

VAPOR PHASE HYDROGEN PEROXIDE FOR DECONTAMINATION IN AGRICULTURE

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Introduction

Institute of Microbiology, Prague, Czech Republic

- **Chemical, biological decontamination**
- **Construction of decontamination equipment**

Cooperations

- **Institute of Chemical Technology, Prague**
- **Institute of Virology, Bratislava, Slovak Republic**
- **Military Research Institute**
- **Army facilities - BSL4 workplace**
- **Private companies**



Decontamination of Large Spaces

Danger of biological threats

- Terroristic attack
- Epidemics or other crisis



Biological Risk

Universal decontamination technology

high efficiency and activity, material compatibility, non-toxicity, odorless, non-staining, resistance to organic material, monitoring capability, environment-friendly use, prolonged reuse life, long shelf life, unrestricted disposal and cost-effectiveness

Decontamination technologies

- heat – effective, easy-to-use, only for movable objects of reasonable size
- irradiation (UV, γ -radiation) – problems with irradiation of large areas
- wet (decontamination solutions) – laborious
- gaseous/vapor – require pre-cleaning

Universal decontamination technology is still actual

Vapor/gaseous Decontamination Technologies

Advantages:

Capable for decontamination of small and/or large areas

Easy-to-use

Applicable at room temperature

Capable to decontaminate complicated spaces (by diffusion)

Vapor/gaseous Decontamination Technologies - Comparison

	ClO ₂	VPHP	Formaldehyde
1. OSHA 8 hr TWA (time weighted average)	0.1 ppm	1.0 ppm	0.75 ppm
2. Odor Detection	Yes	No	Yes
3. Cycle Time (Risk of Exposure)	3-4 hours	4-7 hours	9-15 hours
4. Carcinogen	NO	NO ¹	YES
5. Typical Concentrations	1800 ppm	1000 ppm	8000-10000 ppm
6. Penetration & Distribution	Yes (gas)	No (Vapor)	Yes (gas)
7. Penetrate Water	Yes	No	Yes
Penetrate Oil	No	No	No
Penetrate Grease	No	No	No
8. Emergency Aeration Time	5-30 min	1-6 hours	1 hour + cleanup
9. Residues	None	None	Yes
10. NSF approvals	Yes	No	Yes
11. U.S. EPA approvals	Yes	Yes	No

¹IARC, NTP, and OSHA do not list hydrogen peroxide as a carcinogen. ACGIH lists hydrogen peroxide as an A3 animal

