

Necropsy Kit: Collection & Shipping Instructions For Veterinary Practice

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Veterinary Practice-Based Post-Mortem Sample Collection Overview

If post-mortem sample collection is conducted in a veterinary practice-based setting, we understand that space and resource constraints exist. These instructions address those constraints by explaining the Study goals for a limited necropsy procedure.

1.) A critical milestone for the Study is to confirm an existing morbidity such as a cancerous tumor.

- Please take samples of any observed tumors and place them in formalin jars. Use a separate formalin jar to collect normal-appearing adjacent tissue.
- Place a small sample of each tumor into an RNALater vial for the biorepository. Use a separate RNALater vial to collect normal-appearing adjacent tumor tissue.
- 2.) The Study further evaluates peri-mortem health status if it's feasible to collect blood, urine and fecal samples.
- If the dog is deceased upon arrival or if sample collection will cause undue distress, do not collect these samples.
- If sample collection is feasible, please follow the instructions starting on page 6.
- 3.) The Study aims to collect normal-appearing "core" tissues from the heart, lungs, spleen, kidney and liver.
- Please take core tissue samples and place them in formalin jars.
- Place a small sample of each core tissue into an RNALater vial for the biorepository.
- Please collect bone marrow if there is evidence of a lymphproliferative disorder.

4.) Complete the Death and Necropsy Questionnaire.

- Log into your account at grls.morrisanimalfoundation.org.
- Find the appropriate dog and hit the Death and Necropsy dropdown to start and complete the Death and Necropsy Questionnaire.
- 5.) Complete medical records will be requested from the Study veterinarian and from other clinicians who provided care to the Study dog.
- Please include your gross findings from the post-mortem exam in the medical record.

Clinical Pathology Sample Collection

Premortem

If blood, urine and fecal collection are feasible please collect a set of samples for Zoetis Reference Laboratories (clinical testing) and Azenta Life Sciences (biorepository) as instructed below. If the dog is deceased upon arrival or if sample collection will cause undue distress, do not collect these samples. Skip ahead to Histopathology Sample collection.



If you received a necropsy kit from the Study, sample collection tubes will be pre-labeled. If you are using tubes from your clinic, follow labeling instructions below.

Blood Collection

Collect 13 ml of blood into the lavender-top EDTA tubes provided by the study or from your clinic.

- Label a 3 ml tube with dog name, ID# (094-xxxxx), and date (MM/DD/YYYY).
- Label a 10 ml tube with dog name, ID # (094-xxxxx), and date (MM/DD/YYYY). If you are using a Study kit, it will have the barcoded whole blood sticker (EDTA 094xxxxxN1099).

Collect 40 ml of blood into red-top tubes (NOT serum separator tubes) provided by the Study or from your clinic.



Serum Separation and Collection

Allow serum tubes (red tops) to clot for at least 45 minutes at room temperature, and then centrifuge per your clinic's normal protocol (we recommend 1100–1300 RCF for 10 minutes).

After centrifugation, remove the Vacutainer cap from each serum tube (red top) and using a pipette (DO NOT POUR) transfer the serum into a 3 mL red-top tube and a 10 mL serum transport tube (a red-top can be used).

- Label a 3 ml tube with dog name, ID# (094-xxxxx), and date (MM/DD/YYYY).
- Label a 10 ml tube with dog name, ID # (094-xxxxx), and date (MM/DD/YYYY). If you are using a Study kit, it will have the barcoded serum sticker (SERUM 094xxxxxN2099).

Please read all instructions before proceeding.

Clinical Pathology Sample Collection (Cont.)

Urine Collection

Collect at least 10 ml of urine via cystocentesis or free catch and place into the specimen transport tubes provided by the study or two red-top tubes from your clinic.

- Label one tube with a dog name, ID# (094-xxxxx), and date (MM/DD/YYYY).
- Label a second 10 mL tube with dog name, ID# (094-xxxxx) and date (MM/DD/YYYY). If you are using a Study kit, it will have the barcoded urine sticker (URINE 094xxxxxN3099).



