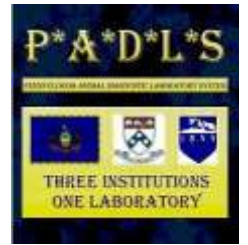




New Bolton Center PADLS Testing & Sample Collection Guide



New Bolton Center PADLS • University of Pennsylvania • 382 West Street Rd., Kennett Square, PA 19348-1692 • PH: (610) 444-5800 FX: (610) 925-6806 • www.padls.org

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Aquaculture

NBC PADLS offers these services to aid the aquaculture industry in diagnostic purposes only. The laboratory does not perform any regulatory testing with respect to aquaculture. Contact your regional veterinarian for coordination of specimen collection.

DEAD FISH ARE NOT GENERALLY SUITABLE FOR A COMPLETE NECROPSY.

The following information is requested for proper collection and submission of specimens to the laboratory.

1. The name, address and phone number of the owner and the submitting veterinarian must be indicated.
2. Live fish delivered or shipped to the laboratory are essential for accurate diagnostic investigation.
3. Fish not submitted alive should be iced immediately after death, placed in a plastic bag, wrapped in newspaper and packed in a Styrofoam shipping box with 2 to 3 frozen ice packs and delivered to the laboratory IMMEDIATELY or shipped to the laboratory by an overnight courier.
4. A detailed history should include the pH of the water, dissolved oxygen levels, feed source (manufacturer), any changes in the fish management, mortality numbers, symptoms and information regarding treatment already given. A description of the water flow and layout of the facility is very helpful.
5. The laboratory reserves the right to determine the suitability of a specimen for testing. If a specimen is unsuitable because of deterioration, the laboratory will request that you resubmit another sample.
6. When in doubt, or if you have any questions, contact the laboratory for assistance.

The methods of shipment to the diagnostic laboratory for fish disease diagnosis are listed from most desirable to least desirable. Specimens can be brought to both NBC and the Manheim Field Office. The impact of fish handling and preservation on disease diagnosis can be summarized:

Shipment method	Microbiology	Parasitology	Histology	Toxicology
Live	+++	+++	+++	+++
Iced	+	+	+/-	+++
Frozen	-	-	-	+++
Formalin	+/-	+/-	-	-

Key	
+++	No effect, excellent specimen for examination
++	Negligible effect, good specimen for examination
+	Moderate effect, specimen may be usable
+/-	Substantial effect, specimen may not be useful
-	Dramatic effect, specimen not useful

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	SET-UP	EST. TURN AROUND	REMARKS
Enteric redmouth <i>Yersinia ruckeri</i>	Bacterial culture	Fish	Live fish, fish iced immediately after death	LIVE – aerated container of their original water ICED – plastic bags in waterproof Styrofoam container w/ cold packs	LIVE – drop off ICED – overnight courier	M-Th	5 days	Fish that have died must be delivered to the laboratory within 2-3 hours after death or they are of no value for testing
Furunculosis <i>Aeromonas salmonicida</i>	Bacterial culture	Fresh water & marine fish	Live fish, fish iced immediately after death	LIVE – aerated container of their original water ICED – plastic bags in waterproof Styrofoam container w/ cold packs	LIVE – drop off ICED – overnight courier	M-Th	5-20 days for atypical strains	Fish that have died must be delivered to the laboratory within 2-3 hours after death or they are of no value for testing
<i>Ceratomyxa shasta</i>	Direct examination, histopathology	Fish (<i>Salmonid spp.</i>)	Live fish, fish iced immediately after death	LIVE – aerated container of their original water ICED – plastic bags in waterproof Styrofoam container w/ cold packs	LIVE – drop off ICED – overnight courier	M-F	1-3 days	See general aquaculture information.
<i>Myxobolus, Myxosoma cerebralis</i> (Whirling Disease)	Plankton Centrifuge Method	Fish (<i>Salmonid spp.</i>)	Sixty (60) fish heads	Whirl bags or freeze & deliver to laboratory	Contact laboratory	Per request	28 days	For diagnostic purposes only.

Clinical Microbiology

TEST/ AGENT	PROCEDURE	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	SET-UP	EST. TURN AROUND	REMARKS
Acid fast stain	acid fast stain	feces swab	sterile container	overnight shipment on gel packs	M - F	1 day	Specimens to be cultured for mycobacteria other than <i>M. paratuberculosis</i> may be referred to NVSL.
		tissue	Culturette	slide mailer			
Aerobic culture (includes Listeria)	aerobic culture	swab fluid	culturette or Amies transport media (latter preferred for <i>Actinobacillus</i> and <i>Haemophilus</i>)	overnight shipment on gel packs	M - F	2 - 5 days	Take specimens from the margin of lesions. If tissue and fluid specimens are not protected by transport medium, ship overnight. Uterine swabs should be taken with a guarded swab commercial collection system. <i>Listeria</i> isolation may require 1-3 months.
		tissue	sterile container				
Anaerobic culture	anaerobic culture	tissue 3.0 cm ³	sterile container, anaerobic transport medium	samples should not be refrigerated	M - F	4 - 10 days	Anaerobic bacteria should be suspected if the infection involves tissue that is normally sterile, a site of recent trauma, or a poorly vascularized area Transport media/swabs available upon request.
		fluid aspirate	Vacutainer (> 3 ml) anaerobic transport medium				
		swab	anaerobic transport medium Amies gel swab				
Antibiotic susceptibility	Minimum inhibitory concentration (MIC) or Disk diffusion	bacterial isolate derived from PADLS culture work	N/A	N/A	M - F	3 - 5 days	Susceptibility reports will be released to veterinarians only. Susceptibility test reports contain <i>in vitro</i> test results only and do not constitute recommendations for treatment. Charges may reflect re-isolation and identification by PADLS.
		characterized bacterial culture	medium slant with growth				
Blood Culture	blood bottle culture (aerobic and anaerobic)	7-10 ml whole blood	SPS Vacutainer or blood collection bottle	overnight shipment on gel packs	M - F	3-7 days	Indicate if patient is on antimicrobial therapy, define drug and dose. Collect specimen following complete surgical preparation. Preferred specimen collected prior to initiation of antimicrobial therapy.

TEST/ AGENT	PROCEDURE	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	SET-UP	EST. TURN AROUND	REMARKS
<i>Campylobacter culture</i>	microaerophilic culture	feces	sterile wide-mouth container	overnight shipment on gel packs	M - F	3-5 days	
		rectal swab-	Amies or Cary Blair transport media				
<i>Clostridium perfringens</i> screen	anaerobe culture PEA and EYA plating	feces or intestinal loop	sterile container	overnight shipment on gel packs	M - F	2-3 days	
<i>Clostridium perfringens</i> toxin typing Toxin detection	ELISA (NBC)	feces or intestinal loop	sterile container	overnight shipment on gel packs	Th W/Sa	2-8 days	PCR can be performed by arrangement. See under Molecular Diagnostics
	PCR (PSU, PVL)	isolated microorganism (feces)	anaerobic BA in AnaPak fecal cup	keep feces frozen			
<i>Dermatophilus</i> exam	Geimsa Stain and/or culture	tissue or swab	sterile container Culturette	overnight on ice/gel packs	M - F	1-7 days	
Gram morphology	Gram stain	heat-fixed slide	slide mailer	none	M - F	1 – 2 days	
Mastitis culture*	Aerobic milk culture	quarter sample; quantitative, bulk tank sample	sterile capped vial freeze immediately (minimum of 10 ml for bulk tank)	overnight on ice/gel packs	M - F bulk tank M-W	2 - 5 days	Please see below for sample collection; call laboratory for additional information.
Mycology	Fungal culture and/or microscopic exam	tissue fluid	sterile container	overnight on ice packs	M - F	1 - 42 days	If dermatophytes are suspected, specimens should not be transported in a sealed container and should not be refrigerated.
		skin scrapings	paper envelope	none			
Quantitative bacterial culture	Dilution Plate Count	feed	1 qt Ziploc® bag	overnight on ice packs	M - F	2 - 5 days	Raw semen samples must arrive at the laboratory within 24 hours of collection in order for examination to be considered valid. Urine samples collected by cytocentesis or from catheterized animals are preferred; however, mid-stream collections are suitable. <i>Samples must arrive at the laboratory on the same day as collection in order for the examination to be considered valid.</i>
		processed semen	sterile container				
		raw semen	sterile container				
		urine	1 L sterile container				
		water, bedding or litter	1 qt Ziploc® bag				

