Journal of Avian Medicine and Surgery 39(2):109–117, 2025

Clinical Report

Medical Management of a Cervical Thymoma by Radiation Therapy in an Indian Ringneck Parakeet (*Psittacula krameri*)

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Abstract: Thymomas are uncommon in birds and management is challenging because of tumor characteristics such as growth location and local invasiveness, limiting surgical and chemotherapeutic options. A 20-year-old female Indian ringneck parakeet (*Psittacula krameri*) was referred for radiation therapy because of an increase in size of a right-sided cervical mass. The mass was tentatively diagnosed as a thymoma after biopsy and incomplete surgical resection 3 months previously. Therapeutic management in this parakeet included 10 treatments of palliative radiation therapy over the course of 1 month. The tumor decreased in size by approximately 60% in length and 40% in width and was palpably softer after radiation therapy, but regrowth was observed 73 days after the final radiation treatment. One dose of chemotherapy with cyclophosphamide was administered at that time, but euthanasia was elected 5 days later due to the parakeet's declining clinical condition and poor prognosis. Radiation therapy was effective in this bird as a palliative treatment in reducing tumor size; however, a modified protocol or a multimodal approach may be needed for longer-term control of thymomas in avian species.

Key words: thymoma, neoplasia, radiation therapy, chemotherapy, avian, Indian ringneck parakeet, *Psittacula krameri*

CLINICAL REPORT

A 20-year-old female Indian ringneck parakeet (*Psittacula krameri*) was referred to the Avian and Exotic Pet Service at the Schwarzman Animal Medical Center (New York, NY, USA) for continued management of a right-sided cervical mass. Three months prior to presentation, the bird was examined by the referring veterinarian because of a 2-day history of decreased activity and a cervical mass noted by the owner. Eight months before this presentation, the bird was considered healthy based on annual examination and results of a complete blood count (CBC), plasma biochemical analysis, and plasma protein electrophoresis that were within reference intervals. On examination of the cervical area by the referring veterinarian, a firm, large right cervical mass was observed, but no measurements were recorded. Results of a cytologic examination of a fine-needle aspirate of the cervical mass revealed a highly cellular aspirate characterized by large numbers of lymphoid cells. The lymphoid population consisted of predominantly small lymphocytes with scant amounts of basophilic cytoplasm and clumped chromatin. Infectious agents were not identified. Resection and biopsy of the mass were attempted 1 week later. At surgery, the mass was described as approximately 1 to 1.5 cm in length, friable, not encapsulated, and adherent to the underlying soft tissue. Multiple sections were biopsied, but the mass could not be completely excised. Biopsy results revealed the tissue was composed of coalescing, densely cellular, nodular aggregates of lymphocytes intermingled with loosely defined packets of polygonal cells with indistinct borders, supported by scant fibrovascular stroma. Anisocytosis and anisokaryosis were mild and mitoses were rare. The histopathologic findings were suggestive of thymoma.

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Figure 1. A 20-year-old Indian ringneck parakeet (*Psittacula krameri*) with a right-sided cervical swelling.

Six weeks after biopsy, the mass measured approximately 2.2 × 2.2 cm (thickness not recorded) on recheck examination at the referring practice. Nine weeks after biopsy, the owner noticed a rapid increase in size of the mass and abnormal sleep posture due to decreased range of motion in the neck. The bird also exhibited the onset of sneezing episodes with epistaxis. On reexamination by the referring veterinarian, the mass measured approximately 3.3 × 2.2 cm. Because of the rapid increase in size of the mass, the bird was referred for radiation therapy.

On initial presentation, the bird was bright and alert and weighed 161 g. On physical examination, a nonpainful right cervical mass was palpated that extended from below the gnathotheca to the base of the neck, with intimate attachment to underlying soft tissues (Fig 1). All other findings of the physical examination were unremarkable.

Based on the history, results of previous diagnostic tests, and rapid regrowth of the mass, therapeutic options and prognosis were discussed with the owner. A computed tomography (CT) study was done the next day to measure the extent of the mass, assess for local tumor invasion, and identify landmarks for radiation therapy. Jugular venipuncture for blood sample collection was not performed because of potential risk due to the size of the mass over the right jugular vein, and the possibility of a vascular tumor.