Fecal Collection

Collect two marble-size samples of feces and transfer each into a separate plastic fecal tube provided by the study or from your clinic.

- Label one vial tube with dog name, ID# (094-xxxxx), and date (MM/DD/YYYY).
- Label the second vial with dog name, ID# (094-xxxxx) and date (MM/DD/YYYY). If you are using a Study kit, it will have the barcoded fecal sticker (FECES 094xxxxxN6099).



Histopathology Sample Collection

For suspected malignant tumors or lesions of interest

- Place a representative tissue sample into a formalin jar, label with the date and "Diseased" tissue type.
 - In the case of multiple metastases, select 2-5 representative lesions to harvest.
- Place a 5 mm cube of diseased tissue into a tube of RNAlater, label with the date and appropriate tissue code, and mark "DISEASED."
- Place a 1 cm cube of normal tissue at least 2 cm away from the tumor/lesion into a separate formalin jar, and label with the date and "Healthy" tissue type.
- Place a 5 mm cube of normal tissue at least 2 cm away from the tumor/lesion into a separate tube of RNAlater, label with the date and appropriate tissue code and mark "HEALTHY."

See Appendix 2 for tissue codes. See histopathology labeling examples on page 11.

In addition to any tumors or lesions, or if no lesions are found, collect samples as described on the next page.

Histopathology Sample Collection (Cont.)

Please collect the five core tissues listed below. Samples may be combined in as few formalin jars as possible, maintaining the ratio of one part sample to 10 parts formalin.

Core Tissues

- **Liver:** 1 cm cube of normal liver.
- **Kidney:** 1 cm cube of normal kidney.
- **Spleen:** 1 cm cube of normal spleen.
- **Heart:** Submit a full thickness 1 cm wide sample of tissue.
- Lung: 1 cm cube of normal lung tissue
- Bone Marrow: Please collect for any lymphproliferative disorder if you have the tools. Recommended locations are the proximal tibia or a rib bone and the recommended tool is a rongeur. For the proximal tibia, scoop out the bone marrow and place into a casssette in formalin. For the rib, cut a section and place the section directly in formalin.

Provide a gross description of these organs in the medical record.

Brain Tissue Collection

Verify with owners before collecting any brain tissue.

Standard Necropsy

If a brain tumor is suspected, and permission has been obtained from the owner, the brain should be removed and placed in a large formalin jar, maintaining the ratio of one part sample to 10 parts formalin. This will be shipped with other Zoetis Reference Laboratories samples for histopathology.

Tissue and Specimen Sample Labeling

Histopathology Labeling Examples

Formalin jars containing a tumor, lesion of interest, or normal tissue adjacent to such lesions should have Zoetis Reference Laboratories Account Number 4761, dog name, Study ID#, tissue type, and date.

Chart: 094-012345		
Smith, Fido		
DATE: <u>12/1/2015</u>		
TISSUE TYPE: Díseased líver		
TPE: <u>piseasea liver</u>		

RNALater vials from the Study kit are pre-labeled. Indicate the tissue type using the numbering scheme in Appendix 2. Indicate whether the tissue is "diseased" or "healthy."

Chart 094-012345	
Smith, Fido	
DATE: 1 <u>2/1/2015</u>	
Tissue type code: <u>८५</u>	
□ DISEASED	

Tissue and Specimen Sample Labeling (Cont.)

Blood, urine and fecal samples will be sent to Zoetis Reference Laboratories for testing, and to the biorepository, Azenta Life Sciences, for storage. Specimen collection tubes from Study kits are pre-labeled and included in "pre-packs" for their destination. If you used collection tubes from your local inventory, label as below.

