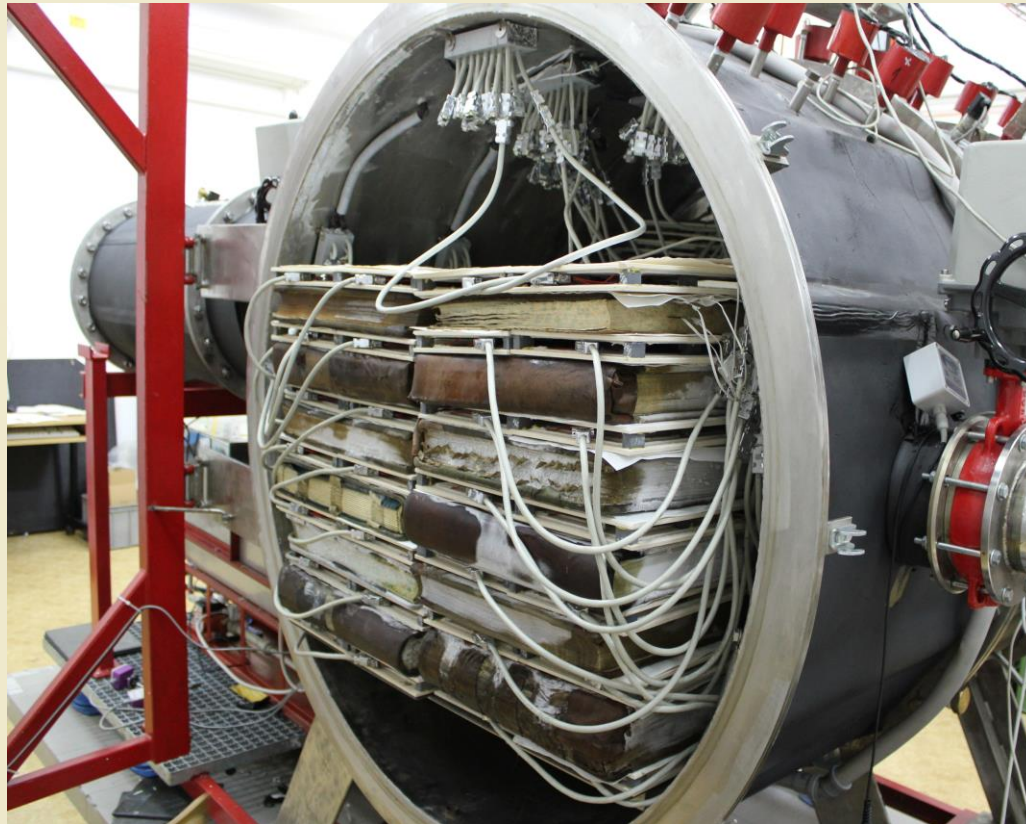
# Disinfection in universal vacuum chamber



# Disinfection in universal vacuum chamber



# Disinfection in universal vacuum chamber



# Disinfection in universal vacuum chamber



Chamber and its content

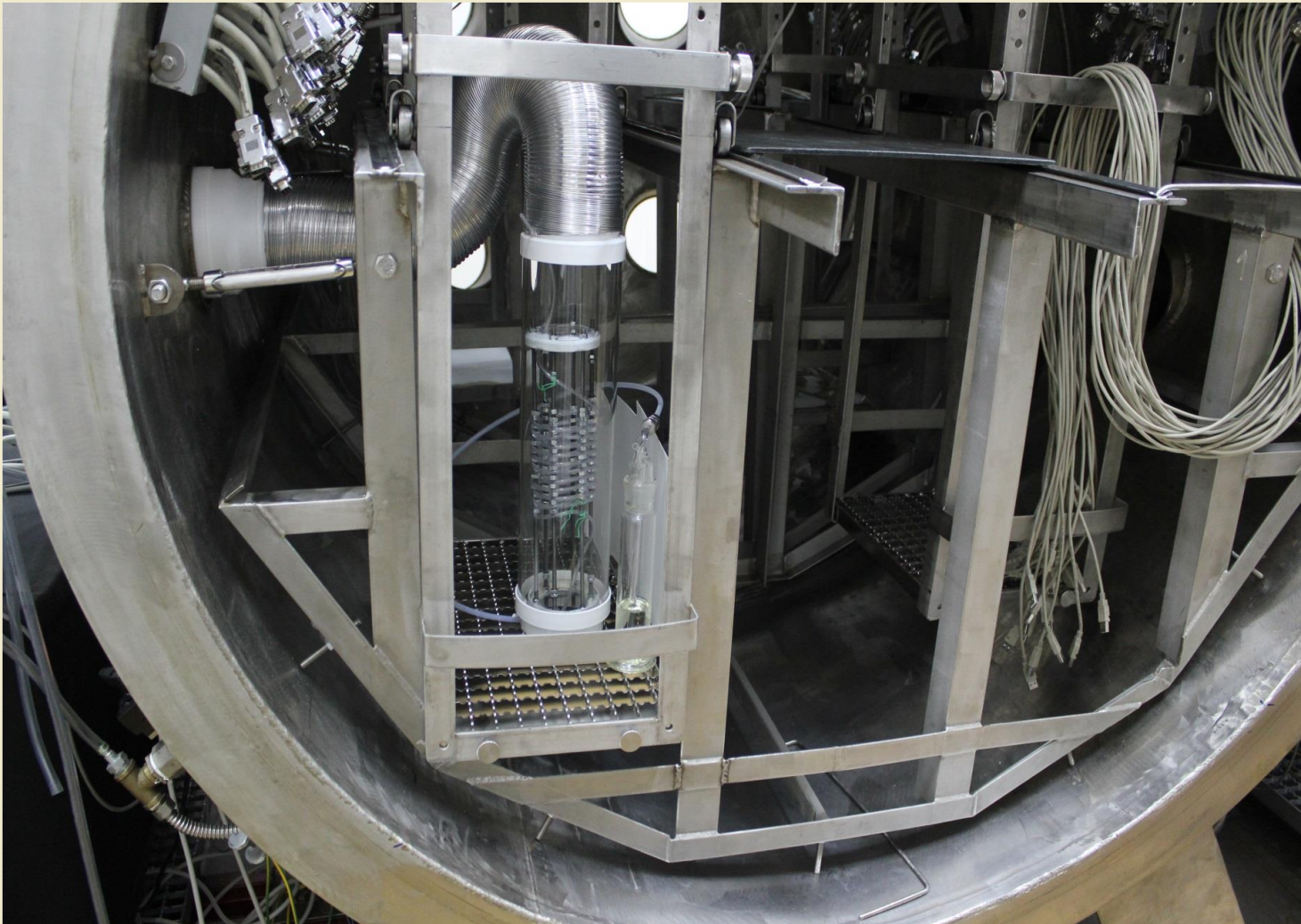
# Disinfection in universal vacuum chamber