Czarneski M. A., Lorcheim K.; *Applied Biosafety* Vol. 16, No. 1, 2011

Generation of Vaporized Hydrogen Peroxide

Current VHP generators

- require electricity
- expensive
- low performance



Generation of Vaporized Hydrogen Peroxide

Portable generator

- No need of electricity supply
- Remote initiations



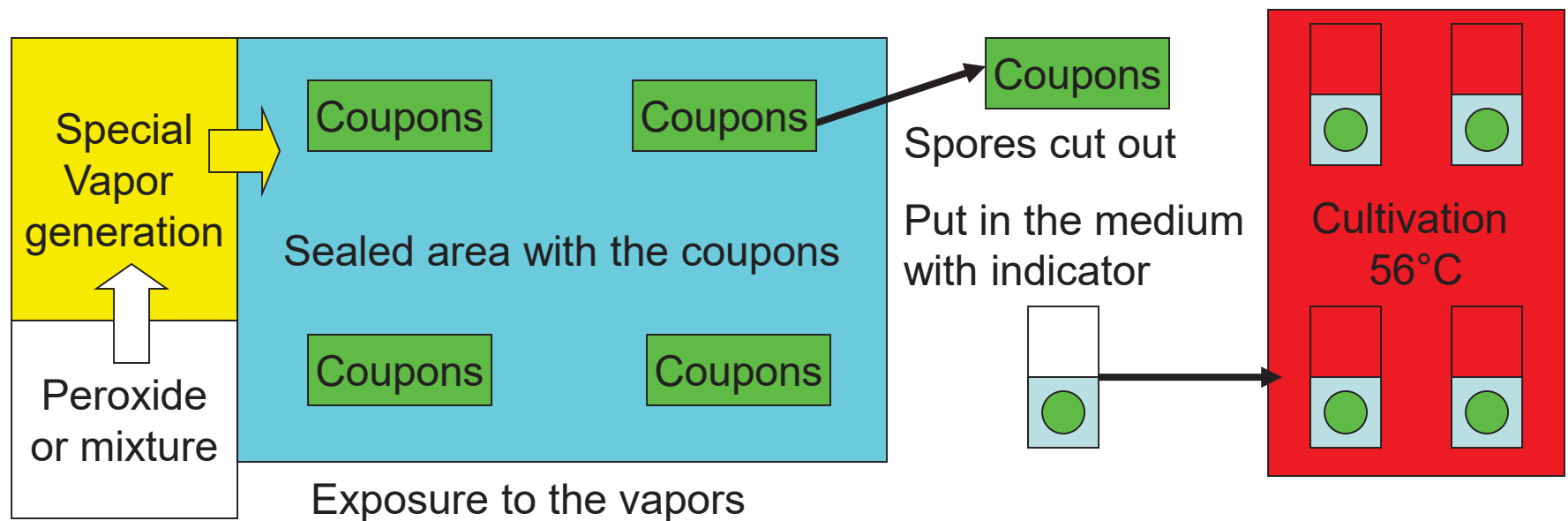
Decontamination of livestock buildings

Livestock buildings:

- **large, complicated space – vapor/gaseous methods are of great advantage**
- **various construction materials – material compatibility of decontamination chemicals**
- **porous materials are present – penetration of chemicals is required**

Penetration test of VHP

- The spores put behind a penetration barrier
- The barrier – filtration paper soaked with blood
- Up to 5 layers arranged



Penetration tests

Number of killed test spots of *Bacillus Stearothermophilus* dependent on used decontaminant, penetration depth and time of exposition
 One test spot = 1×10^6 spores

Decontaminant	penetration depth	time of exposition [h]			
		4	8	12	16
VPHP	1st layer, 0.135 mm	(5/5)	(5/5)	(5/5)	(5/5)
	2nd layer, 0.270 mm	(0/5)	(5/5)	(5/5)	(5/5)
	3rd layer, 0.405 mm	(0/5)	(0/5)	(5/5)	(5/5)
	4th layer, 0.540 mm	(0/5)	(0/5)	(0/5)	(5/5)
MIX 1	1st layer, 0.135 mm	(5/5)	(5/5)	(5/5)	(5/5)
	2nd layer, 0.270 mm	(5/5)	(5/5)	(5/5)	(5/5)
	3rd layer, 0.405 mm	(0/5)	(5/5)	(5/5)	(5/5)
	4th layer, 0.540 mm	(0/5)	(0/5)	(5/5)	(5/5)
	5th layer, 0.675 mm	(0/5)	(0/5)	(0/5)	(5/5)
	6th layer, 0.810 mm	(0/5)	(0/5)	(0/5)	(3/5)
MIX 2	1st layer, 0.135 mm	(5/5)	(5/5)	(5/5)	(5/5)
	2nd layer, 0.270 mm	(1/5)	(5/5)	(5/5)	(5/5)
	3rd layer, 0.405 mm	(0/5)	(2/5)	(5/5)	(5/5)
	4th layer, 0.540 mm	(0/5)	(0/5)	(1/5)	(5/5)
	5th layer, 0.675 mm	(0/5)	(0/5)	(0/5)	(5/5)

Comparison of potentiation mixtures

Nuber of killed test spots of *Bacillus Stearothermophylus* without barier dependent on used decontaminant (pure additivum or mixture with VPHP) and time of exposition One test spot = 1×10^6 spores

Time of exposition [min]	Decontaminant				
	VPHP	ADD 1	ADD 2	MIX 1	MIX 2
5				(1/3)	(0/3)
10				(3/3)	(0/3)
15	(0/3)	(0/3)	(0/3)	(2/3)	(1/3)
20				(3/3)	(0/3)
25				(3/3)	(0/3)
30	(1/3)	(2/3)	(0/3)	(3/3)	(0/3)
35				(3/3)	(2/3)
40				(3/3)	(1/3)
45	(2/3)	(3/3)	(0/3)	(3/3)	(2/3)
50				(3/3)	(2/3)
55				(3/3)	(3/3)
60	(1/3)	(3/3)	(0/3)	(3/3)	(3/3)
75	(3/3)	(3/3)	(0/3)	(3/3)	(3/3)
180	(3/3)	(3/3)	(0/3)	(3/3)	(3/3)

