1. PURPOSE

1.1. The purpose of this Animal Care and Use Procedure (ACUP) is to describe the quarantine and entrance diagnostics recommendations for Passeriformes housed in Cornell facilities. This ACUP is approved by the Cornell Institutional Animal Care and Use Committee (IACUC). Any deviation must be approved by the IACUC prior to its application.

2. SCOPE

2.1. This ACUP is intended for use by all researchers and staff who work with Passeriformes at Cornell University.

3. INTRODUCTION

3.1. The objective of the quarantine period is to protect resident birds from infectious organisms introduced by new birds, and to reduce the risk of zoonotic disease. This is achieved by: 1) isolating incoming birds to observe for clinical manifestations of incubating disease; and 2) diagnostic testing for identification of infectious and/or zoonotic diseases.

3.2. Contact the Center for Animal Resources and Education (CARE) at Cornell University by emailing care@cornell.edu for more information or for assistance.

4. MATERIALS AND EQUIPMENT

4.1. Personal Protective Equipment (PPE; e.g., shoe covers, coveralls, cap, gloves, mask / N95 Respirator).

4.2. Husbandry supplies (e.g., caging, feed, water, food and water containers, enrichment items).

4.3. Disinfecting solution (e.g., 1:10 dilution of bleach or diluted quaternary ammonium compound).
5. PROCEDURES

5.1. Husbandry

5.1.1. Limit quarantine access to essential personnel only.
5.1.2. Handle resident non-quarantine birds before quarantine birds.
5.1.3. Wear proper PPE for quarantine areas, as detailed on facility or animal housing room signage. See ACUP 715 Personal Protective Equipment for examples.

NOTE: When handling wild birds, assume that there will be contact with zoonotic organisms.

5.1.4. Remove and replace soiled food and change water daily.
5.1.5. For non-wild birds requiring a diet change:

5.1.5.1. Facility manager, CARE veterinarian, and PI cooperatively design a diet plan to gradually change diet of incoming birds to that of the destination facility.

5.1.6. Clean and disinfect waterers, feeders, perches, and cages at least once every two weeks. Ensure the equipment is free of organic debris prior to disinfection. Remove disinfectant residue from surfaces (i.e., rinse) prior to bird contact.

5.1.7. Observe birds daily for signs of illness.

5.1.7.1. Report abnormal behavior or appearance to CARE veterinary staff.
5.1.7.2. Bag and store dead birds in a designated refrigerator and inform CARE veterinary staff. Ensure the PI and facility veterinarian are contacted for possible diagnostic work.

5.2. Wild Caught Passeriformes, Quarantine and Testing

5.2.1. General Considerations

5.2.1.1. Wild caught birds must remain in quarantine in an “all-in, all-out” fashion and not be introduced to resident birds unless diagnostic testing is found to be negative for infectious agents and the quarantine period has elapsed. If the birds will not be brought to facilities where other birds reside, diagnostic testing is not necessary outside of protocol requirements and zoonotic concerns.

5.2.1.2. The PI can perform studies while birds are in quarantine, but proper room order and biosecurity practices must be followed (see section 5.1)

5.2.2. Short Term Studies (i.e., housing duration of 2 weeks or less)

5.2.2.1. Maintain birds in quarantine for the entire study / housing period.
5.2.2.2. No testing necessary outside of protocol requirements.
5.2.2.3. House short term birds separate from long term birds. Do not enter the short term bird housing area prior to entering long term bird housing areas.
NOTE: Acclimatization must be followed as stated in the IACUC approved protocol.

5.2.3. Long Term Studies (i.e., housing duration in excess of 2 weeks)

5.2.3.1. Quarantine Housing

5.2.3.1.1. Consult the facility's CARE veterinarian and the study Principal Investigator (PI) for disease specific screening and any applicable treatment. Refer to appendices for examples of pathogens and diagnostic testing.

5.2.3.1.2. Quarantine incoming wild birds for a minimum of 14 days.

5.2.3.1.3. Quarantine incoming wild birds in an area with complete separation from resident non-quarantine.

5.2.3.1.4. If tending to non-quarantine birds on a given day, do this prior to entering the quarantine/isolation area.

5.2.3.1.5. Euthanize sick birds upon arrival. Perform diagnostic evaluation of euthanized birds in consultation with the CARE veterinarian and the study PI. If euthanasia is not possible, separate all sick birds from those remaining healthy and inform the CARE veterinarian in conjunction with the study PI.

