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**OVERVIEW**

**Narver. Antimicrobial Stewardship in Laboratory Animal Facilities, pp. 6-10**

Domain 1: Management of Spontaneous and Experimentally Induced Diseases and Conditions

SUMMARY: There has historically been more of a focus on antimicrobial stewardship in human medical fields as compared to animals, although recently it has been addressed again in animal medical and laboratory settings. There are multiple areas of antimicrobial stewardship including:

* Reduced usage: the idea that antibiotics should only be used following diagnostic testing that confirms the “presence, identification, and antibiotic susceptibility of bacteria”.
* Refined usage: antibiotic use should be guided by the animal species it is used in, the route of administration (enteral vs parenteral for time vs concentration dependent), ease of administration (stress of parenteral administration vs ability to dose sufficiently with enteral administration), target of antibiotic (treat topically rather than systemically if appropriate), and for the shortest appropriate time.
* Replacement: Non-antibiotic preparations such as antiseptics or botanicals, or non-pharmaceuticals such as nail trims and appropriate enrichment for dermatitis can often be effective even when diagnostic testing leads to antibiotic use. The use of aseptic technique should also prevent the need for antibiotics in certain cases.
* Review usage: Antibiotics started prior to diagnosis should be adjusted following diagnosis, antibiotics usage should be reviewed to identify patterns of usage.
* Responsible usage: Antibiotics should be disposed of properly. Handwashing and sanitation are necessary to prevent and control infection. The presence of multi-drug resistant bacteria should be identified and limited. Use of antibiotics that are important for human use should be limited in animals.

QUESTIONS

1.  What are some ways antibiotic usage negatively impacts research (other than antimicrobial resistance)?

2.  What is Procalcitonin?

3.  What is the problem with relying on MIC to dose antibiotics?

4. What is one of the most important infection control activities?

a.   Wearing gloves in susceptible areas

b. Frequent hand washing

c.  Cleaning computer keyboards

ANSWERS

1.  Changes in the microbiota

2.  A biomarker that can be tested for to identify bacterial replication and infection and therefore, indicates the need for treatment with antimicrobials.

3.   Because MIC is based on controlled conditions it may underestimate the required dosage. It also does not necessarily correlate across species and is often based on human studies.

4.  b

**ORIGINAL RESEARCH**

***Reproduction***

**Nishizono et al. Cleavage Speed and Blastomere Number in DBA/2J Compared with C57BL/6 Mouse Embryos, pp. 11-17**

Domain 3: Research

Primary Species: Mouse (*Mus musculus*)

SUMMARY: DBA/2J mice are one of the most important inbred strains used today, but this strain has reproductive problems and obtaining a large colony for research purposes is time consuming and may be cost prohibitive.  This study compared the embryos of C57BL/6J with DBA/2J and found DBA2/J embryos had a slower cleavage speed, and lower cell counts at morula and blastocyst stages. The results of reciprocal in vitro fertilization and male-female reciprocal crosses revealed that these phenotypes were not affected by sperm genome and were recessively inherited.  These findings suggest that DBA/2J mice can serve as a new model for human infertility.

QUESTIONS

1. DBA/2J mice are known

a. To have progressive hearing loss that is reversible at 3 months of age

b. To be extremely susceptible to alcohol and morphine

c. Not to be a good model for eye abnormalities such as glaucoma

d. To be susceptible to audiogenic seizures

2. T or F. DBA inbred is the oldest of all inbred strains of mice, started by Dr. CC Little in 1909.

ANSWERS

1. d

2. T

***Husbandry***

**Michaelis et al. Automated Tracking of Motion and Body Weight for Objective Monitoring of Rats in Colony Housing, pp. 18-31**

Primary Species: Rat (Rattus norvegicus)

Domain 3: Research

Tasks performed at time of certification

T1. Facilitate or provide research support

T2. Advise and consult with investigators on matters related to their research

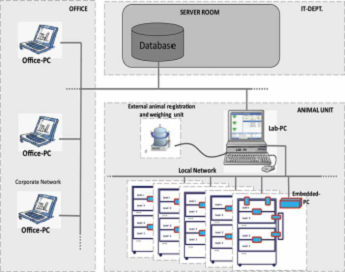
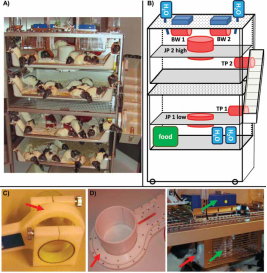
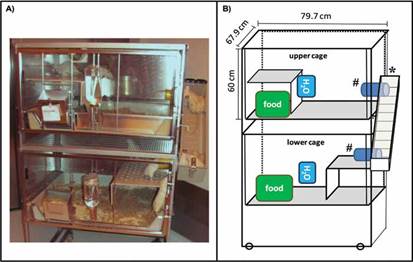
T3. Design and conduct research

