



## Engineering and Work Practice Controls for the Use of Anesthetic Gases During BSL-3 Rabbit Studies

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# The University of Pittsburgh RBL



# Vaccine Study Design

## **20 New Zealand White rabbits**

Vaccinate with BSL-1/BSL-2 agent

Bleed weekly for 4 weeks

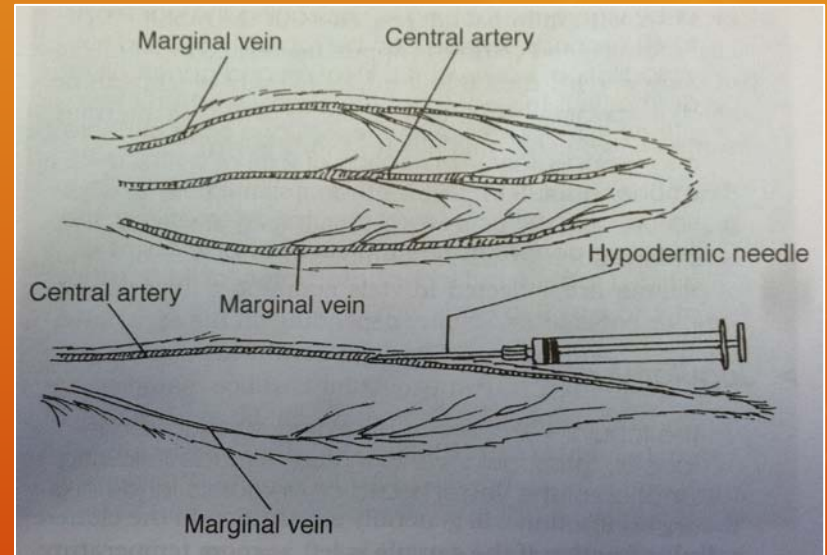
Aerosol challenge with virulent *F. tularensis*

