GUIDELINES FOR HEALTH SCREENING AND SAMPLING OF GALLIFORMS

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Introduction

Upland game birds are captured and relocated within and between jurisdictions to facilitate a variety of management goals. The movement of wildlife carries the risk of transportation of pathogens to novel environments or populations; as well as the potential for naïve animals contracting disease when exposed to novel pathogens after relocation. Disease surveillance is therefore an important component of all relocation projects.

This document is designed to provide guidelines for diagnostic testing procedures that can inform management decisions, while at the same time protecting the health of wild bird flocks as well as reducing the risk of pathogen transmission to domestic poultry flocks. Although these guidelines were developed primarily for use by jurisdictions affiliated with the Western Association of Fish and Wildlife Agencies (WAFWA), some may have broader application.

The gallinaceous species groups covered in this protocol are pheasants, grouse, quail, turkeys, prairie chickens and chukars.

Surveillance Guidance

Surveillance for movements within states/provinces

For movements within a state or province, disease surveillance is recommended to avoid spread of disease to new areas. To minimize stress and injury related to holding birds, movements of birds within a state or province may use pre-translocation disease surveillance testing as a basis for assuring source flock health.

Sample size

If no translocation is planned and sampling is conducted to determine the health status of the population, a minimum of 30 birds should be sampled. For pre-translocation disease surveillance, 33% of the anticipated number to be translocated or a minimum of 30 birds (whichever is greater), is recommended. Alternative sample sizes, based on statistical sample size calculations aimed at confirming freedom of disease with a predetermined level of confidence, may also be chosen by individual jurisdictions. Sample size calculations that adjust for test performance can be performed using online sample size calculators, as for example http://epitools.ausvet.com.au/content.php?page=home. All birds captured for pre-translocation disease surveillance should be immediately released on site following sampling, and test results should be used to determine whether subsequent translocation is appropriate from the tested flock or region. The duration of approval for movement following capture is to be determined by respective jurisdictions.

The agency may elect to conduct disease screening in the receiving population as well, in order to understand disease risks for the translocated birds. Similar sample sizes can be applied for screening the receiving population.

Surveillance for movements between states/provinces

Recipient states or provinces will dictate requirements for testing. The wildlife agencies conducting the proposed transplant should consult the appropriate agency with authority over import and disease testing of wildlife in the jurisdiction receiving the birds, as well as the state

or provincial wildlife veterinarian. The wildlife agency should be prepared to provide any results of pre-transplant screening and discuss which diseases should be tested, which laboratories will be used, how results will be interpreted, and if further diagnostics should be pursued prior to capturing and collecting samples. The agency with authority over import/testing requirements for wildlife in the destination jurisdiction may have final authority when determining whether wild birds will be allowed into the state or province.

Sample size

With transplants of 30 or fewer birds (for all species including solitary or non-flocking species) all birds should be sampled for specific avian diseases, if possible. With captures over 30 birds, a minimum of 30 birds or 33% of the birds to be translocated, whichever is greater, should be sampled. Alternative sample sizes, based on statistical sample size calculations aimed at confirming freedom of disease with a predetermined level of confidence, may be chosen by individual jurisdictions. Live avian sampling methods may include blood and serum evaluation, oropharyngeal, choanal, or cloacal swabbing, and fecal evaluation. Receiving jurisdictions may require a greater number or sampling percentage of birds and request diagnostic testing at specific diagnostic laboratories to maintain continuity of past testing and reference ranges.

"Pre-transplant screening"

In certain situations, "pre-transplant screening" of a subset of the flock prior to translocations may be authorized as test requirements by the receiving jurisdiction. This helps minimize holding times and increases flexibility of trapping days. Similar sample sizes could be applied for screening the receiving population, if deemed necessary.

Disease Screening

Disease agents of primary concern in upland game birds are pathogenic species of *Mycoplasma* and *Salmonella*, as well as avian influenza viruses. These are pathogens that are monitored in both wild and domestic birds, and testing prior to translocation is required by many jurisdictions.

In addition, there may be other diseases of concern in specific locations. A list of various avian diseases that may be relevant to upland game birds is provided at the end of the disease screening section. The list is not meant to be all-inclusive. Which diseases to test for before moving galliform species within or between states and provinces is ultimately up to the jurisdictions involved.

Wild birds may harbor zoonotic pathogens. A person coming into contact and/or sampling wild birds should use proper personal protective equipment to avoid infection with zoonotic diseases or transfer of diseases between birds.