5.2.3.1.6. Do not mix different species of birds within the same enclosure.

5.2.4. Quarantine and Testing of Passeriformes Originating from non-Cornell Institutions

5.2.4.1. Obtain a health history report from the originating institution prior to shipment.

5.2.4.2. Request pre-shipment testing based on PI needs and the pathogen status of the source institution.

NOTE: Stress during shipping can cause latent or occult infections to become clinically evident. Thus, quarantine and diagnostic testing should be performed on all outside institutional birds prior to housing in one of Cornell’s resident bird facilities.

6. PERSONNEL SAFETY

6.1. Medical Emergencies: CALL 911.

6.2. When working with animals wear appropriate PPE, observe proper hygiene, and be aware of allergy, zoonosis, and injury risks. Refer to the CARE Occupational Health and Safety webpage for more information.

7. ANIMAL RELATED CONTINGENCIES

7.1. Post contact information for emergency assistance in a conspicuous location within the animal facility.

7.2. Emergency veterinary care is available at all times including after working hours and on weekends and holidays by calling the CARE (pager 1-800-349-2456)
7.3. Non-emergency veterinary questions and requests for care, email CARE veterinary staff at care@cornell.edu.

8. REFERENCES

8.1. Cornell University Environmental Health and Safety:
https://sp.ehs.cornell.edu/Pages/Home.aspx

8.2. ACUP 542 Maintaining Health and Procedure Records for Research and Teaching Animals:
http://ras.research.cornell.edu/care/documents_k/ACUPs/ACUP542.pdf

8.3. ACUP 715 Personal Protective Equipment:
http://ras.research.cornell.edu/care/documents_k/ACUPs/ACUP715.pdf

8.4. CARE Occupational Health and Safety webpage:
http://ras.research.cornell.edu/care/OHS.html

9. APPENDIX

9.1. Pathogens and Diagnostics

NOTE: Screen at the discretion of the PI or CARE veterinary staff.

9.1.1. Usually for small passeriformes, the following tests and treatment are recommended:

9.1.1.1. Collect fecal pellets (as fresh as possible) from the cage pan in a small tube (such as Eppendorf) and submit for fecal quantitative testing.

9.1.1.2. Obtain cloacal swab using Amies transport or other suitable medium and submit for enteric bacteriology culture (preferably panel 2). For smaller birds, 1mm Dacron swabs are preferred. If the swab tip is too big to be inserted into the vent safely, then collect a fresh fecal sample via aseptic technique from the cage floor and submit it in a sterile vial for enteric bacteriology culture (panel 2).

9.1.1.3. Add Amprolium (Corid) solution to the water supply of birds. See Appendix 9.3 for details.

9.1.1.4. SCATT (moxidectin) application:

9.1.1.4.1. Apply 1 to 2 drops depending on the size of the bird (1 drop per 30g (1.05 ounce) body weight), to the skin of the thigh. Repeat treatment 21 days after first application.

9.1.1.5. Perform complete necropsy if the bird is severely sick or found dead. Below are some helpful tips:

9.1.1.5.1. Before fixing tissue, make quick impression smears of the liver, spleen, kidney and lung, as well as feces and intestine. The lung smear is essentially also a blood smear, as avian lungs are incredibly vascular. To look for blood parasites, perform a Giemsa stain of the lung impression smears. On the intestinal impression
smear, if granulomas are present, they would be a space of negative staining.

9.1.1.5.2. Include the junction between the proventriculus and ventriculus. This is the first place to look for Megabacterium, which will look like fronds of tissue.

9.1.1.5.3. Many enteric pathogens (like Coccidia and mycobacteria) are segmental. Take the duodenum and pancreas together. Take 2-3 pieces of jejunum before Meckel’s diverticulum, 2-3 pieces of ileum, and the ileoceccolic junction and large colon.

9.1.1.5.4. Try to get the spleen as it is the only lymphoid organ you will be able to get. Flip over the ventriculus to find it.

9.1.1.5.5. Signs of dehydration include: renal mineralization, dilation of renal collecting tubules, or a dilated ureter.

