

SURPRISES AT NECROPSY: MAXIMIZING YOUR RETURN FROM POSTMORTEM SAMPLES

David N. Phalen
Avian Reptile and Exotic Bird Hospital
Wildlife Health and Conservation Centre
Faculty of Veterinary Science
University of Sydney, Camden, NSW 2570

INTRODUCTION

In the experience of the author, bird owners are more likely to have a bird that dies necropsied and its tissues examined microscopically than owners of other pet animals. This is probably because most bird owners have other birds and want to know if they are dealing with an infectious disease that might impact the other birds, or to determine if their husbandry practices might have contributed to the bird's death.

When a bird is necropsied, all parties involved, the client, the client's veterinarian and the pathologist, all want the same thing, a diagnosis. There is nothing more unsettling to all parties than to end up with a report where no microscopic lesions are found and the cause of death is not determined. This is inevitable in some cases, because not all causes of death leave behind gross or microscopic lesions. On the other hand, the author and many other pathologists know that this diagnosis is often made because an incomplete set of tissues was submitted for examination or the tissues were inadequately preserved. Other diagnoses are missed because only formalin-fixed tissues are saved, precluding the use of modern molecular diagnostic techniques and even traditional diagnostic techniques such as culture.

The next section of this manuscript is a necropsy protocol that I follow. This protocol has allowed me to consistently examine a necropsy specimen, collecting the appropriate tissues for histopathology, while still collecting useful samples for microbiology and molecular diagnostics. At the end of this paper, examples of diagnostic surprises that have resulted from quality and comprehensive sample collection, are presented. The majority of the cases ($n=205$) reviewed for this report were submitted to the Diagnostic Service of the Avian Reptile and Exotic Pet Hospital in the past three years.

NECROPSY PROTOCOL

Quality of the specimen. Birds decompose rapidly and autolysis severely limits the information that can be obtained with cultures and histopathology. To minimise autolysis, dead birds should be necropsied immediately after death, or chilled and refrigerated until brought in by the client or forwarded to a pathologist. To effectively chill a dead bird, it should be immediately soaked in cold soapy water, wrapped in a wet paper towel, placed in a sealed plastic bag with all air removed, and placed in the coldest part of the refrigerator. Packing the bird in ice after it is wetted is also a very effective way of chilling a bird. When thoroughly chilled, it can be sent on ice overnight to a pathologist. Otherwise the necropsy can be performed by the veterinarian later.

Many samples that reach the pathology laboratory are not of diagnostic quality. One of the most common reasons is a lack of refrigeration of fresh specimens. In most parts of the world, summers are hot. Therefore, a single ice pack will rarely be sufficient to keep even a small bird cold. Although it costs more to ship, be generous with ice packs and send the samples in an insulated container. A

second common problem is when whole birds or large pieces of birds are sent in an insufficient volume of formalin. The more rapidly tissues are fixed the less autolysis. The rule of thumb is that there should be 20 times as much formalin as there is tissue. Therefore, it is best to collect small pieces of tissue which are very thin (maximally 1 cm x 1 cm x 0.5 cm). Stuffing a whole finch into a pill bottle and filling the remainder with formalin will not endear you to your pathologist.

Even when the appropriate sized tissue is collected and the appropriate volume of formalin used, tissues may still not fix. This is because most tissues will settle to the bottom of the specimen container and pile on top of each other. The best practice is to put the specimen container on a shaker table for 24 hours. This is rarely practical for most veterinarians, but swirling the specimen container a few times during the day after the samples are collected is possible and will help them to fix more quickly. Formalin is heavy and is costly to ship, but there are ways around this problem. If you let the tissues fix in your office for 2-3 days, then the tissues can be sent in only the small amount of formalin necessary to keep them moist.

Most pathologists charge by the slide, so there is a tendency to reduce costs by only sending in tissues that appear diseased grossly, or to send a subset of tissues. This is the single most important reason that diagnoses are missed. Most birds have diseases that are multisystemic, but diagnostic lesions may only be found in one organ. Forwarding a complete set of tissues is the only way to assure that if a microscopic lesion is present in the bird, it will be found.