For the CT study, the bird was induced with 1.5 to 3% isoflurane (Piramal Critical Care, Bethlehem, PA, USA) in 2L oxygen/min administered via mask, then intubated with an uncuffed 2.0 Cole endotracheal tube. General anesthesia was maintained with isoflurane between 2% and 3% in 2L oxygen/min. Continuous anesthetic monitoring was achieved using a Doppler placed over the ulnar vein, a multiparameter monitor to measure end tidal CO_2 and pulse oximetry, and nurse evaluations to record these data every 5 minutes. The bird was positioned in dorsal recumbency for the CT imaging in an immobilization Vac-Lok cushion (CIVCO Radiotherapy, Coralville, IA, USA) such that the bird could be as symmetrical as possible for reproducibility. To enhance imaging, iohexol contrast media (2 mg/kg IV; 240 mg/mL; Omnipaque, GE Healthcare Inc, Marlborough, MA, USA) was administered via the left ulnar vein with a 27-gauge needle and an insulin syringe.

Results of the CT imaging revealed a 3.2 × 2.7 × 1.6 cm soft tissue attenuating mass extending along the right aspect of the neck (Fig 2). The mass extended from the base of the skull to the cranial aspect of the shoulder, with partial attenuation of the cervical air sac, and it encased the right jugular vein and abutted against the right aspect of the trachea. It extended cranioventrally along the ventral aspect of the right mandible and dorsally along the dorsal aspect of the cervical spine, where it was closely associated with the spinous processes of the cervical vertebrae. The mass was heterogeneously enhancing, with a hypoattenuating fluid-filled center and a radiodensity of 20 Hounsfield units. The remainder of the scan showed no evidence of pathologic lesions or metastatic disease. Based on these findings, the tentative diagnosis was recurrence of the right-sided thymic mass with local invasion engulfing the vasculature.

Based on the CT results and concern for surgical risks, the owner chose to pursue radiation therapy. A treatment plan for palliative radiation therapy was designed that consisted of 10 radiation sessions with a prescription of 4 Gy each, for a total of 40 Gy. Radiation doses were administered every other day with a calendar layout of Monday-Wednesday-Friday, with slight variation due to patient and owner availability. To reproduce the same position at each treatment session, the same immobilization cushion used for the CT study was used for each session. The treatment

