Journal of the American Association for Laboratory Animal Science
Copyright 2011
by the American Association for Laboratory Animal Science

Vol 50, No 3
May 2011
Pages 337–343

Effect of Sampling Strategy on the Detection of Fur Mites within a Naturally Infested Colony of Mice (*Mus musculus*)

Kelly A Metcalf Pate,[1,*] Kelly A Rice,[1] Roberta Wrighten,[2] and Julie Watson[1]

Fur mites are one of the most common ectoparasites of laboratory mice and traditionally are diagnosed through surveillance of individual colony animals. Although multiple diagnostic modalities exist, few recommendations suggest optimal testing methods, target colony populations, or sampling sites. We compared the fur pluck and sticky paper techniques for the diagnosis of *Myocoptes musculinus* in naturally infested immunocompetent mice and evaluated the effect of mouse age and sampling site on the efficacy of fur plucks. We found that the sticky paper technique was more likely to detect fur mites than were fur plucks. Housing mice individually increased the incidence of false-negative fur pluck tests, whereas sensitivity was equivalent for preweanling and adult mice. The ventral abdomen was the most likely single sampling location to detect evidence of any stage of *Myocoptes musculinus*, but fur mite eggs were overrepresented on the neck. We found that the surface temperature of the murine neck surface was warmer than was the rump and therefore may represent a unique microenvironment for fur mite egg development. Given our findings, we recommend that group-housed adult or preweanling mice should be selected for *Myocoptes musculinus* evaluation and that the ventral abdomen should be sampled. When possible, the postmortem sticky paper technique should be used rather than the antemortem fur pluck method.

Fur mites remain a constant management challenge within modern rodent facilities. In a 2008 survey, 30% and 40% of research institutions self-reported the presence of *Myocoptes musculinus* and *Myobia musculi*, respectively.[4] These numbers are scarcely an improvement over a similar survey conducted 10 y earlier in which fur mite prevalences of 15% in barrier colonies and 40% in conventional colonies were reported.[16] A recent panel discussion on fur mite infestations in mice at the 2010 AALAS conference was well-attended and included representatives from 5 large research institutions in the United States; participants commented on the lack of reliable diagnostic and treatment options.[41]

The exclusion of fur mites from research colonies is arguably no less essential than the exclusion of viral agents. Fur mites have been reported to interfere with established research models by directly potentiating allergic-type hypersensitivity reactions in NC/Kuj mice susceptible to allergic dermatitis and indirectly interfering with normal immune responses to other pathogens such as *Toxoplasma gondii*.[22,38] Furthermore, fur mite infestations have been shown to provoke a Th2 immune response and elevate serum IgE.[17,18] The term 'mite-associated ulcerative dermatitis' (MAUD) was coined to emphasize a perceived correlation between ulcerative dermatitis and fur mite infestation,[7] and infestation with *Myobia musculi* was associated with an increased incidence of ulcerative dermatitis and decreased reproductive efficacy in C57Bl/6 mice that resolved once the infestation was treated.[37] The eradication of fur mites from affected colonies is therefore prudent to maintain the integrity of research results.

Received: 18 Oct 2010. Revision requested: 11 Nov 2010. Accepted: 09 Dec 2010.
[1]Department of Molecular and Comparative Pathobiology, Johns Hopkins University School of Medicine, Baltimore Maryland and [2]Banfield Pet Hospital, Newnan, Georgia.
[*]Corresponding author. Email: kpate5@jhmi.edu

At first glance, fur mites do not appear to be a formidable adversary for modern barrier housing and antiparasitic treatments. Murine fur mites are species-specific noninvasive ectoparasites and include *Myobia musculi*, *Radfordia affinis*, and *Myocoptes musculinus*. Primarily transmitted through direct mouse-to-mouse contact, fur mites complete a life cycle of fur-bound egg to motile nymph to reproductively mature adult in a mere 8 d (*Myocoptes musculinus*) to 23 d (*Myobia musculi*).[1,20,35,36] Control, therefore, should be achievable with the standard barrier techniques of filter-top cages and cage changes in hoods. Acaricides should likewise be effective so long as the drug kills all lifestages or is repeated to disrupt the life cycle. A number of published studies have reported successful colony eradication of fur mites with topical parasiticides (selamectin, ivermectin, moxidectin),[2,12,21,26] oral drugs (ivermectin, moxidectin),[6,25] ivermectin-impregnated feed,[28] injectible ivermectin,[40] environmental agents (chlorpyrifos, dichlorvos),[3,24] and cross-fostering paired with ivermectin treatment.[14]

The dilemma lies in successful identification of mite-infested mice to target eradication efforts to affected colony subsets. Identification of infested mice is desirable because widespread treatment is necessarily more expensive and disruptive than is targeted treatment, and theoretically widespread treatment should not be necessary, given the fur mite's limited mode of transmission.[20,35] Furthermore, widespread acaricide treatment of uninfested mice could be contraindicated due to the potential for adverse effects on mice, humans, and research. For example, ivermectin and moxidectin, although effective against fur mites, can be lethal in neonatal pups and genetically modified mice with defective blood–brain barriers,[19,29,30] and organophosphate insecticides (chlorpyrifos, dichlorvos) can be toxic and carcinogenic to human caretakers.[13] Adverse effects have not been reported for selamectin, but it is expensive and

requires labor-intensive spot-on application directly to the skin of each mouse.[12]

Published guidance on appropriate selection of colony animals to test and diagnostic techniques to use in the detection of fur mites is scarce. As a result, many different detection methods are in use. For example, both Charles River Laboratories and RADIL at University of Missouri examine the mouse pelt under a dissecting microscope, concentrating their efforts on the dorsum. The textbook *Laboratory Animal Medicine* in addition offers that fur mites may be detected antemortem by microscopic examination of fur pluck and skin scrape samples and can be identified postmortem through examination of a sticky surface surrounding the dead mouse (sticky paper technique).[9] At our institution, the sticky paper technique is considered the 'gold standard' for fur mite detection, due to poor success with direct pelt exam, but fur plucks are used for routine evaluation of live colony mice and for investigation of outbreaks.