Examination of the Body. Before pinning out the body, look it over carefully. Examine the eyes, nares, oral cavity, vent, and integument. Fractures of the long bones can often be identified at this point. Weigh the bird, and if it is not already wet, soak it in soapy water and let it drain. Cut its tail and wing feathers at least in half, and pluck out all of its ventral contour feathers from the neck to the vent. This will significantly reduce feather contamination of the internal organs. Pin the bird on a dissecting table in dorsal recumbency so the wings and legs are fully extended. Tattoos in the right or left patagium should be noted. This is also a good time to scan the bird for a microchip.

Integument. Collect representative sections of normal and abnormal feathered skin and non-feathered skin for histopathology. If there are masses or areas of markedly abnormal skin, save and freeze a portion. Impression smears of abnormal skin and crushed feather follicles may also be useful in identifying bacterial and fungal infections. Feather pulp or cysts may be cultured. Never forget to look at the uropygial gland, and if there is any gross abnormality, submit it for histopathology.

Make a U-shaped incision from the point of the shoulder (cranial aspect of the coracoid) to just cranial to the vent through the skin and pull the skin cranially. Care must be taken not to cut too deeply over the abdomen, as the abdominal musculature is very thin. Examine the subcutaneous tissue and underlying musculature for haemorrhage, evidence of previous treatment, and evaluate the amount of body fat. Extend the right incision cranially through the skin of the neck to the ramus of the mandible. With strong shears cut through the right mandible opening the oral cavity and exposing the trachea and oesophagus.

Opening the Coelomic Cavity. At this point the pectoral muscle mass can be evaluated. With the exception of ratite birds, the sternum will have a prominent keel on which the pectoral muscles attach. In cross section, convex bulging of these muscles is normal for a healthy bird, concavity of the muscle mass indicates muscle wasting. Injections are commonly given in the pectoral muscles during therapy. Oedema, swelling, and necrosis are common findings at the sight of these injections. The pelvic musculature is then incised. At this point, the duodenal loop overlying the remainder of the

intestines should be exposed. With the exception of some ducks, the normal healthy liver should not extend beyond the edge of the sternum. The gizzard will either not be apparent, or will extend only slightly under the left edge of the sternum. To remove the sternum, the pectoral muscles are cut on both sides from just ventral to the shoulder parallel with the table caudally back over the ribs. The ribs, coracoid, and clavicle are then cut on both sides with heavy shears while lifting up the sternum. The sternum, with the attached pectoral muscles, is completely separated from the body. As the sternum is being removed, air sac and pericardial lesions may be identified. Collecting fluids and samples for microbiology may be necessary before the sternum is completely removed. With removal of the sternum, the internal viscera from the oesophagus and crop to the cloaca are exposed.

Collecting Samples for Microbiology and Molecular Testing. For microbiological samples to be meaningful, they must be collected immediately after the sternum is removed. Heart blood can be collected for culture by searing the right atrium with a hot spatula and using a needle and syringe to aspirate blood. Liver and spleen cultures can be obtained by searing the organ's surface, and then using the spatula to cut through the capsule in the seared regions. Culturettes or a wire loop can be used to collect a sample for culturing. Abdominal, perihepatic, and pericardial fluids, granulomas, air sacs, and sinuses can also be cultured in a similar manner. Swabs of diseased organs or small pieces of fresh tissue can be saved (frozen) for molecular diagnostic testing. Later, when opening the bowel, samples can be collected for *Salmonella* spp isolation.

Cardiovascular System. One indication of the nutritional status of the bird is fat at the base of the heart. The absence of fat, or serous atrophy of fat, indicates inadequate caloric intake. Unlike the dog or cat heart, the axis of the bird's heart axis parallels that of the body. The aorta of the bird goes to the right (mammals aortas go to the left), and immediately branches into two brachiocephalic trunks. The relatively small carotid arteries branch off the brachiocephalic trunks and extend cranially into the thoracic inlet. The thyroids lie immediately lateral to the carotids in the thoracic inlet. A single parathyroid is located at the caudal pole of each thyroid. It may not be visible in the normal bird. Parathyroids in excess of 1 mm in diameter suggest that the bird was on a calcium deficient diet. In the very young bird, multiple lobes of the thymus will be distributed along the length of the cervical carotid arteries.