TEST/ AGENT	PROCEDURE	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	SET-UP	EST. TURN AROUND	REMARKS
<i>Salmonella</i>	<i>Salmonella</i> culture	feces (10 g)	wide-mouth screw-top container; child resistant prescription vial	overnight on ice/gel packs for all except feed	M - F	Presumptive id: 3 - 4 days 3 - 10-days	Samples should be obtained from the rectum of animals whenever possible. <i>Specimens received in open containers or gloves will be refused.</i>
		tissue (ligated ileum, mesenteric lymph node, etc.)	sterile container				
		exudate swab	Culturette, Amies or Cary-Blair transport medium				
		environmental swab/ Swiffers	condensed evaporated skim milk		M-W	7 - 15-days	
		eggs (200 g)	sterile WhirlPak bag				
		feed	sealed bag				
<i>Salmonella</i> Serotyping	Serotyping	bacterial isolate	Nutrient Agar slant	overnight	M - F	1 - 10 days	Other tests available on bacterial isolate-- Pulsed-Field Gel , Electrophoresis (PFGE), Phage typing Please contact laboratory
Tritrichomonas	Tritrichomonas culture	cow: vaginal wash/cervical mucus	InPouch media sterile capped vial with Diamond's media, or Lactated Ringers Solution (LRS) InPouch media for culture	transport at ambient temperature protected from light LRS - hand carry within 4 hours	M - F	5 - 10 days	Contact lab prior to collection for instructions and transport media Samples collected in Clark's/Weybridge media for venereal Campylobacteriosis cannot be tested for Trichomoniasis.
		bull: preputial wash					
	semen						
	fetal fluids	sterile, capped vial for exam					

* **Mastitis:** Wash the udder and teats and allow to air dry. Clean the teat orifice with alcohol pledgets. Sample in reverse order to reduce contamination. For routine culture, the first stream of milk should be discarded. Hold the collection vial as nearly horizontal as possible and collect 2-3 mL of milk. When collecting composite samples, include an equal volume of milk from each quarter. Freeze samples immediately after collection.

Molecular Diagnostics

General guidance about swab selection for PCR: Cotton/Dacron/Polyester Swabs are acceptable. Calcium alginate and gel swabs should not be used and, where possible, use of wooden swabs should be avoided. Swabs can be submitted dry when indicated or in appropriate medium as recommended below. Call the laboratory running the test for more information.

Legend: * = preferred specimen **BHI** - brain heart infusion broth; **PCR** - polymerase chain reaction; **FFPE** - formalin fixed paraffin embedded; **RRT-PCR** - real time reverse transcriptase polymerase chain reaction

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Avian Influenza (AI) including H5/H7 subtyping	RRT-PCR	Avian: chickens, turkeys, & other gallinaceous birds, wild birds, ducks	Tracheal swab* (for gallinaceous birds), cloacal swab* (for waterfowl), intestine, lung, kidney, trachea, air sacs, sinus	Leak-proof with BHI broth Don't use wooden swabs	Overnight on ice, do not freeze Should be frozen if the samples cannot reach the lab within 48 hrs	1-3 days	Up to 5 swabs can be pooled in a tube containing BHI broth. Available from NBC.
Avian paramyxovirus-1 (APMV-1) including Exotic New Castle Disease	RRT-PCR	Avian: chickens, turkey, & other gallinaceous birds, wild birds, ducks	Tracheal swab* (for gallinaceous birds), cloacal swab* (for waterfowl), intestine, lung, kidney, trachea, air sacs, sinus	Leak-proof with BHI broth Don't use wooden swabs	Overnight on ice, do not freeze	1-3 days	Up to 5 swabs can be pooled in a tube containing BHI broth. Available from NBC.
Infectious Bronchitis Virus	RRT-PCR	Avian	Tracheal swab	Leak-proof with VTM or BI broth Don't use wooden swabs	Overnight on ice, do not freeze	3-5 days	
<i>Mycoplasma gallisepticum</i>	PCR	Avian	Culture broth Plates Tissues Aspirates Swabs	Sterile-capped vial	Overnight on ice packs	1-5 days	Avoid the use of calcium alginate or wooden shaft swabs
<i>Mycoplasma synoviae</i>							
Microbial Sequencing including Viral Bacterial Fungal *MicroSeq™	PCR	All species	Isolate(s) on plate, broth, or in suitable medium *sterile body fluids	Sterile container	Overnight on ice *Overnight frozen	5-15 days	16S or MicroSeq™ for bacterial and fungal ID 28S for Fungal ID-Viral Gene sequencing

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Porcine respiratory & reproductive syndrome (PRRS): Arterivirus, Lelystad agent	RRT-PCR	Porcine	Tissue Serum (1 mL)	Leak-proof container with or without BHI	Overnight on ice Do not freeze	5 -6 days	PCR is run on M-F
<i>Salmonella</i> spp. Including <i>Salmonella</i> enteritidis (SE)	Real time-PCR	All species	Feces (10 g), feed, environmental swabs, fluid (10 mL), tissue, milk filter, eggs	Leak-proof container	Overnight shipment on ice/gel packs, do not freeze	2-3 days	SE PCR satisfies FDA, NPIP or PEQAP test requirements. Call laboratory for details.
<i>Strep. Equi</i> (Strangles)	PCR	Equine	Guttural pouch Tracheal wash/swab Nasal swab Pharyngeal swab	Culturette Container w/ no additives	Overnight on gel packs	Negative result, w/in 24 hrs Presumptive positive – 2 extra business days to confirm diagnosis	

Parasitology

A range of testing services is provided by PADLS laboratories for identification of whole parasite specimens, microscopic ova or larvae, protozoan organisms, ectoparasites, and parasite antigens. In order to provide an accurate and timely identification, we request that samples be submitted according to the guidelines shown below. Use shipping containers that seal tightly. Provide proper temperature requirements for the particular specimen, and forward as quickly as possible. Clearly identify each sample with the animal's ID, owner's name, and veterinarian or practice involved. A full history is always helpful to assist in providing the best possible diagnosis. Please call the laboratory if you have additional questions.

- **Always provide fresh samples, including feces, and ship with frozen gel packs for transit in less than 24 hours.**
- **Parasite ID results will be provided within 14 days. All other samples are processed as workload permits and reported within 7 days.**

TEST/ AGENT	PROCEDURE	TISSUE/ SPECIMEN	CONTAINER	REMARKS
Cryptosporidium	Acid fast stain	1 g feces	Leak-proof, sealed	Smear
Fecal parasite direct smear	Fecal smear	5 g fresh feces	Leak-proof, sealed	
Lungworm screen/sedimentation	Baermann	10 g fresh feces	Leak-proof, sealed	Identifies larvae rather than ova
Fecal flotation	Modified McMaster's	5 g fresh feces	Leak-proof, sealed	Quantification of nematode ova
Parasite identification	Microscopic exam	Parasite	Leak-proof, sealed	Identification of ecto/endo parasites

Pathology

BIOPSY SERVICE: Practitioners are encouraged to use containers and mailers specifically intended for formalin fixed specimens. Label containers appropriately, especially if more than one specimen is submitted. Owner address, patient signalment and a brief history are required information. Submission forms are available online at www.padls.org or by calling the laboratory. Tissues should be placed in 10% buffered formalin. Ten volumes of formalin are required to adequately fix one volume of tissue. Specimens should consist of the lesion site and surrounding tissue. Tissue should not be thicker than 1 cm. Do not allow fresh or fixed tissue to freeze. Formalin will freeze at extremely low temperatures, damaging the tissue. Adding 1 mL of ethanol to each 9 mL of 10% formalin will prevent such freezing. Certain tissues require special attention. Intestines are labile; they should be opened and contents removed with a saline or formalin rinse. Bouin’s fixative is preferred for endometria. Brains are fixed whole in 10 volumes of formalin. Fresh brains should be hemisectioned if viral or bacterial studies are intended—refrigerating one half and saving the other half in formalin.

