Feather and beak dystrophy and necrosis in cockatoos: Clinicopathologic evaluations

Elliott R. Jacobson, DVM, PhD; Susan Clubb, DVM; Charles Simpson, DVM, PhD; Michael Walsh, DVM; Clinton D. Lothrop, Jr., DVM, PhD; Jack Gaskin, DVM, PhD; John Bauer, DVM, PhD; Stephen Hines, DVM; George V. Kollias, DVM, PhD; Paul Poulos, DVM, PhD; Greg Harrison, DVM

SUMMARY

Several species of imported and captive-bred southeast Asian cockatoos with feather and beak disease (FBD) were evaluated. In recently emerging stained feathers from affected birds, intracytoplasmic magenta to basophilic inclusions of various sizes were found in macrophages and basophil-like cells within the pulp and feather epidermis. Occasionally, amphophilic intranuclear inclusions were seen within degenerated feather epidermal cells. On the basis of electron microscopic findings, intracytoplasmic inclusions were not membrane bound and consisted of crystalline arrays of viral particles (17 to 22 nm in diameter). On the basis of size and conformation, viral particles most closely resembled those of parvovirus or picornavirus.

Consistent hematologic or serum enzyme differences were not found among affected or healthy cockatoos. Compared with findings in healthy cockatoos, cockatoos with FBD had significantly lower serum protein concentrations, and results of serum protein electrophoresis indicated that birds with FBD had significantly lower concentrations of prealbumin and γ-globulin fractions. Mean pre- and post-ACTH plasma corticosterone concentrations of cockatoos with FBD were not significantly different from those of healthy cockatoos. In 8 of 9 affected cockatoos evaluated, serum T_4 concentrations before and after thyrotropin stimulation were considered normal.

PATHOLOGIC FEATHER LOSS and degeneration have been seen in various species of pet birds, and a multitude of causes (infectious and noninfectious) have been found or hypothesized to account for these conditions.[1] A specific feather loss and malformation syndrome has been described for captive and free-living sulfur-crested cockatoos *(Cacatua galerita)* in Australia, and for other Australian cockatoos including the galah *(Eolophus roseicapillus)*, little corella *(C sanguinea)*, and Major Mitchell's cockatoo *(C leadbeateri)*.[2] Affected sulfur-crested cockatoos have intracytoplasmic inclusions within macrophages in the pulp and feather epidermis. The inclusions are composed of crystalline arrays of viral particles.[3] Rosskopf et al[4] suspected adrenal insufficiency in a captive sulfur-crested cockatoo in the United States (which was similar in appearance to cockatoos in Australia) that had feather loss and malformation.

The purpose of the present study was to clinically and histologically evaluate feather and beak dystrophy and necrosis in various imported and captive-bred southeast Asian cockatoos.

Materials and Methods

Cockatoos—Several species of recently imported southeast Asian cockatoos (5 lesser sulfur-crested *[C sulphurea]*, 6 moluccan *[C moluccensis]*, 8 umbrella *[C alba]*, 1 citron *[C s citrinocristata]*, 1 triton *[C g triton]*, and 1 Goffin's cockatoo *[C goffini])*, with minimal to severe feather disease (with or without beak necrosis), were evaluated at the College of Veterinary Medicine, University of Florida. One captive-bred lesser sulfur-crested cockatoo with a similar disease also was evaluated. Hereafter this disease will be referred to as cockatoo feather and beak disease (FBD).

Most of the birds evaluated were wild-caught birds of unknown age. Two long-term captive lesser sulfur-crested cockatoos developed FBD 2 to 3 years after purchase. Several birds appeared healthy when imported and did not develop abnormalities until several months later.

The captive-bred lesser sulfur-crested cockatoo (along with a moluccan and a triton), were maintained at the university for up to 1 year after initial evaluation, for visual observation and data accumulation. Eight of the cockatoos with FBC were euthanatized and evaluated histologically, electron microscopically, and angioradiographically.

Hematologic and serum biochemical evaluations—Using manual restraint, blood samples were collected from the jugular veins of 2 lesser sulfur-crested, 1 citron, 3 moluccan, and 2 umbrella cockatoos; samples were collected 3 times from one triton cockatoo during a 4-month period. For comparison, blood samples also were collected from healthy moluccan cockatoos. A portion of each blood sample was collected in EDTA tubes for hematologic determinations (RBC count, WBC count, differential WBC count,

From the College of Veterinary Medicine (Jacobson, Simpson, Hines, Walsh, Gaskin, Bauer, Kollias, Poulos), University of Florida, Gainesville, FL 32610; College of Veterinary Medicine (Lothrop), University of Tennessee, Knoxville, TN 37901-1071; Pet Farm Inc (Clubb), Miami, FL 33166 and All Animal Clinic and Bird Hospital (Harrison), Lake Worth, FL 33463.

Published as University of Florida, College of Veterinary Medicine Journal Series No. 65.

The authors thank Dr. Richard Davis for budgerigar fledgling disease serology.

PCV, and hemoglobin concentration), and a second portion was put in silicone-coated tubes for serum biochemical determinations. The RBC count was determined by use of an electronic cell counter.[a] The WBC count was determined manually, using conventional techniques for avian species.[5] The PCV values were determined, using a microhematocrit centrifuge. Hemoglobin concentrations were determined, using an automated method.[b] Differential WBC counts were determined by counting 100 WBC in each Wright-Giemsa-stained blood film.