Remove the heart and as much of the great vessels as possible. Open the heart by following the blood flow. The right atrium and ventricle are separated by the right atrioventricular valve. This is muscular and not tendinous. The remaining heart valves are identical to those of mammals. Open the lumens of the aorta and brachiocephalic trunks. This is a common location for atherosclerosis.

With the increasing use of ultrasound as a diagnostic tool and our improved knowledge of heart dimensions for various species of birds, a different sectioning of the heart may be indicated. If heart failure is suspected or the heart is misshapen, then transverse sections of the heart may be more helpful in detecting chamber dilation, wall hypertrophy, or wall thinning.

Often only a portion of the heart is submitted for necropsy. If at all possible, submit the entire heart. Sections through the valves, great vessels, and coronary arteries will reveal lesions not always present in the heart walls and the septae.

Digestive System. Before removing any of the digestive system, rotate the proventriculus to the right and visualise, collect, and save the spleen. The normal cockatiel's spleen is seldom more than 4 mm in its widest dimension, while an Amazon parrot's spleen should not be more than 10 mm wide. Spleens from psittacine birds will be round to slightly flattened. A passerine spleen is generally

cylindrical. Splenic enlargement is an important sign of systemic disease and is a common finding in both viral and bacterial infections. Save spleen for histopathology, but also save some frozen. Examine the tongue and its base, and open the entire oesophagus. Most cage bird species will have a crop. The normal crop will be nearly transparent when fully distended. Grasp the oesophagus at the thoracic inlet and sever it from the crop. Carefully dissect it from the lungs, and free by dissection the proventriculus, ventriculus, and intestines. When dissecting the cloaca, examine the dorsal surface for the bursa (young bird), or bursal remnants.

The proventriculus is a thick glandular structure. The ventriculus (gizzard) has a thick muscular wall with a koilin lining. The koilin will often be stained green if the bird has not been eating. Do not remove the koilin. Section the intact ventriculus, including koilin and muscle wall, for histopathology. The duodenum forms a loop. The pancreas has three lobes in most birds, one within the duodenal loop, one lateral to the ascending duodenum, and a smaller transverse lobe adjacent to the ventriculus and the spleen. The pancreas should be light tan-pink in colour. Psittacine birds do not have gall bladders, most passerine birds and many other species do. The jejunum is similar to that of mammals. Caeca are present in many bird species, but not psittacine birds. When they are present, they may be vestigial or extensively developed. The final portion of the digestive system, the colorectum is relatively short in most cage birds. The cloaca, the combined outlet of the digestive, urinary and reproductive systems, should be opened and examined carefully for evidence of erosion or papillomas.

The duodenum should be scraped with a scalpel blade and wet mounts examined for *Giardia* spp and *Spironucleus* spp. *Giardia* spp die soon after the bird does, so will rarely be found in birds which have been dead for more than a few hours. Depending on the species of bird, trichomonads and associated lesions may be found in the mouth, the cervical oesophagus, crop or thoracic oesophagus. Therefore, careful examination of the oesophagus and crop are necessary if trichomonad infections are to be identified. These organisms also die very quickly and are best diagnosed in wet preparations of lesions soon after the bird has died. Opening the entire length of the intestinal tract is indicated to look for worms and mucosal changes. Opened segments of the intestines fix best. Handle them gently so as not to disrupt their architecture.

Sections of bowel can be submitted for culture or frozen and cultured later if indicated. Save diseased bowel for culture or lower bowel if a *Salmonella* spp infection is suspected.