Mycoplasma

Mycoplasmosis is an economically significant chronic respiratory disease of domestic poultry and captive raised upland game birds. There are three mycoplasma organisms that are the subject of prevention and control programs by the poultry industry in the US: *Mycoplasma gallisepticum* (Mg), *M. synoviae* (Ms), and *M. meleagridis* (Mm). Currently, no such program exists in Canada. Each of these organisms causes a distinct clinical form of mycoplasmosis in domestic poultry. Wild turkeys and other galliform birds have the potential to serve as a reservoir for *Mycoplasma* spp. bacteria, and testing programs are important to ensure that relocation or range expansion programs are not implicated in future outbreaks of mycoplasmosis in domestic flocks.

Clinical mycoplasmosis is believed to have been a significant contributing factor in the population decline of a wild turkey flock in Colorado in the early 1980s (Adrian, 1984). Since then, mycoplasma has been detected via serological surveillance at various levels in numerous flocks across the western North America. Few reports of clinical mycoplasmosis have been documented in wild turkeys, although some manifestations of the disease may not be readily apparent. Therefore,

high seroprevalence of specific pathogens may warrant further examination for effects on the flock.

Mycoplasma gallisepticum has been shown to cause significant clinical disease in captive pheasant populations. Conjunctivitis in passerine birds associated with *Mycoplasma gallisepticum* or other *Mycoplasma* spp. bacteria is becoming increasingly significant in western jurisdictions. *Mycoplasma gallisepticum* has been detected in 27 species (15 families) of birds, although conjunctivitis is most common in finches (Dhondt et al., 2014). The disease cycle in passerine birds is believed to be self-sustaining, though screening galliform birds prior to movement may also help to prevent introduction of *Mycoplasma* spp. into novel passerine populations (Dhondt et al., 2014). Chukar partridge and pheasants naturally infected with *Mycoplasma gallisepticum* have displayed moderate to severe swelling of eyelids and mild to moderate tearing along with more classical signs of upper respiratory disease (Cookson 1994). In poultry, gross lesions include a catarrhal exudate in the nasal and paranasal sinuses, trachea, bronchi, and air sacs. Pneumonia and caseous exudate in the air sacs may also be found (Ley and Yoder, 1997).

Mycoplasma synoviae is most commonly a subclinical respiratory infection seen in all ages of domestic poultry. In younger birds, it is known to affect the synovial membranes of joints and tendon sheaths leading to lameness, weight loss, and failure to thrive. It is associated with egg apex abnormalities in domestic poultry, a malformation that decreases hatchability of viable eggs (Feberwee 2009).

Mycoplasma meleagridis is a pathogen that affects domestic (Stipkovits and Kempf, 1996) and presumably wild turkeys and has occasionally been detected in domestic chickens (Béjaoui Khiari et al., 2011; Catania et al., 2014). Clinical disease has not been documented in free-ranging turkey populations, and infection in other upland gamebird species has not been confirmed. Mm is a vertically transmitted disease causing airsacculitis and occasionally leg, joint, neck and feather deformities. Clinical signs are rare in adult birds. Hatchability and chick survival may decrease. Gross lesions include thickened air sacs containing yellow exudates, skeletal and feather defects, and caseous sinusitis (Rhoades 1971).

Public Health Considerations

None. *Mycoplasma* is a host specific pathogen and has no zoonotic potential.

Testing recommendations for *Mycoplasma* spp.

Previous testing protocols were primarily based on serologic assays recommended by the National Poultry Improvement Plan. However, these tests (rapid plate agglutination or hemagglutinin inhibition followed by culture of reactive or suspect birds) are known to yield inaccurate results including false positives, and cross-reaction between tests for Mg, Ms, and Mm (CPW *Mycoplasma* diagnostics investigation 2017). Polymerase chain reaction (PCR) assays may offer improved specificity over serologic assays. Research at the University of Liverpool in the UK has found that PCR is a superior assessment of both active shedding and overall prevalence of Mg in captive pheasants when compared to serologic testing, specifically when compared to the rapid serum agglutination test (Bradbury 2001). Further data are needed, but there is indication that PCR may offer improved results for *Mycoplasma* screening in a variety of wild galliform species.

PCR designed for detection of Ms, Mg, and Mm in domestic poultry (Colorado State University Veterinary Diagnostic Laboratories) have been used to specifically detect *Mycoplasma* species (Mg, Mm, Ms) from choanal swabs of wild turkeys using pools of up to five swabs (i.e., swabs from up to five different birds placed in the same sampling tube). The perceived improvement of these PCR assays over serology was based on improved specificity (PCR consistently detected only one species of mycoplasma from individuals that react to multiple species of mycoplasma when using RPA and ELISA assays), and confidence that

detection of antigen equates to active infection/shedding of the pathogen(s). Mycoplasma PCR has not been extensively evaluated in all species, and interpretation of results may require consultation with the laboratory or others familiar with the assay and species of interest. PCR assays for Mm have limited availability. If PCR testing for Mm is not available, serologic testing for Mm may be required.