Results

- Special barrier was developed**
- The 7 additives was tested**
- The significant results by 2 additives**
- It seems to be a right direction**

Decontamination of livestock buildings

Collection of soil in a swine and poultry farm

Cultivation with sulfamethoxazole

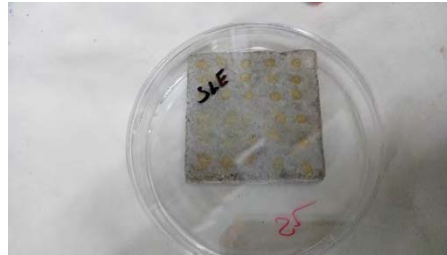
SMX resistant bacterial consortium

Test spots

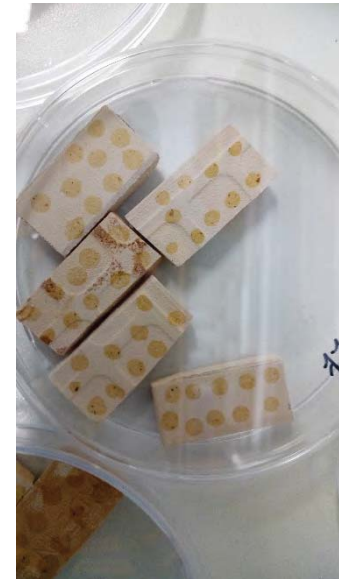
Experimental



carpet



concrete



non-glaze ceramics



wood

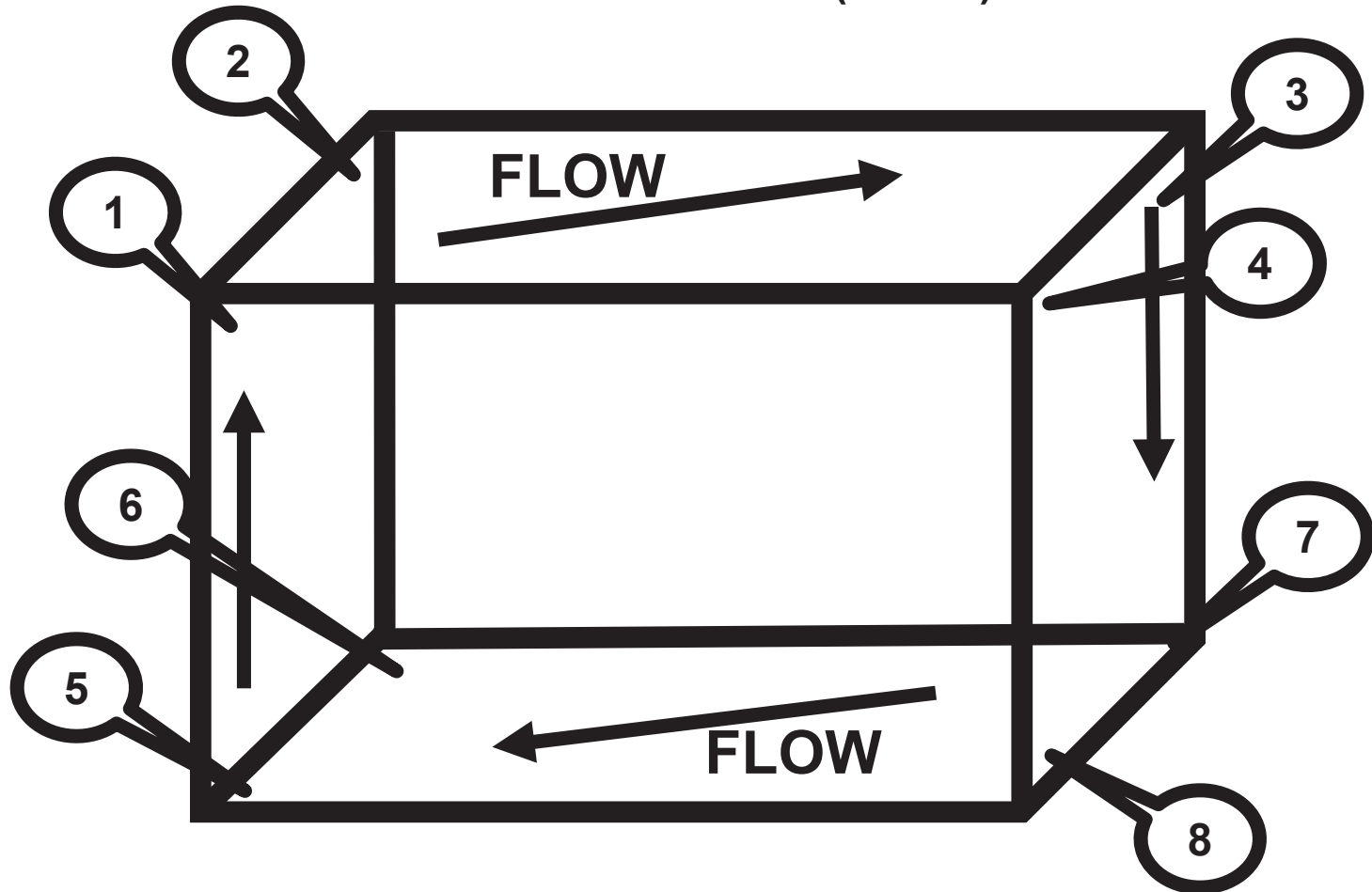


galvanized metal

each spot – 10^7 of CFU

Decontaminated box

Volume: 42.7 cu in (0.7 m³)



Experimental - decontamination

Average temperature: 68 °F

Duration: 1 hour per decon cycle



Characterization of microorganisms

Sulfamethoxazole resistant microorganisms

- Identified by 16s rRNA

Consortium from a swine farm

- *Staphylococcus equorum*
- *Staphylococcus lentus*
- *Bacillus cereus*
- *Lysinibacillus cresolivorans*

Consortium from a poultry farm

- *Lysinibacillus macrolides*
- *Psychrobacter* sp.
- *Kurtia gibsonii*
- *Enterococcus gallinarum*

Results of decontamination

Decontamination of microorganisms from the swine farm

Material	Number of spots	Decon cycles	H ₂ O ₂ consumption	Position in the box	Efficiency	
					24 h cultivation	120 h cultivation
			g H ₂ O ₂ /m ³			
Metal	80	3	345	1 - 8	100 %	100 %
Non-glaze ceramics	160	3	345	1 - 8	100 %	100 %