Urogenital System. The paired kidneys lie in the retroperitoneal synsacral fossa. Each elongate kidney contains two to three divisions. All psittacine birds have three divisions. The kidney is brown and the surface slightly irregular. The roots of the sacral nerves pass through the kidney on its dorsal surface. On the ventral surface of the cranial pole of the kidneys are the paired yellow to pink, flattened, triangular adrenals. In the male, there are paired testicles overlying the adrenals. The testicles have a smooth surface with branching capsular arteries. They are round to bean-shaped and are yellow to yellow-grey. White cockatoos and some other parrot species will have pigmented testicles. Most species have a single ovary that is located at the cranial pole of the left kidney. The juvenile ovary is flattened with a cobblestone surface and is triangular with the base being closest to the heart. An active ovary will have follicles in various stages of development. The oviduct lies on top of the left kidney. When not reproductively active, it is very thin and barely discernable. In hens which are about to lay, the oviduct is long and tortuous, and is as wide or wider in diameter than the intestine. At this stage, the oviduct takes up a considerable part of the abdomen.

Remove the kidney, adrenal, and gonad, by holding the abdominal aorta and dissecting the kidney

free from the synsacrum.

Respiratory System. The nasal passages may be examined by cutting through the skull at the base of the cere. When the mandibles are transected at the base of the eye, the infraorbital sinuses are exposed. Open the trachea from the glottis to the tracheal bifurcation. Closely examine the syrinx and the main stem bronchi for keratin plaques and *Aspergillus* spp lesions. Each lung is tightly adherent to the overlying ribs, and may be dissected free by gently teasing with a blunt object, starting laterally along its thin edge and working medially. The avian lung is orange to pink and spongy, but not expansible. The five paired coelomic air sacs are very thin and best viewed as the sternum is being removed. In most cage birds, the air sacs are transparent.

Musculo-skeletal System. Test the strength of a long bone by breaking it. Crack the tibiotarsus and place it in formalin. This can be saved and demineralised if necessary to evaluate bone marrow. Examine the hock, stifle, and hip joints. Skeletal muscle may contain a number of important lesions that will assist in a diagnosis. Muscle lesions are not always uniform, so submitting sections from multiple skeletal muscles is best.

Nervous System. To remove the brain, separate the spinal cord from the skull at the atlanto-occipital joint. With dull scissors or shears, carefully remove the bone from the roof of the calvarium. When the brain is exposed, dissect its ventral surface with the head held upside down. Allow gravity pull the brain away from the floor of the calvaria. With the exception of the folia of the cerebellum, the avian brain does not have gyri and sulci. Ventrolaterally, the optic lobes are prominent. The pituitary is immediately ventral to the optic chiasm. Examine the eyes prior to dissection for evidence of corneal or anterior chamber disease. The eyes can be dissected free, or fixed *in situ*. By submitting the remainder of the skull in formalin, a wide range of tissues can be examined by the pathologist that would not normally be available.

The spinal cord can be a very important tissue to examine. It is best to dissect the entire vertebral column free of the rest of the body and to section it into 2 cm pieces so the cord will fix. The cord can then be dissected free of the surrounding bone and submitted for histopathology or submitted intact and demineralised before sectioning.

Additional Specimens. Impression smears of the liver and spleen, or any enlarged organ, or diseased surface can be collected. These may be stained by the practitioner, or forwarded, along with the formalin-fixed tissues, to the pathologist. Blood smears made from heart blood can be examined for the presence of bacteria when a septicaemia is suspected, e.g., following a cat bite. Blood smears can also be used to determine if the bird had an elevated white blood cell count at the time of death. Frozen tissues can be banked, or sent on dry ice, if a viral disease is suspected. Similarly, liver, kidney, lung, and gut contents should be saved if an intoxication is suspected.

Archived Tissues. After you finish your necropsy, much of the bird will remain. Most will either be in the formalin pot or the freezer. If you chose to submit only some of the tissues you have collected, keep the remainder of them until you get a diagnosis, so that if necessary, you may submit more.

Newly Hatched Chicks and Embryos. It is generally best to submit an embryo or recently hatched chick undissected in formalin to the pathologist. The coelomic cavity can be opened aseptically and the yolk sac or heart blood cultured. Generally opening the coelom is indicated as this will facilitate the fixation process. The fixation process in the intestines can be facilitated by perfusing them with formalin.