PROCEDURE	SPECIES	TISSUE/SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Biopsy	Domestic livestock, wildlife, zoo animals, avian	Surgical biopsies	Sealed within padded outer mailer	US Postal Service, courier service, or hand deliver	2 days	Unfixed tissue, tissue requiring special handling (e.g. bone, eyes) and special stains require extra time for completion.
Endometrial Biopsy	Equine	Alligator forceps-collected endometrial specimens	Same as biopsy mailers with Bouin’s fixative available	US Postal Service, courier service, or hand deliver	7 days	Reproductive history and rectal findings are required information. An epicrisis (therapeutic recommendation by a theriogenologist) is available only through PADLS-NBC.

Immunohistochemistry (IHC)

Please call laboratory for test availability and handling instructions. There is no extra charge for IHC if multiple tissues from a necropsy are submitted for histopathologic exam. Containers must be leak-proof and appropriately padded. Delivery can be via the US Postal Service, courier service, or by hand. Turn-around time is 7-14 days.

Legend: A – avian, B – bovine, C – camelid, D – canine, E – equine, F – feline, G – caprine, O – ovine, P - porcine

AGENT	SPECIES	TISSUE/SPECIMEN
Avian influenza	A	Trachea, heart, kidneys, intestines, lungs, pancreas
Bovine coronavirus (BCV)	B	Ileum, colon
Bovine respiratory syncytial virus (BRSV)	B	Lung
Bovine viral diarrhea virus (BVD) – ear	B	Ear notch
Bovine viral diarrhea virus (BVD) – tissue	B	Lymphoid, skin, intestine, fetus: chorion, lymphoid
Camelid coronavirus	C	Ileum, colon
<i>Campylobacter spp.</i>		Intestine, fetus: chorion, liver
Infectious bronchitis virus	A	Nasal, trachea, lung, kidney
Canine distemper	D	Lung, lymphoid, gut, kidney, urinary bladder, brain
Canine parvovirus 2	D	Jejunum, ileum, tongue, lymphoid, liver
<i>Chlamydia spp.</i>	O, B, G	Lung, liver, spleen, placenta, cotyledon, fetal tissue

AGENT	SPECIES	TISSUE/SPECIMEN
<i>Coxiella</i>		Chorion
Equine herpesvirus 1	E	Fetus/neonate: lung, liver, thymus, spleen, adrenal, chorion; CNS: brainstem, spinal cord
Feline coronavirus (FeCo/FIP)	F	Ileum, colon, lesions
Feline herpesvirus 1	F	Skin, tongue, lymphoid, lesions
Feline parvovirus	F	Jejunum, ileum, tongue, lymphoid, liver
Helicobacter		Stomach, liver
Bovine herpesvirus 1 (IBR)	B	Nasal mucosa, upper airways, lung; fetus: chorion, lung, liver, adrenal, lymphoid
<i>Lawsonia</i>	E, P	Ileum
<i>Leishmania spp.</i>	D, E, other	Skin, liver, spleen, lymph node, bone marrow
<i>Leptospira spp.</i>	B, O, G, E, P	Kidney, liver, chorion
<i>Listeria monocytogenes</i>	B, O, G, E, P	Caudal brainstem, lung, liver, lymphoid, chorion
Mycobacterium spp.	Various species	Granulomas
<i>Neospora caninum</i>	B, D, other	Brain, heart, skeletal muscle, chorion

AGENT	SPECIES	TISSUE/SPECIMEN
Papillomavirus		Skin lesions
Porcine circovirus 2 (PCV @)	P	Tonsils, lung, lymphoid, ileum, kidney; fetus: chorion, cord, tonsils, lung, thymus, spleen, kidney
Porcine Respiratory & Reproductive Syndrome virus (PRRS)	P	Tonsils, lung, lymphoid, ileum, kidney; fetus: chorion, cord, tonsils, lung, thymus, spleen, kidney
Porcine TGE/PED coronavirus	P	Ileum, colon
Rotavirus		Ileum
<i>Toxoplasma gondii</i>	Various species	Brain, heart, skeletal muscle, chorion
West Nile virus	A, E	Equine: brain, brainstem, spinal cord Avian: intestine, spleen, heart, kidney, brain
<i>Yersinia pestis</i>	Various species	Intestine, lymph node, respiratory, spleen, lesions

Field Necropsy / Diarrhea Panel

Circumstances occasionally require practitioners to conduct postmortem examinations on client premises or in the clinic. Pre-purchased specimen collection kits (complete with instructions, containers, swabs, transport medium, and shipping boxes) are available upon request. All PADLS laboratories are prepared to receive and process packages containing field necropsy specimens.

The US Postal Service and commercial couriers have strict regulations regarding the packaging of potentially hazardous biological and chemical materials. Resistance to crushing and the containment of leaked fluids are prime considerations. Formalin-fixed-tissues should be in screw top containers, the lid taped, and placed within a leak-proof, plastic bag. Excess formalin may be removed if fixation is complete, leaving enough fluid to keep the specimen moist. Do not allow fixed tissues to become frozen from ambient temperatures, dry ice or super-frozen ice packs. Fresh tissues should be double bagged. If delayed in transit, gas distension from putrefaction could cause leakage. Tissues for microbiological procedures should be chilled and not frozen. Specimens for toxicological analysis may be fresh or frozen. To avoid contamination of the specimen selection, the proper container is important (see Toxicology section for details).

PROCEDURE	SPECIES	TISSUE/SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Field necropsy	Agricultural animals	Fresh & formalin-fixed tissues	Styrofoam shipping box as described above	Overnight courier, mail only M-Th	14 days	Field necropsy samples submitted by a referring vet are processed the same as submitted animals. Consult with the case coordinator regarding history and sample/test selection.
Field necropsy	Dog & cat	Fresh & formalin-fixed tissues	Styrofoam shipping box as described above	Overnight courier, mail only M-Th	14 days	Please contact the laboratory prior to submission.
Diarrhea panel	Agricultural animals	20 mL feces	Leak-proof sealed container	Overnight courier, mail only M-Th w/ frozen gel packs	7-10 days	

Necropsy Service

Submitters may send fresh, recently dead animals to the PADLS laboratory of their choice. Deliveries of dead animals or samples may be made 24 hours a day. Live animals may be submitted only by prior arrangement. Arrangements for transportation should be made by the client as soon as possible. Chill carcasses if feasible; do not freeze unless otherwise instructed. Antemortem blood samples (whole blood or serum) should be taken prior to pre-transport euthanasia and should accompany the submission. Referring veterinarians or clients are encouraged to notify the lab and consult with the pathologist before sending the carcass(es) or other samples. A clinical history, written by the referring veterinarian, owner, or flock supervisor should accompany the submission. Individuals submitting wild animals for necropsy must receive permission from the Pennsylvania Game Commission prior to submission of the animal. Insurance cases and cases with the potential for litigation must be identified as such.

Please call for instructions for emergency necropsy submissions during evening and weekend hours. Ancillary tests beyond the gross postmortem examination are at the discretion of the pathologist. The necropsy fee covers most routine tests (e.g., bacteriology, histology, virology, serology, some toxicology, etc.); however, extensive testing may incur additional fees. Tests that cannot be done by PADLS are forwarded to various referral laboratories and the fees charged by the outside laboratory will apply. Turn-around time is usually two weeks. Some tests, e.g., special toxicological assays and virus isolation, may take several more weeks. Submitters will be apprised of the status of their case, and addendums will be issued as information becomes available. Cosmetic necropsies are not performed.