Acct: 4761

Study 094-012345

Smith, Fido

DATE: 12/1/2015

Zoetis Reference Laboratories (Clinical Testing)

- 3 ml purple top EDTA tube
- 3 ml red top serum transport tube
- Urine transport tube
- Fecal transport tube

	Chart 094-012345	
	Smith, Fido	
	DATE: 12/1/2015	
	Tissue type code: <u>85</u>	
MARK ONE	□ DISEASED □ HEALTHY	
		J

Azenta Life Sciences (Biorepository)

- 10 ml purple top EDTA tube
- 10 ml red top serum transport tube
- Urine transport tube
- Fecal transport tube

Sample Packaging & Shipping



Placing a Service Call for FedEx Shipment(s)

Place a service call to FedEx directly at 800.463.3339. Let them know you have packages for pickup. If you have a Study kit, FedEx Clinical Paks are included with shipping stamps applied. If you don't have a Study kit, the Study will reimburse your shipping fees or you can reach out to 855.447.3647 to request a shipping label.

Zoetis Reference Laboratories Shipment

Part I: Clinical Pathology Samples

• Verify all tubes are properly filled, labeled and dated. Place the 3 ml EDTA tube, 3 mL serum tube, one of the urine tubes and the large fecal tube into a biohazard zip-closure bag.

Tubes for Zoetis Reference Laboratories do not have barcodes.

- The Zoetis Reference Laboratories Manifest form will already be prefilled with Morris Animal Foundation's information as well as the Study dog's information.
- The Manifest for clinical pathology will come prepopulated with the test "GRLS Study Bundle."
- Place the manifest into the zip-closure bag containing the necessary samples and seal the bag.
- Place the zip-closure bag inside the shipping box if available.

If you do not have a Zoetis Reference Laboratories Manifest, please reach out to the Study team at 855.447.3647 or grdogs@caninelifetimehealth.org.





Part II: Formalin containers

- Make sure all formalin jars are tightly sealed.
- Tape the lids to the jars.
- Make sure the labels on all formalin jars have the date, the dog's name, the Study ID# and enclosed tissue type(s).
- Place all formalin containers into a zip-closure bag or bags. Include one absorbing sheet per two jars of formalin. Seal the bag(s).
- Complete the green Zoetis Reference Laboratories pathology form. Include your clinic information as well as the clinical history for the Study Dog. **Failure to include the clinical history may delay results.**
- Place the completed Zoetis Reference Laboratories green pathology form and the preprinted Zoetis References Laboratories Manifest (code: Histopathology Simple Multiple Sites), into a zip-closure bag. Make a copy for your records.
- Place the sealed bag(s) of formalin jars and the sealed documents bag into a large zip-closure bag and seal.

If you do not have a Zoetis Reference Laboratories Manifest and pathology form, please reach out to the Study team at 855.447.3647 or grdogs@caninelifetimehealth.org.

Part III: Packaging

- Place the sealed double bag(s) of formalin jars and documents and the clinical pathology samples into the FedEx Clinical Pak, Necropsy Kit box, or any other appropriate size box. Include a frozen ice pack from your supply.
- Close and seal the FedEx Clinical Pak or box. If using a box, ensure that a UN3373 Biological Substances sticker is placed on the outside.
- FedEx Clinical Paks provided in the Study kit contain a FedEx shipping label to Zoetis Reference Laboratories. If you don't have a necropsy kit, the Study will reimburse your shipping expense or send you a shipping label on request.
- Call FedEx at 800.463.3339 to arrange for a pick up.







Part I: Clinical Pathology Samples

- Fill out the Shipment Inventory Form Golden Retriever Lifetime Study and include the patient information, collection date, collection times, sample shipment inventory and any optional comments.
- Place the following three samples into the Styrofoam shipping container if available:
 - Urine
 - EDTA blood (10 mL purple-top tube)
 - Serum (red-top tube or pre-labeled serum transport tube)
- Close the Styrofoam shipping container and place it inside the zip-closure Biohazard bag.
 (If the Styrofoam shipping container is not available, place all tubes directly into the Biohazard bag.)
- Also place the following items inside the zip-closure Biohazard bag:
 - Fecal sample
 - Absorbent paper towel
 - Shipping inventory form (If using a Study kit, all tubes in the biorepository shipment should have a barcode.)