Container for stacking and separation of disinfected books

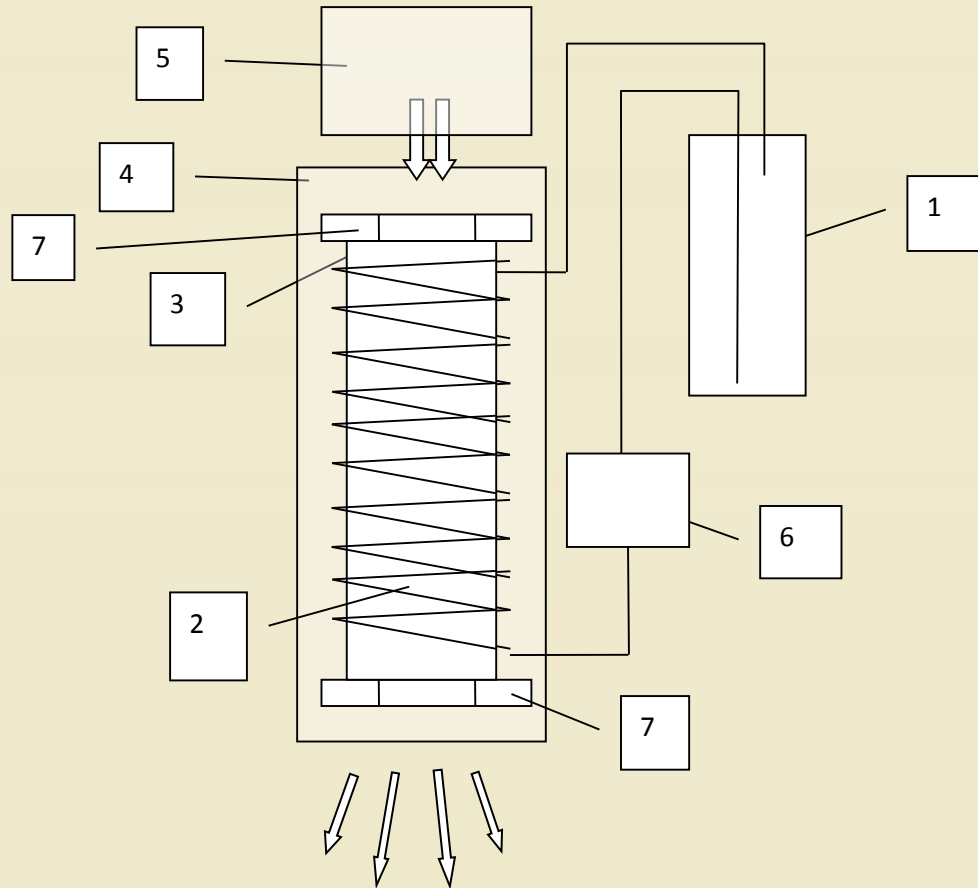




# Vacuum chamber position of EOS vapor generator



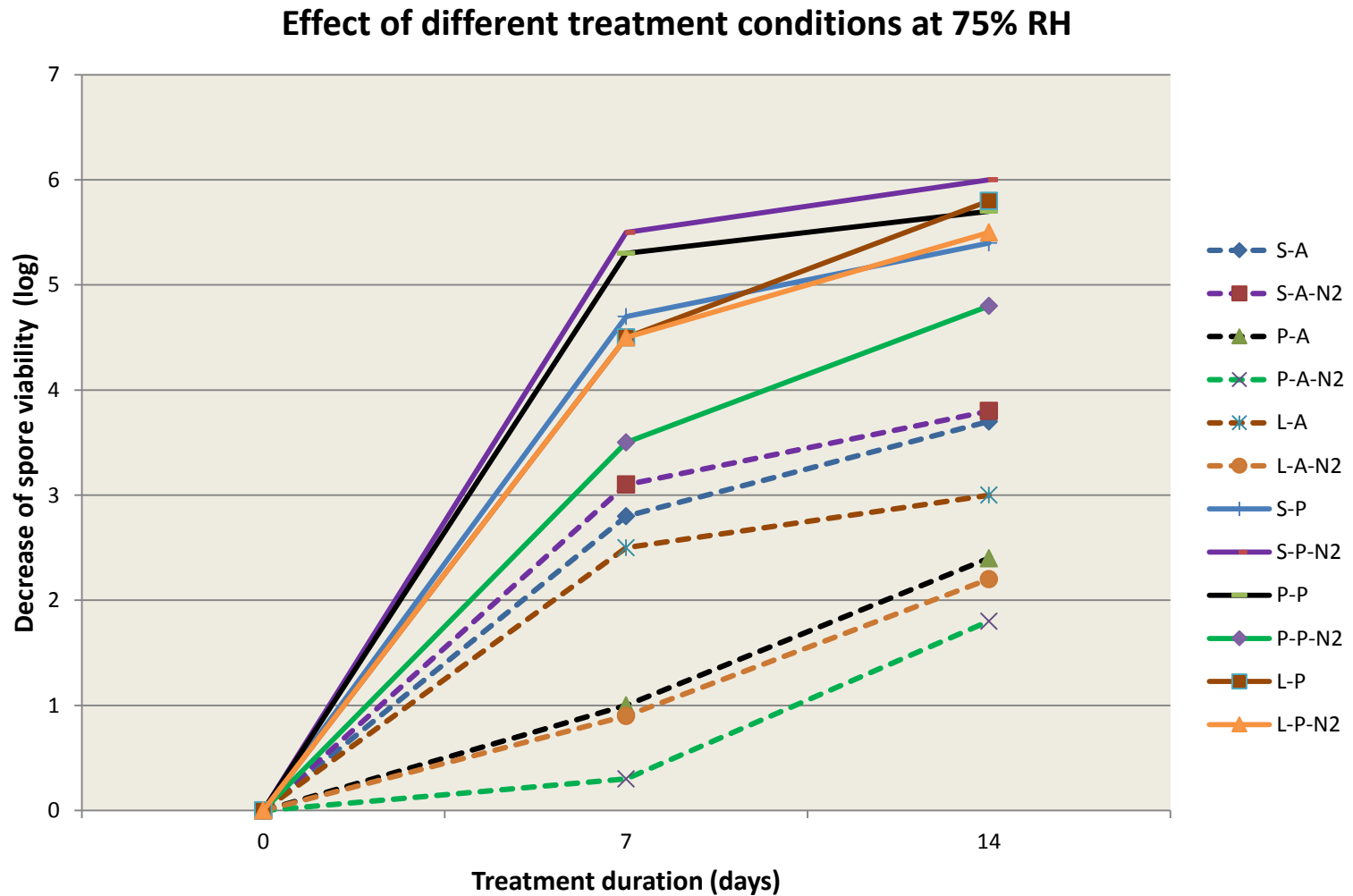
# Generator of EOC vapors



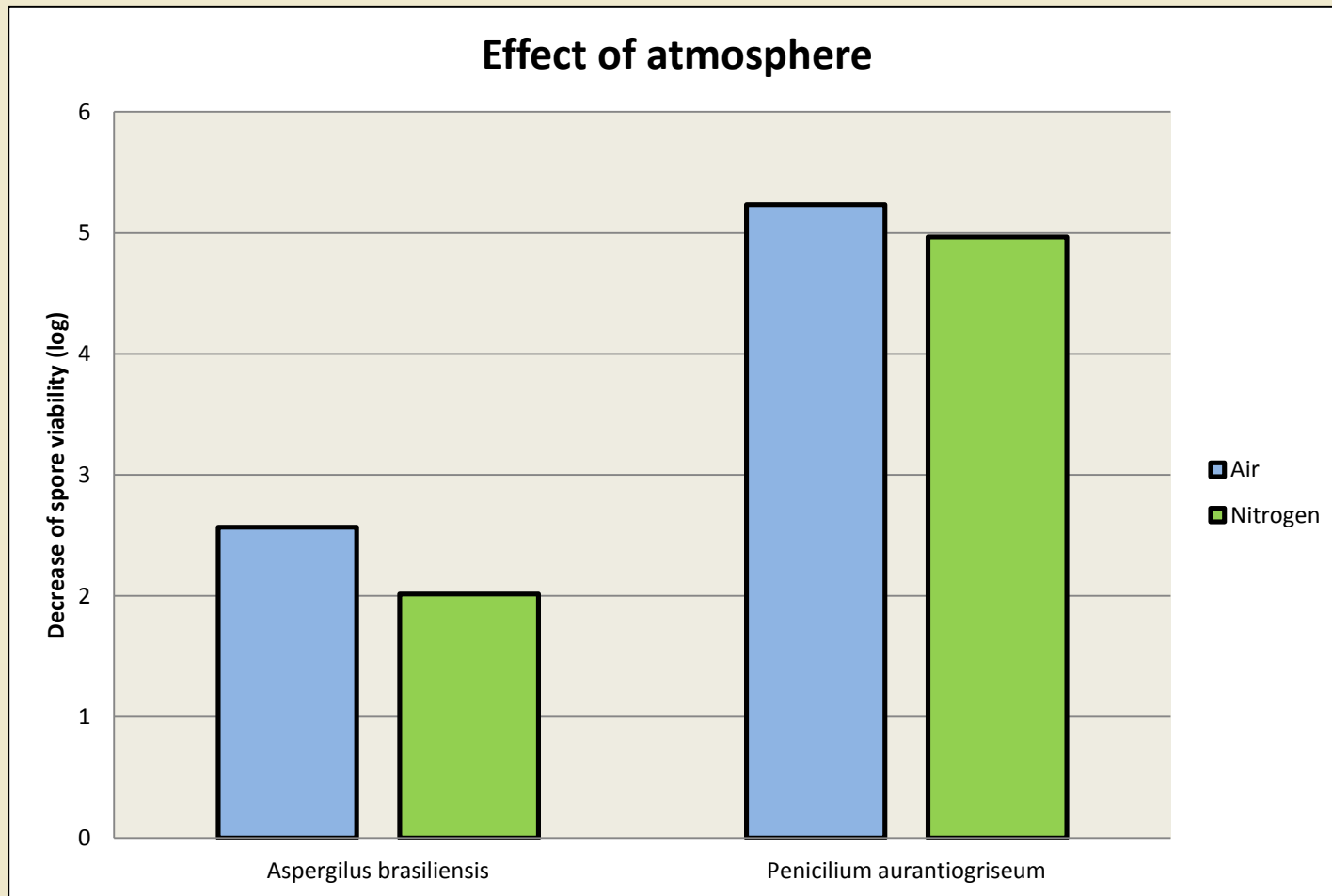
## Legend

1. Reservoir of liquid components of EO
2. PVC tube
3. Wire support of the tube
4. Air duct
5. Fan
6. Liquid EO pump
7. Head of the support with opening