The efficacy of some but not all of these tests has been compared. In one study, the skin scrape followed by direct pelt exam were most sensitive methods; however, the sticky paper technique was not considered.[3] This finding was corroborated by a recent report that the skin scrape is more sensitive than direct pelt exam and that the optimal sampling sites for skin scrape were the head and back; the fur pluck and sticky paper techniques were not evaluated.[27,28] As for which animals to test, although both historic and recent articles report some success in the use of soiled-bedding sentinels for the detection of fur mites,[27,28,34] other investigators report that fur mite contamination of dirty bedding rarely occurs,[20,36] and sampling of colony animals generally is practiced to increase the likelihood of detection.[5] Despite these facts, criteria to consider in the selection of colony animals to test are absent from the literature.

The current study was initiated in response to the failure to detect mites in mice that were housed individually for 7 wk, although these same mice previously had tested positive for *Myocoptes musculinus* prior to individual housing. However, when these same individually housed mice were placed in groups again, fur mites were detectable within 5 wk. These observations led us to hypothesize that singly housing a mouse can result in false-negative fur pluck and sticky paper tests for fur mites. Furthermore, our experiences caused us to question whether we could refine our testing techniques and selection criteria to better detect the presence of *Myocoptes musculinus* within our colony.

We conducted a prospective controlled small-scale study to evaluate the extent to which the population density of mice within a cage affects our ability to detect fur mites in a population of naturally infected, immunocompetent mice. In addition, we compared 1) the postmortem sticky paper technique and antemortem fur pluck method, 2) fur pluck samples harvested from adult and preweanling mice, and 3) fur pluck samples from the neck, rump, and belly. Finally, we assessed the differences in the locations where we detected all 3 fur mite lifestages by evaluating the local body surface temperature in mice and the fur mites' response to a heat gradient.

Materials and Methods

Mice. Experimentally naïve mice on a wildtype mixed B6129 background were donated to this study from a research colony naturally infested with *Myocoptes musculinus*. Mice were housed in compliance with the *Guide for the Care and Use of Laboratory Animals*[15] on autoclaved corncob bedding (Harlan, Indianapolis, IN) with a single cotton nesting square within ventilated cages in a quarantine room under the auspice of the AAALAC-accredited animal care and use program at Johns Hopkins University (Baltimore, MD). Mice received Teklad Global 18% Rodent diet 2018 (Harlan) and reverse-osmosis–treated water by means of an automatic watering system (Rees Scientific, Trenton, NJ) ad libitum. Cages were changed on a 2-wk cycle in changing stations (Lab Products, Seaford, DE) with ample use of a 100-ppm solution of the chlorine dioxide disinfectant Vimoba (Quip Laboratories, Wilmington, DE). Prior to and throughout this study, a soiled-bedding sentinel system was used to confirm that mice were negative for a wide range of excluded pathogens, including Sendai virus, pneumonia virus of mice, mouse hepatitis virus, mouse minute virus, mouse parvovirus 1 and 2, Theiler mouse encephalomyelitis virus, reovirus, epizootic diarrhea of infant mice, lymphocytic choriomeningitis virus, ectromelia virus, murine adenovirus, murine cytomegalovirus, *Mycoplasma pulmonis,* and *Aspiculuris* and *Syphacia* spp. pinworms. In addition, individual colony mouse surveillance for pinworms through anal tape test and fecal float was completed prior to initiation of the study. Before assignment to study groups, all mice were housed in same-sex cages of 4 to 5 siblings, and adult mice were confirmed positive for *Myocoptes musculinus* fur mites by fur pluck evaluation no more than 1 wk earlier. No mice developed any dermatitis or other lesions during the course of this study despite daily health checks, but one adult male mouse unexpectedly was found dead. All animal use was approved by the Johns Hopkins University IACUC.

Diagnostic techniques used throughout this study. *Fur pluck technique.* Approximately 10 mg fur was plucked from 3 sites on each mouse: just caudal to the ears along the dorsal midline (neck); the dorsal tailhead (rump), and the ventral abdomen (belly). These sampling sites were chosen because they represent 3 distinct regions of the mouse, and the base of the tail and abdomen have been reported to harbor large concentrations of *Myocoptes musculinus*.[20,36] Samples from each region were placed on individual sections of clear cellophane tape and adhered to glass slides. Samples were examined under a microscope (Leica, Wetzlar, Germany) at 40× and 100× by 2 independent observers and scored for the presence (or absence) of *Myocoptes musculinus* adults, nymphs, and eggs at each site; scores were consistent between observers. One observer was blinded prior to reading each slide (the other created the numbering scheme to blind the samples), and one observer had 2 y of experience with the diagnosis of fur mites.