Knowledge required to perform these tasks at time of certification as a Diplomate

K1. Biomethodology techniques (e.g., collection of blood and other body fluids and tissues; handling and restraint; administration of compounds and treatments)

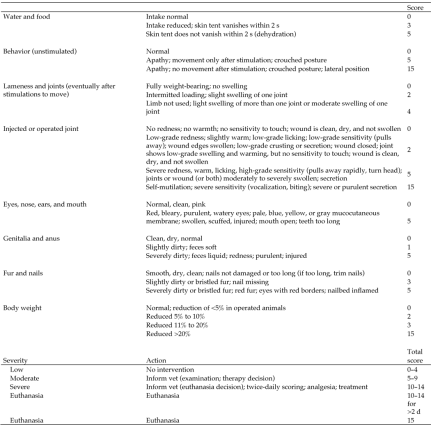
K2. Research methods and equipment (e.g., antibody production; adjuvants; tumor induction; imaging; data collection techniques such as telemetry; observation; behavioral assessment methods)

SUMMARY: These authors from Germany report a unique autoclaveable, modular housing system that allows for a larger group of rats that is more consistent with how wild rats live. Not only does this system improve rat wellbeing with objective data such as remote body weight measurement and continuous movement measurements, but it also showed that rats were more cooperative during handling. Their automated system allowed the detection of individual rats with significant differences from other cohoused rats greater than 2 standard deviations that showed potential health problems. The combination of an objective measure of both body weight in conjunction with locomotion may be an ideal system for early identification of health problems.



QUESTIONS.

1.  What is another name for this scoring system?



2.  Roaming entropy is used to describe what in this paper?

ANSWERS

1.   An ethogram

2.  It is a term that describes the degree which activity is distributed throughout the entire cage area, for example different levels. The authors go on to describe that this relationship suggests that locomotion in a social and complex environment is a marker of an individual animal’s development, even in genetically identical, inbred mice.

***Animal Health Surveillance***

**Gerwin et al. Evaluation of Traditional and Contemporary Methods for Detecting *Syphacia obvelata* and *Aspicularis tetraptera* in Laboratory Mice, pp. 32-41**

Domain 1. K3. Parasitology with emphasis on parasitic diseases that can become established in a colony

Primary Species:  Mouse (*Mus musculus*)

SUMMARY: This study evaluated several methods of detecting mouse pinworm infection. First, several solutions were tested for sensitivity to detect *Aspiculuris tetraptera* eggs by centrifugation or flotation. Fecal centrifugation detected eggs in 100% of samples. When fecal flotation is utilized, based on results reported here the authors recommended solutions with a specific gravity of 1.20 (e.g. sodium nitrate; eggs detected in 90% of samples). Second, Swiss Webster mice intentionally infected with *Syphacia obvelata*(SO) or *Aspiculuris tetraptera* (AT) were tested by several detection methods at specific time points up to 90 days post infection. Methods included fecal concentration (AT only), anal tape test (SO only), direct examination of intestinal contents (cecum and colon), Swiss roll histology (cecum and colon), and PCR analysis (pooled fur swab and feces). Some mice never yielded a positive test result, and were therefore excluded from infection rate calculations. No one test captured all of the positive animals. Combining two tests (PCR and intestinal content examination) detected all positive animals, and is therefore recommended as the testing combination with the highest detection rate/sensitivity.

QUESTIONS

1.  How long does it take *Syphacia obvelata* to complete its life cycle? *Aspicularis tetraptera*?

2.  Where do you find *Syphacia* spp. eggs? *Aspicularis*?

ANSWERS

1.  11-15 days; 23-25 days

2. Skin and hairs of the perianal region of the host; in feces—not on the host

**Curtis et al. Interspecies Variation in the Susceptibility of a Wild-Derived Colony of Mice to Pinworms (*Aspicularis tetraptera*), pp. 42-46**

Domain 1: Management of Spontaneous and Experimentally Induced Diseases and Conditions

