

The Laboratory Pig

Pig Users Wetlab

Administered by
Laboratory Animals Centre
National University of Singapore



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HANDOUT FOR PIGS

Kingdom: Animal

Phylum: Chordata

Class: Mammalia

Order: Artiodactyla (Even-toed ungulates)

Suborder: Hystricomorpha

Family: Suidae

Genus: *Sus*

Species: *scrofa domestica* (Includes domestic, miniature, and some feral swine)

Common Miniature Swine Breeds

- Yucatan miniature and micro swine
 - The only natural occurring miniature swine
 - Supposedly docile
- Hanford
- Gottingen
- NIH
- Sinclair S1
 - Derived from Hormel minipig
 - Born with melanomas that spontaneously regress (incidence at one year is ~85%)

Others: Ohmini, Pitman-Moore, Chinese Dward, Vietnamese Potbellied, Panepinto

Common Commercial Swine Breeds

- Yorkshire
- Landrace
- Duroc
- Major difference between commercial and miniature breeds is the size at sexual maturity:
 - Miniature: 12-45 kg
 - Commercial: 100 kg or more

Uses of Swine in Biomedical Research

- Research involving the cardiovascular system
 - Atherosclerosis, coronary arterial stenosis, infarction, congenital heart disease, testing of stents and grafts
- Nutritional and gastrointestinal models
 - Nutrients absorption and growth, hepatic metabolism, necrotizing enterocolitis, and teaching surgery students
- Renal disease
 - Renal hypertension, vesicular-ureteral reflux, urinary obstruction
- Research and teaching surgery students
 - Laproscopic and endoscopic procedures
- Transplantation research, xenotransplantation
 - Heart, lung, liver, kidney, viscera
 - Derivation of transgenic strains
- Anatomic and physiologic characteristics of skin

- Plastic surgery models
- Other biological models
 - Systemic and dermal toxicology, immunology, diabetes, malignant melanoma, malignant hyperthermia, gastric ulceration, septic and hemorrhagic shock

Procurement

- Purchase swine from AVA approved vendors; those which are pseudorabies, brucellosis, and respiratory tract pathogen free are preferred
- Agricultural sources:
 - Purebred registered herds
 - Closed herds with known lineage
 - Ideally SPF (SFP is a proprietary term in swine, meaning that the herd of animals has been certified by veterinary and gross necropsy examination to be free of the specific diseases of atrophic rhinitis, pneumonia, swine dysentery, lice, mange, pseudorabies, and brucellosis)
 - Note that higher standards are required for animals used in xenograft procedures
 - Non-SPF commercial sources
 - Less genetically “stable”
 - Endemic respiratory disease
 - GI and parasitic diseases are common
 - Cheaper and more readily available
 - Commercial sources:
 - PWG
 - Charles River
 - Ellegaard

SPF Pigs

- National SPF Swine Accreditation Agency (United States)
 - Major organization in the US
- Accreditation involves:
 - Initial depopulation, the repopulation with surgically derived pigs
 - Farm inspection by a trained veterinarian every 90 days
 - Slaughter inspection of market weight pigs (6-9 months) at 90 day intervals
 - Herd must not have used vaccinations or insecticides to achieve disease-free status
 - Certified free of 7 diseases
 - Turbinate atrophy and snout distortions (*B. bronchiseptica* and *P. multocida*)
 - Pneumonia (various viral and bacterial agents)
 - Swine dysentery (*Brachyspira hyodysenteriae*, aka *Serpulina hyodysenteriae*)
 - Lice (*Haematopinus suis*)
 - Mange (*Sarcoptes scabiei* var. *suis*)
 - Pseudorabies (*Herpesvirus suis*)
 - Brucellosis (*Brucella suis*)

Reproduction

- Sexual maturity
 - Domestic breeds: 3-7 months
 - Mini breeds: 4-6 months
- Estrous cycle: 18-24 days (mean 21 days)

- Duration of estrus: 1-5 days (mean 2 days)
- Ovulation: 30-36 hours after the onset of estrus
- Variable interval between onset of estrus and ovulation
 - Sows are usually bred twice to maximize conception rate
- Gestation period:
 - Domestic breeds: 110-116 days (mean 114 days)
 - Mini breeds: Shorter gestation by a few days
- Diffuse epitheliochrial placenta
 - Allows minimal passage of maternal antibodies
 - Colostrum is very important
- Litter size varies greatly:
 - Domestic breeds: 8-12
 - Mini breeds: 4-6
- Weaned at 4-6 weeks
 - Start to eat solid food at 2-3 weeks
- Sows will rebreed as soon as 3-9 days after parturition

