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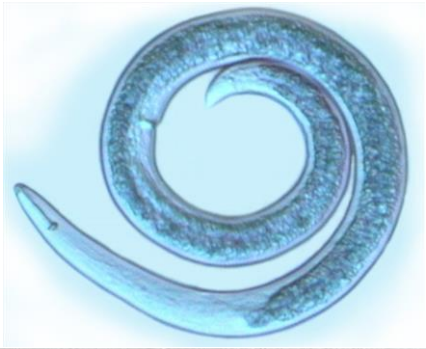
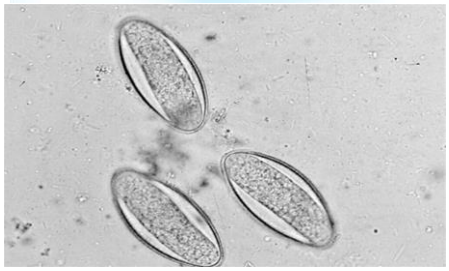
In Focus

Center of Comparative Medicine & Pathology
Research Animal Resource Center
Laboratory of Comparative Pathology





Aspiculuris tetraptera

Adult Image credit: http://www. pasozyty.org.pl/page42.html
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Pinworms- Why the fuss?

Pinworms remain on the list of agents that should be excluded from facilities, due to effects on animal health and research. While rodent pinworms do not pose a zoonotic risk and infections are usually subclinical in immunocompetent mice, heavy parasite burdens may result in unthrifitiness, hepatic granulomas, catarrhal enteritis, rectal prolapse, intussusception, fecal impaction, and perianal irritation. Pinworms in rodents have been shown to influence behavior, gastrointestinal physiology, immunology, growth and hematopoiesis. Pinworm infections may also affect research by limiting the ability of investigators to transfer mice to other institutions.

Inside ~ New Faces in the CCMP

Pinworm Infections in Mice: A Review and Response to an Outbreak

Murine pinworm infections remain a concern for institutions utilizing mice for research. Many institutions experienced pinworm outbreaks recently after receiving infected mice from a well respected and approved vendor. A review of the biology and disease associated with murine pinworms is provided in addition to discussing the outbreak and RARC's response.

Syphacia obvelata and *Aspiculuris tetraptera* are the pinworms of concern for laboratory mice with worldwide distribution[9]. Even though *Syphacia muris*, the common pinworm of laboratory rats, can occasionally infect laboratory mice[8], it is not common and can be identified by the same detection methods used to detect *S. obvelata*. First described in the early 1800s[8], these endoparasites have direct life cycles and transmit infections via ingestion of embryonated (infective) eggs[1,8].

S. obvelata resides in the cecum[8] and has a prepatent (incubation) period of 11-15 days[9]. Similar to the human pinworm, *Enterobius vermicularis*, gravid female worms migrate to the anus, lay up to 350

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Environmental Enrichment for the Laboratory Mouse

The first part of this article published in the Spring/Summer 2014 issue of *In Focus* reviewed the goals of environmental enrichment programs with a focus on providing enrichment to laboratory mice[1-4]. Enrichment can be divided into five major categories: social, occupational, physical environment modification, sensory and nutritional[1,2]. Each category encourages different natural behaviors. In this article, we will review the various types of enrichment that may be provided to laboratory mice.

Group housing is the most effective and efficient method of providing social enrichment to laboratory mice[1-4]. The presence of conspecifics provides complex and varied stimulation daily. Mice naturally form groups called demes, consisting of a single dominant male, one or more females, and other subordinate males. Although these conditions cannot always be replicated in a research setting, mice are a gregarious species and they have an inherent need to interact with each other. Conspecifics may help mice adjust to stress induced by research procedures or the provision of

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Behind the Curtain: All About PPE