Salmonella

Salmonella pullorum, known as Pullorum Disease (PD), is a highly fatal septicemic bacterial disease of domestic poultry. *Salmonella gallinarum* is known as Fowl Typhoid (FT) and is clinically indistinguishable from PD, save for the fact that PD is more frequently associated with disease in young chicks. Clinical signs in chicks and poults include anorexia, diarrhea, dehydration, weakness, and high mortality. Clinical signs in adult birds include anorexia, decreased egg production, fertility, and hatchability, and increased mortality. Lesions include hepatitis, splenitis, typhilitis, omphalitis, myocarditis, ventriculitis, pneumonia, synovitis, coelomitis, and ophthalmitis. These two diseases are primarily transmitted transovarially from hen to egg, though can be horizontally transmitted via a fecal-oral route as well (Shivaprasad 2000).

PD and FT have decreased in prevalence nationwide due to the eradication efforts of the National Poultry Improvement Plan (NPIP Standards 2014). These diseases are still common in many parts of the world, however, and galliform birds are particularly susceptible (Shivaprasad 2000). Because we have insufficient data to rule out the possibility of wild bird reservoirs for PD and FT, screening prior to movement of wild birds is recommended to support the national eradication effort. Movement of birds potentially carrying PD or FT and subsequent exposure of naïve susceptible poultry flocks could have devastating effects on poultry producers.

Testing Recommendations for *Salmonella* spp.

Live animal testing

Testing for both PD and FT is done via serological assay per the NPIP guidelines. The rapid plate agglutination (RPA) assay is the most commonly performed test, though this has been shown to routinely give false positive results in turkeys and other game birds (Shivaprasad 2000). Tube agglutination testing has been shown to be more specific than RPA when testing domestic turkeys (Shivaprasad 2000), and should be considered the gold standard for Salmonella serology in wild turkeys, with all suspect or positive RPA results confirmed by tube agglutination. Less is known about specificity of the RPA and tube agglutination in other species. If possible, individual birds testing seropositive for Salmonella should be necropsied, with tissues cultured for Salmonella. If PD or FT is confirmed in any bird, the state or provincial agriculture veterinarian should be consulted.

Necropsy and culture

Any gross lesions should be noted at necropsy. Tissues should be collected within 48 hours of death, and held at refrigeration until cultured. The culture should be performed as soon as possible after the necropsy. Tissues collected for culture should include: liver, spleen, and ceca as well as any abnormal appearing tissues. An accredited lab familiar with Salmonella isolation should perform culturing. If any birds are culture positive for PD or FT, no birds from that flock should be translocated.

Culture has been performed on cloacal swabs, however this technique is not recommended for confirmation of *Salmonella* as this method lacks sensitivity, particularly when sampling relatively small numbers of birds (Mueller-Doblies 2009).

Public Health Considerations

Salmonella gallinarum is highly host adapted and is not considered to be a serious public health concern. *Salmonella pullorum* occasionally causes

acute, self-limiting enteritis in humans after consumption of highly contaminated meat or eggs.

Considerations for the choice of laboratory

Testing should be done in a certified lab familiar with running the tests. The NPIP website http://www.aphis.usda.gov/animal_health/animal_dis_spec/poultry/ has information on accredited labs for each diagnostic test. The CSU Veterinary Diagnostic Laboratory is currently the only laboratory offering a *Mycoplasma meleagridis* PCR assay.

Avian Influenza

Avian influenzas are type A influenza (RNA) viruses (AIV) in the Orthomyxoviridae family. They are generally classified by their hemagglutinin and neuraminidases subtypes and they may be either low pathogenic (LPAI) or highly pathogenic (HPAI) (Ferro et al., 2012). The vast majority of virulent HPAI outbreaks in North America have been H5 or H7. Outbreaks of any AIV are a concern for poultry industry and wild bird management because increased circulation may lead to the development of HPAI through reassortment or mutation. While aquatic birds (Anseriformes and Charadriiformes) are considered the natural reservoir, AIV have been isolated from 105 wild bird species in 26 families. Susceptibility to AIV subtypes varies widely among bird species. Gallinaceous birds, domestic and wild, are considered highly susceptible. Outbreaks have occurred in farmed quail (*Coturnix coturnix japonica*) and detected in wild bobwhite quail (*Colinus virginianus*) in Texas. Experimental infections have developed in European quail (*Coturnix coturnix*), red legged partridge (*Alectoris rufa*), chukar (*Alectoris chukar*), bobwhite quail, and pheasants (*Phasianus colchicus*).