# Disinfection in the chamber

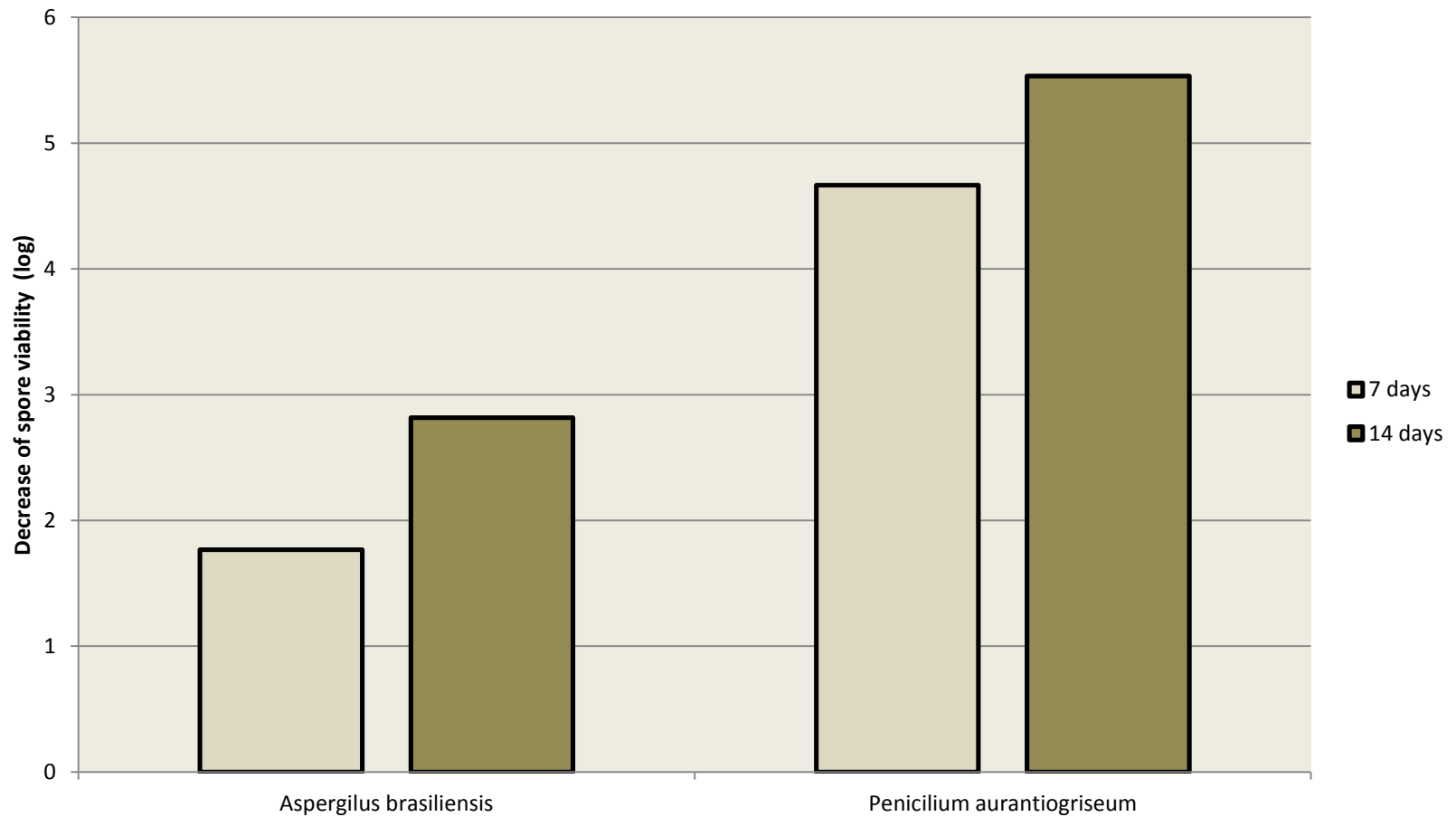


# Disinfection in the chamber



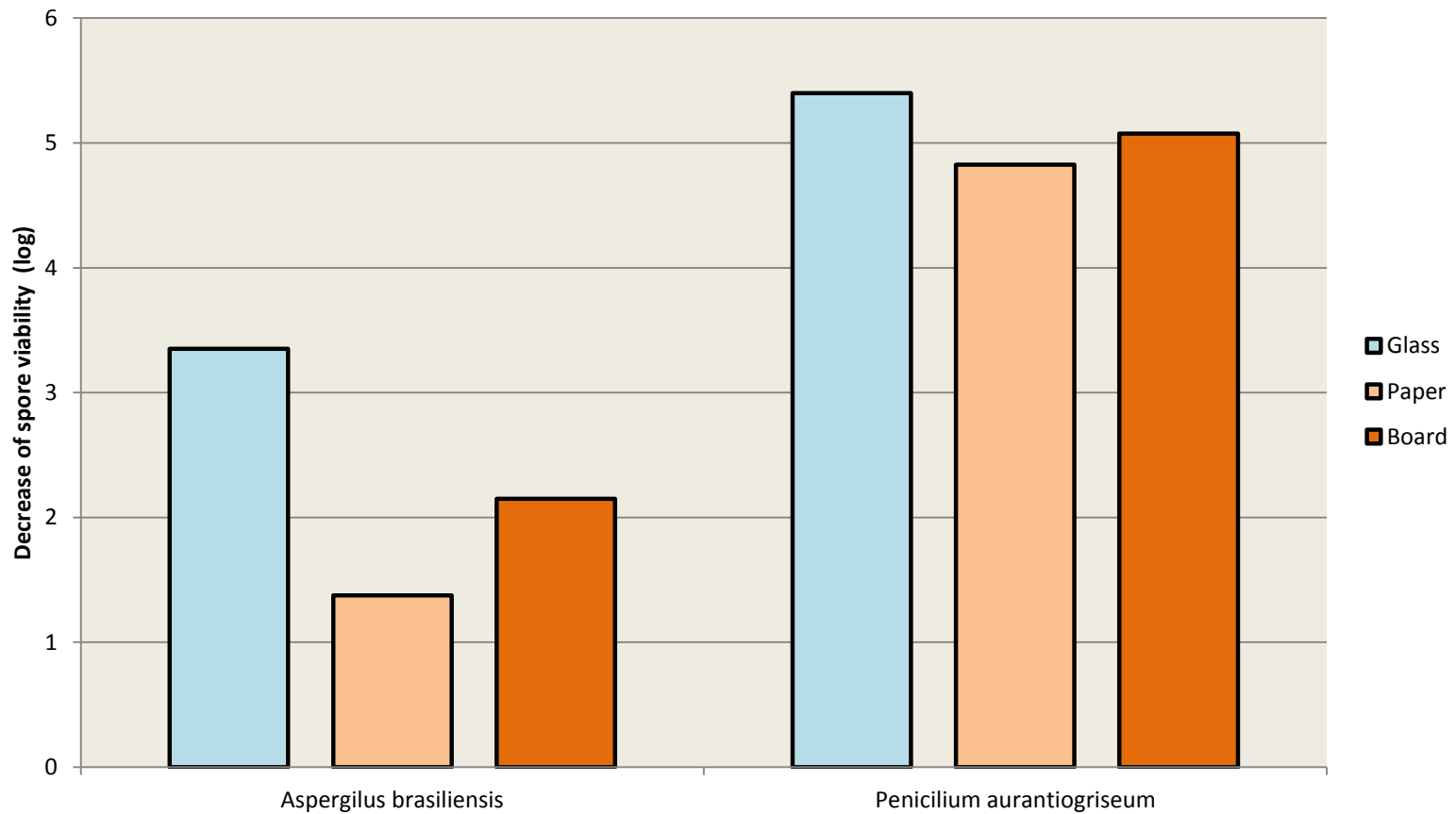
# Disinfection in the chamber

Effect of the treatment duration



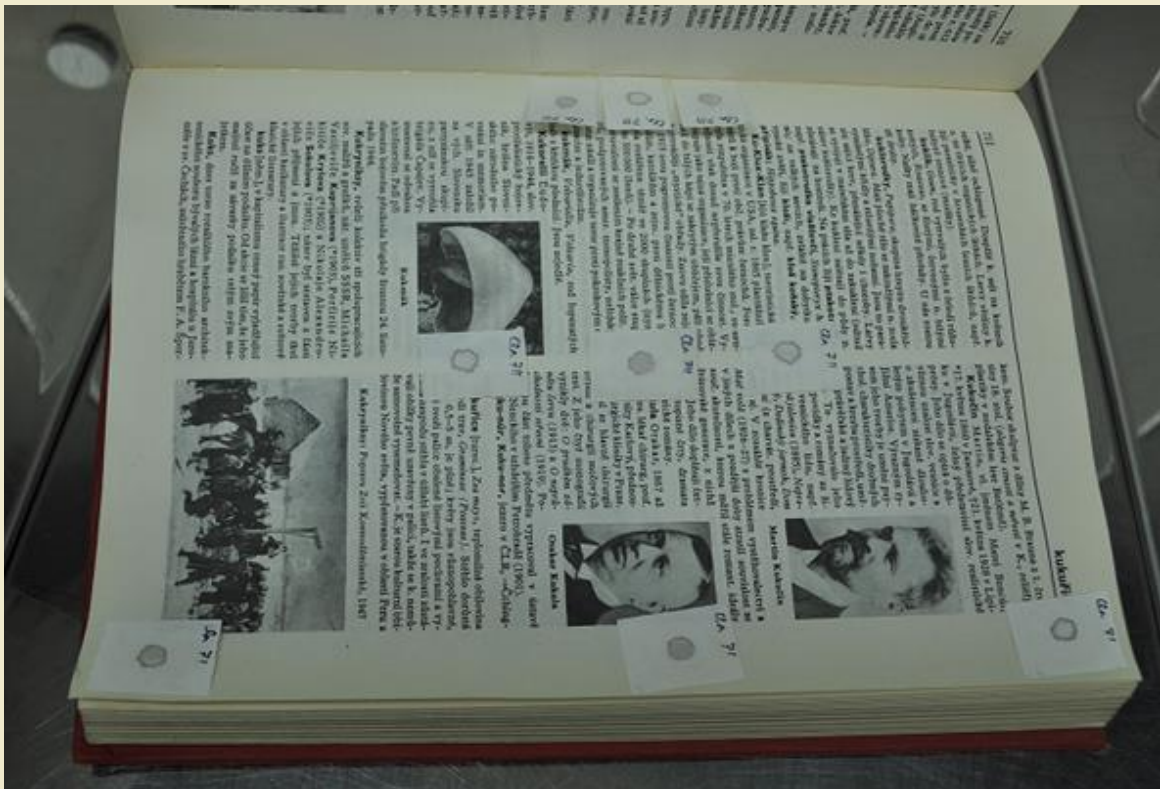
# Disinfection in the chamber

Effect of disinfected material



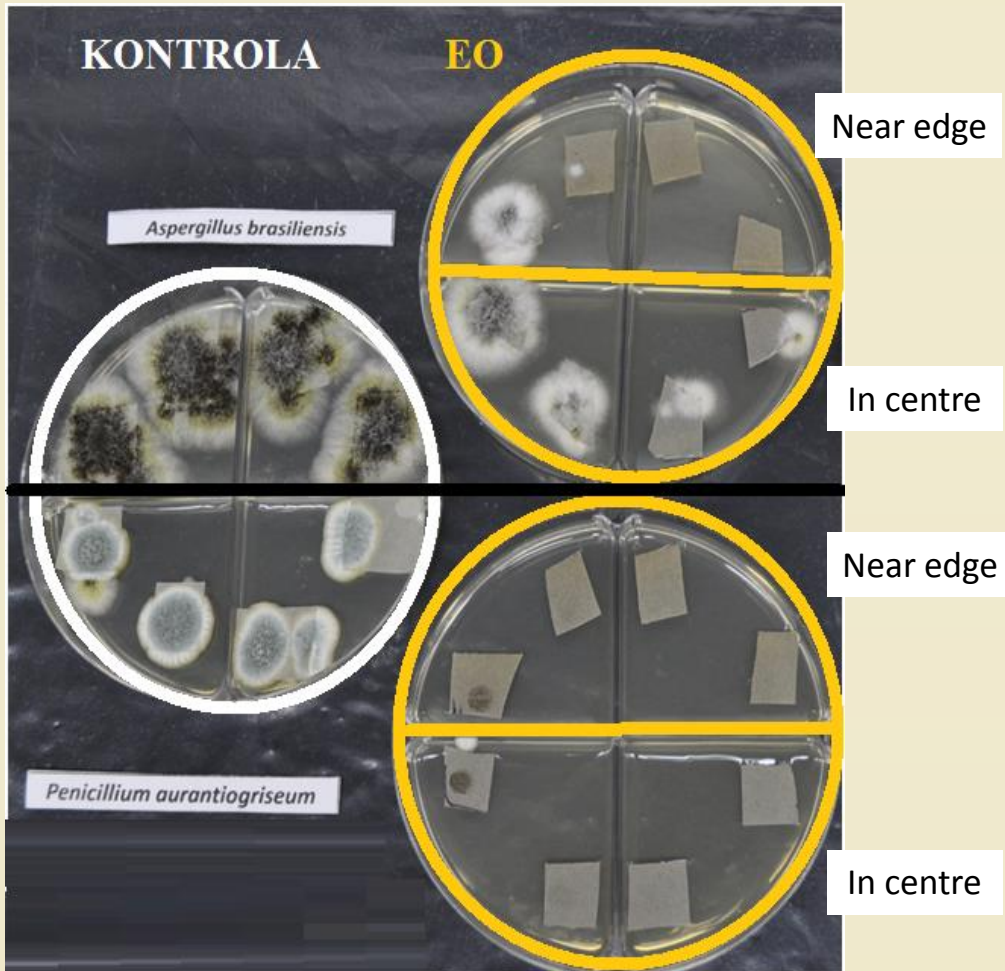
# Disinfection in the chamber

location of the samples in the book



# Disinfection in the chamber

## samples from the book



Spores were inoculated directly on pages of the book.

After disinfection inoculated pieces were cutted out.

In the upper part of the figure is control sample and two disinfected samples of *A. brasiliensis*.