Part II: RNAlater tubes

- Place RNAlater tubes into the small padded envelope(s). Include one absorbing sheet per envelope and seal the envelope(s).
- Place the sealed padded envelope(s) and completed Submission Form(s) for Tissues in RNAlater into a biohazard zip-closure bag and seal the bag.



Part III: Packaging

- Place the plastic bag containing the biorepository specimens, the biohazard bag(s) containing the RNAlater samples, and a frozen ice pack from your supply into the provided FedEx Clinical Pak pre-addressed to Azenta Life Sciences and seal the envelope by removing the clear seal to expose the adhesive strip.
- FedEx Clinical Paks provided in the Study kit contain a FedEx shipping label to Azenta Life Sciences. If you don't have a necropsy kit, the Study will reimburse your shipping expense. Please contact the Study Team for shipping information or to request a shipping label.
- Call FedEx at 800.463.3339 to arrange for a pickup.

Sample Reporting

Test results from Zoetis Reference Laboratories will be available to the patient's registered Study veterinarian at morrisanimalfoundation.org within 7-10 business days. From the homepage, click on the GRLS icon in the upper right corner, and scroll down to log in. The results are posted under the "Lab Results" dropdown. The results will come as two separate reports - one report for the histopathology and a second for the clinical pathology results.

A Death and Necropsy Questionnaire is requested whenever a Study dog passes, regardless of cause of death.

A hard copy of the questionnaire can be found at the end of this booklet (Appendix 3) for notetaking purposes.

Log on at grls.morrisanimalfoundation.org to complete a Death and Necropsy Questionnaire (DNQ) form for your patient.

You can access the form by selecting the appropriate patient from your portal page and accessing the Death and Necropsy Questionnaire dropdown that appears after a dog is marked as Enrolled Deceased in our system by MAF staff.

If you have any questions, or if you do not have an account, please do not hesitate to email the Study team at grdogs@caninelifetimehealth.org or call toll-free at 855.4GR.DOGS (855.447.3647). We are here to help!

Appendix 1: Presumed Cause of Death

Cancer/Neoplasia		Behavioral	Cardiovascular/Respiratory	
Adrenal Tumor Basal Cell Tumor Bile Duct Tumor Bladder Tumor Brain/Spinal Cord Tumor Eye Tumor Heart Tumor Heart Tumor Hemangiosarcoma Histiocytic Sarcoma Kidney Tumor Leukemia Liver Tumor Lung Tumor Lymphoma Mammary Tumor Mast Cell Tumor	Melanoma Multiple Myeloma Nasal Tumor Osteosarcoma Pancreatic Tumor Perianal Adenocarcinoma Prostate Tumor Soft Tissue Sarcoma Squamous Cell Carcinoma Stomach/Intestinal Tumor Testicular Tumor Thyroid Tumor	Aggression Anxiety Cognitive Dysfunction (Senility)	Arrhythmia Cardiomyopathy Congestive Heart Failure Heartworm Infection Pneumonia Pulmonary Hypertension Pulmonic Stenosis Subaortic Stenosis Valvular Disease	
Derm	atologic	Ear-Nose-Throat	Endocrine	
Atopy Dermatitis Sarcoptic Mange		Epistaxis Hearing Problem Otitis Externa/Media/Interna Upper Respiratory Infection	Addison's Disease (Hypoadrenocorticism) Cushing's Disease (Hyperadrenocorticism) Diabetes Insipidus Diabetes Mellitus Hypercalcemia Hyperparathyroidism Hypoparathyroidism Hypothyroidism Pancreatic Insufficiency	