Primary Species: Mouse *(Mus musculus)*

SUMMARY: Pinworms are common parasites in wild and laboratory rodents. Despite nonpathogenicity, infections can modulate Th2-associated immune response making these rodents unsuitable for some types of research. In laboratory mice, *Aspiculuris tetraptera*, is the most common pinworm with prevalence of 0.19% in diagnostic samples submitted in North America. Compared to wild-derived mice, research indicated that laboratory mice are more resistant to *A. tetraptera* infection, but, among laboratory mice species, young and male mice appear more susceptible to infection. Since little information is available in terms of the *A. tetraptera* susceptibility to wild-derived mice, this study hypothesized that susceptibility to *A. tetraptera* infection in 6 wild-derived mice (*Mus spretus, Mus spicilegus, M. m. musculus* (PWK/PhJ), *M. m. domesticus* (LEWES/EiJ and WSB/EiJ), *Mus macedonicus*) would be species- or subspecies-dependent. According to the results, *M. spicilegus* and *M. spretus* showed the highest susceptibility to pinworm infection. Difference from observed in *M. musculus* in the past, no differences in susceptibility according to age, sex or number of mice per cage were observed with the strains tested. Further work is needed to understand the effect of *A. tetraptera* infection in the immunomodulation, behavior and growth of the wild-derived mice strains reported in this study.

QUESTIONS

1. In a recent report, *A. tetraptera* susceptibility was increased in which wild-derived mice?

a. *A. tetraptera*

b. *Mus spretus, Mus spicilegus*

c. *M. m. domesticus* (LEWES/EiJ and WSB/EiJ),

d. *Mus spretus, Mus macedonicus*

2. In the same study, increased susceptibility to *A. tetraptera* infection in the tested wild-derived mice strains was observed between:

a. Age

b. Gender

c. Number of mice per cage

d. None of above

 ANSWERS

1. b

2. d

**Clark et al. Cross-Foster Rederivation Compared with Antibiotic Administration in the Drinking Water to Eradicate *Bordetella pseudohinzii*, pp. 47-51**

SUMMARY: *Bordetella pseudohinzii* is the provisional name of a proposed novel species of *Bordatella* that has been associated with oropharynx colonization in mice and the potential to produce mild to moderate rhinitis and therefore confound research data of associated systems.

In an effort to eradicate *B. pseudohinzii* from a mouse colony three methods of eradication were evaluated:  cross-fostering of newborn pups from infected dams to non-infected mothers, treatment of animals with sulfamethoxazole and trimethoprim (TMS) via drinking water, and treatment of mice with tetracycline in the drinking water.  Presence or absence of the pathogen was determined by fecal PCR for up to 4 weeks post-treatment.  Of these three methods cross-fostering produced animals that were negative for the pathogen via testing by PCR for 24 out of 29 litters up to 4 weeks after cross-fostering.  In contrast TMS treatment produced only 1 out of 12 cages negative for the pathogen at 2 weeks after treatment ended, and tetracycline had only 3 out of 12 cages negative at 2 weeks after treatment ended.

Cross-fostering appears to be the superior method of eliminating *B. pseudohinzii* in a mouse colony.  The authors also reported anecdotally that combining antibiotic treatment of pregnant dams with subsequent cross-fostering has further improved efforts to eliminate the pathogen.

QUESTIONS

1.  What is the primary site of infection and histopathological changes associated with *Bordatella pseudohinzii* infection in the mouse?

2.  Between cross-fostering, antibiotic treatment with sulfamethoxazole and trimethoprim (TMS), or treatment with tetracycline, which method was the most effective at eliminating *Bordatella pseudohinzii* from a mouse colony.

ANSWERS

1.  Upper respiratory tract, specifically the nasal cavity.

2.  Cross-fostering was the superior method for eliminating *Bordatella pseudohinzii.*

**Reichard et al. Pilot Study to Assess the Efficacy of Ivermectin and Fenbendazole for Treating Captive-Born Olive Baboons (*Papio anubis*) Coinfected with *Strongyloides fulleborni* and *Trichuris trichiura*, pp. 52-56**

Domain 1: Management of Spontaneous and Experimentally Induced Diseases and Conditions

Secondary Species:  Baboon (*Papio spp.*)