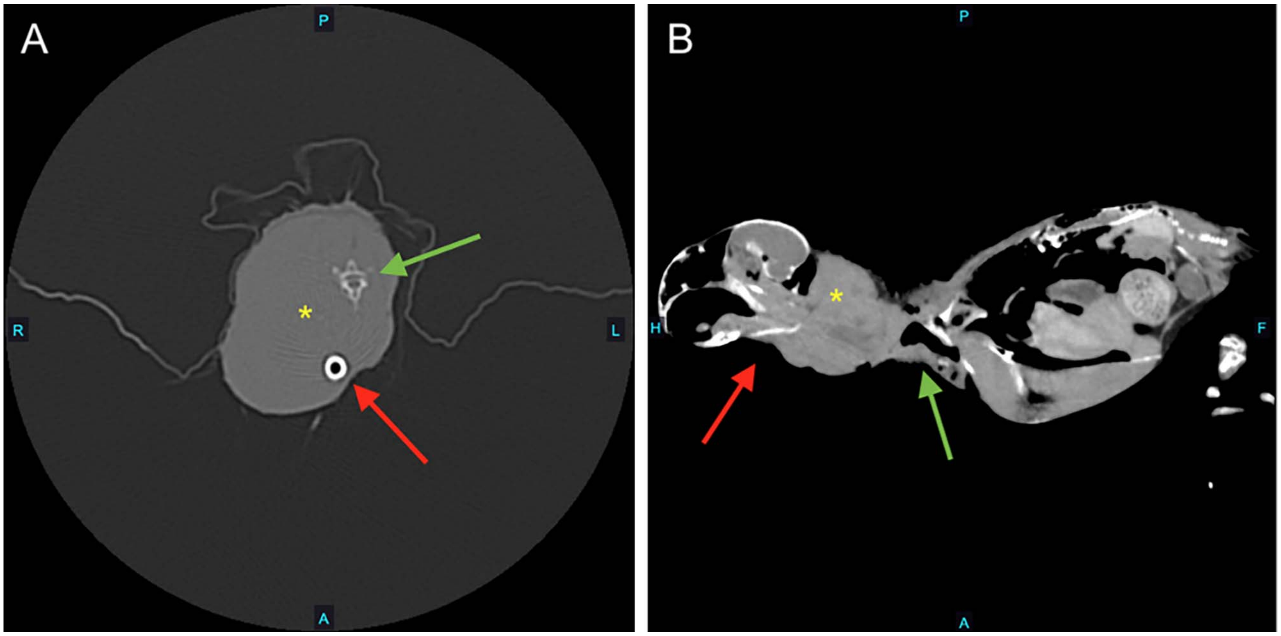

Figure 2. Axial and sagittal images from the computed tomography scans of the 20-year-old Indian ringneck parakeet (*Psittacula krameri*) described in Figure 1. (A) Axial view showing the cervical mass (yellow asterisk), the trachea with an endotracheal tube in place (red arrow), and the cervical spine (green arrow). There is moderate displacement of the trachea to the right of midline. (B) Sagittal view showing the extent of the mass (yellow asterisk). The cranial margin (red arrow) and caudal margin (green arrow) of the mass are visible just overlying the crop.

plan focused on avoiding the eyes and brain. To assure complete tumor dose coverage, the esophagus, crop, trachea, and cervical spine could not be avoided because of the intimacy of these organs to the tumor. The only portions of the trachea and esophagus that could be spared were the portions caudal to the tumor (Fig 3).

Radiation treatment was delivered via 2 obliqued, bilateral opposing fields following a computerized plan to spare the eyes and brain. Because the large mass

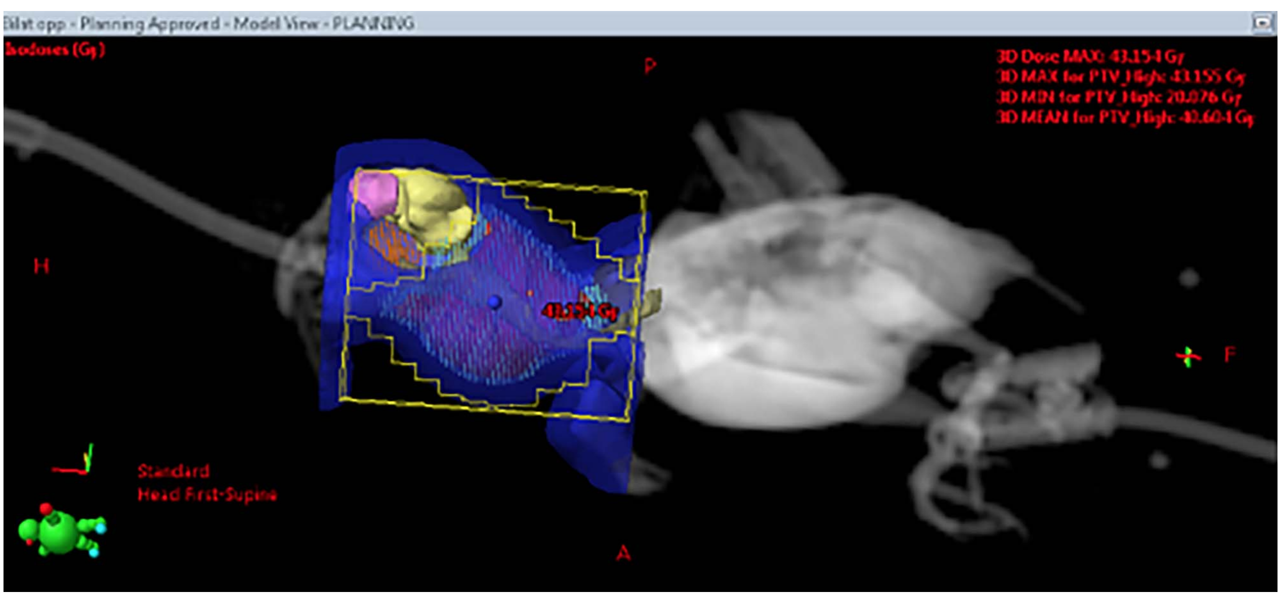

Figure 3. Absolute isodose levels with presence of bolus in the 20-year-old Indian ringneck parakeet (*Psittacula krameri*) described in Figure 1. The bird is positioned in left lateral recumbency with the head obliqued toward a sternal recumbent position.

occupied most of the cervical region, a Varian 120 multi-leaf collimator (Varian Medical Systems, Palo Alto, CA, USA) was used to shape around the targeted area, thus blocking the adjacent organs at risk. Portal imaging was performed with a kilovoltage verification imager (Theraview, Acceletronics, Inc, Exton, PA, USA) before each treatment to assure accurate positioning and intended sparing of the right eye and brain.