Growth and Development

- 2N=38
- Lifespan: 15-25 years
- Birth weight
 - Domestic breeds: 1-2 kg
 - Mini breeds: 0.5 kg
- Newborn pigs:
 - Do not have brown fat
 - Cannot mobilize glycogen and lipid stores for thermogenesis
 - Therefore, require an external heat source, e.g. heat lamp
- Piglets:
 - Mobile shortly after birth
 - Nurse almost hourly in the first few hours of life
- Hematocrit
 - Physiologically low in neonatal swine; require iron dextran injections

Nutrition

- Swine are true omnivores
 - Will consume a wide variety of foods
- Commercial pig food
 - Designed to enhance growth rate
 - May contain food additives such as antibiotics and growth stimulants
- Ad libitum feeding
 - May produce obesity without any gain in nutritional value
- Miniature swine diets
 - Lower in protein, higher in fiber than commercial diets
 - Control obesity through fixed quantity feeding
 - Does not cause nutritional disorders if fed to domestic swine, but general condition and weight should be monitored
- Presurgical fasting
 - Fasting 8-12 hours is said to empty the stomach and small intestine
 - Fasting 48-72 hours is required to empty the colon

- Bedding and chewable objects should be removed from the cage of an animal being fasted for surgery
- Because of the risk of “salt-poisoning”, water should not be withheld except if absolutely necessary (e.g., stomach surgery)

Behavior

- Social, intelligent animals
 - Prefer to be in contact with other members of their own species
- Can be housed in social groups
 - Animals housed together should be of equal size
 - Dominance fighting:
 - Can occur when new animals are mixed into the group
 - Dominant animals will bite the tails and ears of submissive animals, especially at feeding time
 - Leads to dominance hierarchy, especially large boars
- Can be cannibalistic to sick or injured animals
 - Always house ill or post-surgical animals singly
- Singly housed animals
 - Should be able to see animals in other pens
- Cannot bend to groom
 - Will scratch against the side of the cage
- Site for defecation
 - At the opposite end of the cage from where they are given food and water
- Enjoy rooting with their snouts
 - Best to provide toys that can be rooted or thrown
 - They also like to pull chains

Husbandry

- Flooring
 - Should provide secure footing
 - Become stressed with slippery flooring, and may develop stress symptoms such as stress ulcers
 - Should provide a surface for wearing down their hooves
 - If not, hooves must be trimmed every 3-6 months
 - Concrete or seamless epoxy floors should have grit added to provide for secure footing
 - Best used with deep bedding of wood shavings or straw to provide material for rooting behavior
 - plastic-coated metal grid floors provide good sanitation but swine will pull the plastic off the metal with the first sign of a tear
 - fiberglass slatted-rail floors can also be used provide good sanitation; not easily damaged
 - spacing between grids should be about 1/2 inch to keep hooves from getting caught in between
- Best housed in pens, not cages
- Enclosure should be sturdy
 - Should be free of anything tearable/breakable
 - Pigs good at manipulating any loose-fitting area of the cage with their snouts
 - Chain link fencing can be used for small swine
 - Sturdy aluminum or stainless steel bars for sides

- Edges should be rounded
 - Vertical bars are preferred
- Easily sanitized stainless steel or Teflon feeders
 - Preferable to rubber feeders
- Readily use automated watering systems
 - If only bowls are used, they should be secured to the side of the cage to prevent spillage resulting in water deprivation and “salt poisoning”
- Provide durable toys/balls
 - Keeps them busy and satisfies the rooting instincts
- Wash cages without bedding once or twice daily
 - Swine should not be wetted during this procedure
- If deep bedding is used:
 - The cage can be spot cleaned of feces daily
 - Change the bedding once or twice weekly
 - Tend to stay cleaner with bedding
 - Main disadvantage to bedding is that swine will eat it when fasted
- Some swine develop dry skin
 - Must be rubbed with moisturizing oils
 - Critical in summer for those housed in outside runs