SUMMARY: This study evaluated the efficacy of combined treatment with ivermectin and fenbendazole for treating captive olive baboons (*Papio anubis*) infected with *Strongyloides fülleborni*and *Trichuris trichiura*, 2 common nematode parasites of these NHP. Infected baboons were treated for a total of 9 wk with ivermectin (400 μg/kg IM twice weekly) and fenbendazole (50 mg/kg PO once daily for 3 d; 3 rounds of treatment, 21 d apart). Five baboons naturally infected with both *S. fülleborni*and *T. trichiura*(*n*= 4) or *S. fülleborni*alone (*n*= 1) received the combination therapy; an additional baboon infected with both parasites served as a nontreated control. The efficacy of IVM–FBZ was measured as the reduction in fecal egg counts of *S. fülleborni*and *T. trichiura*as determined by quantitative fecal flotation examination after treatment of baboons with IVM–FBZ. All baboons treated with IVM–FBZ stopped shedding *S. fülleborni*and *T. trichiura*eggs by 8 d after treatment and remained negative for at least 161 d. The nontreated control baboon shed *S. fülleborni*and *T. trichiura*eggs throughout the study period. Results indicate that the IVM–FBZ regimen was efficacious for treating olive baboons infected with *S. fülleborni*and *T. trichiura*.

QUESTIONS

1. True or False: *Strongyloides fülleborni*and *T. trichiura*are zoonotic organisms?

2.  In which locations of the gastrointestinal tract do *Strongyloides fülleborni*and *T. trichiura*infect?

3.   What are clinical signs associated with *Strongyloides fülleborni*infection in immunocompetent hosts? Immunocompromised hosts?

4.   What are clinical signs associated with *T. trichiura*in baboons?

ANSWERS

1.  True

2.  *Strongyloides fülleborni*infects the duodenum and upper jejunum of baboons and other OWP, and *Trichuris trichiura*infects the large intestine and cecum.

3.  Immunocompetent- well, tolerated, clinical signs rare. Immunocompromised (or young)- bronchopneumonia, pulmonary hemorrhage, diarrhea, listlessness, anorexia, emaciation, and reduced growth rate

4.   Clinical signs of trichuriasis in baboons are rare but include diarrhea, lethargy, abdominal pain, weight loss, inappetance, dehydration, and intussusception

**Hodo et al. Lack of *Trypanosoma cruzi* Infection in Urban Roof Rats (*Rattus rattus*) at a Texas Facility Housing Naturally Infected Nonhuman Primates, pp. 57-62**

Domain 3: Research

T3: design and conduct research

SUMMARY: This study was conducted as many natural cases of Chagas diseases caused by a protozoan parasite, Trypanosoma cruzi, has been reported in NHP facilities across the southern United States due to probable infected vectors and wildlife.

To identify the modes of transmission of Chagas disease and role of wildlife reservoirs in the facility, Post mortem examination of Roof rats carcass (*Rattus rattus*) were collected. Heart and clotted blood within the ventricles were collected. In addition, vector surveillance for kissing bugs was also done.

RESULTS:

1. Two independent PCR assays was done on the collected heart and blood from the 145 rat carcass for detection of T. cruzi and other trypanosomes. The 145 hearts and 61 blood samples were negative for Trypanosoma cruzi.

2.   The vector surveillance yielded no kissing bugs during the study period.

Results suggested that roof rats are unlikely to be important local reservoirs of T. cruzi at the said facility. Further investigation of transmission dynamics and more comprehensive vector surveillance must be done.

QUESTIONS

1.  True or False: Chagas disease causes inflammatory response, cellular damage, and fibrosis in the cardiac tissue.

2.  What is the primary known vector of Chagas disease?

3.  True or False: Congenital transmission of Chagas disease is documented.

ANSWERS

1.       True

2.       Kissing bugs (triatomine)

3.       True

***Anesthesia***

**Kang et al. Use of Liposomal Bupivacaine for Postoperative Analgesia in an Incisional Pain Model in Rats (*Rattus norvegicus*), pp. 63-68**

Domain 2

Primary Species: Rat (*Rattus norvegicus*)

SUMMARY:Bupivicaine has been commonly used for perioperative analgesia but has not been commonly used in the postoperative state due to its short duration of action.  In this study, the authors evaluated the use of a commercially-available (for humans) slow-release liposomal formulation of bupivacaine to assess its ability to attenuate postoperative mechanical and thermal hypersensitivity in an incisional pain model. This substance is reported to have an efficacy period of 48-72 hours in humans.