For each radiation treatment, the patient was induced with isoflurane administered via face mask, then intubated and maintained under general anesthesia as previously described. Although the bird was positioned in dorsal recumbency for the CT study, positioning was difficult to reproduce at the initial treatment. For more accurate reproducibility, the bird was placed in left lateral recumbency with the head obliqued sternally and the radiation fields set up for dorsoventral and ventrodorsal directions (beam angle 0° and 180°) while matching the portal verification images to the planned digital reconstructed radiograph. Target volumes included in the plan were gross tumor volume (GTV) and planning target volume (PTV). Because thymomas are typically benign tumors, a clinical target volume was not applied. The margin assigned around the GTV to produce the PTV was only 3 mm, as a wider margin was not possible because of patient size. This margin could be applied only in directions away from the skin surface; where the tumor was abutting skin, the margin was no more than 1.5 mm to include skin thickness. When targeting superficial structures, an equivalent tissue-dense material of appropriate thickness must be applied to the surface for the dose buildup to reach 95–100%. This tissue-dense material is referred to as a bolus and typically carries a density of water (1 g/cm^3). A 0.5-cm bolus was applied around the neck region of the patient to allow the dose to build up closer to the prescription dose at the skin surface. Radiation was administered at a dose of 4 Gy as planned. After the first radiation treatment (day 0 of treatment), meloxicam (1 mg/kg PO q12h × 10 days; Metacam, Boehringer Ingelheim, Ridgefield, CT, USA) was prescribed to treat potential inflammatory adverse effects of radiation.

Minimal adverse effects were seen during the initial sessions. The size of the mass was measured intermittently during the course of radiation to assess response to treatment. On the fourth radiation treatment (day 13) the mass measured approximately 2.0 × 2.0 × 1.2 cm, and the patient was doing well at home. Before the sixth radiation session (day 20), the bird appeared tachypneic on physical examination and had bilateral serosanguineous nasal discharge. A nasal flush was performed, and a large mucoid plug was flushed from each naris. Samples of the mucoid plug were submitted for aerobic bacterial culture and sensitivity testing. Doxycycline (100 mg/kg SC; Vibravenous, Pfizer, Capelle aan den Ijssel, Netherlands) and an additional single dose of meloxicam (0.5 mg/kg IM) were administered, and radiation treatment was postponed. An *Acinetobacter* sp was isolated on culture that was resistant to cephalosporins and chloramphenicol. On follow-up examination on day 22 of treatment, the bird exhibited wheezing, with dried bloody debris on the plumage around the nares and visible mucosal ulceration of the choana. Radiation therapy was postponed again and robenacoxib (7 mg/kg IM; Onsior, Elanco US Inc, Greenfield, IN, USA) was administered. After 1 week without radiation, the bird was reevaluated on day 27, at which time the nasal discharge, upper airway wheezes, and choanal ulceration appeared resolved. Subsequently, the sixth radiation dose was administered following similar anesthetic and radiation protocols as previously performed. After recovery from anesthesia, a nasal flush was repeated. A moderate amount of mucus was flushed, and doxycycline was administered at the previous dose and site of administration. At the next radiation session (seventh treatment, day 29), the skin was hyperemic at the cranial aspect of the tumor, but no other abnormalities were observed. Robenacoxib was administered at the previously prescribed dosage and route. Radiation proceeded as scheduled (day 30, day 34, day 36) until 10 sessions were completed.

Two weeks after completing radiation, the mass measured 2 × 1 cm (depth not recorded), approximately 60% of the length and 40% of the width measured on CT at presentation. The mass was palpably softer and more compressible. Respiratory signs had resolved, and the only relevant finding on examination was yellow skin pigmentation over the tumor.