Carcasses are incinerated/rendered after necropsy and no parts of the body can be returned.

Disposal of carcasses not submitted for necropsy is by arrangement only and will incur a fee. Necropsy of animals in a state of advanced autolysis is at the discretion of the pathologist.

PROCEDURE	SPECIES	TISSUE/SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Mammalian necropsy	Domesticated livestock, confined wildlife, zoo animals	Necropsy animals should be submitted recently dead or terminal. Multiple animals are desirable as indicated on the fee schedule	Small carcasses should be bagged & boxed with frozen gel packs	Client delivery as soon as possible after death, or alive (only w/ prior arrangements)	14 days	Cases requiring removal of the spinal cord require 3 weeks for completion & additional fees.
Dog/Cat necropsy	Dog, cat	Please call for availability and instructions				
Avian necropsy	All avian species	1-10 fresh dead &/or live birds representative of the condition	Rigid, leak-proof, container w/ frozen gel packs (dead birds)	Hand deliver or overnight courier. Mail only M-Th. (do not ship live birds)	1-3 days	Live birds should be hand delivered in appropriate containers with sufficient ventilation & space. Some tests, e.g. special toxicological assays & virus isolation, may require several more weeks for completion.

PROCEDURE	SPECIES	TISSUE/SPECIMEN	CONTAINER	SHIPPING*	EST. TURN AROUND	REMARKS
Aquaculture necropsy	Fin fish	1-10 fish (live preferred)	Bucket with water sample & sufficient oxygen supply	Client deliver of live fish	72 hours*	Live fish should be hand delivered in appropriate container(s) w/ sufficient oxygen supply.
		Dead fish are NOT generally suitable for a COMPLETE necropsy. Dead fish need to be iced immediately after death, placed in a plastic bag, wrapped in newspaper & packed in a Styrofoam shipping box w/ 2-3 frozen ice packs.	Rigid leak-proof container w/ frozen ice packs (dead fish)	Pack dead fish in Styrofoam cooler & deliver to the laboratory IMMEDIATELY or ship by an overnight courier only M-Th (Do not ship live fish)	*ancillary testing may require several additional weeks for completion	Call lab before shipping.
Exotic necropsy	Reptiles, amphibians, exotic mammals	Necropsy animals (1-10) should be submitted recently dead or terminal. Refrigerate – do NOT freeze dead specimens	Live animals should be in enclosed transportation containers w/ adequate ventilation. Small carcasses should be bagged & boxed w/ frozen gel packs in a leak-proof container in insulated box or cooler for fresh dead	Client delivery as soon as possible after death, or alive. Ship only w/ prior arrangements by an overnight courier only M-Th. Venomous reptiles must be presented DEAD.	72 hours* *ancillary testing may require several additional weeks for completion	Live animals, except venomous reptiles, should be hand delivered in appropriate containers w/ sufficient air supply. Cases requiring the removal of the spinal cord require 3 wks for completion & additional fees. Call lab before shipping.

Avian Serology

The majority of the tests conducted by PADLS are on serum samples from commercial poultry flocks being tested for avian influenza in association with the Live Bird Market System (LBMS), PA Monitored Flock Program, and National Poultry Improvement Plan (NPIP) testing requirements. NPIP certification is also available for Salmonella and Mycoplasma species. Please contact BAHDS for additional information on this program. The remainder of the tests are associated with:

1. Regulatory testing required for interstate shipment, sales and PA exhibitions
2. Surveillance testing for diseases, e.g., avian influenza
3. Diagnostic testing from birds submitted to the laboratory for necropsy
4. Flock serologic profiling.

Avian serology testing requires appropriate amounts of uncontaminated, non-hemolyzed sera. Sample collection technique and timely submission significantly impact laboratory results. Poor quality sera may be reported as non-testable. The following outline is provided to assist poultry servicemen/veterinarians in proper blood collection/serum submission. Contact the laboratory for proper submission forms and blood collection tubes.

1. Collect 2 to 3 mL of blood aseptically from wing vein.
2. Transfer blood to a **clean, dust-free** tube (most blood tubes equal 5 mL).
3. Stopper and place in a horizontal position for several hours, at room temperature until the serum separates from the clot. Alternatively, specimen tubes which will be hand-delivered may be sealed with tape and placed in a semi-horizontal position for several hours until the serum separates.
4. After serum separation, the specimens should be left in an upright position at room temperature or in the refrigerator overnight until transport to the lab.
5. Submit these specimens to the laboratory as soon as possible, or pour the serum into a clean, sealed vial.
6. Submit a minimum of 1.0 mL of serum when requesting multiple tests.
7. Fill out the proper specimen submission form, include all requested information.
8. If possible, the samples should be sent refrigerated using an overnight courier service or, alternatively, hand delivered to the lab.
9. Contact the laboratory to make special test arrangements for export or sale samples or when holidays interrupt the normal test schedule.
10. The laboratory reserves the right to determine the suitability of a specimen for testing.

Codes: **AGID** Agar Gel Immunodiffusion Assay; **ELISA** Enzyme Linked Immunosorbent Assay; **HI** Hemagglutination Inhibition Assay **IFA** Indirect Fluorescent Antibody Test; **RPA** Rapid Plate Agglutination

NOTE: Turnaround time (business days) is based on one-time testing with a negative result. Suspect samples will be retested requiring additional time. The turnaround time for some ELISA testing is dependent on the number of samples submitted to the lab. Samples may be held until an adequate number of samples are available for efficient use of test reagents. Contact the laboratory if a more rapid turnaround is required.

TEST/ AGENT	PROCEDURE	TISSUE/ SPECIMEN	SHIPPING*	SETUP	EST. TURN AROUND	REMARKS
Avian influenza (AI)	AGID	0.5 mL serum Eggs	Gel pack (serum)	M-F	2 days	See Virology (Avian) section for virus isolation information
Infectious bronchitis virus (IBV)	ELISA	0.5 mL serum	Gel pack	M-F	2-14 days	See Virology (Avian) section for virus isolation information
Infectious laryngotracheitis virus (ILT)	ELISA	0.5 mL serum	Gel pack	M-F	2-14 days	Used for epidemiological surveillance of broilers over 6 weeks old outside of endemic areas
<i>Mycoplasma gallisepticum</i> (MG)	ELISA, RPA	0.5 mL serum	Gel pack	M-F	1-14 days	See Molecular and Clinical Microbiology Sections for agent detection information. DO NOT FREEZE
<i>Mycoplasma synoviae</i> (MS)	ELISA, RPA	0.5 mL serum	Gel pack	M-F	1-14 days	See Molecular and Clinical Microbiology Sections for agent detection information. DO NOT FREEZE
<i>Salmonella pullorum</i> (Typhoid)	RPA, Tube agglutination	0.5 mL serum	Gel pack	M-F	2 days	Samples must be received no later than 2 pm Thurs. for tube agglutination tests

Mammalian Serology

Properly performed serology requires appropriate amounts of uncontaminated serum. Specimen collection technique and timely submission significantly impact the quality of the laboratory results. The following outline is provided to assist in the submission of specimens for optimal results. Check individual tests under “remarks” for special handling requirements.