The Occupational Health and Safety Administration (OSHA) mandates that institutions provide a safe workplace for employees. In a vivarium, ensuring personnel safety involves the use of PPE (personal protective equipment). PPE includes a wide variety of items, such as gloves, lab coats, bonnets, face shields, eye shields, shoe covers, respirators and of course, the ubiquitous disposable gowns used within RARC's vivaria. The PPE requirements for a given area can vary widely based on species, use of hazardous materials, the type of facility and animal housing utilized (barrier), and even personal health history. In barrier facilities such as LIC, ZRC, the new BRB, and the RRL barrier, PPE is donned at the barrier entrance with the same PPE utilized between most rooms in the barrier. In non-barrier operated vivairia, such as Cornell's main facility and a large part of the RRL, PPE is donned and removed inside each room. The PPE requirements for a particular area are always posted near the entrance to that area. Policies regarding PPE in the vivarium are developed by RARC in conjunction with institutional employee health and environmental health and safety groups. Appropriate PPE is supplied by RARC for use within the vivaria.

People frequently think of bites, scratches, and zoonotic diseases as the major safety risk associated with handling lab animals. While these do exist, the risk of zoonotic disease transmission from specific pathogen free (SPF) laboratory animals is quite small. And while PPE may provide some protection from bites and scratches, the best prevention for those injuries is the use of appropriate handling and restraint techniques (including anesthesia when appropriate) by trained personnel. The most significant health concern for people working with laboratory animals is actually lab animal allergies.

Laboratory animal allergies (LAA) are common among individuals with

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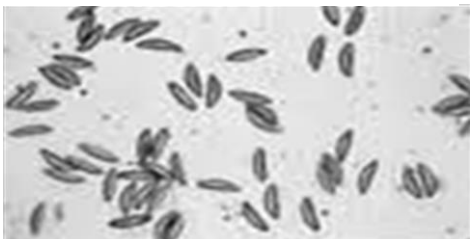
Syphacia obvelata

Adult Image credit: http://www.vet-parasitology.com/oxyurida.php
Egg Image credit:
http://jaxmice.jax.org/jaxnotes/archive/487f.html



Please Welcome Dr. Heather Martin

Dr. Heather Martin joined the CCMP as a Senior Clinical Veterinarian in September. Heather has a wealth of experience in both academia and industry. Prior to veterinary school, Heather worked in a therapeutic ultrasound lab for several years where she developed an interest in laboratory animal medicine. Pursuing that interest she attended Kansas State University's College of Veterinary Medicine. Upon graduation from Kansas State in 2006, Heather completed postdoctoral training in comparative medicine at the Massachusetts Institute of Technology. She is a Diplomate of the American College of Laboratory Animal Medicine.

Dr. Martin will oversee the provision of clinical care to animals housed at the Rockefeller Research Laboratory (MSKCC), the S-building (WCMC) and the Belfer Research Building (WCMC), as well as Weill Cornell's Research Animal Surgical Facility. Heather enjoys collaborating with and supporting investigators with their research utilizing animal models and is excited to join MSKCC and WCMC.

Pinworm Infections in MIce

Cont. from pg. 1

'sticky' eggs, which adhere to the perianal area, and die[8]. Eggs embryonate in 5-20 hours[8]. Naive mice are infected after ingestion of these eggs from either the perianal area or the environment[8].

A. tetraptera resides in the colon and has a longer prepatent period of ~24 days after which females begin laying eggs intermittently[8]. Females can live for 3 more weeks, resulting in a lifespan of approximately 45-50 days[8]. In contrast to *S. obvelata* eggs, *A. tetraptera* eggs are passed in the feces[8] and embryonate in 5-8 days[8]. *S. obvelata* and *A. tetraptera* can be distinguished morphologically as well as by the shape and size of their eggs[1,8,9].