Testing recommendations for avian influenza

Testing for AIVs is typically done by PCR. Initial PCR screening tests for avian influenza viruses are conducted to determine if any AIV viruses are present. If an AIV is detected, samples are then sent to the National Veterinary Services Laboratory to determine the subtype of AIV and whether it is of high or low pathogenicity. Oropharyngeal and cloacal swabs can be collected for AIV PCR. Oropharyngeal swabs are preferred for gallinaceous birds, but cloacal swabs may be considered if oropharyngeal swabs are not feasible. Additional serological testing is available to evaluate exposure to AIVs. Serology should be utilized and interpreted with caution since the tests are not validated in wildlife. Avian influenza is a reportable disease and birds testing positive on either PCR or serology must be reported. The following tests are commonly applied to test for avian influenza. Specific test requirements will be determined by the individual jurisdiction.

- Oropharyngeal and/or cloacal swab submitted in one tube of BHI media for rRT-PCR for AIV matrix. Follow up testing for H5 and H7 if positive (possibly other H types H3, H4).
- Serology exposure to AIV proteins recognizing all subtypes via indirect ELISA. H5 and H7 AIV detections are reportable to OIE.
- Other serologic tests include Agar Gel Immunodiffusion test (AGID) and competitive ELISA (cELISA).

Sample Collection and Preparation for Mycoplasma, Salmonella, and Avian Influenza Testing

Swabs

Choanal swab for Mycoplasma PCR

- Swab the choana (palatine fissure), avoiding contact with the rest of the mouth. Gently rotate swab in choanal cleft, collecting mucus and cells.
- Dip swab in brain-heart infusion (BHI) tube and rotate swab 5-10 times.
- When removing swab from tube, press swab against side of tube repeatedly until no more liquid comes from the swab.
- Discard swab.

Oropharyngeal swab for AIV PCR

- Swab the oral cavity and opening of the trachea, avoiding the esophagus, and bring the swab up through the choanal cleft.
- Dip swab in BHI tube and rotate swab 5-10 times.
- When removing swab from tube, press swab against side of tube repeatedly until no more liquid comes from the swab.
- Discard swab.

For Mycoplasma and avian influenza, up to 5 swabs (1 swab per bird) may be pooled in one BHI tube. Label BHI tubes (e.g. pool 1, pool 2, etc.) and record which individual birds contributed to each pool. Do not add samples from different populations into the same pool to aid interpretation in the unlikely event of a positive result. For other diseases check with the testing laboratory before pooling samples. Pooling may decrease the sensitivity of the test below desired detection limit.

Cloacal swabs for PCR

- Separate the feathers to expose the cloaca. Gently insert the swab and rotate swab it in the cloaca, collecting mucus and cells.
- Dip swab in BHI tube and rotate swab 5-10 times.
- When removing swab from tube, press swab against side of tube repeatedly until no more liquid comes from the swab.
- Discard swab.

Serum

Venipuncture

- Volume: no more than 1% of the body weight of the bird should be drawn for disease sampling. Check with lab regarding volume needed for each test
- Venipuncture sites in order of preference:
  - Jugular vein (right side is usually larger): Wet down area with alcohol and hold off jugular vein at the thoracic inlet. It is best to insert the needle at a shallow angle with the bevel down, which will allow gentle lifting of the vein while maintaining blood flow and reduce the risk of lacerating the other side of the vessel. The jugular vein may be difficult to access in birds with well-developed cervical air sacs.
  - Medial metatarsal vein: often useful in waterfowl and galliforms. On the inside of the leg.
  - Cutaneous ulnar (basilic/wing) vein: On the inside of the elbow. Prone to forming hematomas that may limit flight, use with care.
- After drawing blood, it is important to apply firm, gentle pressure over the venipuncture site to ensure that excess bleeding under the skin does not occur.

Sample Handling and Storage

- Remove the needle from the syringe prior to injecting the blood into collection tubes in order to minimize hemolysis.
- If drawing blood using a tuberculin syringe with attached needle, cut the needle off with scissors before emptying into sample tubes. For birds less than 100 gm, sample size may be extremely limited and that many blood samples will be placed directly into Eppendorf tubes without anticoagulant since serum yields the best results for multiple diagnostic tests.
- Place tube on side or at an angle to facilitate clotting.
- All samples should be stored cool (NOT frozen) with, but not directly contacting, ice packs in a cooler. Blood and swabs should be driven or mailed overnight to the diagnostic laboratory. Please provide a list of individual birds and swab pools with each submission.