Sticky paper technique. Mice were euthanized by cervical dislocation and placed immediately in lateral recumbency onto the center of adhesive covers for 96-well plates (Falcon, Franklin Lakes, NJ). After overnight incubation at room temperature, mice were removed from the sticky paper, and the paper was placed against a dark background and scored for the presence (or absence) of adult and nymph *Myocoptes musculinus* under a dissecting microscope (Nikon SMZ1000, Nikon, Melville, NY) by 2 independent blinded observers. This technique has also been referred to as the 'dorsal tape test' and 'cellophane tape test' in previous literature.[9,39]

Experiment 1: Effect of housing density, sampling method, mouse age, and sampling site on *Myocoptes musculinus* detection. *The effect of housing density.* Our preliminary data suggested that singly housed mice were less likely to support detectable fur mite populations than were group-housed mice. We attempted to replicate that preliminary finding through a prospective controlled study. Group-housed mice (n = 18; age, 8 w) were confirmed positive for *Myocoptes musculinus* by fur pluck and then housed either singly (4 cages total: 2 males, 2 females) or in a group of 3 (5 cages total: 2 groups of 3 males, 3 breeding trios

of 2 females with 1 male; 1 male from a breeding trio was not evaluated due to unexpected death). All adult mice were evaluated 5 wk later for fur mites by fur pluck and then immediately euthanized and examined by sticky paper technique.

Effect of sampling method. We hypothesized that fur plucks were a less sensitive detection method than was the sticky tape test. To directly compare the sensitivity of the fur pluck and sticky paper techniques, data on both techniques from the prospective controlled study on the effect of housing density were compared.

Effect of mouse age. We sought to clarify whether the fur pluck method was more sensitive for sampling of preweanling compared with adult mice. Adult mice (n = 11; 3 males, 8 females; age, 13 wk; one male of a trio was not evaluated due to unexpected death) were confirmed positive for *Myocoptes musculinus* by fur pluck and then housed as breeding trios for 5 wk with 8 to 10 preweanlings per cage; all adults and 13 of those 2-wk-old preweanlings were evaluated for fur mites by fur pluck. All mice included in this analysis were confirmed positive for fur mites immediately after fur pluck sampling by the sticky paper technique.

Effect of fur pluck sampling site. We evaluated whether the choice of neck, rump, or belly as fur pluck site influenced the sensitivity of this assay and compared the fur mite lifestages that we found at each site. Mite-infested mice (n = 74) were evaluated by fur pluck for fur mites in this portion of the study. Samples taken from the neck, rump, and belly were evaluated for the presence of *Myocoptes musculinus* adults, nymphs, or eggs as described earlier. Only mice that were positive for fur mites at one or more sampling site were included in this analysis. Adults were distinguished from nymphs by their larger size, and adults had 8 legs whereas nymphs had 6.

Experiment 2: The role of temperature in *Myocoptes musculinus* lifestage niche choice. *Comparison of mouse body-surface temperatures by anatomic location.* Data from experiment 1 revealed that the proportion of adults, nymphs, and eggs differed among fur pluck collection sites. We hypothesized that each of these anatomic locations may represent a microenvironmental niche on the mouse and that each fur mite lifestage may have a requirement for different environmental conditions. We measured surface body temperatures at the neck, rump, and belly as a representative niche environmental condition. Focal body surface temperatures of 29 live adult mice were determined by using a commercially available human temple thermometer (DIT, BestMed LLC, Golden, CO) applied directly to the skin through parted fur. The precision of this method was tested by taking 3 consecutive measurements of each site on the first 5 animals and finding no difference between consecutive measurements. Sites measured were representative of the fur pluck sites used in this study: the neck just caudal to the ears, the rump immediately cranial to the tail, and the ventral midabdomen.

Response of* Myocoptes musculinus *to a heat gradient. To evaluate whether heat affects *Myocoptes musculinus* travel choices, an adult mouse positive for *Myocoptes musculinus* on fur pluck was euthanized by cervical dislocation immediately after fur pluck evaluation. Skin samples 0.5 cm in diameter were harvested immediately from each of the fur pluck sampling sites (neck, rump, and belly) by using scissors, and all 3 samples placed in a group in the center of 3-cm diameter circles of construction paper in the center of a 10-cm culture dish (Corning P420 Hotplate Stirrer, Corning, Lowell, MA), half of which was maintained on a hot plate at 98.3 °F (average mouse body surface temperature) and the other half on an unplugged hot plate at room temperature. The remainder of the culture dish floor was covered in adhesive paper to capture mites moving off the paper. Controls consisted of similar samples harvested from mice positive for mites and placed on a culture dish on an unplugged hotplate at room temperature or on a hotplate at 98.3 °F; these control dishes were bisected by a line to delineate 2 random sides prior to the start of this assay. After 12 h, mites on each half plate and on controls were quantified under a dissecting microscope (Nikon SMZ1000, Nikon).

Statistics. The likelihood that different mouse parameters (cage population, mouse age) and sampling parameters (evaluation technique, location of fur pluck sample) affected the detection of fur mites was calculated by constructing 2 × 2 contingency tables and applying the Fisher exact test (chosen rather than a χ test, due to the small sample size). Comparison of surface body temperature at different anatomic locations was achieved through ANOVA followed by a posthoc Tukey multiple comparison test. Prism 4.0 (GraphPad, La Jolla, CA) was used for all data analysis and figure construction. Results were considered statistically significant if the P value was less than 0.05 and if the 95% confidence interval did not span 1.0.