Appendix 1: Presumed Cause of Death Cont

Eye	Gastrointestinal	Hematologic	
Cataract(s) Corneal Ulcer Glaucoma Keratoconjunctivitis Sicca (KCS) Pigmentary Uveitis Progressive Retinal Atrophy/Degeneration Trauma/Injury Uveitis (Other Than Pigmentary)	Bloat with Torsion (GDV) Bloat without Torsion Chronic Colitis Food Allergy/Sensitivity Gastritis/Gastroenteritis Gastrointestinal Foreign Body Inflammatory Bowel Disease Megaesophagus Pancreatitis	Hemophilia Immune-mediated Hemolytic Anemia Immune-mediated Thrombocytopenia Pancytopenia Von Willebrand Disease	
Infectious	Musculoskeletal	Nervous	
Babesia Ehrlichia Fungal infection (specify) Influenza Leishmania Leptospirosis Lyme disease Rocky Mountain Spotted Fever	Bone Fracture(s) Cruciate Ligament Rupture Elbow Dysplasia Growth Deformity Hip Dysplasia Immune-mediated Polyarthropathy Intervetebral Disc Disease Osteoarthritis Osteochondrosis Dessecans (OCD) Panosteitis Patellar Luxation Rheumatoid Arthritis Spondylosis Trauma/Injury	Cervical Spondylomyelopathy Degenerative Myelopathy Epilepsy Laryngeal Paralysis Limb Paralysis Lumbosacral Stenosis Meningitis Myasthenia Gravis Steroid-responsive Meningitis-arteritis	
Reproductive	Toxicosis	Trauma	
Dystocia Mastitis Prostate Abscess Prostatitis Pyometra	Anticoagulant Rodenticide Chocolate Ethylene Glycol (Antifreeze)	Bite Wounds Hit By Car	
Urinary	Other	Unknown	
Acute Renal Failure Chronic Renal Failure Cystitis Bladder Stones Crystalluria Ectopic Ureter Glomerulonephritis Incontinence Kidney Infection/Pyelonephritis Kidney Stones	Specify		

Appendix 2: Tissue Coding

Code	Description	Additional Indications
70	Other tissue, source	Tissue: Diseased Healthy
71	Adrenal Gland	Left Right Both
72	Bone	None
73	Bone Marrow	None
74	Brain	None
75	Colon	None
76	Duodenum	None
77	Esophagus	None
78	Еуе	Left Right Both
79	Gonads	Left Right Both
80	Heart	None
81	lleocecocolic Junction	None
82	lleum	None
83	Jejunum	None
84	Kidney	Left Right Both
85	Liver	None
86	Lung	Specify Lobe:
87	Lymph Node	Left Right : Axillary Mesenteric Prescapular Mandibular Popliteal Other:
88	Oral Cavity	None
89	Pancreas	None
90	Parathyroid Gland	None
91	Prostate	None
92	Rectum	None
93	Skeletal Muscle	None
94	Skin	None
95	Spinal Cord	None
96	Spleen	None
97	Stomach	None
98	Thyroid	None
99	Urinary Bladder	None

Appendix 3: Death and Necropsy Questionnaire

Use this copy of the questionnaire to take notes regarding the end of life visit. Then log in at grls.morrisanimalfoundation.org to enter these notes into the database.

GENERAL INFORMATION -

-				
Study ID: 094	_			
Dog Sex Status:				
	emale 🗖 Intact Male 🗖 Neuter			
Date of Death:// \	Was the dog euthanized? \Box Ye	s 🗖 No		
Drug Name:				
 Unknown Fatal-Plus Sol Euthasol Other: 	ution 🗖 Beuthanasia-D Specia	I		
Amount administered and uni	ts:	ml 🛛 🗖 Not noted		
Drug concentration and units		mg/ml 🛛 Not noted		
What was the primary organ s	system involved in the cause of	death? Select only one.		
Cardiovascular	Hepatic	Urinary		
Dermatologic	matologic 🛛 Musculoskeletal 🗖 Reprod			
Endocrine	locrine 🗖 Neurologic 🗖 Unknown			
Gastrointestinal	Ear/eye/nose/throat			
Hematopoietic/lymphoid	Respiratory			
What was the primary pathop	hysiologic involved in the caus	e of death? Select only one.		
Cardiovascular	Inflammatory Traumatic			
Congenital	Metabolic	Unknown		
Degenerative	generative 🗆 Neoplastic 🗖 Other:			
Infectious	Toxic			