Interpretation of test results

Results from testing, particularly serology results, must be interpreted with caution. Interpretation should be conducted on a case by case basis while considering all aspects of the situation. For this reason, we do not provide specific guidance on interpretation of test results and recommend that agencies consult their wildlife health specialist(s) as well as the agency with authority over import/testing requirements for wildlife in the destination jurisdiction for guidance on interpretation. In cases where multiple tests will be run, agreement on reconciliation of conflicting results should be established in advance.

Guidelines on holding and transportation

Temporary holding and transport

Often birds have to be held prior to obtaining disease testing results or due to time needed to achieve the desired transport number. If no larger temporary holding facility is available on-site, portable plastic or cardboard carriers, or pet carriers may be used for holding birds for up to 48 hours, but ideally less than 24 hours. Cardboard boxes, including those with wax or plastic coating, should be disposed of after use unless being used with the same group of birds. Plastic boxes may be disinfected for re-use (see cleaning and disinfection protocols below). Keep in mind that birds should be kept in a cool, quiet area with adequate ventilation. If long transports (>6 hrs) with warmer temperatures are anticipated, fluids may administered by veterinary staff via subcutaneous or gavage tube administration. Fresh (green) grass clippings also may be used to provide hydration and some nutrition to birds held pending release.

Reducing Disease Incidence and Mortalities in Holding Facilities

Birds that are temporarily housed in pens prior to release or translocation should be provided appropriate habitat and space, a clean and quiet environment to minimize stress and disease incidence. Further, the enclosures should be predator-proof to minimize stress and mortalities from outside sources. It is important to recognize early signs of disease and to intervene swiftly and appropriately to reduce mortality.

Density of animals

The following table (from ANR Publication 8155, University of California-Davis Extension Service) gives recommendations on pen sizes for different game species for those being held for greater than 24-48 hours.

Table 1. Suggested pen size and bird density
*Double the space required for each species if growing pens do not have adequate cover.

Pen size			
Species	Width (ft)	Length (ft)	Bird density (sq ft/ bird)
Chukar	50 or 100 (15 or 30 m)	150 (45 m)	3-5 (0.28 - 0.46 sq m)
Pheasant	50 or 100 (15 or 30 m)	150 (45 m)	10-12*
Quail	50 (15 m)	150 (45 m)	3-4 (0.28-0.37 sq m)

Cleaning and disinfection

The goal of cleaning and disinfection is to minimize or destroy disease-causing agents such as viruses, bacteria, fungi, and parasites. When possible, the pathogens of concern should be identified in order to better target the disinfection process.

Cleaning involves the physical removal of organic material (i.e. feces, feed, carcasses, etc.) and disinfection. It is important to remove the organic material prior to disinfection as some pathogens can survive the disinfection process in feces, food byproducts, wood, leaves, etc. In pens with natural ground cover, it may not be practical or possible to remove all of the organic material. Organic material should be reduced and removed for pen sanitation between captive flocks/groups of birds.

Disinfection involves reducing or killing the pathogens. Drying out the facility and exposing the area to natural sunlight may decrease pathogen loads, but may not suffice as the sole method of disinfection. There are several types of chemical disinfectants. Some good choices in avian settings include Vircon®, Roccal®, and 10% sodium hypochlorite (bleach). Treated surfaces should be rinsed thoroughly before coming into contact with birds. Avian parasites are minimized by treating the animals using insecticides or parasiticides if indicated, removing wet organic material, and rotation of pen use. (Morishita and Gordon, 2002).

Minimizing stress

It is important that pens are designed in an area away from heavy foot traffic, loud noises, and activity in order to minimize the stress to the birds. This is especially important with housing wild-caught birds since they are unaccustomed to captivity.

Minimize the use of loud equipment, and keep personnel entering the enclosure to the minimum needed to take care of housing and feeding. If animals are handled ensure handlers are quiet during the process. Covering the head and body with a towel, sock, or cloth bag can also reduce stress of handling.

Predator and rodent-proofing housing (pens)

One effective technique involves burying hardware cloth 12 inches into the soil substrate around the coop to protect the captive birds from rodents and other burrowing animals. This will not keep out all rodents or snakes, but will eliminate most of the larger predators.