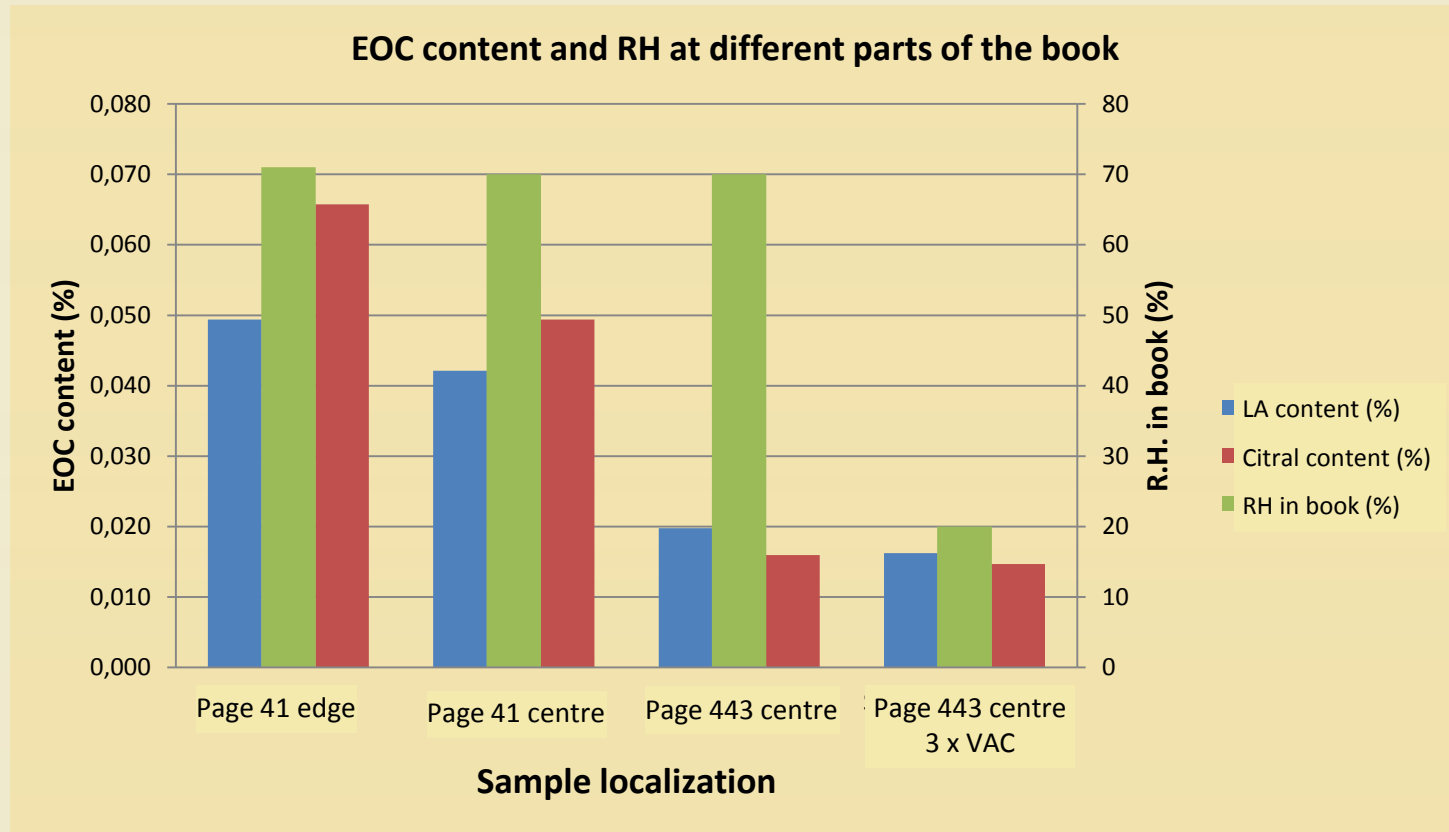
In the lower part is control sample and two disinfected *P. aurantiogriseum* samples.

Each set of four samples was inoculated with decreasing approximate number of spores  $10^6$ ,  $10^5$ ,  $10^4$  a  $10^3$  (from the left).