1. Aseptically collect adequate amounts of blood for the number of tests requested.
2. Collect blood in a clean, tightly sealed, leak proof tube or container (red top tube or serum separator tube).
3. Leave tubes in a horizontal position at ambient room temperature (not below freezing).
4. Submit specimens as soon as possible. Store specimens refrigerated if submission is delayed.
5. Pack samples securely to prevent breakage. Packages should contain ice packs if samples will be in transit more than 24 hours or during warmer months of the year. Use courier service whenever possible as the routine delays which occur in mailing can result in deterioration of sample quality.
6. Fill out submission forms completely and legibly and submit with samples. Be sure to indicate the tests to be performed as well as clinical history and purpose of test, (e.g. sale, diagnostic, etc.). Number all specimens to match forms using consecutive numbers only. Submissions should be signed by an accredited veterinarian.
7. Whenever possible, send clear serum which has been aseptically transferred to a sterile red top serum tube. We will accept clotted whole blood samples with the understanding that they may have to be reported as hemolyzed or non-testable if they arrive in poor condition.
8. The test schedules are flexible. In some instances setup is based on demand with a minimum and maximum time shown. Contact the serology section of the laboratory to which you are submitting specimens in advance when you need a group of animals tested within certain time constraints. Allow 1-2 days for transport of samples between PADLS laboratory locations.
9. Acute serum specimens will be tested within the same run as convalescent specimens when submitted together.
10. Turn-around time is defined in working-days as the time elapsed between the date (and time) that the specimens are received in the laboratory until the date (and time) that the results are recorded in the Laboratory Information Management System (LIMS), or, the time the testing is completed. Suspect specimens will be retested requiring additional time.

Codes: **AGID** - agar gel immunodiffusion

Export testing or special needs: Contact the PADLS laboratory prior to sample collection to schedule all necessary tests. Large volume export testing especially requires advance notification due to limited availability of some reagents and to allow scheduling of technical time to ensure timely reporting of results.

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	SETUP	EST. TURN AROUND	REMARKS
Bovine Leukemia (BLV, bovine leukosis)	AGID	Bovine	1 mL serum	As needed	2 days	Please contact lab before shipping large groups of (300+) samples to assure availability of testing. The turn-around time for large groups of samples submitted upon request may be 1-5 days.
Equine Infectious Anemia (EIA)	AGID (Coggins) ELISA	Equine	1 mL serum	M-F	1-2 days	For same day ELISA results, samples must be received by noon. Results for samples received after noon will be available the next day.

Toxicology

Enclose a detailed history with submissions, including clinical signs, lesions, tentative diagnosis, a list of drugs administered, and the response to previous treatments. A complete history may enable us to decide which additional tests are warranted.

In situations involving small animals, reptiles, or birds where the amount of sample indicated below may not be available, please submit as large a sample as possible. Each individual specimen must be in a separate, clean, clearly labeled container. Containers should be freezable, such as plastic bags. For liquids, use clean glass/plastic containers. **Purity of sample is of extreme importance in toxicology.**

Do not use preservatives with the exception that EDTA or heparin should be used as anticoagulants for whole blood samples. Serum should be separated from the clot. Containers should be as full as possible, but allow for expansion if samples are to be frozen. If “source materials” are obtained, mail them in a separate package. **Do not mail them in the same package as the tissue samples.** Information regarding their availability, means of exposure, and an estimate of amount ingested should be included. Enclose the label or a list of all chemicals and concentrations on the label.

If a topical or injected toxicant is suspected, submit a large area of tissue from the suspected absorption site. Many poisons produce microscopic changes in tissues. Therefore, always submit sections of tissues in 10% formalin for histopathologic examination in addition to the fresh or frozen tissues required for chemical analysis.

If other analyses are desired, or if you have questions, please call the laboratory. At present, samples are kept 6 months after date of submission unless we are specifically advised to the contrary.

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Aflatoxins	LC/MS	see mycotoxins					
Algal toxins (anatoxin, microcystins, nodularin)	LC/MS	Any	100 mL water	Clean glass bottle	Cold w/ ice packs	7 days	
			5-10 g liver	Whirl-pak or Ziploc® plastic bag			
<u>Anion panel (water)</u> Chloride, fluoride, nitrate, nitrite, phosphate, sulfate	Ion chromatography	Any	100 mL water	Use bottles provided by an extension agent, public health official, or an empty distilled water bottle	Chilled/frozen in insulated container w/ gel packs	1-3 days	
<u>Anion panel (biological fluids)</u> Chloride, nitrate, nitrite, phosphate, sulfate	Ion chromatography	Any	2-5 mL serum 1-2 mL ocular fluid	Clean well-sealed container	Chilled/frozen in insulated container w/ gel packs	1-3 days	

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Anion – single	Ion chromatography	Any	2-5 mL water or serum 1-2 mL ocular fluid	Clean well-sealed container	Chilled/frozen in insulated container w/ gel packs	1-3 days	
<u>Anticoagulant rodenticide screen</u> Brodifacoum, bromadiolone, chlorophacinone, coumafuryl, difenacoum, difethialone, diphacinone, pindone, valone, warfarin	HPLC	Any	20 g liver	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days	Stomach contents may not be useful due to length of time required for clinical signs to develop after a single exposure. Literature reports indicate blood may contain higher toxin concentrations than serum, although both have been used successfully
			Bait				
			2 mL whole blood	EDTA or heparin Vacutainer	Chilled (not frozen) in insulated container w/ gel packs		
Avitrol 4-aminopyridine	GC/MS	Any	Crop contents, suspect bait	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days	
Bromide (single anion)	Ion Chromatography	Any	1 mL serum or drug solution	Take sample in red top vacutainer tube & remove serum from clot Ship in clean vacutainer tube or plastic vial	Chilled/frozen in insulated container w/ gel packs	1-3 days	Therapeutic monitoring for treatment with NaBr or KBr or verification of drug solution concentration
Calcium	ICP/MS	Any	Any	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	1-3 days	This is a useful adjunctive test to confirm ethylene glycol toxicosis.
Carbamate Insecticides	GC/MS w/ LC/MS when needed	Any	Serum, liver, baits, stomach contents	See organic chemical screen	See organic chemical screen	5 days	Screens for carbamate pesticides
Chloride	Ion Chromatography	Any	Serum, ocular fluid or water	Clean well-sealed container	Chilled/frozen in insulated container w/ gel packs	1-3 days	

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Chlorinated hydrocarbon insecticide screen	GC/ECD	Any	5 g liver, bait, fat or brain	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5-7 days	
Cholinesterase, blood	Spectrophotometry	Any	1 mL whole blood	EDTA tube	Chilled	2-5 days	Cholinesterase activity is often used to assess significance of exposure to organophosphate, and less reliably, carbamate pesticides.
Cholinesterase, brain or retina	Spectrophotometry	Any	1 eye (cattle or sheep only) ½ brain (right or left hemisphere). For small rodent or bird, submit entire brain	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	2-5 days	Cholinesterase values may vary among different brain regions; most laboratories use an entire half in order to assure consistency of results. Cholinesterase activity is often used to assess significations of exposure to organophosphate, and less reliably, carbamate pesticides.
Copper	ICP/MS	Any	Liver biopsy 100 mg wet-weight fresh tissue	Whirl-pak or Ziploc® plastic bags or clean well sealed plastic or glass tubes	Chilled/frozen in insulated containers w/ gel packs	3 days	Useful for nutritional assessment of copper status. Copper levels in the blood have variable diagnostic reliability.
			2 mL serum	Red top tube			
			2 mL plasma	Purple or green top tube			
Cyanide	Test strip	Any	1-5 mL whole blood	EDTA tube	Chilled	1-3 days	Cyanide is volatilized by stomach acid, so stomach contents must be frozen as quickly as possible & shipped in an airtight container
			10 g stomach / rumen / crop contents / feed	Air-tight container	Frozen in insulated container with gel packs		
			200 g forage or feed	Plastic bag			
Drug Analysis (Single Drug)	LC/MS	Any	2-3 mL serum	Serum or red top tube	Chilled in insulated container w/ gel packs	5 days	Call lab for drug availability
			10 mL urine	Plastic tube or container			
			10 g liver	Plastic container or Ziploc® bag			