The bird was reevaluated approximately 2 months later (day 112). At that time, the tumor was noticeably enlarged and was firmly attached to underlying tissues. An ultrasound examination was performed, and the results showed a well-vascularized soft tissue mass measuring 2.7 × 1.9 cm on the right craniolateral neck, with a large blood vessel suspected to be the jugular vein intimately associated with the deep margin of the mass. A CT study was scheduled for the next day to evaluate local invasion or metastasis to plan for potential surgical excision. Before induction of anesthesia, bilateral epistaxis was observed and the CT was cancelled. Yunnan Baiyao (½ capsule sprinkled in food q24h; Kunming, Yunnan, China), an

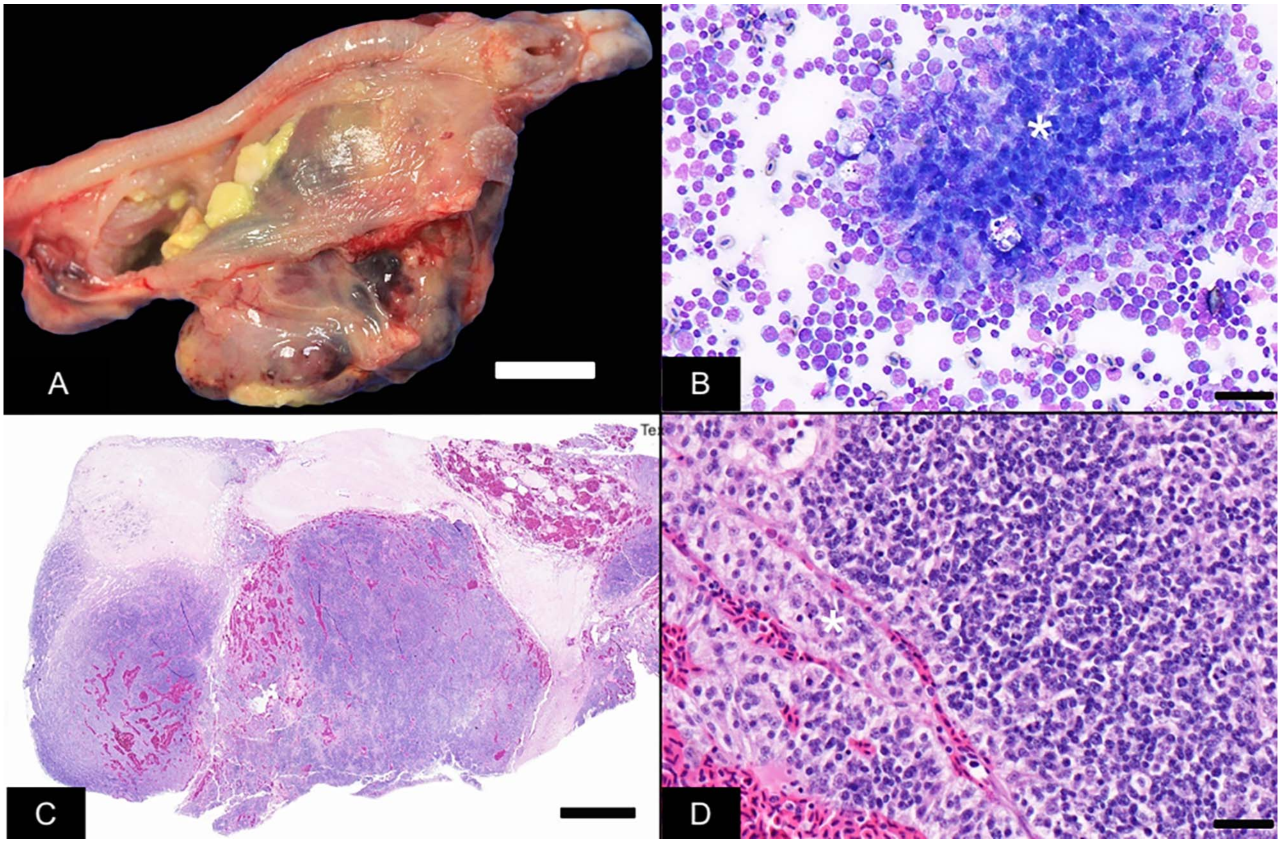

Figure 4. Gross, cytologic, and histologic appearance of a right-sided cervical mass in the 20-year-old Indian ringneck parakeet (*Psittacula krameri*) described in Figure 1. (A) A 3 × 3 × 2.3 cm, subcutaneous, firm mass was present on the right side of the neck with dilation of the proximal esophagus dorsal to the mass (bar = 1 cm). (B) Cytologic evaluation (touch imprints) of the mass showing variably sized lymphocytes interspersed by aggregates of epithelial cells (asterisk) and scattered foamy macrophages (Wright-Giemsa; bar = 20 μm). (C) Low-magnification histologic appearance of the cervical mass. Lobules of densely cellular basophilic regions (lymphocytes) are interspersed by less cellular, eosinophilic regions (epithelial cells or necrosis) (hematoxylin and eosin; bar = 1 mm). (D) High-magnification histologic appearance of the cervical mass. Aggregates of epithelial cells (white asterisk) contain pale eosinophilic, abundant cytoplasm. Numerous lymphocytes (right side of image) comprise most of the mass (hematoxylin and eosin; bar = 20 μm).

herbal supplement believed to aid with coagulation, was prescribed and a single injection of robenacoxib (6 mg/kg IM) was administered. The Yunnan Baiyao was discontinued after 13 days of treatment due to the patient exhibiting decreased appetite. A medical oncology consultation was pursued to plan chemotherapy.

On recheck examination 1 week later (day 119), the mass measured approximately 3.4 × 2.3 × 1.4 cm. A blood sample was collected from the medial metatarsal vein and submitted for a CBC before treatment. Abnormal results included low hematocrit (38%; reference interval, 45%–54%),[1] leukocytosis (40 900 cells/μL; reference interval, 8000–14 000 cells/μL)[1] with heterophilia (15 500 cells/μL; reference interval, 4480–7616 cells/μL)[1] and lymphocytosis (23 700 cells/μL; reference interval, 3584–6272 cells/μL),[1] and a decrease in the number of estimated thrombocytes. Chemotherapy was initiated with cyclophosphamide (200 mg/m^2 SC; cyclophosphamide for injection USP, Sandoz, Princeton, NJ, USA). The owners were instructed to restart the meloxicam and Yunnan Baiyao treatments as previously prescribed.