# Disinfection in the chamber

## EOC and RH in the book after disinfection

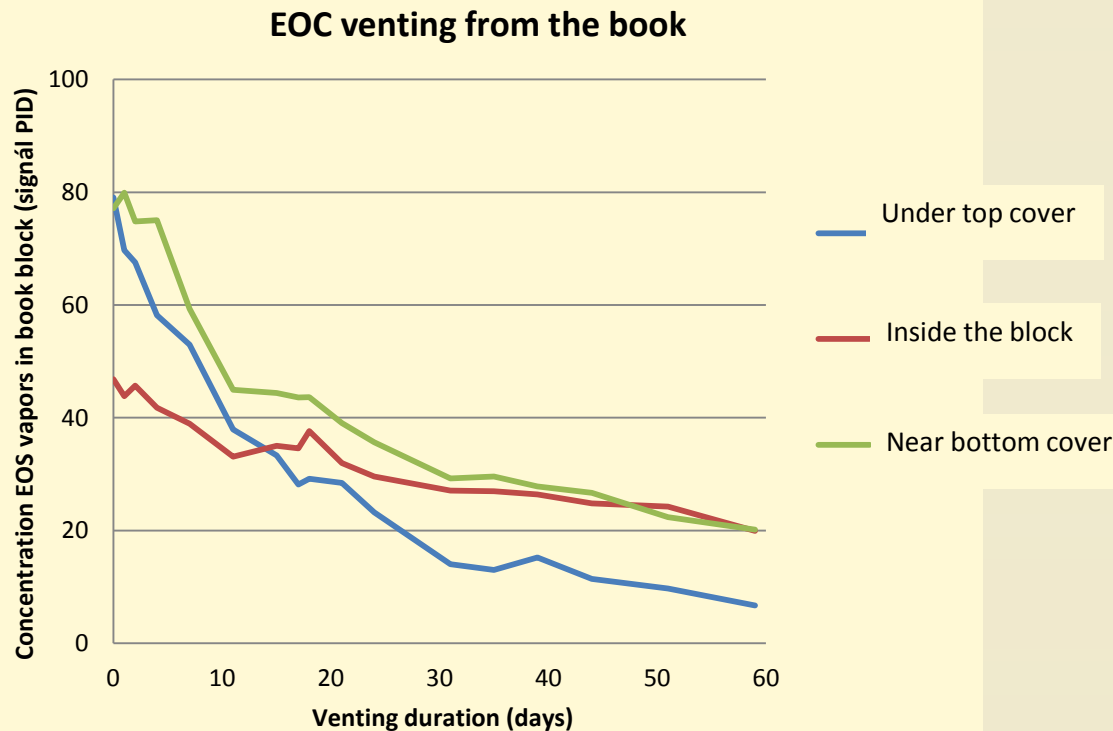


# Disinfection in the chamber

free desorption of EOC from the book

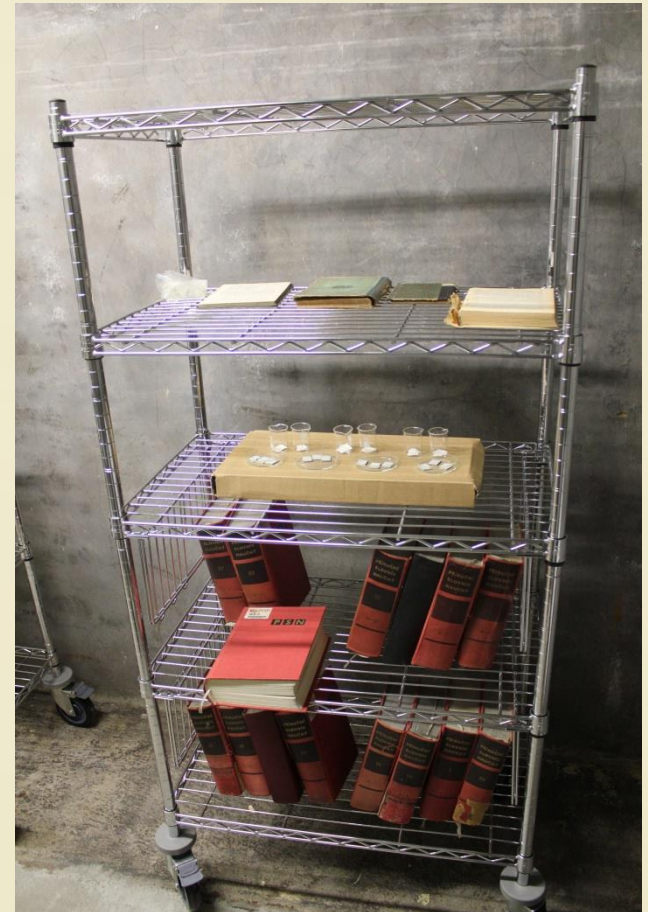
The book lies on laboratory table

The size of the book  
26 x 20 x 6 cm



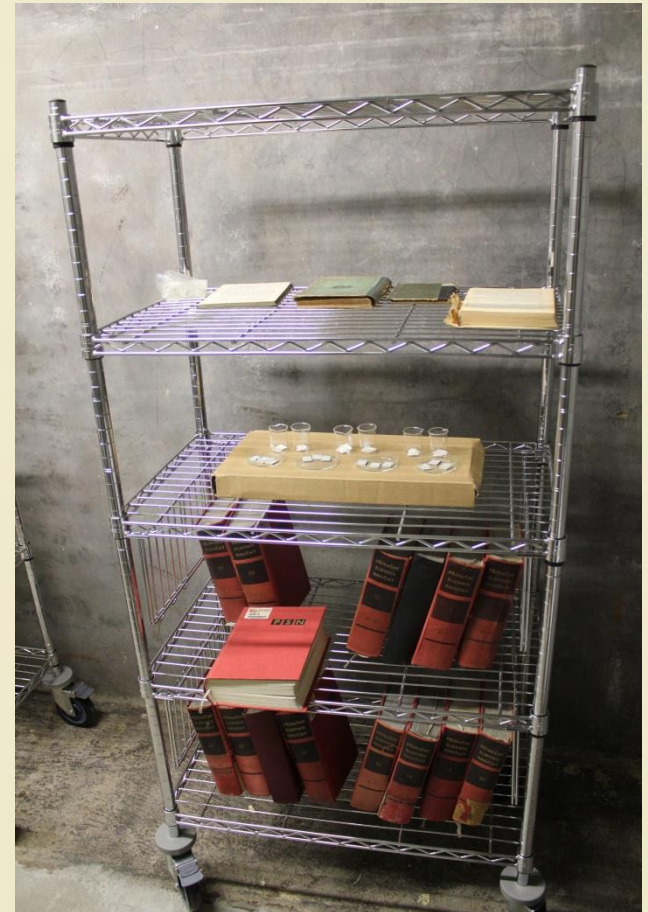
# Model depository

$$2 \times 5 \times 3 \text{ m} = 30 \text{ m}^3$$



# Model depository

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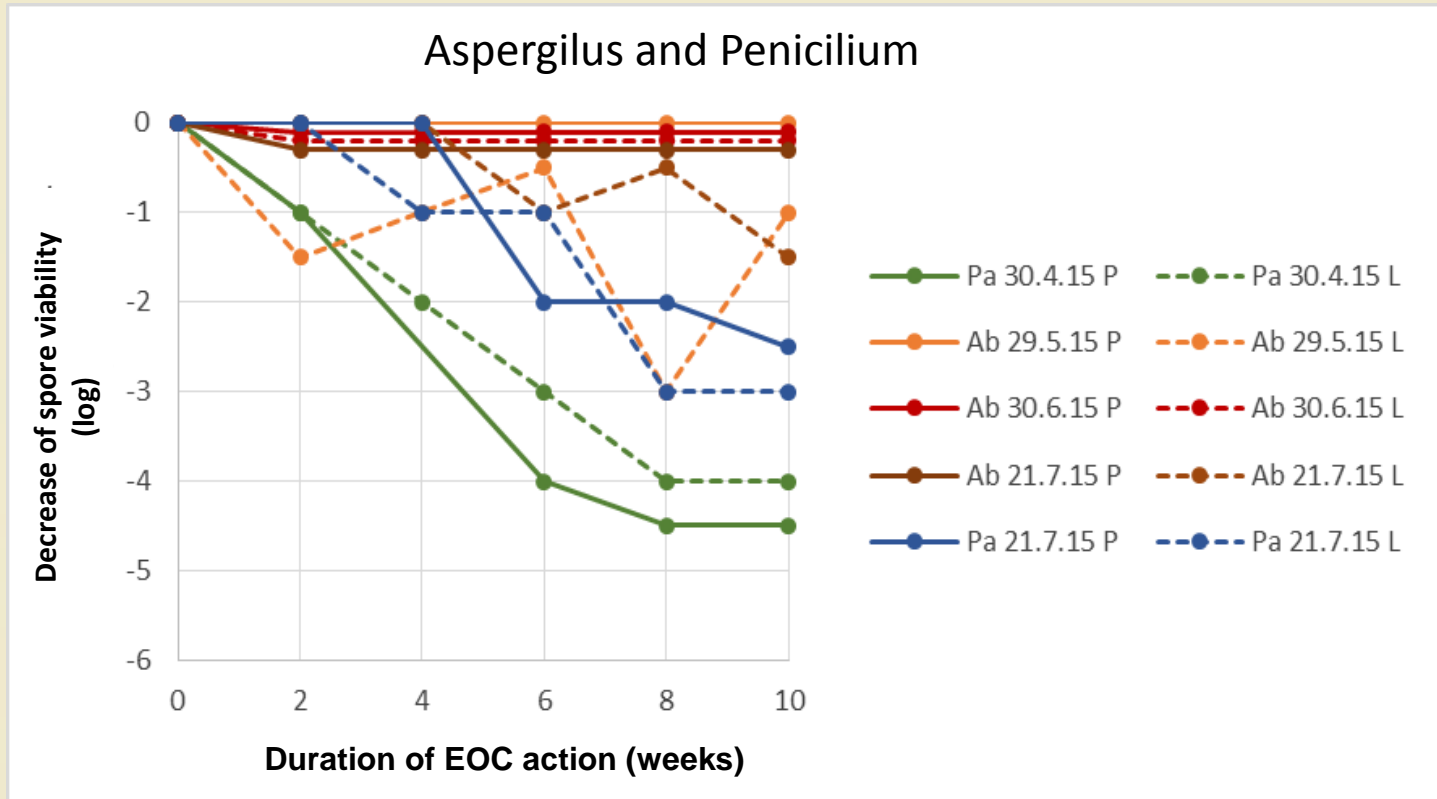
# Model depository

## EOC vapor generator



# Model depository

EOC vapors fungicidal effect on paper substrate



Fungicidal effect is possible only for *P. aurantogriseum*.  
With *A. brasiliensis* we can find only fungistatic effect

# Model depository

## Experiment no. 1

Controll

EOC in depository



### Test 1

Rozpůlené knihy a lepenky byly minimálně 4 týdny skladovány jedna polovina v modelovém deponitáři a odpovídající druhá polovina v kontrolním prostředí.