Rats and mice are universal pests that cannot always be completely excluded from pens. Using poisons can be problematic because of presence of an abundance of alternative, attractive feed, and the possibility of penned birds ingesting the poison. Traps are a better solution, although the process is more time consuming. Proper placement and an ample supply of traps are important. Place traps across paths used by rats or mice including between obstacles, next to walls, or rafters, etc. Runway sets are more effective than randomly placed baited traps (Morishita and Gordon, 2002).

Recognizing and minimizing disease

Although there may be occasional mortalities observed in confined aviaries, a cluster or spike in dead birds should be a red flag that something is wrong. Any carcasses should be removed from the pen and properly disposed of. If there is no obvious evidence of predation or trauma, the freshest carcasses should be placed in a bag and saved in a cooler or refrigerator and shipped as soon as possible to a veterinary diagnostic laboratory. A complete history should accompany the shipment with as much pertinent information as possible (number of birds in the pen, number of mortalities, other potential contributors, etc.). If mortalities are predominantly originating from one pen, initiate a quarantine; which consists of not moving or introducing new birds to the pen, not sharing equipment between pens; and setting up a disinfectant footbath for personnel entering and leaving the area. Occasionally signs of disease will be more subtle, such as watery feces, sneezing or snicking, labored breathing, nasal or ocular discharge, neurologic signs like circling, or severe weight loss (usually recognized when handling).

- Additional biosecurity measures:
    - Minimize new bird introductions to the pen
    - Group animals by collection date and location (when possible) and age
    - Require personnel working with the game birds to wear dedicated clothing and footwear for the pens; especially if they have poultry or other birds at home
    - Deceased birds not submitted for testing should be incinerated or buried (if disease suspected) or disposed of in a routine manner for other causes of death

Guidelines for euthanasia

Euthanasia should be conducted according to the American Veterinary Medical Association Guidelines for Euthanasia of Animals (Available at:https://www.avma.org/KB/Policies/Pages/Euthanasia-Guidelines.aspx).

Other diseases

Many other diseases can affect upland game birds, or are of concern to the domestic poultry or turkey industry. Below is a list of some other diseases that potentially could be of importance in certain locations and that should be taken into account if deemed necessary by the state or provincial wildlife or agricultural veterinarians.

Parasitic diseases

Upland game bird flocks can carry a variety of parasitic diseases that should be considered and treated on a case by case basis prior to the translocation.

Internal parasites

Protozoa (cryptosporidia, coccidia, and eimeria): Coccidiosis is known to affect many species, but its significance at a population level appears to be minimal. However, small mortality events have been documented. Examination of fecal specimens by direct smear of fresh feces mixed with LRS or normal saline, and flotation should detect significant infections. In many gallinaceous species, cryptosporidia reside in the respiratory tract and cause tracheitis, air sacculitis, coughing, and dyspnea; excessive mucus in the respiratory tree may be seen at necropsy and oocysts may be seen in feces.

Flagellated protozoa: *Trichomonas gallinae* is a flagellated parasite of the esophagus. Not a significant pathogen in many species but has caused mortality in a number of doves. Other *Trichomonas* spp. may be found in the intestinal tract. *Histomonas meleagridis* is a flagellated protozoa that causes blackhead disease. The parasite is carried by *Heterakis gallinarum*, which is a nematode commonly found in the ceca of chicken, turkeys and other birds, and also have been found in earth worms (Davidson 2006). Clinical signs include ruffled feathers, drooping wings, and passing of sulfur colored droppings. Best method of diagnosis of *Trichomonas gallinae* is direct smear of feces and oropharyngeal swab. *T. gallinae* may be cultured via the In pouch®

system used for *Tritrichomonas foetus* diagnosis and incubating at a slightly higher temperature (101°F/ 38.3°C).

Helminths: Nematodes, cestodes and flukes may all be found in the intestinal and respiratory tracts, and associated organs of gallinaceous birds (e.g. *Aulonocephalus linidquisti*, *Subulua bumpti*, *Rhabdometra odiosa*, *Rillietina* spp., *Dispharynx nasuta, Heterakis gallinarum, Capillaria* spp., *Syngamus trachea, Prosthogonimus* spp.). Most do not appear to cause significant disease in the adapted host but transfer of such parasites to a new ecosystem or host species could be problematic. Intestinal parasites should be screened for using fecal flotation.

Miscellaneous: Hematozoa generally cause minimal pathology in most but have caused problems in some ecosystems (Hawaii). Microfilaria indicate infection with one or more filarid worms in the air sacs, coelom, heart or eye. Many of the filarids found in the coelomic cavity do not appear to cause significant mortality or morbidity. *Oxyspirura* spp. (eyeworms) has indirect life cycle with a cockroach as an intermediate host species. Examination of blood smears and careful examination of the eyes should be able to detect the parasites in the population.