Results

Preliminary observations on the effect of housing density on the detection of *Myocoptes musculinus*. Preliminary data showed that 6 mice (age, 15 wk) that had previously tested positive for *Myocoptes musculinus* by fur pluck all tested negative by fur pluck after being singly housed for 7 wk. Two of these mice were euthanized after fur pluck and were negative by sticky paper technique. However, when 3 of the 4 remaining mice were group-housed subsequently as a trio, fur mites became detectable (by fur pluck and sticky paper technique) in the adults and their offspring within 5 wk, whereas the sole singly housed mouse continued to test negative by fur pluck and sticky paper techniques. These findings suggested that the initial fur mite infestation had not disappeared as a result of decreased housing density but rather had fallen below the level of detection. We then hypothesized that the number of mice within a cage could affect the sensitivity of fur pluck and sticky paper diagnostic techniques.

Experiment 1: The effect of housing density, sampling method, mouse age, and sampling site on *Myocoptes musculinus* detection. *Effect of housing density.* After 5 wk, all singly housed mice (n = 4) were negative for fur mites by fur pluck whereas, at least one positive mouse was identified in each of the 5 cages that contained 3 adult mice. A total of 57% (8 of 14) of group-housed mice were positive for fur mites by fur pluck. Therefore, on a cage basis, a fur pluck sample from a cage containing a single mouse was less likely to detect *Myocoptes musculinus* fur mites than were fur pluck samples from each member of a cage with multiple mice (Fisher exact test; 95% confidence interval, 1.618 to 6059; P = 0.0079). All mice, regardless of housing density, were positive for fur mites by sticky paper technique.

Effect of sampling method. In this prospective experiment, singly housed mice were all negative for fur mites by fur pluck but were all positive by sticky paper. For group-housed mice, the sticky paper technique was 1.75 times more likely to detect *Myocoptes musculinus* than was the fur pluck method (Fisher exact test; relative risk, 1.75; 95% confidence interval, 1.112 to 2.755; P = 0.0159). Fur pluck had a sensitivity of 57% for the detection of fur mites (8 of 14 mice positive for mites) as compared with sticky paper (all 14 mice positive for mites). During the course of this study, no mice that were positive by fur pluck were found to be negative by sticky paper if tested at

the same time, indicating a positive predictive value of 100% for the fur pluck assay.

Effect of mouse age. Fur plucks detected 53.8% (7 of 13) of infested preweanling mice and 45.5% (5 of 11) of infested adult mice (Fisher exact test; 95% confidence interval, 0.5217 to 2.690; $P = 1.000$). All preweanling and adult mice were positive by sticky paper technique.

Effect of fur pluck sampling site. The belly was the most sensitive single site for the detection of any stage of *Myocoptes* spp. by fur pluck [χ^2 test, $P < 0.0001$; followed by Fisher exact tests comparing neck with belly (95% confidence interval: 1.038 to 1.253; $P = 0.009$) and rump with belly (95% confidence interval: 1.225 to 1.672; $P < 0.0001$)], but no difference was noted when the sensitivity of the belly was compared with the sensitivity of the neck and rump combined (Fisher exact test comparing the belly and the combined results of the neck and rump for each animal; 95% confidence interval, 1.007 to 1.178; $P = 0.0630$; Figure 1 A). Although adult mites were 3 times more likely to be found on the rump or belly than on the neck (Fisher exact test; relative risk, 3.015; 95% confidence interval, 2.031 to 4.476; $P < 0.0001$), mite eggs were 3 times more likely to be found on the neck than on the belly (Fisher exact test; relative risk, 2.701; 95% confidence interval, 2.047 to 3.565; $P < 0.0001$) or rump (Fisher exact test; relative risk, 2.957; 95% confidence interval, 2.005 to 4.361; $P < 0.0001$; Figure 1 B).

Comparison of mouse body surface temperature by anatomic location. The surface temperature of the mouse neck is significantly (ANOVA followed by posthoc Tukey multiple comparison test, $P < 0.01$) warmer than that of the mouse rump. The average adult mouse body surface temperature was 98.25 °F. The difference between neck and rump surface temperature ranged from −0.02 to 0.90 °F, with an average temperature disparity of 0.18 °F. The average neck temperature was 98.33 °F whereas the rump averaged 98.15 °F (Figure 2).

The response of fur mites to a heat gradient. Nine fur mites were found on the warmed paper, but none were present on the room temperature side of the sticky paper (Figure 3). On both control plates, an equal number of *Myocoptes musculinus* (2 on each side for the room-temperature control, 3 for 98.3 °F) were found on each 'side' of the sticky paper. No replicates were completed because only one mite-infested mouse was available for euthanasia at the time of this assay.