Traditionally, in live animals *S. obvelata* is diagnosed via cellophane anal tape tests[1,8,9] and *A. tetraptera* is diagnosed via fecal flotation[1,8,9]. While antemortem testing may be the simplest, it is generally less sensitive and can result in more false negative results than postmortem testing methods[2]. Direct evaluation of cecal and colonic contents at necropsy is considered to be the most accurate diagnostic method[4]. Recently, PCR has been introduced to augment traditional testing methods[4,5]. Due to the potential for false negative diagnostic results, all newly received animals from non-RARC-approved institutions/vendors are treated for pinworms while in quarantine.

Little is known about the survival of pinworm eggs in the environment. *S. obvelata* appears to be more fastidious than the other murine pinworms and has failed to embryonate in most media[8]. While eggs of *S. obvelata* were reported to be viable for less than 48 hours at room temperature[8], *S. muris* eggs were shown to be viable for almost a month[3]. However, pinworm eggs have not been documented in the environment in modern facilities utilizing microisolator caging[7]. The need for environmental treatment is debatable. While *S. obvelata* has been successfully eradicated from rodent colonies with oral anthelmintic treatment alone[7], there are no published reports of eradication of *A. tetraptera* without concurrent decontamination of the environment[10].

Earlier this summer RARC was informed by one of our principal animal suppliers that mice in one of their production barrier units tested positive for *S. obvelata*. They determined that any mice shipped from that unit for the preceding 3 months could have been infected. RARC's Biosecurity Veterinarians promptly determined that mice from these shipments were received into four rooms in three vivaria at both MSKCC and WCMC. Animals that were still present from these shipments were tested extensively. Only one shipment of mice received into a single room tested positive for *S. obvelata*. However, quarantine was established for all four rooms until repeated negative tests results were confirmed over the ensuing months. Investigators and staff were required to don extra personal protective equipment (PPE) upon entering and instructed not to enter any other rooms in the facility after being in pinworm suspect rooms. In addition, transfers of animals from all four rooms were suspended. All animals from the confirmed positive shipment were moved to the rodent quarantine facility. All animals in the infected room and those moved to quarantine were treated for pinworms with fenbendazole impregnated feed for eight weeks.

Through the timely response of the Biosecurity Veterinarians as well as the cooperation of the affected research staff, pinworms were believed to have been eliminated from the vivaria allowing investigators to continue their research with confidence in the health status of their mice. While RARC has a high level of confidence that the infection was eradicated, increased surveillance testing will continue through February 2015.

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Environmental Enrichment, *Cont. from pg. 1*

Lab mice, "like" their wild counterparts wood mice, *Apodemus sylvaticus*, prefer social groups.
Image credit:
http://www.culture24.org.uk/science-and-nature/art82478

Natural or commercially nutritional supplements provide enrichment offering diversity and fostering foraging behavior.
Image credit: http://www.shutterstock.com/video/clip-340447-stock-footage-the-mouse-gnaws-carrots-on-a-white-background.html

husbandry[1-3]. The benefits of group housing may nonetheless be negated by overcrowding. An increased density can lead to higher levels of stress, aggression, and health problems[1]. Additionally, movement of mice between cages can disrupt dominance hierarchies and lead to aggression. Therefore, maintaining an appropriate animal density and social order is crucial for preventing the development of maladaptive behaviors and assuring quality animal care.

Occupational enrichment refers to enrichment that encourages behaviors that mice would engage in for a large proportion of their time in the wild[1]. Examples include nest building and foraging. This type of enrichment promotes species-typical behavior, is the most likely to improve animal welfare, and provides animals with a measure of control over their environment[1-3]. There are many materials that may encourage nest building. The materials provided and the amount of effort needed to shape the nest will affect its quality. Some common nesting materials include compressed cotton squares (Nestlets™, crinkled paper, and tissue paper. Cotton squares encourage natural behavior by having mice manipulate the material and also provide insulation for thermoregulation. Squares can be compressed to different densities, with the least compressed being easier to manipulate and providing higher quality nests[5]. Crinkled paper also provides insulation and has the added benefit of reducing eye lesions in nude mice and other strains lacking eyelashes that, when present, serve as a barrier to small particles and short fiber debris. Tissue paper can also be used by mice to build nests, though this is not their preferred material and nest quality tends to be lower[6]. Also, its high dust content can be problematic for mice without lashes[1]. Little work has been published on the benefits of foraging to mice, yet anecdotal evidence demonstrates that further work should be performed on this subject. When provided with foraging boxes or bags containing nesting material or treats, mice will work to remove these items from the boxes and use them[1].