Veterinary Care

- Give a minimum of 72 hours to adjust to new environment
 - Do physical exam, screening test for parasites
- Change diet gradually over several days
 - Increase fiber if stress-induced diarrhea develops
- Periodic physical exam for long-term housed adult swine
 - Includes weight and parasite check
- Vaccination program — basis:
 - Risk assessment
 - How animal will be used in research
 - Housing conditions, e.g., proximity to new animals of unknown herd status

NACLAR

- Relative humidity of 30-70%
- Temperature: 16-27 °C
 - Post-surgical or ill animals may need a warmer environment
- Standard laboratory animal practice is to provide 10-15 air changes per hour with 100% fresh outside air
 - aids in reducing odor and ammonia (contributory to respiratory disease)
- Pigs are basically noisy
 - House away from quieter animals such as cats, rodents, rabbits
- Size of enclosure for one pig
 - <15 kg: 0.72 square metres
 - Up to 25 kg: 1.08 square m
 - Up to 50kg: 1.35 square m
 - Up to 100 kg: 2.16 square m
 - Up to 200 kg: 4.32 square m
 - >200 kg: > 5.4 square m

For group housed pigs, pens may be slightly smaller per pig (NACLAR Guidelines page 48)

- Intrapecies separation might be essential if animals:
 - Where obtained from multiple sites or sources (commercial or institutional)
 - Differ in pathogen status
- Specific attention should be paid to:
 - Management of effective environmental temperature
 - Exposure to sun, ventilation, vapor pressure, floor condition, area per pig, manure management, and quantity and quality of feed and water.
- No particular daily photoperiod is necessary for growing pigs
 - Breeding and lactating pigs respond best to 16 hr light:8 hr dark
- Pigs should be observed, their well-being should be assessed, at least twice each day
- Pigs are by nature social animals.
 - Agricultural research that proposes to house growing pigs individually or in isolation from other swine should be approved by the IACUC. Routine, total isolation should be avoided, but individual housing with at least some social contact (olfactory, some tactile) is acceptable.
- Animal care personnel in swine research and teaching facilities should not be in contact with swine elsewhere unless strict bio-security precautions are followed.

Restraint and Handling

- Agricultural methods such as hog tying and suspension via rear legs are inappropriate for use in laboratory settings
 - Stressful; make animals timid and potentially aggressive
- Easily trained to accept sling apparatuses for minor procedures such as venipuncture and physical exam
- May be herded and restrained against the side of the cage with hand-held panels
- Can be trained to walk with a leash and harness
- Belly scratch can sometimes do the trick
- Respond well to food treats for training
 - Food may be used to calm them during long-term restraint in slings
 - They particularly like carrots, candy, donuts and cookies

Identification

- Ear tags
- Ear notching
- Paints and markers
- Hair clipping

Selected Zoonoses

- *Balantidium coli*
 - Ciliated protozoan in cecum and colon of swine
 - Transmission is by ingestion of cysts or trophozoites passed in feces
 - Commensal in pigs, bloody diarrhea in humans
- *Erysipelothrix rhusiopathiae*
 - G+ bacillus
 - Infects via ingestion of contaminated feed and water or via skin wounds
 - In pigs causes chronic arthritis, urticarial rhomboid lesions, acute bacteremia, acute coagulopathy and hemolysis

- In humans usually causes a local skin lesion, but occasionally may cause endocarditis and sepsis
- *Leptospira interrogans* and *L. borgpetersenii*
 - G— spirochete with prolonged carrier state
 - Pigs can be infected with any of the pathogenic serovars; pomona and bratislava are most common
 - Infectious via mucous membranes, through milk, and via abraded skin
 - Chronic infection in pigs causes mostly abortions, stillbirths, and birth of weak piglets
 - Acute, infection in pigs usually causes mild transient anorexia, pyrexia lethargy
 - In humans, various strains can cause hemorrhage, liver failure, renal failure and death
- Salmonellosis
 - G— rod with prolonged carrier state
 - *S. choleraesuis* may cause sepsis in pigs (dyspnea, icterus, late diarrhea)
 - *S. typhimurium* and *S. choleraesuis* may each cause diarrhea and fever in pigs
 - Wide variety of clinical disease in humans (esp. *S. typhimurium* definitive type 104)
- *Streptococcus suis*
 - G+ coccus commonly carried in upper respiratory, genital and GI tracts of pigs and other animals
 - Incidence of human and pig clinical disease is low, but clinical disease is often severe
 - Meningitis, septicemia, arthritis, pneumonia, endocarditis
 - Most frequently transmitted to humans via skin abrasions or cuts; washing with soap kills it
- In non-SPF pigs, swine influenza and brucellosis may occur