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS	
Drug screen, prepurchase	ELISA GC/MS LC/MS	Equine	5 mL serum	Serum or red top tube	Chilled in insulated container w/ gel packs	5 days for screen If positive, confirmation will take 5 more days	Corticosteroids, tranquilizers, non-steroidal anti-inflammatories	
			10-20 mL, urine	Plastic vial				
<u>Electrolytes panel</u> Magnesium, calcium, chloride, phosphorus, potassium, sodium	ICP/MS & IC	Any	Ocular fluid, serum	Serum or red top tube w/ clot removed from serum samples	Chilled in insulated container w/ gel packs	3 days		
Ethylene glycol (antifreeze) screen	GC/MS	Any	10 g kidney	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	3-5 days	Kidney calcium concentration is a useful confirmation in suspected ethylene glycol toxicosis.	
			10 g bait					
			10 mL urine	Plastic vial				
			2 mL serum	Serum or red top tube				
Fat Content	Solvent extraction	Any	10 g liver	Plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days		
Fumonisin	LC/MS	see mycotoxins						
<u>Ionophore screen</u> Monensin, narasin, salinomycin, lasalocid, laidlomycin, maduramicin	LC/MS	Any	400 g feed or GI contents	Plastic bag	Dry chilled or frozen	5 days		
Iron	ICP/MS	Any	5 g liver	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	3 days		
LC/MS Screen	LC/MS	Any	10 mL urine	Plastic vial	Chilled/frozen in insulated container w/ gel packs	5 days minimum	This screen tests for a wide variety of pesticides and drugs. For specific questions, please call the laboratory.	
			100 g rumen or stomach contents	Whirl-pak or Ziploc® plastic bag				
			10 g liver					
			2-3 mL serum	Serum or red top tube				

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
LC/MS confirmation of GC/MS or LC/MS screens	LC/MS	Any				3-7 days following test request	
Lead (blood)	See mineral panel/screen						
Macrolide endectocides Ivermectin, moxidectin, doramectin, eprinomectin, selamectin	LC/MS	Any	5 g brain, liver, fat, feed	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in container w/ gel packs	5 days	Call laboratory if specimen is not one of those listed.
			50-100 mL milk	Plastic container			
Melamine & cyanuric acid	GC/MS	Any	20 g feed	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in container w/ gel packs	5-7 days	
Mercury	See mineral panel/screen						
<u>Pesticide screen:</u> Metaldehyde	See organic chemical screen						
<u>Mineral panel screen</u> (heavy metals) Arsenic, cadmium, lead, selenium, thalium	ICP/MS	Any	5 g liver &/or kidney	All solid environmental samples: Whirl-pak or Ziploc® plastic bags (no metal)	Water, tissues, serum/plasma, urine: chilled/frozen in insulated container w/ gel packs	3 days	
			5 g stomach / rumen / crop contents				
			5 g suspect material				
			100 mL water	Consult laboratory			
			100 g feed	Whirl-pak or Ziploc® plastic bag			
			2-5 mL whole blood	EDTA vacutainer preferred			

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
<p><u>Mineral panel screen</u> (complete) – tissue, feed, biological fluids, 16 elements, selenium, but no mercury</p> <p>Arsenic, cadmium, calcium, cobalt, copper, iron, lead, magnesium, manganese, molybdenum, phosphorus, potassium, selenium, sodium, thallium, zinc</p>	ICP/MS	Any	100 g feed	All solid environmental samples: Whirl-pak or Ziploc® plastic bag (no metal)	Water, tissues, serum, plasma, urine chilled in insulated container w/ gel packs	3 days	The postmortem specimens of choice are liver & kidney. Liver is the sample of important elements (e.g. Ca, Cu, Zn, Fe, Mg, Mn, Mo), whereas kidney is best for evaluation of toxic elements (e.g. As, Pb, Hg, Tl). It is suggested that both samples be analyzed for optimal evaluation of sudden death or unthriftiness where clinical and/or diagnostic findings are inconclusive. For live animals both serum & whole blood should be submitted. Avoid contact w/ rubber if zinc is of interest. Herd-based testing is available.
			5 g suspect material				
			5 g stomach / rumen / crop contents	Tissue samples: Whirl-pak or Ziploc® plastic bag			
			5 g liver &/or kidney				
			100 mL water	Consult laboratory			
			2-5 mL urine	Plastic container			
			2 mL serum & 2 mL whole blood	Serum/plasma royal blue vacutainer tubes			
<p><u>Mineral panel screen</u> (tissue or biopsy nutritional metal screen)</p> <p>Calcium, cobalt, copper, iron, magnesium, manganese, molybdenum, selenium, zinc</p>	ICP/MS	Any	5-30 g liver or 100 mg wet weight liver biopsy	Whirl-pak or Ziploc® bag	Chilled/frozen in insulated container w/ gel packs	3 days	
<p><u>Mineral panel screen</u> (biological fluid metal screen)</p> <p>Calcium, copper, soluble iron, magnesium, total phosphorus, selenium, sodium, potassium, zinc</p>	ICP/MS	Any	3 mL serum	Take sample in royal blue top vacutainer tube, remove from clot, ship in clean royal blue top tube or plastic vial	Chilled/frozen in insulated container w/ gel packs	3 days	The test provides total phosphorus (Pi) & nonprecipitable iron, rather than the total phosphorus & total iron reported in serum mineral analysis. The test thus eliminates the need for a separate Pi analysis & removes the interference of hemolyzed cells on serum Fe. Herd-based testing is available.

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
<u>Mineral panel</u> (ocular fluid) Calcium, chloride, magnesium, sodium, phosphorus, potassium	ICP/MS	Any	1-2 mL ocular fluid	Plastic vial or red top tube	Chilled/frozen in insulated container w/ gel packs	3 days	
<u>Mineral screen: lead</u> (blood)	AA	Any	1 mL whole blood	Preferred: EDTA tube Second best: heparin tube	Chilled in insulated container w/ gel pack	1-3 days	
			Tissues	Plastic bag or container			
<u>Mineral screen: mercury</u>	AA	Any	1 mL whole blood	Preferred: EDTA tube Second best: heparin tube	Chilled (not frozen) in insulated container w/ gel pack	7 days	
			5 g liver &/or kidney	Tissues in Whirlpak or Ziploc® plastic bag			
			5 g suspect material				
<u>Mineral screen: selenium</u> (blood, milk, serum, tissue)	ICP/MS	Any	2 mL whole blood	Lavender top tube	Chilled/frozen in insulated container w/ gel pack	7 days	Preferred sample is whole blood – send cold or frozen. Serum – spin down & separate before freezing Defined feed – please state the expected level of selenium (<10ppm). Results are intended for clinical or diagnostic use only.
			2 mL serum	Royal blue serum tube or red top serum tube			
			5 g liver	Plastic bag			
			5 mL milk	Plastic container			

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
<u>Mineral screen:</u> single element, not mercury or selenium	ICP/MS AA	Any	100 g feed	Environmental samples: Whirl-pak or Ziploc® plastic bags (no metal)	Water, tissues, serum, plasma, urine chilled in insulated container w/ gel packs	3 days	
			5 g suspect material				
			5 g each liver, kidney	Tissue samples: Whirl-pak or Ziploc® bag			
			5 g stomach / rumen / crop contents				
			100 mL water	Consult laboratory			
			2-5 mL urine	Plastic container			
			2 mL serum & 2 mL whole blood	Serum/plasma – royal blue top vacutainer tubes			
<u>Mineral screen:</u> single element, non-routine	ICP/MS	Any	100 g feed	Environmental samples: Whirl-pak or Ziploc® plastic bags (no metal)	Water, tissues, serum, plasma, urine chilled in insulated container w/ gel packs	3 days	
			5 g suspect material				
			5 g each liver, kidney	Tissue samples: Whirl-pak or Ziploc® bag			
			5 g stomach / rumen / crop contents				
			100 mL water	Consult laboratory			
			2-5 mL urine	Plastic container			
			2 mL serum & 2 mL whole blood	Serum/plasma – royal blue top vacutainer tubes			
<u>Mineral screen:</u> selenium (feed)	ICP/MS	Any	100 g feed (defined or undefined)	Plastic bag	Chilled/frozen in insulated container w/ gel pack	7 days	Defined feed – please state the expected level of selenium (<10ppm). Results are intended for clinical or diagnostic use only.
Monesin	see Ionophore screen						