Carpet	160	4	460	1 - 8	100 %	100 %
Wood	24	4	248	1 - 8	100 %	100 %
Gypsum	160	3	186	3, 4	12.5% of spots reduced of 1 order to 10 ⁷	87.5% of spots reduced by 1 order to 10 ⁸
				1, 2	81.5% of spots dropped by 7 orders	
				5	6% of spots - 100% efficiency	6% of spots - 100% efficiency
Concrete	240	3	345	1.2.3.4.	50% of spots reduced by 2 orders to 10 ⁶	0 %
				5.6.7.8	50% of spots reduced by 5 orders to 10 ³	0 %

Results of decontamination

Decontamination of microorganisms from the poultry farm

Material	Number of spots	Decon cycles	H ₂ O ₂ consumption	Position in the box	Efficiency	
			g H ₂ O ₂ /m ³		24 h cultivation	120 h cultivation
Non-glaze ceramics	80	3	345	1 - 8	100 %	100 %
Wood	80	3	345	2, 4 only	100 %	87.5 %

Material compatibility

Wood



Before

After

Non-glaze ceramics



Before

After



Before



After

Decontamination by VHP

Cons

- **High efficiency and activity**
 - **Material compatibility**
 - **Non-toxicity**
 - **Odorless**
 - **Environment-friendly use**
 - **Long shelf life-time**
 - **Unrestricted disposal**
 - **Cost-effectiveness**
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- Porous materials can be decontaminated**
 - The higher dosage required**
 - The penetration can be an issue**
 - The efficient distribution in the decontaminated area is crucial**

Acknowledgment

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