Bleed every 2-3 days for up to 28 d

Euthanize and necropsy

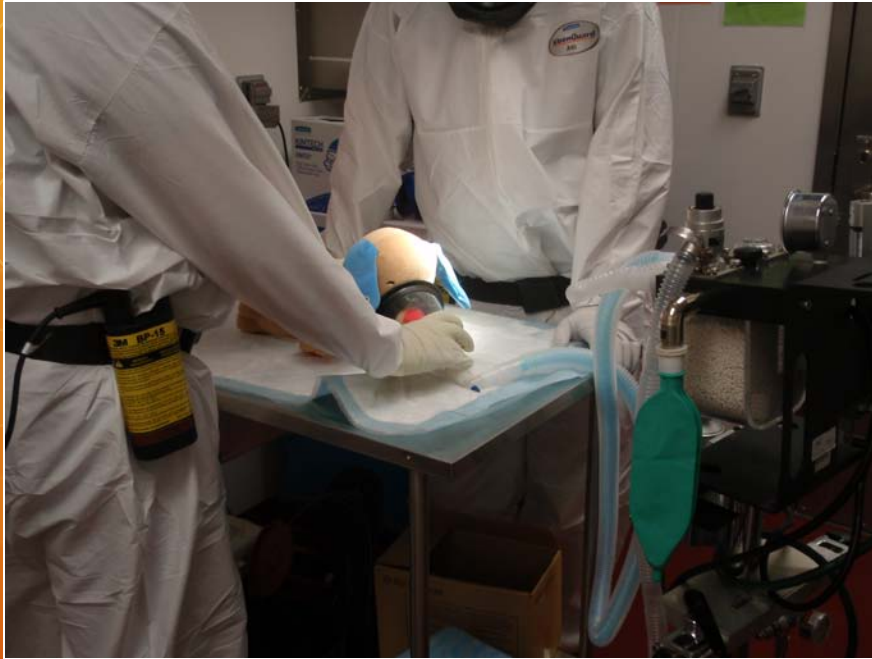


# Rabbit Blood Sampling via Ear Vein/Artery

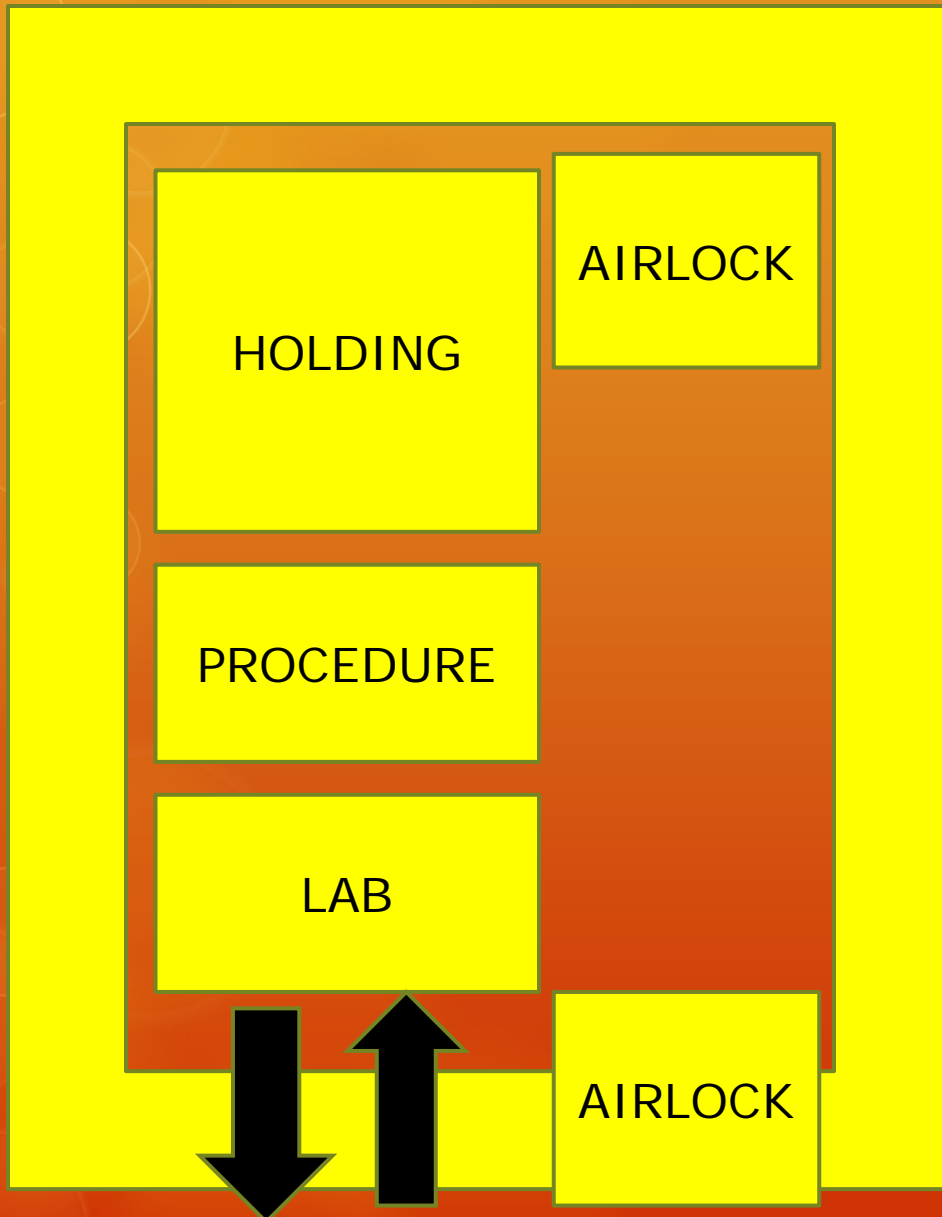


*The Laboratory Rabbit, CRC Press, 1996*

# Rabbit Blood Sampling using Isoflurane



# Animal Suite Layout





# HEPA and Organic Vapor/HEPA filters



# Chem Disk Monitoring Badges for HAG





# Chem Disk monitoring for isoflurane



Monitoring employee exposure



Monitoring the room level

Session #	Technician #	Location of Monitor	Exposure to isoflurane (PPM)
1	1	Employee exposure	None Detected (<0.6 ppm)
1	1	Room level	73
2	1	Employee exposure	0.95
2	1	Room level	500
2	2	Employee exposure	1.2
2	2	Room level	Not performed

# Portable Engineering Control



Active Scavenge System, Vetquip, Queensland, Australia

# Large Animal Anesthesia SOP

- Don combination organic vapor-HEPA filter on PAPR
- Post signage in PAPR room and procedure room that combo filter is required
- Connect scavenging pump to exhaust line of anesthesia circuit
- Connect absorbent charcoal canister to outlet of scavenging pump
- Ensure charcoal canister is new or has adequate capacity for procedure
- Turn on evacuation pump
- Refill anesthesia canister using pouring assembly and report/clean up all spills immediately
- Perform anesthesia circuit leak test
- Place properly-sized nose cone with sealing gasket on animal, ensure proper fit, and then turn on anesthesia canister
- Reduce flow rate when animal appears to be properly sedated
- Turn off anesthesia canister when blood sampling completed
- Flush system by pressing purge button at least twice
- Remove nose cone from animal
- Turn off scavenging pump
- Weigh the charcoal canister and record or dispose if at capacity
- Report any problems to supervisor