List any factors contributing to the cause of death: _____

NECROPSY _____

Was a necropsy performed? Yes No Necropsy performed by: Registered study veterinarian Other veterinarian Veterinary Pathologist / Under Supervision of Veterinary Pathologist		
If not the Study Veterinarian, please fill ou	t the following:	
Veterinarian Name:		
Clinic Name:		
Clinic Email:		
Clinic Address:		
Clinic Phone Number:	Clinic Fax Number:	
Date of Necropsy:// Approximate number of whole hours between time of death and time of necropsy: hrs □ Unknown		
Do you have any gross findings? □ Yes	⊐ No	
If yes, please complete the Gross Necropsy Fin	ndings section. Otherwise, skip ahead to page 28.	
GROSS NECROPSY FINDINGS		
Skin Normal Abnormal Not Evaluated 	Comments if Abnormal:	
Subcutaneous Fat - Quality Normal Abnormal Not Evaluated 	Comments if Abnormal:	
Left Eye - Dissected Evaluation Normal Abnormal Not Evaluated 	Comments if Abnormal:	

Right Eye - Dissected Evaluation

Normal Abnormal Not Evaluated Comments if Abnormal:

Thyroid Gland

□ Normal □ Abnormal □ Not Evaluated Comments if Abnormal:_____

Parathyroid Glands:

Normal Abnormal Not Evaluated Comments if Abnormal:

Esophagus:

Normal Abnormal Not Evaluated Comments if Abnormal:

Thoracic Cavity - In Situ:

Normal Abnormal Not Evaluated Comments if Abnormal:

Heart:

Discrimination Not Evaluated Comments if Abnormal:

Pericardial fluid present?:	Yes 🗖 No	Not evaluated	
Estimated volume of fluid in milliliters:mL			
Color of pericardial fluid:			
Black	Clear	Yellow	
Brown	Red	🗆 Other:	
Clarity of pericardial fluid: 🗖 Clear 🗖 Opaque 🗖 Other			
Lungs:			
Normal Abnormal Not Evaluated Comments if Abnormal:			
Abdominal Cavity - In Situ:			

Normal Abnormal Not Evaluated Comments if Abnormal:

Abdominal fluid present? Normal Abnormal Not evaluated Estimated volume of fluid in milliliters:mL Color of pericardial fluid:			
Black		Clear	Yellow
Brown		Red	Other:
	pericardial flu Bile DUrine		ther:
Duodenun	ı		
Normal	Abnormal	Not Evaluated	Comments if Abnormal:
Pancreas • Normal	Abnormal		Comments if Abnormal:
Jejunum • Normal	- Abnormal	Not Evaluated	Comments if Abnormal:
lleum • Normal	Abnormal	Not Evaluated	Comments if Abnormal:
Cecum Dormal	Description Abnormal	Not Evaluated	Comments if Abnormal:
Colon Dormal	- Abnormal	Not Evaluated	Comments if Abnormal:

Liver

□ Normal □ Abnormal □ Not Evaluated Comments if Abnormal:

Spleen

Normal Abnormal Not Evaluated Comments if Abnormal:

Right Kidney

Normal Abnormal Not Evaluated Comments if Abnormal:

Right Adrenal Gland

Normal Abnormal NotEvaluated Comments if Abnormal:

Left Kidney

Normal Abnormal Not Evaluated Comments if Abnormal:

Left Adrenal Gland

Normal Abnormal Not Evaluated Comments if Abnormal:

Urinary System (Bladder, Prepuce, Vulva, etc.)