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
<u>Mycotoxins:</u> Aflatoxins, DON, DAS, fumonisins, ochratoxin, T-2 toxin, zearalenone	LC/MS	Any	1 kg feed, silage	If dry, submit in plastic bag or carton; if high moisture, ship frozen	Insulated container w/ gel packs if high moisture & frozen	7-10 days	Representative sampling is extremely important for all deed analyses as mycotoxins are generally present in isolated spots. Collect feed from 8-12 different locations from a feed lot or storage bin. Mix it thoroughly & submit a kilogram for testing.
Nitrates/Nitrites (feed)	Test Strip	Any	Feed, forage	If dry, submit in plastic bag or carton; if high moisture, ship frozen	Insulated container w/ gel packs if high moisture & frozen	3 days	
Nitrates/Nitrites (serum, ocular fluid)	Ion chromatography	Any	5 mL serum or blood	Royal blue or red top serum tube	Chilled/frozen in insulated container w/ gel packs	3 days	Blood from live animals w/ clinical signs should be examined for methemoglobin.
			Aqueous humor	Plastic tube			
			Whole eye	Whirl-pak or Ziploc® bags			
Nonsteroidal Anti-inflammatory Screen	LC/MS	Any	2 mL serum	Red top tube	Chilled/frozen in insulated container w/ gel packs	5 days	Includes: flunixin, ibuprofen, indomethacin, ketoprofen, meloxicam, mefenamic acid, oxyphenylbutazone, phenylbutazone, salicylic acid, tolmetin, diclofenac
			5 g liver	Whirl-pak bag			
			10 mL urine	Plastic tube			
Organic Chemical Screen	GC/MS	Any	5 mL serum	Red top tube	Chilled or frozen on gel packs	3-5 days	This screen tests for a wide variety of pesticides, drugs, pollutants, & industrial chemicals. For specific questions, please call the laboratory.
			10 mL urine	Plastic vial			
			100 g rumen / stomach / crop contents, liver	Whirl-pak or Ziploc® plastic bag			
			400 g suspect material				
			1 L water	Distilled water bottle			
PCBs (total)	GC/ECD	Any	5 g tissues or biological samples, preferably liver, fat or brain	Plastic or Whirl-pak bag	Chilled/frozen in insulated container w/ gel packs	5 days	Quantitated Aroclor 1260

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
pH	pH meter	Water, biological fluids	100 mL water, GI contents	Clean plastic container	Chilled on gel packs	1 day	
Plant identification	Examination	Any	Whole plant, incl. roots, or rumen / GI contents; if weed in hay, submit representative sample of suspect material only	Plastic bag if fresh	Pressed or chilled if fresh	5-7 days	Contact laboratory before submittal. When submitting a fresh plant, roots should be wrapped in wet newspaper & the entire plant placed in a plastic bag. When chilled, the plant will arrive in good condition.
Selenium	See mineral panel/screen						
Starlicide (3chloro-p-toluidine HCl)	GC/MS	Any	Crop contents, suspect bait	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days	
Strychnine	GC/MS	Any	400 g suspect material	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days	
			100 g rumen, stomach or crop contents				
			100 g liver				
			10 mL urine	Plastic tube			
			3 mL serum	Red top tube			
<u>Tranquilizer Screen</u> Xylazine, medetomidine, ketamine, diazepam, acepromazine, hydromorphone, midazolam, fluphenazine, detomidine	LC/MS	Any	2 mL serum	Red top tube	Chilled/frozen in insulated container w/ gel packs	5-7 days	
		5 g liver	Plastic or Whirl-pak bag				
Vitamin E	HPLC	Any	4 mL serum	Royal blue or red top serum tube	Chilled/frozen in insulated container w/ gel packs	7 days	Protect from light
			5 g liver	Plastic bag			Serum: spin down & separate before freezing

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Vomitoxin (deoxynivalenol DON)	LC/MS	see mycotoxins					
Warfarin	HPLC	see anticoagulant screen					
Zearalenone	LC/MS	see mycotoxins					
Zinc (serum)	ICP/MS	Any	1 mL serum	Take sample in royal blue top vacutainer tube; remove from clot, ship in clean royal blue top tube or plastic vial	Chilled/frozen in insulated container w/ gel packs	1-3 days	Zinc will leach out from other rubber-stopper tubes & invalidate the assay for zinc. Please obtain the sample in a royal blue (trace metals) vacutainer tube (clot type), separate it from the clot, & transfer it to a clean royal blue top tube or plastic vial. Hemolyzed samples should be avoided.
Zinc phosphate	GC/MS	Any	Stomach contents	Whirl-pak or Ziploc® plastic bag	Chilled/frozen in insulated container w/ gel packs	5 days	
			Bait				
			Feed				

Avian Virology

PADLS provides diagnostic virology service for avian, mammalian and aquatic species. In the avian virology laboratory, virus isolation is conducted routinely in cell culture and embryonated chicken eggs (ECE). Assays used to identify viruses following isolation include, but are not limited to, hemagglutination (HA), hemagglutination inhibition (HI), direct/indirect Immunofluorescence assays (FA/IFA), agar gel immunodiffusion (AGID), monoclonal antibody based Dot-ELISA, molecular PCR/real-time polymerase chain assays (PCR/rtPCR) and electron microscopy. Although virus isolation and identification is prearranged on a weekly schedule, emergency designated submissions are processed immediately upon arrival. The cell culture section maintains a variety of cell lines and SPF certified ECE to provide isolation and identification of viruses from a variety of avian sample types. Tissues for virus isolation should be collected as aseptically as possible and placed in sterile tubes or whirlpak bags containing BHI. BHI is available from PADLS laboratories at no cost.

Note: A virus isolation and identification workup typically takes 1 to 3 weeks. Some viral pathogens, however, may require multiple serial passages in ECE or tissue culture to allow for amplification/identification or to confirm a negative result. Hence, some virus isolations may require more than a month to resolve.

Legend: * = preferred specimen, **VI** - virus isolation, **BHI** – brain heart infusion

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Fowl Adenovirus Type 1 (FAV-1)	VI	Chickens, quail, turkey	Cloacal swab, feces, intestines, kidney, liver, lung*, pharynx, tracheal swab	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	7-14 days	Test is set up M-F BHI is available upon request
Avian Influenza	VI	Chickens, ducks, pheasant, quail, turkey, wild birds	Air sacs, cloacal swab, intestine, lung, sinus, trachea*, tracheal swabs	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	7-14 days	Test is set up M-F. Serotyping is sent to NVSL. PCR can be performed by arrangement. See under Molecular Diagnostics. Tracheal swabs are preferred for PCR.
Infectious Bronchitis Virus (IBV)	VI	Chickens	Cecal tonsil, tracheal / cloacal swabs*, trachea, lung, kidney*, oviduct	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	7-21 days	PCR can be performed by arrangement. See under Molecular Diagnostics.
Infectious Bursal Virus Disease (IBDV)	VI	Chickens, turkey	Bursa*, cecal tonsil, spleen, kidney	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	7-14 days	PCR can be performed by arrangement. See under Molecular Diagnostics.
Infectious Laryngotracheitis Virus (ILT)	VI	Chickens, guinea fowl, peafowl, pheasant	Lung, trachea*, tracheal exudates	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	7-14 days	Test is set up M-F
Paramyxovirus Type I: Newcastle Disease	VI	Caged/wild birds, chickens, pigeons, turkey	Air sac, brain, kidney, liver, lung, spleen, tracheal / cloacal swabs*	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	3-14 days	Test is set up M-F

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Poxvirus	VI	Caged/wild birds, chickens, turkey	Nodular lesions, scab w/ underlying epithelium*, upper respiratory tract	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	10-14 days	Test is set up M-F
Reovirus Viral Arthritis	VI	Chicken	Cloaca, feces, intestines, lung, spleen, synovial joint fluids*, tendon sheath	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	3-14 days	Test is set up M-F

Mammalian Virology

The Mammalian Virology Laboratory provides isolation and identification of viruses from a variety of specimens from different species. Virus isolation is conducted in cell culture on a weekly basis. Virus identification is carried out using immunology and/or molecular assays. Electron microscopy (negative staining or thin section) is employed to identify viruses in tissue specimens that are difficult to propagate, fail to replicate in cells, or must be identified by morphology. The virology laboratory does not accept specimens from canine or feline species unless they are considered wildlife species or involved in a zoonotic problem. Tissues for virus isolation should be collected as aseptically as possible. Viral transport media is available at no cost upon request from PADLS laboratories.