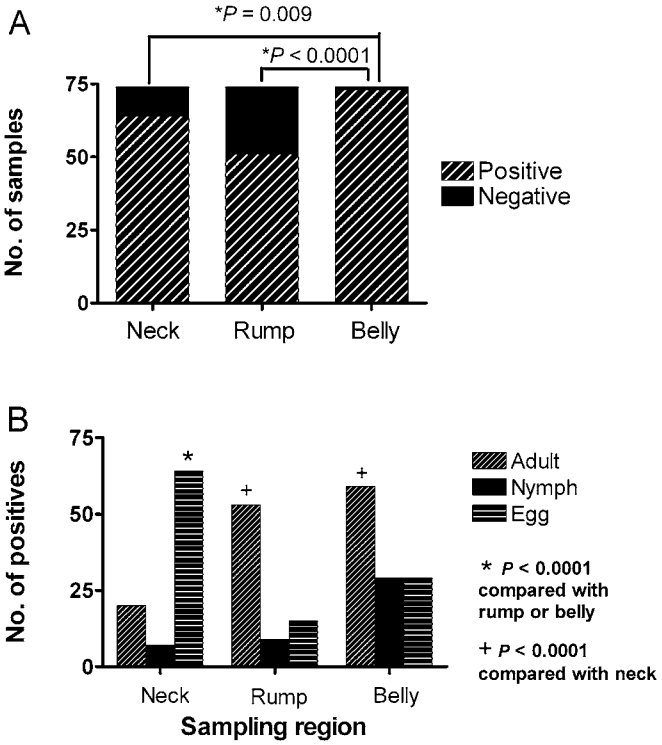

Figure 1. The effect of fur pluck sampling site on the detection of *Myocoptes musculinus* and different fur mite lifestages. (A) Comparison of the efficacy of sampling site for the detection of fur mites by fur pluck. (B) Comparison of *Myocoptes musculinus* lifestages found at each fur pluck sampling site.

Discussion

In this prospective controlled small-scale study, we found that housing mice infested with *Myocoptes musculinus* in individual cages resulted in false-negative results when mice were evaluated by the fur pluck technique. In addition, we showed that the sticky paper technique is more sensitive for mite detection than are concurrently harvested fur plucks. Although we were unable to detect a difference in the likelihood of fur plucks from adult and preweanling mice to detect mites, we showed that the belly is the most likely single sampling site in which to detect *Myocoptes musculinus* by fur pluck. We discovered that fur mite eggs are overrepresented in fur pluck samples from the neck. We demonstrated that the surface temperature of the murine neck is significantly warmer than is the rump and show preliminary evidence that *Myocoptes musculinus* may be able to sense and move toward a heat source.

Our failure in our preliminary study to detect fur mites by either fur pluck or sticky paper testing techniques on individually housed mice that previously were fur-pluck-positive has not been previously reported. Infestation status was confirmed by returning a subset of these fur-pluck negative mice to group housing and subsequently again detecting mites by fur pluck. Interestingly, in our prospective study singly housed mice remained positive by sticky paper technique even though they were negative by the less sensitive fur pluck while cages of group-housed mice remained positive by both assays. Our preliminary findings involved singly housing the mice for 7 wk, but in the prospective study, mice were singly housed for 5 wk only. Perhaps if the mice in the prospective study were individually housed longer, the sticky paper technique might also have failed to detect mites in singly housed mice. Our study suggests detecting *Myocoptes musculinus* in individually housed mice may be more difficult than for group-housed mice. This fact should be taken into consideration when deciding how to best monitor a colony, especially given that many sentinel mice are either housed individually or in pairs. Investigation into whether the sensitivity of the fur pluck assay can further be increased by taking multiple samples over time could further inform colony sampling strategies, and further study is needed to determine the best cage population size to optimize the likelihood of detecting fur mites when screening a mouse colony.

The most likely explanation for the lower sensitivity of the fur pluck or sticky paper technique in singly housed mice is a reduction in fur mite load on individually housed mice. Population density of mice within a cage has the potential to alter cage microenvironmental and murine physiologic parameters. Increasing mouse density raises temperature, ammonia, and carbon dioxide levels within the cage.[31,32] It also has the potential to alter immune function, either through increasing corticosterone levels, resulting in immune suppression, or by increasing the responsiveness of T cells to antigen.[11,23] Fur mite populations may respond to these changes in environment or immune function. Indeed, it has been suggested that the

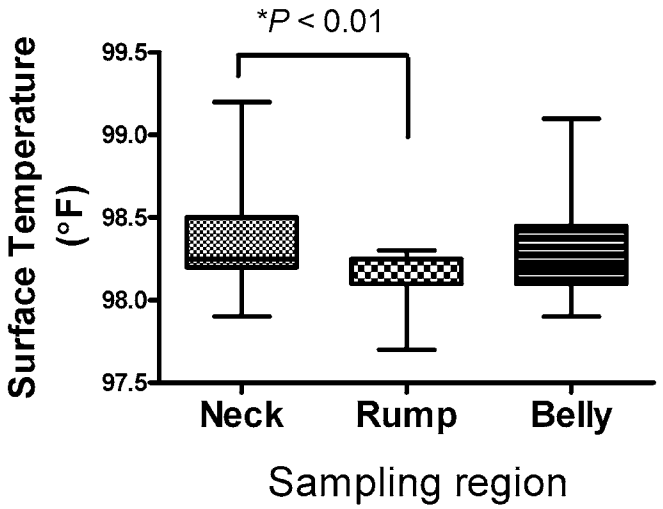

Figure 2. Comparison of mouse body surface temperature by anatomic location. Focal body surface temperatures of 29 live adult mice were measured at the neck, rump, and belly by using a commercially available human temple thermometer.