Five days later (day 124), the bird was reexamined because of lethargy, hyporexia, wheezing, and a mildly elevated respiratory effort. One-view standing radiograph showed questionable hepatomegaly and the cervical mass. Because of a poor prognosis and quality of life, the owners elected euthanasia and consented to necropsy.

At necropsy, the mass on the right side of the neck was firm and measured 3 × 3 × 2.3 cm, with an additional smaller (1.4 × 0.7 × 0.6 cm), firm, tan nodule adhered to the skin of the neck. At the left side of the thoracic inlet, a free-floating, 2.5 × 1.5 × 0.8 cm, yellow, soft mass was present. The proximal esophagus dorsal to the large mass was dilated and contained soft yellow ingesta (Fig 4A). Cytologic evaluation of the right-sided masses showed myriad, variably sized lymphocytes interspersed by aggregates of epithelial cells containing moderate amounts of basophilic cytoplasm and round to oval central nuclei with inapparent nucleoli, mixed with scattered foamy macrophages and bacteria (Fig 4B). Histologic evaluation of the 2 firm, right-sided masses showed numerous lymphocytes with scant eosinophilic cytoplasm, round nuclei, and moderately coarse chromatin intermixed with variable amounts of epithelial cells containing abundant, vacuolated eosinophilic cytoplasm and round to oval nuclei with finely stippled chromatin. These findings were consistent with a type B thymoma exhibiting epithelioid morphology (subtyped as type B1 [lymphocyte rich], Fig 4C and D).[2] Foci of coagulative necrosis with cyst formation were present within the mass, along with hemorrhage and accumulation of golden brown hemosiderin pigment.

Cytologic and histologic evaluation of the soft, free-floating material in the left thoracic inlet showed macrophages, heterophils, and large numbers of coccoid bacteria with frequent intracellular bacteria, consistent with bacterial cellulitis. An overt cause for this change was not found; however, given the presence of severe heterophilic inflammation with intracellular bacteria and inflammation of the esophageal adventitia overlying the thymoma, a previous esophageal perforation was considered. Few parabronchi in the lungs contained inflammation with intralesional keratin, suggestive of acute or peracute aspiration from the oral cavity or upper gastrointestinal tract. Additional findings were focal myositis of the syringeal muscles, minimal lymphofollicular hyperplasia of the air sacs, grade I–II atherosclerosis in the aorta and arteries of the brain,[3] cholangiohepatitis with vacuolar changes and extramedullary hematopoiesis in the liver and spleen, interstitial nephritis, and focal lymphocytic pancreatitis that was considered possible metastasis from the thymoma.