9.2. *Chlamydophila psittaci*, Screening and Treatment:

9.2.1. At the discretion of CARE veterinary staff, perform one of the following screening procedures on incoming shipments to identify the zoonotic potential of the colony. Do not introduce wild-caught birds to resident birds prior to diagnostic screening and applicable treatment of positive birds.

NOTE: Chlamydiosis is a reportable disease due to its potential as a zoonotic agent.

9.2.2. Small (10-200 gram) wild passeriformes do not pose a significant risk for exposure to *C. psittaci*. Examples of wild passeriformes in this size category at Cornell include finches, canaries, sparrows, and chickadees.

9.2.2.1. **PCR Testing** - Test of choice due to greater sensitivity

9.2.2.1.1. Test all clinically diseased birds individually, or a representative sample of the diseased birds, for *C. psittaci* using pharyngeal, choanal or conjunctival swabs. Collect feces or fecal swabs from small, delicate birds. Take samples on 3 consecutive days, store in the appropriate transport medium, and process all 3 samples at the same time.

9.2.2.1.2. Separate all clinically diseased and / or PCR positive birds from healthy birds whenever possible, and treat for at least 30 days (see treatment below). Extend the quarantine period for these birds by 2 months. Retest birds by PCR at the end of the extended quarantine. If these birds remain positive despite treatment, either:

9.2.2.1.2.1. Extend quarantine for an additional 2 months and administer alternative treatment.

9.2.2.1.2.2. Euthanize the bird.

9.2.2.2. **Serial Sentinel Necropsy** - Perform a complete necropsy with histopathology, including Giemsa, Gimenez, or Machiavello stain of the
spleen, on a representative sample from the shipment, at two time points after arrival.

9.2.2.3. **Serological Tests** - This testing is not recommended as PCR is a more reliable method for *C. psittaci* detection.

9.2.3. **Treatment**

9.2.3.1. Euthanize all birds positive for *C. psittaci*. If euthanasia is not possible, treat the birds with one of the following:

9.2.3.1.1. **Tetracyclines** - Treatment of choice

9.2.3.1.1.1. **Oxytetracycline**

9.2.3.1.1.1. IM can be used for larger birds, but muscle necrosis at the injection site is a possible side effect.

9.2.3.1.1.2. **Doxycycline**

9.2.3.1.1.2.1. **IM Injection**: 75-100 mg/kg IM q5-7 days for 4 weeks, and then every 5 days for the duration of a 45 day treatment.

9.2.3.1.1.2.2. **Feed**: 1000 mg/kg of doxycycline in the feed

9.2.3.1.1.2.3. **Medicated Drinking Water**: 200-800 mg/liter; very stable in water and is well accepted by birds

**NOTE**: Sick birds may not eat or drink; thus, injectable administration of oral gavage may be necessary. Tetracycline impregnated pellets and crumbs are available.

9.2.3.2. **Enrofloxacin** in the feed at 250-1,000 ppm; used as an alternative treatment to tetracyclines.

**NOTE**: Prophylactic treatment of healthy birds is not recommended due to the increased risk of antibiotic resistance. Additionally, tetracyclines are only effective against actively metabolizing organisms, and not in treating latently or persistently infected birds.

9.3. **Coccidiosis (*Isospora* spp.), Treatment**:

9.3.1. Consult with a CARE veterinarian as to whether treatment is necessary.

9.3.2. Administer 50 mg/L of Amprolium in the drinking water for the first 5 days of the quarantine period, to reduce the incidence of clinical coccidiosis.

9.3.3. Repeat the coccidiostat treatment for the last 5 days of the quarantine period (just prior to transfer out of quarantine).

9.3.4. Dose birds individually if water is not consumed voluntarily due to illness.
NOTE: Atoxoplasmosis is a parasitic disease primarily of passerine birds, especially canaries, finches, sparrows, and starlings. It is caused by species of the coccidian protozoan Atoxoplasma, a host-specific parasite. Identification of atoxoplasma oocysts is notoriously difficult because of structural similarity to those of Isospora spp and sporadic shedding of the organism by infected birds. Diagnosis can also be made on the live bird by peripheral blood smear examination and by cytologic examination of liver or spleen imprints or aspirates.

10. HISTORY

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