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# Emerging Rodent Pathogens and Their Role in the Rodent Sentinel Program

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# Outline

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- What organisms to include
- Testing Methodologies
- Biosecurity
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# Introduction

- A health monitoring program should be a dynamic entity of any laboratory animal program. Health status can directly impact research results and should be protected through routine monitoring of animals within the colony.

- The design should include appropriate sample size and organisms monitored, as well as reliable testing methodology.

- Procedures should be in place to reduce introduction of pathogens, including appropriate biosecurity measures and approval of animal sources.

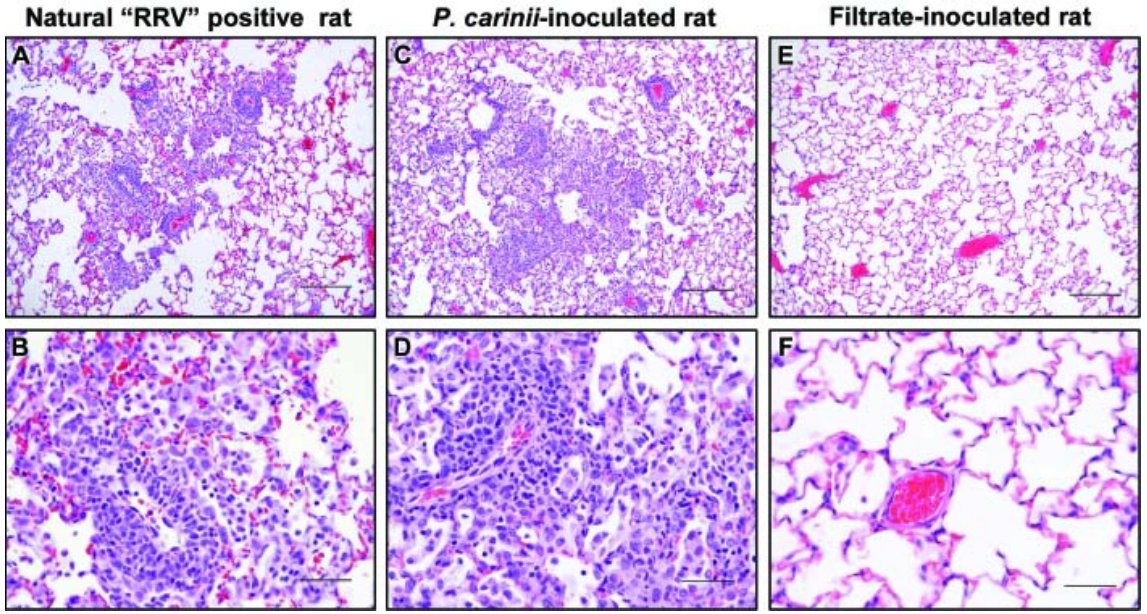
- Formulas exist to calculate proper sample size based on known organism prevalence and colony size, but what organisms should be monitored? How does one account for emerging pathogens?

# Novel organisms – Murine Norovirus[5]

- Murine Norovirus
  - Discovered in 2003 in immunocompromised mouse colony; clinical symptoms elicited in immunodeficient mice
  - Immunocompetent mice can be infected but may not display clinical symptoms

# Novel organisms – *Pneumocystis carinii*[6]

- *Pneumocystis carinii*
  - Determined in 2010 as causative agent of idiopathic lung lesions in rats
  - Example of known disease with no causative agent identified; now easily monitored within colonies




Livingston et al. 2011

# Sample testing size

- ILAR formula (1976)
  - Statistical analysis
  - Infectivity of organism
  - Production and husbandry procedures

- Number of animals sampled depends on nature of infection and husbandry and caging

- As prevalence of infection decreases, sample size required to detect the infection increases

# Sample testing size

**Table 1 Calculation of the number of animals to be monitored**

Diseases with an infection rate of 50% or more (Sendai, MHV) require far fewer animals to detect their presence than diseases with low infection rates.