UNEXPECTED FINDINGS IN UNCOMMONLY SUBMITTED TISSUES

In the end, the ability to diagnose a disease process in a bird depends on examining the tissues that are impacted by the disease. The fewer tissues submitted the less likely that a disease diagnosis will be made. The tissues I find that are least often sent as diagnostic specimens are the head, skin, muscle, bone marrow, endocrine tissues, central nervous and upper respiratory systems, representative portions of the heart and its vessels, and some portions of the digestive tract. Lesions in these tissues can vary from common to rare but without them a diagnosis may not be made.

Another issue that may not be considered by a practitioner is that clinically relevant material that can be gained from a necropsy. In addition to determining the cause of death, other clinically relevant information can be derived from microscopic examination of the tissues. In many cases there is disease present in birds that the practitioner may not recognize. Recognizing the extent of the lesions associated with various diseases can help the practitioner to formulate improved treatment plans when presented with birds with similar diseases in the future.

Head. The head contains a remarkable wealth of tissue types including skin, muscle, bone, respiratory epithelium, oral mucosa, the infraorbital sinuses, salivary glands, nerves, and the many structures of the eye. I have found examination of these tissues to be particularly helpful in pigeons, finches, and chickens.

Lesions caused by viral, bacterial, fungal, parasitic and nutritional diseases are common found in the head. Lesions caused by poxviruses generally occur around the eye lids and commissures of the mouth. They can also occur on the oral mucosa. In my experience, pigeon herpesvirus lesions are often confined to the oral and respiratory mucosae of the head. These ulcerated lesions are generally covered by bacteria. They must be very painful to the bird and would prevent them from eating or drinking. Pigeon circovirus inclusions are infrequently seen in cells within lesions of the oral and upper respiratory epithelium but if the bursa is not submitted, this may be the only place where inclusions are found..

Upper respiratory infections caused by *Chlamydophila psittaci*, mycoplasmas, and other bacteria are common in pigeons, some canaries and finches, cockatiels and chickens. The lesions seen in the infraorbital sinuses and respiratory passages, especially when combined with those found in the trachea, as is the case in poultry, provide valuable clues in determining which pathogen is causing disease.

Trichomoniasis can result in the ulceration of the oral cavity and the build up of caseous plaques. In some species the organisms penetrate the underlying tissues causing coagulation necrosis. These lesions are typically profoundly heterophilic.

A surprising number of lesions can be found in salivary glands. Squamous metaplasia of the salivary glands of the oral mucosa and in advanced cases the respiratory epithelium of the nasal passages, may be the only lesion that can confirm vitamin A deficiency. Spiral bacteria are found in the salivary glands of some cockatiels and have been associated with disease. A lymphoplasmacytic inflammation of salivary glands is a regular finding in finches and canaries. The cause of these lesions is yet to be determined.

The skin on the face and the eyelids may be the only skin that is submitted with a necropsy. Bacterial and fungal infections of the cere are relatively common in pigeons and may not be obvious in a gross necropsy specimen. *Cnemidocoptes* spp infections of the cere are generally obvious at post mortem

specimen, but are sometimes only found histologically.

Eyes are very rarely submitted for histology. Eyelid and conjunctival sections can be very valuable diagnostic specimens. Conjunctivitis is a common component of *Mycoplasma* spp and *Chlamydophila* spp infections and these may be the only lesions related to these infections in the bird. Mycobacteria and fungi can also localise in the eye lid causing granulomas and *Encephalitozoon hellem* infections cause a chronic active conjunctivitis and keratitis. Examination of the globes in some cases reveals disease, such as uveitis, that would not have been recognised in the live bird. Diagnosis of lymphoid leucosis was made in a chicken because of characteristic ocular lesions.

Trachea and Syrinx. Characteristic tracheal lesions are particularly common in chickens with *Mycoplasma gallisepticum* infection and can be present in chickens with *Avibacterium* (previous *Haemophilus*) *paragallinarum*, other bacterial infections of the upper respiratory tract and infectious with Infectious Laryngotrachetis Virus. *Mycoplasma* lesions can also extend into the trachea in pigeons.