External parasites

External parasites consist of flies (hippoboscid), mites, ticks, fleas. Infestation of nests can lead to abandonment and loss of young. Ticks infesting birds are often larval soft ticks and severe infestations can cause significant anemia. External parasites can contribute to declines in body condition and increase stress levels as a result of skin irritation and pruritus. Careful physical examination should be able to detect the presence of external parasites in the population.

Parasite Testing
Collect fresh feces from traps, transport boxes or cloacae and examine by standard direct and fecal floatation methods; collect oropharyngeal swabs and examine by saline wet mount and culture via In pouch® system, examine blood smears and conduct careful physical exams.

Mitigation
Treat birds based on the results of diagnostic testing or as a preventative measure when diagnostic testing is not available. Treatment recommendations are based on label dosages for domestic species. Use of these drugs in non-domestic species should be considered off label and requires a valid veterinary-client-patient relationship. In addition, relevant regulations concerning withdrawal times should be observed if the pharmaceuticals are administered immediately prior to or during the hunting season. Coccidia/eimeria: amprolium 0.012%-0.024% for 3-5 days in water, sulfadimethoxine 0.05% for 6 days in water, sulfamethazine 0.1% in water for 2 days. Flagellate protozoa: metronidazole 50 mg/kg/day for 5 days; ronidazole 6-10 mg/kg/day for 14 days. Intestinal helminths: ivermectin 0.2mg/kg IM or PO, levamisole 25-30 mg/kg PO or 0.03% to 0.06% in water, pyrantel pamoate 4.5 mg/kg PO, fenbendazole 20-50 mg/kg PO. Cestodes: praziquantel 5-10 mg/kg PO or IM. Other internal helminths: ivermectin 0.2 mg/kg PO or IM. External parasites, systemic treatment: Ivermectin 0.2 mg/kg PO or IM, moxidectin 0.2mg/kg PO or topically. External parasites, topical treatment: pyrethrin sprays, 5% carbaryl powder.

Broad spectrum mitigation strategy: amprolium or sulfamethazine in water, ivermectin PO or IM, and topical treatment with carbaryl or pyrethrin.

Viral Diseases

Avian pox is caused by avipoxviruses. Infections result in coalescing, proliferative, degenerate, and necrotizing dermatitis on the head and legs (Davidson 2006). The oral cavity and esophagus can also be affected (wet form) (Davidson 2006).

Species: Turkeys, grouse, chukars, pheasants, quail, and many other avian species.

Turkey rhinotracheitis is caused by an avian metapneumovirus. The virus causes acute respiratory tract infections of domestic turkeys and has been detected in domestic pheasants, Muscovy ducks, and guinea fowl (OIE 2009). Further, the virus has been detected in some wild

bird populations in the United States (Shin, Njenga et al. 2000). Clinical signs in domestic chicken and turkeys include nasal discharge, coughing, ruffled feathers, airsacculitis, pneumonia, hepatitis, and pericarditis (OIE 2009).

Species: Turkeys, pheasants, other gallinaceous birds.

Lymphoproliferative disease (LPD) is caused by an oncogenic avian retrovirus of domestic turkeys that has historically been restricted to Europe and Israel, but recent studies have shown that LPD is widespread in wild turkeys in the US (Thomas, Allison et al. 2015). Most wild turkeys do not develop clinical signs but affected birds may show ruffled feathers, anorexia, diarrhea, and reduced activity (Davidson 2006). Post mortem findings in affected birds may include nodules in various organs and enlarged spleen (Davidson 2006).

Species: Turkeys.

West Nile virus is a mosquito borne arbovirus that can cause meningoencephalitis, hemorrhages, myocarditis, and splenomegaly in many bird and mammalian species (Davidson 2006). The virus is now considered endemic across the US (Petersen, Brault et al. 2013).

Species: Corvids are especially susceptible, but mortalities have been reported in sage grouse (Walker, Naugle et al. 2007), other grouse species, pheasants, quail, chukars and a variety of other galliformes as well as other bird species. (Source of species list: NWHC https://www.nwhc.usgs.gov/disease_information/west_nile_virus/affected_species.jsp)

Marble spleen disease/ pheasant adenovirus is caused by avian adenovirus (group II), and affects farmed gamebirds such as pheasants, guinea fowl, peafowl, and chukar partridges (Fitzgerald and Reed 1989). The virus causes cute respiratory disease and post mortem findings are characterized by an enlarged mottled spleen pulmonary congestion (Fitzgerald and Reed 1989).