DISCUSSION

This report describes radiation therapy for management of a thymoma in an adult Indian ringneck parakeet. Radiation therapy proved a useful treatment modality to provide brief tumor control with moderate adverse effects associated with treatment. However, as a sole therapeutic option, radiation therapy was not effective in achieving long-term control. An extensive literature search using key words *avian*, *thymoma*, and *radiation* yielded no results. To our knowledge, this is the first report of radiation therapy as a treatment modality for thymoma in a psittacine species.

Thymomas are uncommon in birds, with only a few case reports and brief mentions in textbooks describing thymomas in various species.[2,4–14] In mammals, thymomas are common in dogs, cats, rabbits, goats, and people.[15–21] Thymomas typically are considered benign, but thymic epithelium has been described as a malignant component of the tumor in mammals.[22] Generally, thymomas in mammals are classified into 2 categories: 1) noninvasive, which are usually well encapsulated, or 2) locally invasive to intrathoracic structures (thoracic wall, vena cava, pericardium). In mammals, therapeutic approaches include surgical excision, radiation therapy, and chemotherapy, with surgical excision and radiation generally being the best therapeutic options.[23–28] Surgical options in birds differ anatomically from those in mammals because the thymus is located in the right cervical region in birds rather than intra- or extrathoracic as in most mammals. Like thymomas in people, thymomas in birds are classified based on their histologic appearance and cell population, as standardized by the World Health Organization.[29] In avian species, tumor growth with local invasion of the surrounding tissues, including the jugular vein and esophagus, is the main complication, which makes surgical excision a high-risk procedure.[2,14] In mammals, thymomas can metastasize and have been linked to myasthenia gravis.[30–32] Metastatic disease is uncommon in avian species but was reported in a scarlet macaw (*Ara macao*).[2]

In avian species, thymomas manifest with unilateral cervical swelling and the onset of related clinical signs such as hyporexia, regurgitation, and decreased passive range of motion of the cervical spine.[2,4,8,10,13,14] Differential diagnoses for the clinical signs and cervical mass effects described include inflammatory swelling due to a foreign body, localized trauma, ingluvitis, granuloma due to bacterial or fungal infection, and other tissue abnormalities such as cysts or other neoplasms. The main neoplastic differential to rule out is thymic lymphoma, which has been reported in a Java sparrow (*Lonchura oryzivora*) and shares macroscopic and microscopic similarities.[33]

The initial diagnostic workup for cervical swelling in birds ideally includes a CBC and a biochemical profile. However, in this bird, jugular venipuncture to collect an adequate blood sample at the initial examination was considered risky because of the large size and possible vascularity of the mass. Other venipuncture sites were attempted but adequate blood samples for biochemistry analysis could not be obtained. Fine-needle aspiration with cytologic evaluation and incisional or excisional biopsy are the diagnostic techniques most commonly used to identify this tumor. Computed tomography is the diagnostic imaging modality of choice because the improved detail allows differentiation of soft tissues and is used in surgical and radiation planning. Imaging modalities such as radiographs and ultrasound may not provide adequate detail but can prove useful in initial diagnosis.

Common therapeutic approaches for treatment of thymomas in mammals can lead to complications for avian species. Surgical excision has been performed in birds with mixed results because of incomplete excision or compromised surgical technique due to involvement of cervical vasculature and the esophagus.[2,8] Chemotherapy for thymomas has been reported in both avian and mammalian species, and excision or debulking ± radiation therapy is common.[15–21,27,34] This bimodal approach attempts to control leftover neoplastic cells, but it is not the optimal approach for these tumors. Chemotherapeutic treatment was elected for this case based on the recommendations from the hospital's oncology service because of safety, published literature, and potential efficacy. In rabbits, the preferred therapeutic approach is radiation therapy, which differs from that of other mammals.[23–25] Radiation therapy is challenging in birds because of the structures intimately attached to or surrounding the tumor. Moreover, radiation can lead to adverse effects, such as prolonged surgical healing secondary to skin inflammation. Radiation side effects tend to be inflammatory in nature and localized to the radiation site. Palliative therapy with corticosteroids has been reported in avian species.[35] Although there are concerns, radiation therapy has been used in avian patients for several tumor types, with varied success.[36–38] To our knowledge, there are no other reports of radiation therapy being used to treat avian thymomas. In this Indian ring-necked parakeet, radiation therapy yielded only short-term positive results. Early radiation-related inflammation could have precipitated the clinical signs seen in this bird, such as epistaxis due to localized vasculitis. We were unable to determine if the esophagitis present in this case was an adverse effect of radiation, a sequela of aspiration, or caused by the tumor itself. In one report of surgical excision of a thymoma in a burrowing owl (*Athene cunicularia*), esophagitis was also a postmortem finding, but whether it was related to the tumor or surgical excision was not clarified.[8] Thymomas are linked with multiple immune-mediated, paraneoplastic, and inflammatory conditions. In the patient described here, it remains unclear if the inflammatory findings on postmortem evaluation were related to a paraneoplastic syndrome. Goals of therapy in this case were to halt tumor growth and/or tumor size so that surgical excision could be pursued.