Adult male Sprague Dawley rats underwent isoflurane anesthesia and received a 1 cm longitudinal incision (through skin and fascia) on the plantar aspect of the left hind paw. The five treatment groups were as follows:

* Saline 1 ml/kg SQ 5 minutes prior to skin incision and then q 12 hours for 2 days post-op
* Buprenorphine 0.3 mg/ml, 0.05 mg/kg SQ 5 minutes prior to skin incision and then q 12 hours for 2 days post-op
* Bupivicaine 5 mg/ml, 2 mg/kg SQ local infiltration immediately adjacent to skin incision once 5 minutes prior to skin incision
* Liposomal bupivacaine 13.3 mg/ml, 1 mg/kg SQ local infiltration immediately adjacent to skin incision once 5 minutes prior to skin incision
* Liposomal bupivacaine 13.3 mg/ml, 6 mg/kg SQ local infiltration immediately adjacent to skin incision once 5 minutes prior to skin incision

The saline group showed no mechanical or thermal hypersensitivity attenuation at any time post-op in this rat incisional pain model. The buprenorphine group showed mechanical attenuation for 2 days and thermal attenuation for 1 day (recall it was administered BID through day 2 so its effect rapidly dissipated shortly after the cessation of its administration).  The authors referred to previous studies in which the buprenorphine dose needed attenuate mechanical hypersensitivity was found to be lower than that needed for thermal hypersensitivity. The bupivicaine group (single injection) had no mechanical attenuation and thermal attenuation for 4 days. The authors referred to previous studies in which the mechanical attenuation achieved by bupivacaine may, depending on the study, last from about 30 minutes to 2 hours and the mechanical attenuation in this study was assessed for the first time at 2 hours post-administration so may have already dissipated. The lower dose of liposomal bupivacaine (1 mg/kg, single injection) showed mechanical and thermal hypersensitivity attenuation for 4 days. The higher dose of the liposomal bupivacaine (6 mg/kg, single injection) showed mechanical attenuation at 1 day and thermal attenuation for 4 days. The authors postulated that the higher volume needed to achieve the 6 mg/kg dose may have caused tissue distention and irritation and, resulted in the discrepancy seen in the mechanical hypersensitivity as compared to the lower dose (and thus lower volume) group.

Currently, liposomal bupivacaine is only approved for the human market, and thus it is expensive to administer to a small veterinary patient such as a rodent given the manufacturers recommendation that the vial be discarded 4 hours after being opened. However, it is currently under review by the FDA as a veterinary drug which could make its use much more financially feasible particularly in small veterinary patients such as rodents.

QUESTIONS

1.   How was the mechanical hypersensitivity testing conducted?

2.   How was the thermal hypersensitivity testing conducted?

ANSWERS

1.  Von Frey monofilaments

2.   Assessed as withdrawal time of the affected foot relative to the application of heat (generated by a 50 W lightbulb) with a 20 second cutoff time set to prevent tissue injury

***Experimental Use***

**Boivin et al. Physiologic, Behavioral, and Histologic Responses to Various Euthanasia Methods in C57BL/6NTac Male Mice, pp. 69-78**

Domain 2: Management of Pain; Task 4: Euthanize.

Primary Species: Mouse (*Mus musculus*)

ONE-LINE SUMMARY: After studies using male C57BL/6NTac mice, the authors determined that using CO2 with or without isoflurane is an acceptable euthanasia method compared to intraperitoneal injection with pentobarbital-phenytoin, which is the “gold standard” for euthanasia or rodents.

SUMMARY: In this study, the authors asked whether inducing general anesthesia with the inhalant anesthetic isoflurane prior to euthanasia with CO2is an improvement over using CO2 only.  They then compared both isoflurane and CO2 with sodium pentobarbital-phenytoin administered intraperitoneal (“gold standard” for rodent euthanasia).

They used 16 week old male C57BL/6NTac mice and measured behavioral data, cardiovascular data, activity data and ACTH hormone levels (as an indication of stress).  They obtained sporadic evidence that pentobarbital-phenytoin euthanasia may be less stressful than CO2euthanasia.  This was only based on ACTH results, as the cardiovascular, behavioral and activity data were not significantly different.  Given their data, they concluded that the use of CO2with or without isoflurane anesthesia was an acceptable euthanasia method for use in mice.

They also looked at any pathologic alterations in the lungs that may be related to the use of isoflurane or CO2.  They found that the lungs of CO2-euthanized mice had increased perivascular and peribronchiolar edema.  So their recommendation was that an alternative method of euthanasia should be used when studies rely on analysis of the lungs.

QUESTIONS

1.  True/False. The 2013 AVMA Panel on Euthanasia states that when using CO2as a method of euthanizing rodents a controlled chamber-replacement rate of 20% to 30% is necessary.

2.  How does the Canadian Council on Animal Care differ from the AVMA guidelines on euthanasia with respect to CO2 euthanasia?