Administration of Medication

- Oral
 - Place into gelatin capsules if foul taste mix with food
 - If restrained in a sling, can use balling gun or stomach tube
- Parenteral
 - Rear leg: gluteal, semimembranosus or semitendinosus muscles
 - Avoid sciatic nerve
 - Neck: seems to be less painful than the leg
 - In small swine, neck injections will be IM
 - In large swine, neck injections will be SC because of layers of fat

Venipuncture

- Use 20 gauge 1.5" needle for swine <50 kg
- Cranial vena cava
 - To avoid injury to vagus nerve, insert needle on the right side of neck, lateral to manubrium sterni, at 30-45 degree angle toward left shoulder
- Jugular vein: lies deep
- Auricular vein: useful for IV catheters in ill swine
- Femoral vein: often used for catheters during anesthesia
- Cephalic vein: fixed; no rolling
- Lateral saphenous vein
- Subcutaneous cranial abdominal vein
 - For potbellied pigs
 - On the ventral portion of the abdomen, dorsolateral to the mammary chain

- Apply firm pressure behind the elbow joint, along the thorax
- Advantages: ease of collection, large volume of blood, safety
- Possible sequelae: infection thrombophlebitis, inadvertent peritoneal cavity puncture

Anesthesia

- Endotracheal intubation: challenging
 - Small jaw opening, long snout, large tongue
 - Swine have a narrow laryngeal passage, with easily traumatized vocal cords and blind folds
 - the lateral folds of the larynx can easily be ruptured and the tube passed into the SC tissues
 - Laryngospasm is common
 - Usually intubated in either dorsal or sternal recumbency
 - Standard laryngoscope blades, 195 mm or longer, are sufficient for swine less than 50 kg; for larger animals, usually need 3-5 cm extension

Unique Features of Swine During Anesthesia

- Swine may be more susceptible to hypothermia because of their relatively hairless skin
- Quite susceptible to anesthetic-induced cardiac arrhythmias
 - More in domestic than in miniature breeds
 - Especially with xylazine and/ or halothane
 - Especially during heart/ coronary artery manipulation
 - Fatal arrhythmias may be prevented with bretylium 3-5 mg/kg slow IV injection every 30 minutes during cardiac manipulation
- Lidocaine 0.3 mg/kg/h can also be used as a continuous IV infusion
- If ventricular fibrillation occurs, electrical defibrillation is best used

Malignant Hyperthermia

- A genetic condition in certain breeds of domestic swine
 - In Yorkshire, Landrace, Pietrain
- Not reported in mini swine
- Extremely muscular with decreased subcutaneous fat
- Autosomal recessive defect in the gene that codes for the ryanodine receptor calcium channel in skeletal muscle
 - The channel is hypersensitive, activated by lower concentrations of agonists and releasing higher amounts of calcium from the sarcoplasmic reticulum than is normal
 - A triggering agent causes acute increase in intracellular calcium, leading to sustained muscle contraction and heat release
 - Accelerated oxygen and muscle glycogen consumption
 - Excessive heat, acids, potassium, CO₂ and muscle proteins are released into the blood
 - Thermogenesis and peripheral vasoconstriction cause hyperthermia
 - Rapid rates of aerobic metabolism desaturate oxygen
 - Rapid depletion of intramuscular glycogen and ATP causes almost immediate rigor mortis after death
- Induced either by stress (Porcine Stress Syndrome)
 - Also induced by certain anesthetics or paralytics
 - succinylcholine, gallamine, acepromazine, ketamine, halothane, isoflurane
- DNA screening test is available

- To differentiate normal, heterozygous and homozygous individuals
- Several countries tightly control breeding to increase the number of heterozygotes
- can be prevented with prophylactic IM or IV dantrolene:
 - 5 mg/kg
- Signs
 - Elevated CO₂ (first sign), tachycardia, hyperthermia, skeletal muscle rigidity, metabolic acidosis
- Rapidly fatal unless triggering agent is discontinued
 - Give dantrolene and intensive supportive care
 - Fluids, bicarb, oxygen, cool baths
- Swine lungs are sensitive to overventilation
 - Pulmonary tissue ruptures easily
 - Tidal volume for swine is 5-10 ml/kg with ventilatory pressure of 18-22 cm H₂O