The physical environment in which mice are kept may also help improve their welfare[1-4]. Increasing cage size has not been shown to necessarily benefit laboratory mice; however, housing density does influence the level of aggression observed as overcrowded mice are more likely to display signs of aggression[1,4]. Increasing cage complexity improves learning and memory, brain physiology and responses to distress[1]. One disadvantage of increasing cage complexity is that it may incite aggression by providing highly valued resources worth fighting for, and by creating elevated platforms from which mice can attack each other[1,6,7]. Accessories that can be added to the cage environment include shelters and tubes[1-3,6]. Although plastic shelters are more durable than paper shelters, they have been shown to increase aggression between mice[6-8].They may be beneficial for female or singly housed mice, but their use should be carefully evaluated for groups of male mice or aggressive strains. Tubes may also be used as they allow multiple animals to group together as if burrowing and reduce wire-gnawing behaviors[1]. In any case, shelters should provide multiple entry points so that submissive mice are not trapped by more aggressive ones.

Sensory stimulation using olfactory cues is provided by the mice themselves as a way to mark territory and establish a dominance hierarchy[1]. Transferring a small amount of nesting material from the nest during cage change can reduce aggression between mice, as the olfactory cues are transferred as well, reducing the need to re-establish a hierarchy. Other types of sensory stimulation have not been evaluated.

Finally, nutritional enrichment may also improve the welfare of mice by providing a diversity of feeds for them to consume[1]. This type of enrichment cannot be provided to animals on special diets, or on food consumption studies, as this may confound study results. Natural types of feed that can be provided include seeds, mealworms or grains. Mice find these items highly desirable and spreading them around a cage can stimulate foraging behaviors. However, these naturally sourced foods can sometimes contaminate facilities with infectious organisms or affect research by exposing mice to pesticides, fungal toxins, or unknown impurities. Alternatively, commercially processed treats such as crumbles, pellets, etc.. can provide the same benefits but are controlled and typically formulated to minimize biosecurity risks[1,4].

Providing enrichment to laboratory mice is synonymous with providing quality care. This improves the lives of the animals used in biomedical research and helps maintain the validity of scientific data[7,9]. A team approach involving researchers, husbandry and veterinary staff, and administrators is needed to maintain a high standard of care and prevent or minimize unwanted outcomes.

References (see left inset)

Environmental Enrichment for the Laboratory Mouse

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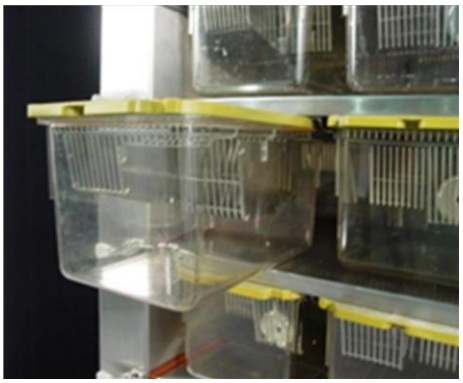
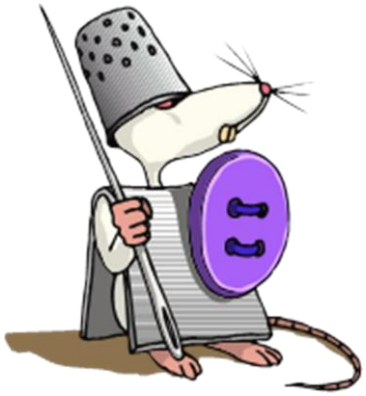
Behind the Curtain: All About PPE *Cont. from pg. 1*