Note: For some specimens, virus isolations may require multiple serial cell passages for amplification and expression of viable and measurable virus in a specimen. Hence, certain virus isolations may require 28 days or longer to process.

Legend: * = preferred specimen, **BHI** – brain heart infusion, **FA** – fluorescent antibody, **IHC** – immunohistochemistry, **VI** – virus isolation

TEST/ AGENT	PROCEDURE	SPECIES	TISSUE/ SPECIMEN	CONTAINER	SHIPPING	EST. TURN AROUND	REMARKS
Bovine viral diarrhea (BVD)	IHC	Bovine	Ear notch ¼ - ½ inch. Full thickness ear punch or ear notch ½ inch full thickness.	Red-top tube in formalin	Standard shipping	5-10 days	Samples in formalin should be shipped within a week of collection.
Porcine respiratory & reproductive syndrome: Arterivirus, Lelystad agent	FA VI	Porcine	Lung, serum, tonsil	Leak-proof container w/ BHI	Overnight shipment on ice Do not freeze	FA: 2-3 days VI: 14-21 days	VI is set up on T, PCR is run on F. PCR can be performed by arrangement. See under Molecular Diagnostics.

Proper Sample Collection

General Guidelines

1. Samples should be collected aseptically and placed in sterile plastic bags (e.g., Whirl-pak) or heat sterilized containers. Seal tightly. Do not use chemically disinfected containers, or plastic gloves or sleeves.
2. Label all submissions with the location (tissue) and species of origin. The same bacterial species may be highly significant or a meaningless contaminant, depending on the tissue and/or species from which the sample was obtained. Also, depending on the tissue/species of origin, different culture conditions may be necessary to isolate and identify specific pathogens.
3. If possible, specify the test(s) you want done, and the pathogens you suspect, particularly in the case of specimens with normal bacterial flora (feces, intestinal contents, skin, or oral mucus membranes). If we don't know what you're looking for, we may not inoculate the proper media to find it.
4. It is best to collect other samples before opening the gastrointestinal tract. Tissue samples (lung, liver, spleen, kidney, etc.) should be 5 g or larger to allow surface searing in the laboratory to reduce contaminants. Use screw top containers for fecal samples. Fecal samples should not be submitted in stoppered tubes, as fermentation will dislodge the stoppers.
5. Place each sample in a separate container to prevent cross-contamination. If the intestine is to be cultured, tie off both ends of a segment and place it in a separate container.
6. Except in the case of abortions, please separate samples that are to be examined by different laboratory sections. If a specimen is to be examined by both the virology and bacteriology sections, the specimen should be divided, each piece placed in a separate container, and labeled with the source of the tissue and the desired laboratory service.
7. Fluids for culture (e.g. body cavity fluids, pericardial fluid, joint aspirates) should be submitted in a sealed sterile tube, in as large a volume as is available (up to ~10 ml), since the concentration of organisms may be very low in these samples. Fluids may be submitted in blood culture bottles or Isolator tubes (keeping in mind that Isolator tubes must be processed in the lab within 24 hrs) for highest sensitivity. Fetal fluids (thoracic or peritoneal fluids, or heart blood) to be examined for *Leptospira* sp. by FA test are best submitted in a sealed sterile tube to which 10% buffered formalin is added at a rate of 1.5 ml per 20 ml fluid. Never submit fluid in syringes, which tend to leak in transit and contaminate packaging. Never submit fluids or other specimens for bacteriologic culture in EDTA (purple top) Vacutainer tubes, as EDTA is highly toxic to many bacterial species.
8. Milk samples should be submitted in screw top tubes frozen or placed on ice packs. Less than 1 ml is required. Large volume milk samples submitted frozen may lead to delayed analysis.
9. Specimens for isolation of anaerobic pathogens require special care. Anaerobic bacteria are very susceptible to oxygen exposure. Tissue or fluid specimens are preferred, and if swabs are the only practical sample, the Port-a-Cul system is preferred. Cultures for *Clostridium* sp. in parenchymatous organs ordinarily provide no significant information concerning the cause of death if the samples are taken more than one hour after death.
10. Some specimens, such as porcine nasal swabs for *Bordetella* sp. isolation must be delivered to the laboratory within 12 hours of collection. Fastidious organisms such as *Campylobacter* spp. require special media for transport to the laboratory. Where there is any doubt as to what samples to collect and how to transport them - - CALL THE LABORATORY FIRST!
11. When collecting large numbers of samples (e.g. >30 milk samples for mastitis diagnosis or fecal samples for Johne's disease diagnosis), please call the laboratory for scheduling. This permits the laboratory to have personnel and media available for prompt processing.
12. In general, it is advised to keep specimens cold from the time they are collected until they arrive at the laboratory. Specimens should be shipped in insulated containers with a sufficient number of ice packs to last 48 hours. Specimens arriving in the laboratory in a decomposed state will not be processed, since processing and culture of these tissues lead to meaningless or erroneous results.

13. For cases where bacteremia is suspected and blood culturing is requested, blood culture systems should be inoculated with the proper amount of blood collected aseptically. Single bottle blood culture systems are recommended.
14. Select samples judiciously! See the following list for guidelines, and call the laboratory if questions remain.

Blood Collection

To avoid hemolysis draw blood slowly and as cleanly as possible into the appropriate tube. Avoid repeated jabs with the needle and hematoma formation. Fill container to the appropriate level and avoid vigorous mixing. Do not freeze whole blood, unseparated serum, or unseparated plasma. Keep in mind whether the sample will be a single blood draw or repeated multiple times. For multiple blood draws separated by a period of weeks, a maximum of 1% of the animal's body weight can be removed. If blood must be drawn more frequently than once every two weeks, a total of 0.5% of the animal's body weight can be removed each week with this total volume being spread out over the entire week if needed.

Container	Initial Contents	Processing	Final Contents
Red/Tiger Top	Clotted blood	Let sit to allow clot formation. Centrifuge.	Serum
Lavender (purple) Top	EDTA whole blood	Gently invert to mix. May or may not centrifuge immediately.	EDTA Plasma (centrifuged) EDTA Whole Blood (not centrifuged)
Green Top	Heparin whole blood	Gently invert to mix. May or may not centrifuge immediately.	Heparinized Plasma (centrifuged) Heparinized Whole Blood (not centrifuged)
Blue Top	Citrate whole blood	Gently invert to mix. Centrifuge immediately.	Citrate Plasma

Tissue Samples

Tissue samples should be taken with a disposable or clean, dry device. Each sample should be placed in a separate sterile container. Refer to the table in the testing section for requirements for specific tissues. It might be necessary to take multiple samples of the same organ if there are lesions in multiple locations.

Culture Swabs

Swabbing techniques, and the swabs themselves, need to provide an efficient collection of sample, its subsequent preservation, and ultimately the release of the target cells. Use universal precautions for collecting and handling all specimens. Whenever possible, collect all culture specimens prior to administration of any antimicrobial agents. Avoid contamination with indigenous flora.

Slide Preparation

Using a sterile cotton swab, gently remove some of the exudate from the animal surface. Place the exudate on a glass microscope slide and, pressing firmly, smear the exudate across the slide. Use an open flame source (hand-held flick-style cigarette lighters work well) and gently heat the underside of the slide to attach the exudate to the slide.