Avian species have been thought to be more resistant to radiation than other species, but there are few data on the sensitivity of the avian head and cervical structures to radiation. Results of 1 study on radiation dose calculation and efficacy of radiation delivery to the choanal area of military macaws (*Ara militaris*) showed the actual amount of radiation delivered was 8–10% less than the calculated goal.[40] Information is also lacking on long-term adverse effects of radiation in avian species. Because of this information void, a semiconservative approach for radiation therapy was used in this bird, with an every-other-day protocol for 10 sessions.

With high-energy radiation treatment >1 megavoltage (MV), an increasing amount of soft tissue is necessary for radiation penetration before achieving the maximum dose absorption. Afterwards, the dose absorbed gradually decreases because the radiation energy is transferred into deeper tissues. As such, the surface tissues are spared from significant dose absorption. With a 6-MV energy beam, 100% dose absorption is reached at approximately 1.5-cm depth-of-water-equivalent-density matter, with approximately 95% dose absorption reached at about 1 cm.[41] When targeting superficial structures, an equivalent tissue-dense material of appropriate thickness, a bolus, must be applied to the surface for the dose buildup to reach 95–100%. A 1-cm bolus to overcome this 6-MV photon property would be extremely cumbersome because the bolus weighed more than the covered structure and interfered with accurate positioning. Therefore, because of the small size of this patient, a thin bolus of 0.5 cm was chosen, which provided reasonable coverage, better reproducibility, and the ability to manipulate the patient during positioning. Ideally, the aim is to have the GTV covered by 95% of the prescription dose (40 Gy), with the goal of having 95% coverage of both. To achieve this, a full 1-cm bolus would have been needed in this bird to allow the dose to cover tissue up to the skin surface, because tissue between tumor and skin was minimal for most of the target circumference. As such, we also prescribed a dose of 36 Gy (90% of the GTV's prescription dose) to the PTV. This is routinely done in human medicine; however, a GTV margin is typically assigned 100% of the prescription dose. Comparing the percentage of PTV with the total dose prescribed to the GTV in this case resulted in approximately 85.5% of the GTV's prescription dose. Because of the thin bolus used in this bird, only a fraction of the prescribed dose to the tumor was delivered to the skin. On necropsy, the tumor was intimately adhered to the skin; therefore, it would have been ideal to have delivered a higher dose to these tissues. Another factor that impacts the prescription of radiation for birds is their unique anatomic features, including thin skin, adjacent air sacs, and size or location of the target tissue which may cause concern for iatrogenic radiation of adjacent tissues.[37,40]

Radiation therapy achieved brief tumor control in this Indian ringneck parakeet, indicating radiation therapy could be effective in treating a small thymoma or as adjunct therapy with surgical excision, with the added precaution of potential prolonged skin healing. Based on the results in this case, a multimodal approach that combines surgical excision or chemotherapy with radiation therapy may achieve tumor control as described in other species.[23,26,28] Concurrent chemotherapy with radiation therapy for other neoplasia has been described in birds with positive results for achieving tumor control and extending life expectancy.[42–44] Further studies are required to understand radiation effects in tumor control and adverse effects in avian species.

Acknowledgments: We thank Drs Karen Cantor and Rebecca Mattucks for their contribution to this case, Ms Amanda Ramkissoon for histologic preparation of the samples, and Dr Michael Wiseman for review of the cytologic specimens.

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