# Engineering Controls



BSC in lab

Downdraft table



Drop-down 'snorkel'  
in imaging suite

# Aerosol Challenge



# Study Design

## **Load Chem Disk badges with gases at Assay Tech**

Store half; ship half for VHP treatment at Pitt RBL

Obtain 24-hour confirmatory results for VHP efficacy

Ship VHP-treated badges back to Assay Tech

Blind analysis of all badges for measured levels of gases

<b>Halothane, ~200 ug</b>	<b>Halothane, ~300 ug</b>	<b>Halothane, ~25 ug</b>	<b>Halothane, ~50 ug</b>
<b>Isoflurane, ~100 ug</b>	<b>Isoflurane, ~200 ug</b>	<b>Isoflurane, ~15 ug</b>	<b>Isoflurane, ~30 ug</b>
1-Stored	6-Stored	11-Stored	16-Stored
2-Stored	7-Stored	12-Stored	17-Stored
3-Stored	8-Stored	13-Stored	18-Stored
4-Treated with VHP	9-Treated with VHP	14-Treated with VHP	19-Treated with VHP
5-Treated with VHP	10-Treated with VHP	15-Treated with VHP	20-Treated with VHP



# VHP in Class III Biosafety Cabinet



# Results: Badges 1-10

<u>Halothane ~200 ug</u>	<u>Isoflurane ~100 ug</u>
1-Stored: 220 ug	1-Stored: 110 ug
2-Stored: 240 ug	2-Stored: 120 ug
3-Stored: 240 ug	3-Stored: 130 ug
4-Treated: 240 ug	4-Treated: 120 ug
5-Treated: 260 ug	5-Treated: 130 ug
Mean-Stored: 233.33 +/- 11.55	Mean-Stored: 116.67 +/- 5.77
Mean-Treated: 250 +/- 14.14	Mean-Treated: 125 +/- 7.07
Student's T-test: <b>No significant difference</b>	Student's T-test: <b>No significant difference</b>

<u>Halothane ~300 ug</u>	<u>Isoflurane ~200 ug</u>
6-Stored: 320 ug	6-Stored: 160 ug
7-Stored: 360 ug	7-Stored: 170 ug
8-Stored: 360 ug	8-Stored: 170 ug
9-Treated: 310 ug	9-Treated: 150 ug
10-Treated: 360 ug	10-Treated: 170 ug
Mean-Stored: 346.67 +/- 23.09	Mean-Stored: 166.67 +/- 5.77
Mean-Treated: 335 +/- 35.36	Mean-Treated: 160 +/- 14.14
Student's T-test: <b>No significant difference</b>	Student's T-test: <b>No significant difference</b>

# Results, Badges 11-20

<u>Halothane ~25 ug</u>	<u>Isoflurane ~15 ug</u>
11-Stored: 23 ug	11-Stored: 12 ug
12-Stored: 25 ug	12-Stored: 15 ug
13-Stored: 25 ug	13-Stored: 16 ug
14-Treated: 26 ug	14-Treated: 16 ug
15-Treated: 27 ug	15-Treated: 17 ug
Mean-Stored: 24.33 +/- 1.15	Mean-Stored: 14.33 +/- 2.08
Mean-Treated: 26.5 +/- 0.71	Mean-Treated: 16.5 +/- 0.71
Student's T-test: <b>No significant difference</b>	Student's T-test: <b>No significant difference</b>

<u>Halothane ~50 ug</u>	<u>Isoflurane ~30 ug</u>
16-Stored: 48 ug	16-Stored: 33 ug
17-Stored: 50 ug	17-Stored: 34 ug
18-Stored: 54 ug	18-Stored: 36 ug
19-Treated: 48 ug	19-Treated: 34 ug
20-Treated: 50 ug	20-Treated: Not detected
Mean-Stored: 50.67 +/- 3.06	Mean-Stored: 34.33 +/- 1.53
Mean-Treated: 49 +/- 1.41	Mean-Treated: N/A
Student's T-test: <b>No significant difference</b>	Student's T-test: <b>Not performed</b>



# Ongoing Monitoring Results- Rabbit Tularemia Study

<u>Session:</u>	<u>Category:</u>	<u>Measured iso, PPM:</u>
1	Employee exposure	ND(<0.6 ppm)
1	Employee exposure	None detected
2	Employee exposure	None detected
2	Employee exposure	None detected
3	Employee exposure	None detected
5	Employee exposure	None detected
1	Room level	15
1	Room level	9.8
2	Room level	21
2	Room level	14
3	Room level	ND (<0.6 ppm)
3	Room level	12
4	Room level	13
4	Room level	12
5	Room level	7.9
6	Room level	9.2

# Rabbit Blood Sampling Continues to Evolve...



# So Does Small and Large Animal Monitoring

<b><u>Species</u></b>	<b><u>Procedure</u></b>	<b><u>Location</u></b>
Rats, Mice, Ferrets	Euthanasia and Necropsy	Procedure Room, Laboratory
Mice, Ferrets, Old World & New World Primates	Preclinical Imaging	Imaging Suite
Ferrets	Infection	Laboratory
Rabbits	Blood Sampling	Procedure Room
New World Primates	Telemetry Implantation	Procedure Room



# Thank You

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