Although mite infections may cause lesions in the lung in finches and other birds, many times they are only found in the trachea. *Aspergillus* spp infections may also only be found within the syringeal lumen. If the head is not submitted, squamous metaplasia of the respiratory epithelium of the trachea may be the only indication of vitamin A deficiency.

It has been the author's observation, that voice changes are one of the signs that may be present in lorikeets with clenched claw syndrome. In the one case that I have had syrinx submitted, a significant myositis was present in its muscles.

Digestive Tract. The digestive track extends from the oral cavity to the cloaca and all sections should be collected. The crop is an important tissue to submit as bacterial, fungal and trichomonad ingluvitis is common, especially in pigeons. Columbid and less commonly Psittacid herpesvirus and circoviruses will also cause lesions in the crop. The cervical and thoracic oesophagus are rarely submitted tissues, and can also have significant lesions. In budgerigars and finches, trichomonad lesions are often localised to a narrow section of the thoracic oesophagus. The author has recognised a colonisation of the cervical oesophagus by spiral bacteria in a Gouldian finch, a lesion that has not been previously described. Bacterial infections and neoplasms can also occur along the entire length of the oesophagus.

The entire stomach should be submitted and representative portions of the proventriculus, isthmus and ventriculus sectioned. Cryptosporidiosis is a very common lesion of Gouldian finches. The organisms, however, may not be uniformly distributed within the proventriculus and may only be found near the isthmus. Likewise, *Macrorhabdus ornithogaster* may only be present in a narrow zone of the isthmus, but in more advanced cases may extend into the proventriculus and into the koilin of the ventriculus. Other yeast infections may penetrate the koilin and in severe causes cause ventricular ulceration. Other yeasts can also be found colonising the lumen of the proventriculus, but this is uncommon. Invasive bacterial infections of the ventriculus also occur. Metastatic mineralization secondary to high calcium diets can occur in the mucosa of the proventriculus and the koilin layer of the ventriculus. They are often found in one and not the other.

Neoplasia can also occur in either the proventriculus and ventriculus. The mucosa of the proventriculus is a common target for the tumours caused by Marek's disease. In the cases reviewed for this study, ventricular papillomas were found in the ventriculus of one budgerigar. Similar lesions

of the oral and cloacal mucosae are caused by some genotypes of Psittacid herpesvirus-1.

Lymphoplasmacytic proventriculitis was identified in a number of finches. The cause of this lesion was not determined. The ventriculus and to a slightly lesser extent the proventriculus are the organs where Proventricular Dilatation Disease lesions are most commonly found.

The microenvironment created by the different levels of the intestines generally means that intestinal lesions are localised to one section of the intestine and are not diffuse. This means that either the entire intestinal tract or representative sections must be examined if intestinal lesions are expected to be found. Viruses and bacteria, including *Salmonella* spp, *Clostridium* spp, *Spironucleus* spp and *Giardia* spp. can all cause intestinal disease, but lesions are often confined to limited segments of the intestine. Most nematodes and cestodes are only found in certain sections of the intestine.

The cloaca is an often neglected tissue that is uncommonly submitted for histopathology. Lesions here are infrequent, but are often significant. Ulcerative cloacitis is a common lesion in parrots and other species with salmonellosis. The cloaca may also be the only location where mucosal papillomas are found. In ostriches, the bursal tissue is diffusely distributed across the dorsal wall of the cloaca. Ulcerative lesions of the cloaca found in ostrich chicks have been associated with an infectious bursal disease-like virus.

In this study some unusual intestinal lesions were identified. Villous clubbing was identified in the duodenum of a pigeon suggesting that it had an enteric virus. Villous clubbing, blunting, and fusion were identified in galahs and corellas with *Spironucleus* spp infections. These lesions were also found in birds with similar signs that were not infected with *Spironucleus* spp, also suggesting a possible viral origin of them. In two lorikeets with clenched-claw syndrome papillomatous changes were found in the jejunum raising the question as to whether they could be caused by a herpesvirus.