Normal Abnormal Not Evaluated Comments if Abnormal:

Reproductive System

□ Normal □ Abnormal □ Not Evaluated Comments if Abnormal:____

Skeletal Muscles

□ Normal □ Abnormal □ Not Evaluated Comments if Abnormal:____

Bones Identify specific bones in comments.

Normal Abnormal Not Evaluated Comments if Abnormal:

Bone Marrow

□ Normal □ Abnormal □ Not Evaluated Comments if Abnormal:

Stomach

Normal Abnormal Not Evaluated Comments if Abnormal:

Synovial Fluid

Normal Abnormal Not Evaluated Comments if Abnormal:

Lymph Nodes

Normal Abnormal Not Evaluated Comments if Abnormal:

Brain

Normal Abnormal Not Evaluated Comments if Abnormal:

Spinal Cord

Normal Abnormal Not Evaluated Comments if Abnormal:

Nervous System Other Lesions

Normal Abnormal Not Evaluated Comments if Abnormal:

Tumors, Masses, or Other Lesions of Interest

Normal Abnormal Not Evaluated Comments if Abnormal:

Additional General Gross Necropsy Findings

Comments:

SUPPLEMENTAL INFORMATION —

Do you have photographs?

Yes
No

Do you have radiographs?

Yes
No

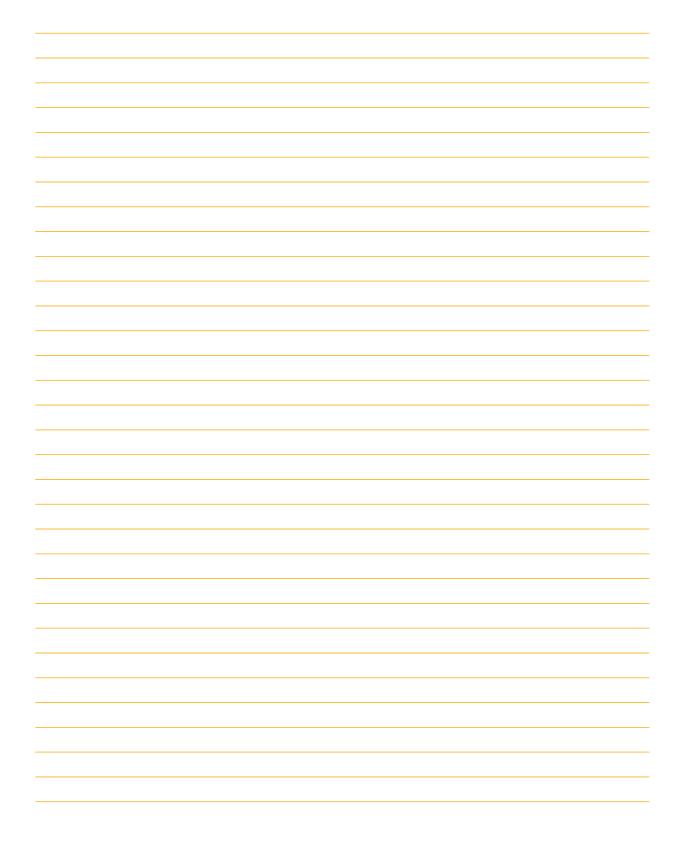
Do you have clinical pathology results?