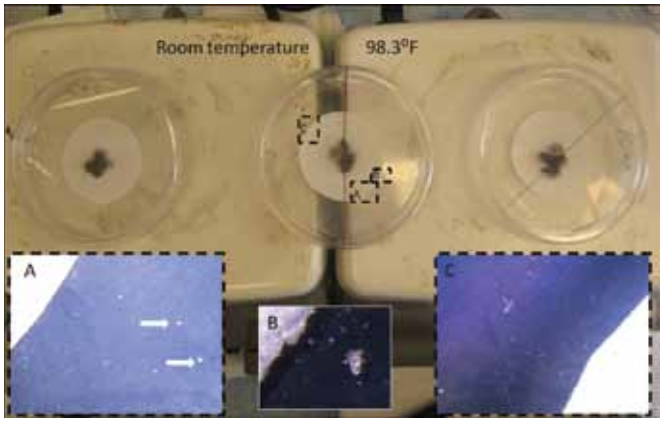

Figure 3. The response of fur mites to a heat gradient. (A) All 9 adult and nymph mites exposed to the heat gradient were found on the sticky paper over the 98.3 °F hot plate. Two representative mites (arrows) are shown; magnification, ×2. (B) A single representative mite as viewed under the dissecting microscope; magnification, ×10. (C) No adult or nymph mites were found on the sticky paper over the room temperature hot plate when exposed to a gradient.

longer mice are infested, the smaller the population of *Myobia musculi* they support, presumably due to the development of host immunity.[10] The availability of multiple hosts therefore would increase the chance that a single mouse would support larger mite populations, particularly if breeding provides a supply of naïve hosts. Alternatively, it is possible that fur mites optimally may require more than one mouse host to complete their life cycle; perhaps mites reproduce less efficiently in the cages of individually housed mice. More thorough quantification of mite load in response to changes in microenvironmental parameters and assessment of mouse immune status may help clarify whether such factors play a role in our findings and in the ecology of *Myocoptes musculinus*.

This report is the first published account of a direct comparison of sticky paper and fur plucks for detection of *Myocoptes* spp. We found that sticky paper is more likely to detect fur mites than are concurrent fur plucks. In the past, fur plucks have been compared with skin scrapes and direct pelt exams with comparable sensitivity to that reported here (75% previously reported when compared with skin scrapes as the gold standard;[3] 57% reported here when compared with the sticky paper technique). Further studies are needed to determine whether the sticky paper technique proves superior to direct pelt exam or skin scrape. However, given that even sticky paper did not detect fur mites in singly housed mice in our preliminary study, we suspect that more sensitive tests for fur mites are needed sorely, regardless of how the sticky paper technique compares with other currently available tests. A PCR or serologic assay could serve as a valuable diagnostic tool in the future.

Infestations of *Syphacia obvelata*, the common murine pinworm, are less severe in the presence of a functional adaptive immune response and decrease in intensity as the host ages.[33] In light of this finding, we hypothesized that preweanling mice, which have relatively naïve adaptive immune systems, would be infected with greater numbers of *Myocoptes musculinus* than would adults. However, we did not detect a difference in the sensitivity of fur plucks from 2-wk-old preweanlings and 13-wk-old adults.

We found that the mouse's belly is the most sensitive single site to sample to detect any stage of *Myocoptes musculinus* by fur pluck. This observation is consistent with a previous study, in which *Myocoptes musculinus* were found predominantly in the inguinal and abdominal regions of mice.[20] Current diagnostic practices often concentrate sampling efforts on the dorsum of the mouse. This emphasis is not inappropriate, as we found that samples from the neck and rump, when considered together, offer equivalent sensitivity to those from the belly alone. However, the need for a full-body examination including both the dorsum and inguinal areas for *Myocoptes* spp. has been emphasized,[36] and our findings support this recommendation to add the ventral abdomen to the standard list of sampling sites.

Our data showed that different *Myocoptes musculinus* lifestages were localized to distinct regions on the body of the mouse. Evidence of a shift in regional niche on a single host as part of the life cycle of an ectoparasite is unusual. Many ectoparasites, such as ticks, undergo niche switches as a necessary part of their development, but these changes usually involve entirely leaving one host species for another environment.[8] Lifestage-specific niche changes within a single host, however, are not uncommon for endoparasites, such as *Syphacia obvelata*, which spend adulthood within the cecum and colon then migrate to the perianal area to lay eggs.[1] Further studies on the movement of adult and nymph mites on the mouse are needed to clarify the role of microenvironmental niches on the *Myocoptes musculinus* life cycle.

Interestingly, the preferred location of the fur mite eggs (the neck) proved warmer than the rump. It is possible that eggs require the slightly higher temperature for optimal development, and it is tempting to speculate that the eggs develop faster in group-housed mice due to elevated cage temperature. In fact, in a group cage, the opportunity for greatly elevated localized body temperatures could be optimized by behavior such as huddling. If this situation was the case, it would provide yet another potential explanation for increased mite populations on group-housed mice compared with singly housed mice. The incubation requirements of *Myocoptes musculinus* eggs require further exploration. Consistent with previously reported findings,[20] our preliminary study using hotplates as heat source suggested that fur mites will move toward a heat source maintained at mouse body temperature when given a choice between it and an identical source maintained at room temperature. Further studies are needed, however, to truly determine whether *Myocoptes musculinus* are thermotaxic and whether *Myocoptes* spp. of different stages can distinguish be-

tween minute changes in temperature such as those observed at different surface locations on the mouse.