Cardiovascular System. Diseases similar to those found in other species can also be found in the hearts of birds, so veins, arteries, heart valves, and myocardium should be examined. Atherosclerosis is most commonly found in the aorta, brachiocephalic trunks, and the carotid arteries. Amyloidosis is most often found in the liver, but in some cases can be confined to vessels. Thrombi can be found in any vessel, but are particularly common in pulmonary arteries. Hyperplasia of the smooth muscle cells in the media of the arteries and veins of the lungs can be an indication of heart failure.

Bacterial infections of the heart can be confined to the pericardium, pericardial space and the epicardium or can result in myocardial lesions. Bacterial endocarditis is relatively rare, but when it does occur is found on the left atrioventricular valve.

Myocardial degeneration and lymphoplasmacytic myocarditis were significant but uncommon lesions found in this review. Myocardial degeneration was found in a flock of pigeons and was thought to be due to exposure to a toxin or as the result of a vitamin E or selenium deficiency. Lymphoplasmacytic myocarditis was seen in a number of species including finches and pigeons, but its cause was not identified.

Heart dilation and hypertrophy are generally recognised during the gross post mortem. In one of the hearts examined during this study, however, myocyte hypertrophy identified microscopically was the only evidence that the bird had a hypertrophic cardiomyopathy.

Endocrine Tissues. Endocrine tissue is uncommonly submitted for histopathological examination, but may be the only tissue in the body containing diagnostic lesions. In this study, goitre was identified in a budgerigar and a pigeon. Thyroiditis was not found, but this lesion occurs sporadically in birds. Parathyroids are also essential if the calcium status of the bird is going to be determined. The pituitary is virtually never submitted, unless a pituitary tumour is suspected, and thus little is known of the prevalence of pituitary disease. Increased examination of the head may allow for a more frequent examination of the pituitary gland.

If the entire kidney is collected then the adrenal can be sectioned with the cranial pole of the kidney. The most common lesion of the adrenal in this study was hypertrophy and was believed to be associated with stress. A granulomatous inflammation of the adrenal was found in one budgerigar and this lesion contained acid fast organisms.

Immune System. The spleen is a critical organ to submit for histopathology. The bursa and the thymus should be submitted when tissues from a young bird are to be submitted. In the pigeon, the bursa is often the only tissue to contain inclusion bodies of the pigeon circovirus. Similarly, inclusions caused by the Beak and Feather Disease virus can also be found in the bursa of parrots. The bursa is also susceptible to bacterial infection and abscess formation and will involute early in some diseases, possibly secondary to stress. The thymus is rarely submitted unless an entire hatchling or embryo is provided. It has been the author's experience that this tissue is rarely found in sick nestling parrots, finches and pigeons. Whether it involutes early as the result of disease or normally involutes at a very young age in these birds is unknown.

Central nervous system: Brain and spinal cord should be submitted in all birds that exhibited neurologic signs. In this study, lesions consistent with avian encephalitis virus infections were found in one chicken, paramyxovirus inclusion bodies were found in the brain and spinal cord of a canary and a severe meningoencephalitis was found to be a common lesion in pigeons with salmonellosis. A massive brain infarction was identified in a red-tailed black cockatoo with a systemic bacterial disease.

Embryos and Recently Hatched Chicks: Freshly dead embryos and recently hatched chicks make excellent samples for histopathology because they can be fixed intact or with their body cavity opened and virtually every organ system can be examined if serial sections of the body are obtained. In a single three day old pigeon chick a degenerative myopathy of the muscles of the neck, bacterial overgrowth of the crop, bacterial invasion of the bursa and a diffuse severe degeneration of the thymus were identified.

CONCLUSION

All parties involved want a diagnosis when a bird dies. The best way to obtain a diagnosis is to form a partnership between the clinician and pathologist. A complete and thorough gross necropsy must be performed and a complete set of tissues submitted for microscopic examination. Many tissues are rarely submitted to pathologists, yet these often contain the only diagnostic lesions. Increasing the range of tissues submitted to pathologists will result in many more instances of determining the cause of death or disease.

RECOMMENDED READING

Schmidt RE, Reavill DR, Phalen DN. (2003). *Pathology of Pet and Aviary Birds*, Iowa State Press, Ames, Iowa.