The most significant health concern for people working with laboratory animals is laboratory animal allergies



The proper use of engineering controls such as individually ventilated caging systems significantly decrease airborne exposure to rodent allergens.
Image credit: http://www.thorenaquatics.com/Thoren_Caging/training.html



PPE depends upon the type of housing utilized-
BARRIER vs. NON-BARRIER.
What facilities do you enter?
How can you tell??

In barrier facilities *PPE is donned at the barrier entrance*, with the same PPE utilized between most rooms in the barrier

occupational exposure, and can become quite debilitating. An estimated 10-44% of all laboratory animal workers will eventually develop LAA, with up to 10% developing occupation-related asthma. The numbers are even higher in those who have pre-existing allergies (73%)[1]. The major antigens inducing allergic responses to rodents are mouse and rat urinary proteins that can be aerosolized anytime a rodent cage is opened. These particles are ~50um in size, about the limit of detection by the naked eye. The majority of these particles are too small and present at concentrations too low to be visualized. Very small quantities in the nanogram range, or concentrations in the range of parts per billion, are sufficient to elicit a reaction in sensitized individuals.[2]

There is also an increased risk of allergies in family members of exposed workers with no direct animal contact.[3] Krop *et al* (2007) found significant amounts of rodent allergens in the mattress dust in the homes of lab animal workers, indicating these allergens are brought home by employees. This study concluded that human hair is a major carrier of rodent allergens, as the employees wore protective clothing and gloves but did not use a hair covering. PPE in the form of a bonnet or hair-covering mitigates this risk, providing a disposable layer that is removed on exiting the animal facility. The same principle can be applied to street clothing worn in the animal facility. Disposable gowns are required for all RARC animal rooms; bonnets are required for many RARC facilities and are provided for voluntary use in all rooms.

Besides lowering allergen exposure, PPE helps to decrease risk of exposure to various hazards used in animal research. These may include biological, chemical, and radiation hazards. Animals that receive human cells or tissues (xenografts) may harbor human pathogens potentially found in these tissues. Therefore more stringent PPE requirements, in compliance with OSHA's Bloodborne Pathogen Standard, are required in all xenograft rooms. The use of chemical hazards or radionuclides also requires additional PPE (and training) compared to standard rodent housing rooms.

In the vivarium, PPE is used for two principal reasons. In addition to protecting pesonnel, it also protects the animals. It is an important part of RARC's biosecurity program, which keeps colonies free from excluded rodent pathogens. (This list of excluded pathogens is available upon request.) These agents may, in addition to causing disease, have physiologic effects that can confound research even without causing clinical signs. The proper use of PPE minimizes the risk of exposure of animals to excluded agents. PPE use is particularly important for protecting immunodeficient animals as these animals can develop significant clinical disease from common commensal organisms normally found on human skin or in the environment. PPE prevents research animals from being directly exposed to clothing or skin, minimizing the risk of staff or their clothing acting as fomites carrying pathogens. Therefore PPE protects personnel as well as protecting the animals.

Despite its importance, PPE is actually the last line of defense. In addition to PPE, engineering controls and work practices help keep animals and people safe. Two engineering controls utilized by RARC are the individually ventilated caging systems and change stations/biosafety cabinets present in every rodent housing room. In one study, biosafety cabinet use decreased airborne exposure to rodent allergens 2-fold.[4] Utilizing a biosafety cabinet is an example of a work practice that will decrease your exposure to rodent allergens and decrease the environmental load of such particles for everyone working in the room. Taken together, engineering controls, work practices and PPE minimize the risks of exposure to allergens, hazardous materials, or diseases that can occur when people and research animals interact.

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Have a great fall
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