3. According to rodent studies, how much time is required for corticosterone levels to increase in response to a stressful event?

a. Immediately

b.   4 mins

c.  14 mins

d. 40 mins

ANSWERS

1.  False. A controlled chamber-replacement rate of 10% to 30% is necessary.

2.  Canadian Council on Animal Care – recommend the use of anesthetics prior to CO2 euthanasia.

AVMA Panel on Euthanasia – does not require the use of anesthetics before CO2 euthanasia in rodents

3.  b. 4 mins

**McDonnell-Dowling et al. Consequences of Oral Gavage during Gestation and Lactation on Rat Dams and the Neurodevelopment and Behavior of Their Offspring, pp. 79-83**

Domain 3: Research, Biomethodology techniques

Primary Species: Rat (*Rattus norvegicus*)

SUMMARY: Oral gavage is a common route of administration for drugs in preclinical testing. There is a growing body of evidence that this technique causes stress, but little was known about the effect on neurodevelopment and behavior pups if the dam was treated during gestation or lactation. Pregnant Sprague-Dawley dams either had no treatment or oral-gavage of distilled water once daily from gestational day 7 to postnatal day (PND) 21. Oral gavage had no effect on common reproductive or developmental parameters: body weight gain, duration of gestation, litter size, pup survival, eye opening, pinna unfolding, fur appearance and anogenital distance. Pups of gavaged dams had longer body lengths than control pups on PND 7 and 14, but reduced forelimb grip performance on PND 14 and 17. It is likely that habituation to handling and gavage over the time period lessened the stress on dams and effect on pups. While oral gavage seems to be an acceptable technique during pregnancy and lactation, it may be desirable to add both a no treatment and sham treatment control (e.g. saline gavage) in pharmacologic studies since there are some demonstrated effects of treatment on pups.

QUESTIONS

1. Which two parameters were found to be significantly affected in the treatment group vs the untreated group?

a. Pup body length

b. Pup weight gain

c. Dam adrenal weight

d. Forelimb grip performance

e. Anogenital distance

2. Deficiencies in pup forelimb grip performance suggest deficiency in development of which brain area?

a. Cerebrum

b. Cerebellum

c. Hippocampus

d. Brain stem

ANSWERS

1. a and d

2. b

**Laffins et al. Evaluation of Infrared Thermometry in Cynomolgus Macaques (*Macaca fascicularis*), pp. 84-89**

Domain 3: Research

Primary Species: Macaques *(Macaca spp.)*

SUMMARY: The authors of the study compared temperature data from sedated cynomolgus macaques (*Macaca fascicularis*) to evaluate differences between rectal, infrared (inguinal and chest), and implanted telemetry techniques. The objective was to demonstrate the diagnostic equivalence of the infrared device with the other approaches. Body temperature data were obtained from 205 (137 male, 68 female) cynomolgus macaques under ketamine (10 mg/kg IM) sedation over a 3-mo period during scheduled physical examinations. Infrared measurements were measured 5 cm from the chest and inguinal areas. The authors evaluated 10 (9 functional devices) in sedated cynomolgus macaques (5 male, 5 female) implanted with telemetry units in a muscular pouch between the internal and external abdominal oblique muscles. The authors determined that the mean body temperature acquired by using telemetry did not differ from either the mean of inguinal and chest infrared measurements but did differ from the mean of temperature obtained rectally. In addition, the mean rectal temperature differed from the mean of the inguinal reading but not the mean of the chest temperature. The authors then confirmed that infrared thermometer can be used to replace standard rectal thermometry.

QUESTIONS

1. What made this study different from other IR studies published before?

a. Nothing

b. Measurements were taken 15 cm from the chest

c. Measurements were taken 5 cm from the chest

d. None of the above

2. For accurate temperature readings by IR thermography animals should be?

a. Clipped and sedated

b. Animal sedated

c. Animal conscious when taking temperatures

d. All of the above

ANSWERS

1. c

2. a

**Johnston et al. Using a Cageside Device for Testing Glycosylated Hemoglobiin in Cynomolgus Macaques (*Macaca fascicularis*), pp. 90-94**

Domain 3: Research

Primary Species: Macaques (*Macaca spp.*)

SUMMARY

Introduction: Daily glucose measurements do not provide accurate measures of long-term average blood glucose concentrations. The best method for assessing long-term glycemic control is the measurement of HbA1C levels, which reflect the attachment of glucose to hemoglobin and thus the average of a person’s blood glucose levels over the RBC lifespan (approximately 3 mo). Therefore, optimal treatment of diabetes in human patients involves control with insulin combined with routine HbA1C monitoring. The most widely studied NHP that develop spontaneous diabetes are macaques, and the most extensive research involving the development, characteristics, and comorbidities of diabetes has been conducted in cynomolgus and rhesus macaques. Because HbA1C values are strongly correlated with blood glucose levels and the risk of developing complications, this indicator is a useful parameter to screen for and monitor diabetes in nonhuman primates. The aim of this study was to validate a cage side handheld device that is used in human medicine for use in cynomolgus macaques that would only require a single drop of blood, displays results in 5 min, and drastically reduces the per-sample cost.