Species: Pheasants, chukars, guinea fowl, peafowl, chicken, turkeys.

Avian adenoviruses: *Quail Bronchitis* is caused by an avian adenovirus and mainly affects young, domestic Bobwhite Quail. Clinical signs include respiratory distress, coughing, sneezing, rales, and nasal or ocular discharges (Fitzgerald 2007). Post mortem findings may include airacculitis, hepatitis, and necroses in the liver and spleen. Mortality rates may reach 100% in birds < 2 weeks of age, but is lower in adult birds (Fitzgerald 2007). *Inclusion body hepatitis* causes acute disease characterized by diarrhea, decreased appetite, lethargy and low to moderate mortality rates (<10 – 30%) (Fitzgerald 2007).

Species: *Quail bronchitis*: Northern bobwhite quail. *Inclusion body hepatitis*: Northern bobwhite quail, Gambel's quail.

Reticulendotheliosis is caused by an avian retrovirus of the genus gammaretrovirus. Clinical signs in affected birds include weight loss, paleness, ataxia, tremors, circling, and abnormal feathering. Neoplasias may include the liver, spleen, intestine and heart (Davidson 2006).

Species: Turkeys, chicken, quail, ducks, geese, and likely many other species

Newcastle disease is caused by avian paramyxovirus serotype 1 (PMV-1). In domestic poultry, infections are divided into highly virulent (velogenic), moderately virulent (mesogenic), and of low virulence (lentogenic). Velogenic and mesogenic infections (classified as virulent Newcastle disease, vNDV) are reportable. Clinical signs include respiratory disease, nervous signs such as tremors, paralyzed wings and legs, twisted necks, and paralysis. Post mortem lesions include petechial and hemorrhages on serosal and mucosal surfaces. Mortality rates can be very high with vNDV outbreaks (NWHC 2017). Pigeon paramyxovirus (PPMV1) is an avian paramyxovirus 1 that affects doves and related species but has been identified in chickens and grey partridge in the United Kingdom

Species: Galliform species are considered susceptible, but outbreaks of NDV in wild birds have only occurred in double crested cormorants and columbiformes. The disease has also been detected in captive reared pheasants and Hungarian partridges (NWHC 2017).

Infectious laryngotracheitis is caused by the Gallid herpesvirus I. Clinical signs include coughing, gasping, rales, severe dyspnea, tracheitis and conjunctivitis. Post mortem findings may include hemorrhages in the trachea and larynx and intranuclear inclusion bodies in the tracheal epithelium (OIE 2014)
Species: Domestic chicken, pheasants, partridges, and peafowl.

Bacterial diseases

Ulcerative enteritis is caused by infection with the bacterium *Clostridium colinum* and results in weight loss, bloody diarrhea, and high mortality. Postmortem findings are characterized by ulceration of the intestine and focal necrosis in the liver (Davidson 2006).

Species: Quail, chicken, turkeys, pheasants, grouse, and other gallinaceous birds.

Avian tuberculosis is caused by *Mycobacterium avium*. Clinical signs include chronic, progressive weight loss and weakness. Post mortem findings may include white to yellowish nodules in the liver, spleen, and intestines (NWHC 2017).

Species: Any bird species.

Avian cholera is caused by the bacterium *Pasteurella multocida*. Clinical signs are often acute and include depression, ruffled feathers, diarrhea, increased respiratory rate, and mucoid discharge from the mouth. Post mortem lesions may include hyperemia and congestion, enlarged liver and spleen, and necrotic foci in liver and spleen (NWHC 2017).

Species: Avian cholera is especially common in waterfowl and is uncommon in upland game birds (NWHC 2017), but has been diagnosed in grouse, turkeys, pheasants, quail, and a variety of other species.
Chlamydiosis is caused by the bacterium *Chlamydia psittaci*. The disease may be inapparent to highly fatal, depending on the strain of *Chlamydia*, physiological condition of the bird, route of exposure, and presence of other stressors. Clinical signs may include nasal and ocular discharge, ruffled feathers, diarrhea, and respiratory distress. Post mortem findings may include spleno- and hepatomegaly and fibrinous polyserositis (NWHC 2017).

Species: Chlamydiosis has been reported in captive turkeys, quail, pheasants, chukars, and peafowl, but is generally detected infrequently in wild gallinaceus birds (Kaleta and Taday 2003)

Public Health Considerations: *Chlamydia psittaci* has zoonotic potential.

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