We undertook this study with the aim of optimizing criteria for the detection of *Myocoptes musculinus* in naturally infested immunocompetent mice. According to our findings, we recommend testing preweanlings or adult mice that are group-housed. Postmortem tests using the sticky paper technique are more sensitive than are fur plucks and should be used whenever possible, particularly if mice have been singly housed. For antemortem detection of *Myocoptes musculinus* using fur plucks, the belly is the most likely to yield positive results if a single sampling site is used and should be sampled in addition to the neck and rump. Our finding that different mite lifestages are more prevalent within distinct microenvironments on the mouse indicates that we still have much to learn about the ecology of *Myocoptes musculinus*. Further studies are needed to determine whether our recommendations extend to other fur mite species or to *Myocoptes musculinus* when in the presence of another mite species. However, even with optimized sample selection criteria, more sensitive assays for fur mites are needed.

Acknowledgments

We thank the laboratory of David Ginty for the donation of mite-infested mice; Cory Brayton, Nadine Forbes, David Huso, and Joseph Mankowski for use of laboratory space and equipment; Sarah Poynton, Bob Adams, Alton Swennes, Eric Hutchinson, and Joseph Mankowski for valuable commentary and discussion; and Kathryn Flemmen, Eric Hutchinson, Ginny Goodwin, and Eric Syversen for technical assistance. This study was supported by NIH Training Grant 2T32RR007002-34A1, and additional support was provided by Robert J Adams and Johns Hopkins Research Animal Resources.