Methods: The study population comprised 38 (29 male and 9 female) adult cynomolgus macaques. Eight of these animals are part of a confirmed type 2 diabetic colony (13 to 26 y). In each animal, approximately 4 mL blood was collected from a peripheral vein and placed into an EDTA anticoagulant tube. The samples were tested for HbA1C percentage by using boronate affinity technology. After the 4-mL sample was collected, a single drop of capillary blood was collected and used to run the cage side test (A1CNow+ System, PTS Diagnostics, Indianapolis, IN). The cage side test uses both immunoassay and chemistry technology to measure A1C and total hemoglobin, respectively. The test results are expressed as A1C%, obtained by dividing the amount of A1C by the total hemoglobin in the sample and multiplying by 100. The correlation between the 2 measures was examined to determine whether the A1C readings differ between them.

Results and Discussion: In 7 of the 9 confirmed diabetic animals, the cage side device gave an HbA1C result of greater than 13%, which is the upper limit of the device, thus limiting the usefulness of these values in our analysis. The values for the remaining 2 of the diabetic animals were 12.8% and 9.4%; the commercial lab values for those animals were 9.3% and 7%, respectively. The nondiabetic animals had cage side HbA1C values of 4.9% to 6.4%, compared with values of 3.9% to 4.7% from the commercial lab. The correlation of the 2 methods was highly correlated. Overall, the differences between the 2 measures were consistent, with the cage side HbA1C values always higher. For the nondiabetic animals, the average difference between the measures was 1.39%. However, the difference between the measures in the diabetic range was 2.4% at the 9.4% HbA1C level and 3.5% at the 12.8% HbA1C level, suggesting that the relationship is not constant. Applying the 1.39% difference to the 5.0% cutoff for diabetes (that is, a cutoff of 6.39% for the cage side test) indicated that 2 (6.7%) of our 30 animals would have been misclassified as diabetic. Therefore, the authors felt that the HbA1C test using handheld device may help to rule out nondiabetics and indicate which animals require additional testing for a range of intermediate value.

QUESTIONS

1. This article mentions that macaques are classified as diabetic once they exhibit fasting glucose levels greater than \_\_\_\_\_\_\_, HbA1C greater than \_\_\_\_\_\_\_, and weight loss.

a.  200 mg/dL and 3%

b.  150 mg/dL and 5%

c.   250 mg/dL and 3%

d.  100 mg/dL and 3%

2. Besides HbA1C measurement, what other test is used to monitor long-term glycemic control in NHPs?

a.  Fasted plasma glucose

b.   Urine glucose

c.   Fasted plasma insulin concentrations

d.  Serum fructosamine

e.  Urine ketone

 ANSWERS

1. b

2. d

**CASE REPORTS**

**Jorgensen et al. Calculation of Glucose Dose for Intraperitoneal Glucose Tolerance Tests in Lean and Obese Mice, pp. 95-97**

Domain 3: Research

Primary Species: Mouse (*Mus musculus*)

SUMMARY: Glucose tolerance tests are frequently used in nonclinical research with laboratory animals.  Information regarding how rodent glucose doses should be calculated when obese and lean animals are compared is not well documented.  With typical approaches, obese mice receive larger glucoses doses than lean animals potentially leading to overestimation of glucose intolerance in obese animals.  In this case study, intraperitoneal glucose tolerance tests were performed in mice with diet-induced obesity and their lean controls, with glucose doses based on either total body weight or lean body mass.  Results were as follows: blood glucose AUC was increased significantly in obese mice by 75% and 87% when glucose was dosed according to lean body mass and total body weight, respectively; mice with diet-induced obesity were approximately equally glucose intolerant between both dose calculations.  The authors recommend calculating glucose dose according to lean body mass to eliminate concern regarding overdosing obese animals.