Anesthetic Monitoring

- ocular reflexes
 - Not reliable in assessing anesthetic depth in pigs
- If the pig is adequately anesthetized, it should have a relaxed jaw tone, absent pedal withdrawal reflex, and absent ear twitch reflex

Signs of Pain in Pigs

- Anorexia
- In general, look for anything out of the ordinary
- Restlessness, unwillingness to lie down
- Squealing upon palpation of painful area
- Excessive squealing and running when handled (normal behavior if handled by a stranger)
- Aggression in adults
- Unwillingness to move
- Hiding

Swine Diseases

- Vesicular diseases
 - Foot and Mouth Disease: Picornavirus
 - Swine Vesicular Disease: Picornavirus
 - Exanthema: Calicivirus
 - Stomatitis: Rhabdovirus
- Fever diseases
 - Classical Swine Fever (AKA: Hog Cholera): Pestivirus
 - African Swine Fever: Poxvirus
- Others
 - Pseudorabies (Aujeszky's disease): Herpesvirus
 - Anthrax: *Bacillus anthracis*
 - Teschen/Talfan: Enterovirus
 - Rabies
 - Tuberculosis: *Mycobacterium bovis*

Euthanasia

Euthanasia should be performed in a way that minimizes pain and distress as much as possible. Therefore, euthanasia recommendations are based on producing rapid unconsciousness in the animal, followed by death. The *2000 Report of the AVMA Panel on Euthanasia* classifies certain

methods of euthanasia as acceptable. These methods consist of those that reliably cause death in a humane fashion when used alone in a conscious animal. Conditionally acceptable methods are those that do not reliably cause death in a humane fashion, or those for which little scientific documentation is available. Unacceptable methods are those that are either inhumane, or that cause significant risk to personnel.

The perception of pain or distress requires functioning cortical and subcortical brain structures. If these brain structures are rendered nonfunctional by anesthesia or other means, then the euthanasia method becomes less important, since no pain or distress will be experienced by the animal. Therefore, certain methods of euthanasia are acceptable in anesthetized animals, but not in conscious animals.

Procedures

1. In general, animals should be euthanized using the method and agent specified on the protocol.
2. The euthanasia technique and agent chosen must be considered acceptable for the species being euthanized. Personnel should consult an LAC veterinarian regarding acceptable euthanasia techniques. Personnel considering use of euthanasia methods not recommended in this document should also contact an LAC veterinarian for guidance.
3. Pentobarbital is the most commonly recommended agent for euthanasia. An acceptable dose of pentobarbital initially depresses the cerebral cortex, leading to rapid loss of consciousness, and then depresses the brainstem leading to respiratory and cardiac arrest. Pentobarbital euthanasia causes only minimal, transient pain associated with venipuncture. Commercially available pentobarbital mixtures are available for euthanasia and are acceptable for use; some contain lidocaine or phenytoin. The proper dose of a commercially available euthanasia mixture should be calculated based on the pentobarbital component. Neuromuscular blocking agents may not be used in combination with pentobarbital.

Swine euthanasia:

- a. Sedation with Telazol 3-10 mg/kg IM, and then pentobarbital 100 mg/kg IV.
- b. Sedation with ketamine 15-25 mg/kg IM, and then pentobarbital 100 mg/kg IV.
- c. IP, IM or intracardiac injection of pentobarbital is not acceptable.

Handling during Euthanasia

1. In addition to choosing the correct method of euthanasia, NUS personnel must also handle animals in a humane fashion during euthanasia, to minimize animal anxiety. Domestic animals should be handled gently to calm the animal. Non-domestic species may need to be tranquilized or anesthetized to minimize distress prior to handling for euthanasia. Personnel performing euthanasia should be technically proficient in the method of euthanasia, and should understand the physiological consequences of the method of euthanasia.
2. Although the aim of euthanasia is to produce minimal pain and distress to the animal, animals may sometimes still become frightened. Vocalizations and pheromones or other odors released by a frightened animal may distress other animals in the room. Therefore, other animals should not be present during euthanasia.

Confirmation of Death

Before disposing of a euthanized animal, NUS personnel must confirm that death has occurred. This is usually accomplished by ensuring that vital signs have ceased for several minutes. Thoracotomy may be used to affirm that the heart has stopped beating.