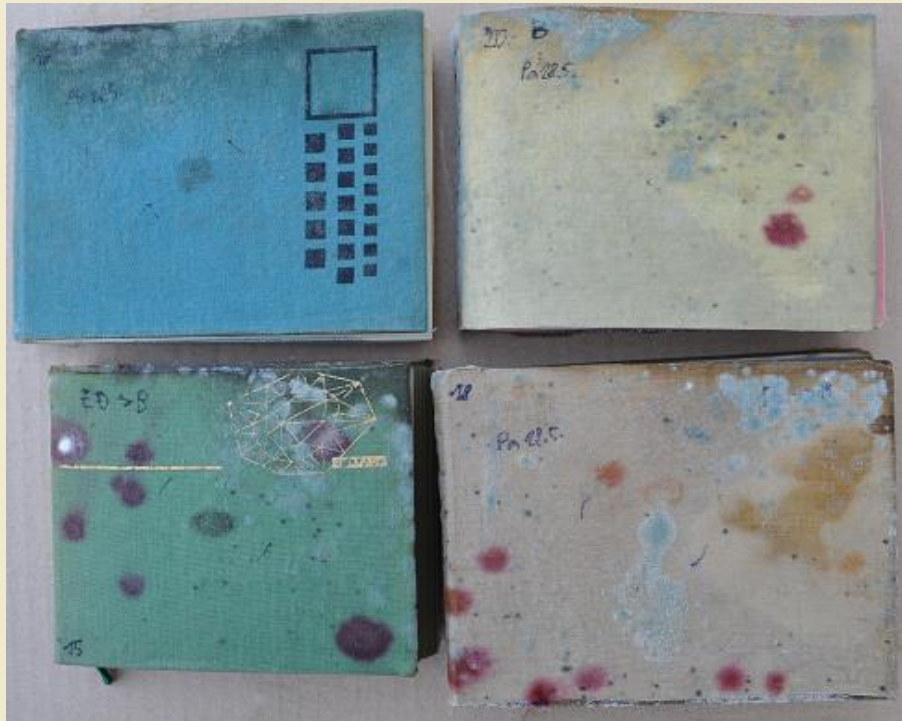
Poté byly vloženy do PVC obalů (propustných pro páry EO), smočeny vodou, uzavřeny v obalech a dále skladovány ve sledovaném prostředí (deponitář s EO / kontrola).

V následujícím období byl pozorován pozvolný nárůst plísní (pocházejících z přirozené kontaminace knih a inokulace testovacích spor) po 1, 2 a 3 týdnech.

# Model depository

## Experiment no. 2 (4 weeks in EOC)

Control (1 week after wetting)



Stav kontrolních knih po 1 týdnu namočení (bez přítomnosti EO).

Všechny čtyři vzorky mají na sobě již vyvinutou sporující plíseň (modro-zelené spory), která se v následujících dnech bez překážek rychle šířila do okolí.

EOC in depository (3 weeks after wetting)



Stav vzorků z modelového deponitáře s parami EO po 3 týdnech namočení.

Po 3 týdnech v mokru se pouze na jedné ze čtyř knih se objevila plíseň. Vytvořila již spory, ale obtížně a zpomaleně se šíří do okolí, protože desky knih do sebe absorbovaly EO. Červené pigmentové skvrny neznámého mikroorganismu se zde neobjevily.



# Model depository

## Experiment no. 3

Book covers laying 3 weeks on moist stack of filter paper standing in water



Control stored out of the depository



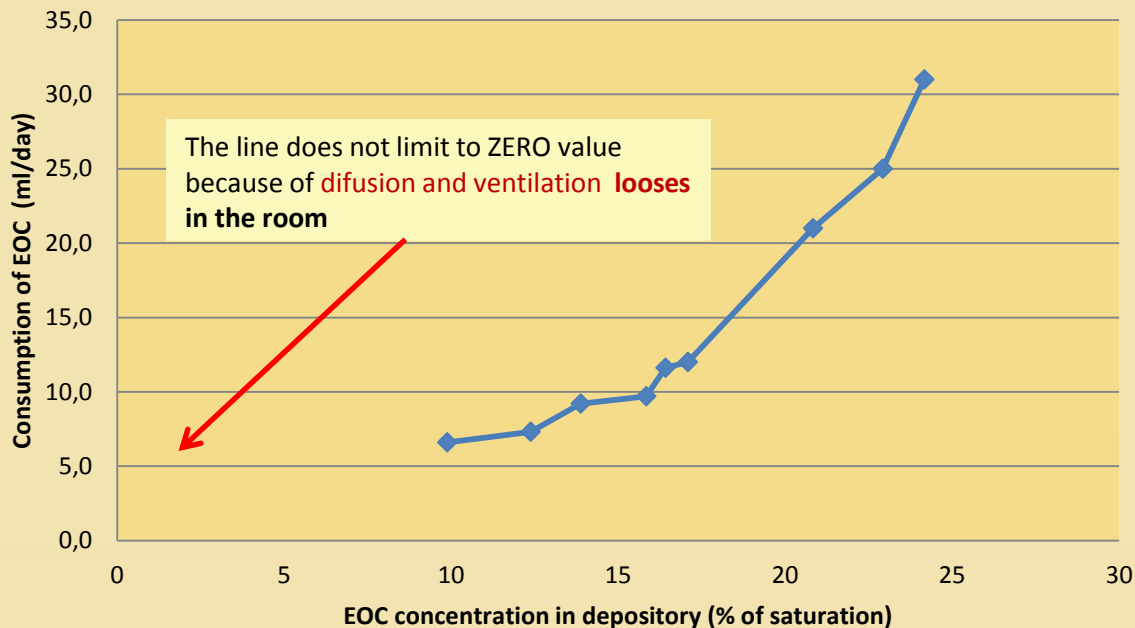
Sample stored 6 weeks in depository with EOC

This is the simulation of book fallen into spill of water in flooded depository. RH of atmosphere surrounding the book was 70 -75% with intensive air circulation. White squares are distances on which lays the load preventing curving the book cover and save its contact with the underlying moist stack of filter paper.

# Model depository

## Practical data

### EOC consumption in 30 m<sup>3</sup> depository



Volume: 30 m<sup>3</sup>

Temperature control:  
21 – 22°C (2 heating units)

RH control: 70 – 75%  
(humidifier + opened vessel with water)

Intensive air circulation  
(two fans in the room)

Price EOC: 618 Kč/kg

30 m<sup>3</sup>

At 15% of saturation we need 10 g EOC/day it is cca **6,18 Kč/day and 2256 Kč/yr**  
1000 m<sup>3</sup> depository **needs 75 190Kč/yr** (similar ventilation and difusion)

The used concn. is not plesant for a longer stay of person

*Thank you for the attention*

•Duration of EOC action (weeks