Yes
No

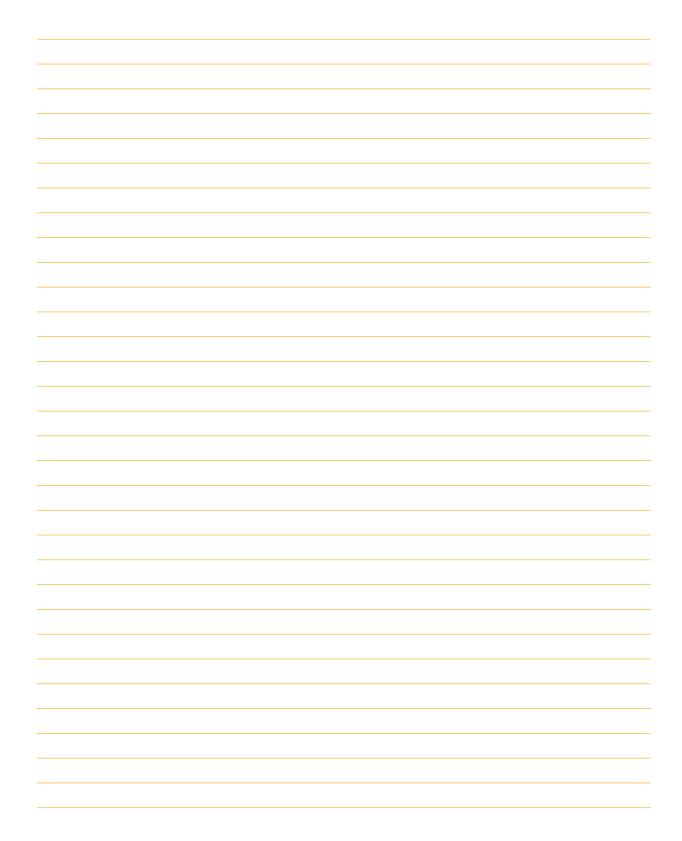
Additional Remarks/Comments:: _____

Thank you again for supporting the Golden Retriever Lifetime Study.

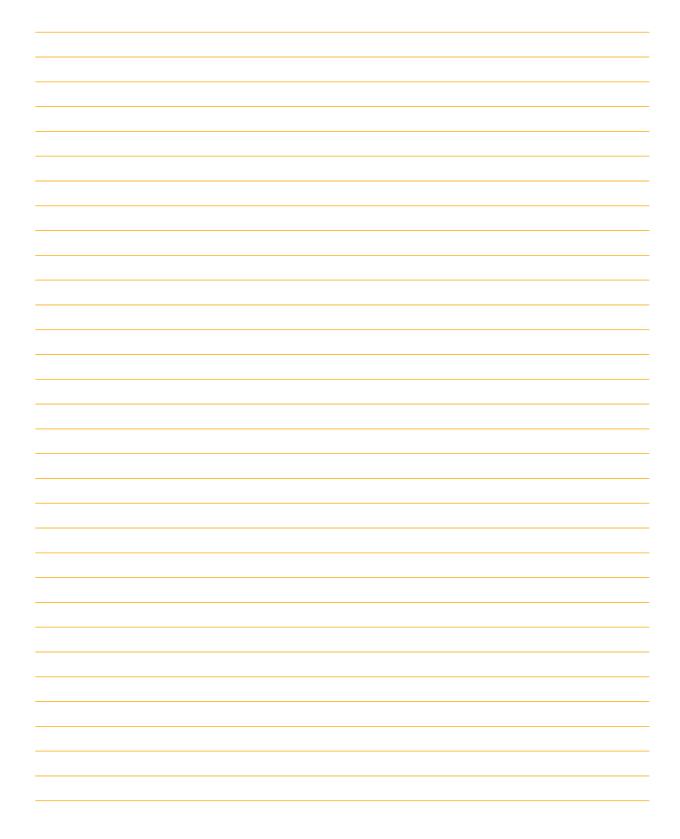
Notes



Notes



Notes



About Morris Animal Foundation

Morris Animal Foundation is a nonprofit organization that invests in science to advance animal health. The foundation is a global leader in funding scientific studies for companion animals, horses and wildlife. Since its founding in 1948, Morris Animal Foundation has invested in studies that have led to significant breakthroughs in diagnostics, treatments, preventions and cures to benefit animals worldwide. Learn more at morrisanimalfoundation.org.



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GOLDEN CHAMPIONS

Mars Veterinary

Please contact the Study team at 855.447.3647 for advice or assistance with any sample submission or to request replacement or additional supplies.