**Assumptions**

1. Both sexes are infected at the same rate
2. Population size > 100 animals
3. Random sampling
4. Random distribution of infection

The sample size is calculated from the following formula:

$$\frac{\log 0.05}{\log N} = \text{Sample size}$$

$N$ = percentage of non-infected animals
0.05 = 95% confidence level

Relation of sample size to prevalence rate

| | Sample sizes at different confidence levels | | |
|---|---|---|---|
| Suspected prevalence rate (%) | 95% | 99% | 99.9% |
| 10 | 29 | 44 | 66 |
| 20 | 14 | 21 | 31 |
| 30 | 10 | 13 | 20 |
| 40 | 6 | 10 | 14 |
| 50 | 5 | 7 | 10 |

Example: 10 animals should be monitored to detect at least one positive animal if the suspected prevalence rate of an infection is 30% (confidence level: 95%)

Nicklas et al. 2002

# Sentinel Type

- Contact Sentinels
  - Most reliable
  - Susceptibility to organisms can be strain specific

- Dirty Bedding Sentinels
  - Susceptibility to organisms can be strain specific
  - Many organisms do not transmit well via soiled bedding

- Colony resident
  - Randomly selected
  - Represent true susceptibility of population (if immunocompetent)

# Other sample types

- Tissue samples
- Blood samples
- Fecal samples
- Environmental samples

# What organisms to include

- FELASA recommendations
- Vendor health reports
- Organisms within the research scope
  - May desire animals negative for organism being studied

# Testing Methodologies - Serology

Pros

- Inexpensive
- Easy to collect blood sample
- Can assay multiple organisms, multiple antigens
- Highly sensitive and specific

Cons

- Can take several weeks for seroconversion
- Does not indicate if infection is active or historical
- False positives can occur with older animals

# Testing Methodologies - PCR

## Pros

- Highly sensitive and specific
- Multiplex approach possible
- Detect active infection or environmental contamination
- Use multiple sample types

## Cons

- High cost
- Short duration of shedding may necessitate frequent sampling
- DNA may be degraded
- Susceptible to contamination

# Testing Methodologies - Microbiology

## Pros

- Relatively low cost
- Tests for wide range of organisms
- Detects active infection

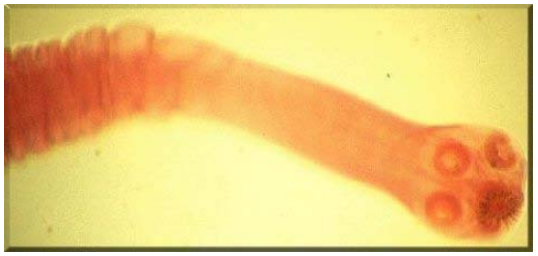
## Cons

- Susceptible to contamination
- Requires careful sample handling
- Fastidious organisms can be difficult to detect
- Variable characteristics of some organisms

# Testing Methodologies - Parasitology

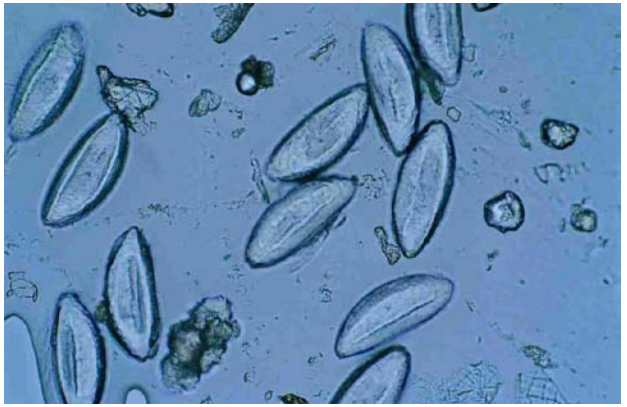
## Pros

- Relatively low cost
- Tests for wide range of organisms
- Detects active infection



## Cons

- Relatively insensitive
- Variable shedding of some organisms
- Dependent on skill of technician

# Biosecurity

- Correlates to success of health monitoring program
  - Entry procedures
  - Sterilization of supplies
  - Cage changing
  - Animal procedures

- Proper review of animal sources entering facility
  - Quarantine if needed

- Implement biosecurity auditing program within one's facility to ensure compliance and health of animals

# Conclusion

- Proper sampling size, sentinel selection, and test methodology lead to protection of research results by reducing variation due to health status

- Novel organisms may be more quickly discovered and therefore controlled or eradicated through proper health monitoring program implementation

- Health monitoring is an integral component of proper animal research and measures should be taken to ensure accuracy and integrity of the health monitoring program.

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