References

1. **Baker DG.** 2007. Flynn's parasites of laboratory animals. Ames (IA): Blackwell Publishing.
2. **Baumans V, Havenaar R, van Herck H, Rooymans TP.** 1988. The effectiveness of Ivomec and Neguvon in the control of murine mites. Lab Anim **22:**243–245.
3. **Burdett EC, Heckmann RA, Ochoa R.** 1997. Evaluation of 5 treatment regimens and 5 diagnostic methods for murine mites (*Myocoptes musculinus* and *Myobia musculi*). Contemp Top Lab Anim Sci **36:**73–76.
4. **Carty AJ.** 2008. Opportunistic infections of mice and rats: Jacoby and Lindsey revisited. ILAR J **49:**272–276.
5. **Compton SR, Homberger FR, MacArthur Clark J.** 2004. Microbiological monitoring in individually ventilated cage systems. Lab Anim (NY) **33:**36–41.
6. **Conole J, Wilkinson MJ, McKellar QA.** 2003. Some observations on the pharmacological properties of ivermectin during treatment of a mite infestation in mice. Contemp Top Lab Anim Sci **42:**42–45.
7. **Dawson DV, Whitmore SP, Bresnahan JF.** 1986. Genetic control of susceptibility to mite-associated ulcerative dermatitis. Lab Anim Sci **36:**262–267.
8. **Estrada-Pena A.** 2008. Climate, niche, ticks, and models: what they are and how we should interpret them. Parasitol Res **103 Suppl 1:**S87–S95.
9. **Fox JG, Anderson LC, Loew FM, Quimby FW.** 2002. Laboratory animal medicine. San Diego (CA): Academic Press.
10. **Friedman S, Weisbroth SH.** 1975. The parasitic ecology of the rodent mite *Myobia musculi*. II. Genetic factors. Lab Anim Sci **25:**440–445.
11. **Fullwood S, Hicks TA, Brown JC, Norman RL, McGlone JJ.** 1998. Floor space needs for laboratory mice: C56BL/6 males in solid-bottom cages with bedding. ILAR J **39:**29–36.
12. **Gonenc B, Sarimehmetoglu HO, Ica A, Kozan E.** 2006. Efficacy of selamectin against mites (*Myobia musculi*, *Mycoptes musculinus*, and *Radfordia ensifera*) and nematodes (*Aspiculuris tetraptera* and *Syphacia obvelata*) in mice. Lab Anim **40:**210–213.
13. **Hodgson E, Rose RL.** 2007. Human metabolic interactions of environmental chemicals. J Biochem Mol Toxicol **21:**182–186.
14. **Huerkamp MJ, Zitzow LA, Webb S, Pullium JK.** 2005. Cross-fostering in combination with ivermectin therapy: a method to eradicate murine fur mites. Contemp Top Lab Anim Sci **44:** 12–16.
15. **Institute for Laboratory Animal Research.** 1996. Guide for the care and use of laboratory animals. Washington (DC): National Academies Press.
16. **Jacoby RO, Lindsey JR.** 1997. Health care for research animals is essential and affordable. FASEB J **11:**609–614.
17. **Jungmann P, Freitas A, Bandeira A, Nobrega A, Coutinho A, Marcos MA, Minoprio P.** 1996. Murine acariasis. II. Immunological dysfunction and evidence for chronic activation of Th2 lymphocytes. Scand J Immunol **43:**604–612.
18. **Laltoo H, Van Zoost T, Kind LS.** 1979. IgE antibody response to mite antigens in mite-infested mice. Immunol Commun **8:**1–9.
19. **Lee VK, Tiwary AK, Sharma-Reddy P, Lieber KA, Taylor DK, Mook DM.** 2009. Moxidectin toxicity in senescence-accelerated prone and resistant mice. Comp Med **59:**227–233.
20. **Letscher RM**. 1970. Observations concerning the life cycle and biology of *Myobia musculi* (Schrank) and *Myocoptes musculinus* (Koch). [MS thesis] College Station (TX): Texas A and M University.
21. **Mook DM, Benjamin KA.** 2008. Use of selamectin and moxidectin in the treatment of mouse fur mites. J Am Assoc Lab Anim Sci **47:**20–24.
22. **Morita E, Kaneko S, Hiragun T, Shindo H, Tanaka T, Furukawa T, Nobukiyo A, Yamamoto S.** 1999. Fur mites induce dermatitis associated with IgE hyperproduction in an inbred strain of mice, NC/Kuj. J Dermatol Sci **19:**37–43.
23. **Nicholson A, Malcolm RD, Russ PL, Cough K, Touma C, Palme R, Wiles MV.** 2009. The response of C57BL/6J and BALB/cJ mice to increased housing density. J Am Assoc Lab Anim Sci **48:**740–753.
24. **Pence BC, Demick DS, Richard BC, Buddingh F.** 1991. The efficacy and safety of chlorpyrifos (Dursban) for control of *Myobia musculi* infestation in mice. Lab Anim Sci **41:**139–142.
25. **Pollicino P, Rossi L, Rambozzi L, Farca AM, Peano A.** 2008. Oral administration of moxidectin for treatment of murine acariosis due to *Radfordia affinis*. Vet Parasitol **151:**355–357.
26. **Pullium JK, Brooks WJ, Langley AD, Huerkamp MJ.** 2005. A single dose of topical moxidectin as an effective treatment for murine acariasis due to *Myocoptes musculinus*. Contemp Top Lab Anim Sci **44:**26–28.
27. **Ricart Arbona RJLN, Wolf FR.** 2010. Treatment and eradication of murine fur mites: II. Diagnostic considerations. J Am Assoc Lab Anim Sci **49:**583–587.
28. **Ricart Arbona RJLN, Wolf FR.** 2010. Treatment and eradication of murine fur mites: III. Treatment of a large mouse colony with ivermectin-compounded feed. J Am Assoc Lab Anim Sci **49:**633–637.
29. **Schinkel AH, Smit JJ, van Tellingen O, Beijnen JH, Wagenaar E, van Deemter L, Mol CA, van der Valk MA, Robanus-Maandag EC, te Riele HP, Berns AJM, Borst P.** 1994. Disruption of the mouse *mdr1a* P-glycoprotein gene leads to a deficiency in the blood–brain barrier and to increased sensitivity to drugs. Cell **77:**491–502.
30. **Skopets B, Wilson RP, Griffith JW, Lang CM.** 1996. Ivermectin toxicity in young mice. Lab Anim Sci **46:**111–112.
31. **Smith AL, Mabus SL, Muir C, Woo Y.** 2005. Effects of housing density and cage floor space on 3 strains of young adult inbred mice. Comp Med **55:**368–376.
32. **Smith AL, Mabus SL, Stockwell JD, Muir C.** 2004. Effects of housing density and cage floor space on C57BL/6J mice. Comp Med **54:**656–663.
33. **Stewart PW, Chapes SK.** 2003. Role of major histocompatibility complex class II in resistance of mice to naturally acquired infection with *Syphacia obvelata*. Comp Med **53:**70–74.
34. **Thigpen JE, Lebetkin EH, Dawes ML, Amyx HL, Caviness GF, Sawyer BA, Blackmore DE.** 1989. The use of dirty bedding for detection of murine pathogens in sentinel mice. Lab Anim Sci **39:**324–327.

35. **Watson DP.** 1960. On the adult and immature stages of *Myocoptes musculinus* (Koch) with notes on its biology and classification. Acarologia **2:**335–344.
36. **Watson DP.** 1961. The effect of the mite *Myocoptes musculinus* (C L Koch 1840) on the skin of the white laboratory mouse and its control. Parasitology **51:**373–378.
37. **Weisbroth SH, Friedman S, Scher S.** 1976. The parasitic ecology of the rodent mite, *Myobia musculi*. III. Lesions in certain host strains. Lab Anim Sci **26:**725–735.
38. **Welter A, Mineo JR, de Oliveira Silva DA, Lourenco EV, Vieira Ferro EA, Roque-Barreira MC, Maria da Silva N.** 2007. BALB/c mice resistant to *Toxoplasma gondii* infection proved to be highly susceptible when previously infected with *Myocoptes musculinus* fur mites. Int J Exp Pathol **88:**325–335.
39. **West W, Schofield JC, Bennett BT.** 1992. Efficiency of microdot technique for the administration of topical 1% ivermectin for the control of pinworms and fur mites in mice. Contemp Top Lab Anim Sci **31:**7–10.
40. **Wing SR, Courtney CH, Young MD.** 1985. Effect of ivermectin on murine mites. J Am Vet Med Assoc **187:**1191–1192.
41. **Zielinski Mozny N, Colby LA, Gaudette AA, Cotroneo TM, Mook DM, Smith PC, Wolf FR**. 2010. Fur mite infestations in mice: detection, control, and treatment strategies, p 49. National Meeting Program. Memphis (TN): American Association for Laboratory Animal Science.