QUESTIONS

1. What does DIO stand for?

a. Diet-induced obesity

b. Diabetes-induced obesity

c. Dopamine-induced obesity

d. Digoxin-induced obesity

2. How was glucose tolerance assessed in mice in this case study?

a. PO

b. IP

c. SQ

d. IV

ANSWERS

1. a

2. b

**Yu et al. Breeding and Rearing Naked Mole-Rats (*Heterocephalus glaber*) under Laboratory Conditions, pp. 98-101**

Domain 4: Animal care

Tertiary Species: Other Rodents - Naked Mole-Rat (*Heterocephalus glaber*)

SUMMARY: Naked mole-rats (NMR) demonstrate high longevity, tumor resistance, hypoxia tolerance and pain insensitivity, making them a widely used model in aging, cancer, neurobiology and other fields. Breeding NMR in captivity is challenging because they are a eusocial species; only a single dominant female and 1 – 3 males reproduce. They have a long gestation period (70 days) and in captivity give birth 2 – 4 times annually compared to once per year in the wild. This low reproductive efficiency limits their use in research. The average litter is 11 pups. The aim of the study was to assess the reproductive performance of NMR in a novel closed colony system.

Seven NMR colonies were obtained and housed in clear plastic cages containing wood-shaving bedding. Each housing unit consisted of activity chambers with connected runways, feeding and resting chambers and defecation chambers. Housing room conditions were maintained at approximately 29°C, humidity of 40 - 70%, and a light: dark phase of 2 – 4 h: 18 – 22 h. Diet included apples, potatoes, sweet potatoes, pumpkins or cucumbers in the morning and afternoon. No water was provided. Cages were cleaned and sterilized every 1 – 2 weeks.

A closed-colony mating system was introduced. Offspring (N1 generation) were removed from their natal colonies after weaning (1 – 2 months) and placed in a new cage. At 5 months, juveniles were re-housed as single sex groups by natal colony until sexual maturity (12 months for males and 10 months for females) when they were paired with non-natal animals and placed in a new cage. Thirty N1 pairs were established. The offspring of the N1 pairs (N2 generation) were bred and paired in the same way as the N1 animals. Forty N2 pairs were established. No other NMR were present to help nurse and care for pups.

In all 3 colonies (original, N1 and N2) average annual parity (% of successful pairs), litter size, numbers of pups weaned and survival rate post-weaning were evaluated. The results showed no significant difference between the 3 colonies in these parameters.

The weights of 60 (30 males and 30 females) N1 and 60 (30 males and 30 females) N2 offspring were measured twice weekly from 10 to 32 weeks. At 20 weeks, females in the N1 generation showed a significant increase in average weight compared to males (P<0.05).

A novel closed-colony strategy comprising of a pair of breeders and single litter of pups produces offspring more quickly and also facilitates experimental observations of individual animals.

QUESTIONS

1. The non-dominant colony animals are irreversibly suppressed reproductively and cannot successfully mate and give birth to offspring. (T/F)

2. The NMR is the longest lived rodent and in captivity has been known to survive for approximately:

a. 28 years

b. 15 years

c. 35 years

d. 10 years

e. 40 years

ANSWERS

1. F

2. a. 28 years

**POSITION STATEMENT**

**“Functionally Appropriate Nonhuman Primate Environments” as an Alternative to the Term “Ethologically Appropriate Environments,” pp. 102-106**

SUMMARY: The American Society of Primatologists (ASP), the Association of Primate Veterinarians (APV) and the American College of Laboratory Animal Medicine (ACLAM) have proposed the term “functionally appropriate nonhuman primate environments” as an alternative to the previously used term “ethologically appropriate environments (EAE)” to better describe environments for nonhuman primates involved in biomedical research.

The reasons for this change are:

* The term “ethologically appropriate environments” does not provide additional clarification beyond that in current regulatory and scientific language.
* The reasons nonhuman primates are kept in human care should be incorporated into determinations of appropriate environments.
* The behavioral flexibility of NHP is ignored.
* Duplication of the wild is not necessary for promoting welfare.
* Naturalistic appearance of environments not always are the most effective.
* Confusion in applying the term ethology to the captive setting. Since the construct of EAE has not yet been put into use, and it has a number of weaknesses as just described, it is timely to propose an alternative term that has a number of advantages over that phrase.

QUESTIONS

1. What do you understand with functional environment?

2. Which of these statements is false?

a. This approach recognizes the importance of both the physical and social environments.

b. Behavioral scientists work closely with biomedical scientists and veterinarians

c. The use of objective scientific analysis is required to identify what characteristics of the environment are the most useful in promoting species-typical behavior.

d. The new term apply to the environments of NHP involved in biomedical research, and it may apply to other settings.

ANSWERS

1. A functional environment could include elements of the natural environment (grass, trees), or it could have more basic structure, but still elicit appropriate behavior from the animals and improve wellbeing, particularly if they are housed in socially- relevant configurations with adequate environmental stimulation.

2. d is false