Serum aspartate transaminase (AST), alkaline phosphatase (ALP), and lactic dehydrogenase (LDH) activities and calcium, phosphorus, glucose, cholesterol, and uric acid concentrations were determined, using an automated analyzer method.[c]

Serum was collected from each sample and was electrophoresed on cellulose polyacetate strips at 200 V for 20 minutes, stained with ponceau-S in 7.5% trichloracetic acid, and rinsed with 5% acetic acid. After clearing the strips in 40% aqueous N-methyl pyrrolidine, the strips were dried for 20 minutes at 90 C. The separated proteins were densitometrically quantitated, using a scanning densitometer (ACD-15)[d] at 525 nm.

Serologic evaluations—Serum samples from 1 moluccan, 3 Goffin's, 1 lesser sulfur-crested, and 1 umbrella cockatoo, all with FBD, were assayed for antibodies against budgerigar fledgling disease (BFD) papovavirus. Blood from the jugular vein was placed in a silicone-coated tube and centrifuged at 5,000 rpm for 5 minutes; the serum was removed and stored until assayed. Serum antibody titers against budgerigar papovavirus were determined by use of serum neutralization. Titers ≥1:20 were considered positive.

Endocrinologic evaluations—Five healthy lesser sulfur-crested cockatoos and 10 cockatoos with FBD (5 lesser sulfur-crested, 2 little corella, 1 moluccan, and 2 Goffin's cockatoos) were evaluated for their response to ACTH. A pre-ACTH blood sample was collected from the jugular vein of each bird, and plasma was collected from each sample. Each bird was given 15 IU of ACTH[e] (0.6 ml) IM, and a post-ACTH blood sample was collected 2 or 4 hours after ACTH administration. All samples were collected within 2 minutes after initiation of manual restraint. Plasma samples were frozen at −20 C until assayed for corticosterone, using a previously described radioimmunoassay.[6]

Nine cockatoos with FBD (7 lesser sulfur-crested cockatoos, 1 moluccan, and 1 triton cockatoo) were evaluated for their response to thyrotropin. A prethyrotropin blood sample was collected from the jugular vein of each bird, and serum was collected from each sample. Each bird was given 1 IU of thyrotropin[f] IM, and a blood sample was collected 4 to 6 hours after thyrotropin administration. All blood samples were collected within 2 minutes after initiation of manual restraint. Serum samples were frozen at −20 C until assayed for thyroxine (T_4), using a previously described radioimmunoassay.[7]

Microscopic evaluations—Recently emerging abnormal feathers (ie, feathers that had macroscopic abnormalities) from 9 cockatoos with FBD (4 moluccan, 1 triton, and 4 lesser sulfur-crested cockatoos) were biopsied. Birds were anesthetized with ketamine HCl (3 mg/kg of body weight, IV). A minimum of 6 recently emerging feathers were biopsied by making a full-thickness circumferential incision around the base of a feather follicle. The biopsy specimen included the feather and its associated follicle. For comparison, recently emerging feathers from 6 healthy moluccan cockatoos also were biopsied. The incision site was sutured with 5-0 chromic gut. Feathers were fixed in 10% neutral-buffered formalin (NBF) for 24 hours, longitudinally sectioned in half, and fixed for an additional 24 hours in NBF. All such fixed biopsy specimens were embedded in paraffin, sectioned at 7 μm, and stained with hematoxylin and eosin (H&E); additional specimens were stained with Giemsa, Feulgen, and methyl green pyronin.

Contour feathers from a healthy moluccan cockatoo and from a moluccan cockatoo with FBD were prepared for scanning electron microscopy by fixation in Trump's solution for 1 hour, postfixation in 1% osmium tetroxide for 1 hour, dehydration, placement in a critical point dryer[g] for 45 minutes, mounting, placement in an evaporation vacuum device,[h] and coating for 6 minutes with gold and palladium. Feathers were examined with a scanning electron microscope.[i]

For cytologic evaluation, recently emerging feathers from birds with FBD were removed from corresponding follicles and split longitudinally, the pulp was removed, and impression smears were made. After air drying and fixation in absolute methanol for 30 seconds, impression smears were stained by use of the Wright-Giemsa method. The smears were examined microscopically.

Complete necropsies were conducted on 4 umbrella, 1 citron, 1 moluccan, and 2 lesser sulfur-crested cockatoos with FBD. All tissues were placed in NBF, embedded in paraffin, sectioned at 7 μm, and stained with H&E. Additional sections of liver from 2 lesser sulfur-crested cockatoos were stained with periodic-acid Schiff (PAS).

For electron microscopy, recently emerging abnormal feathers from a moluccan cockatoo and a triton cockatoo with FBD were biopsied. The feather follicle and proximal pulp were sectioned into small (1 mm) cubes and fixed in 3.5% buffered glutaraldehyde, then fixed in 1% osmium tetroxide, and embedded in araldite. Thin sections (60 to 70 nm) were placed on grids, stained with uranyl acetate and lead citrate, and examined electron microscopically.[j] Liver sections from a lesser sulfur-crested cockatoo with FBD were similarly processed and examined.

Microbiologic evaluations—Necrotic material from beak lesions of 4 lesser sulfur-crested cockatoos was cultured on blood agar and on MacConkey's agar (both incubated at 38 C) and on Sabouraud's agar (incubated at 22 C).

Multiple affected feathers from a triton and a moluccan cockatoo were removed and split longitudinally, and the pulp collected aseptically. Pulp was placed in tubes containing Eagle's minimal essential medium, 10% bovine fetal serum, penicillin (100 U/ml), and streptomycin (100 μg/ml). The mixture was inoculated directly into cultures of cockatiel embryo fibroblasts and chicken embryo fibroblasts incubated at 38 C.

Angioradiographic evaluations—Three cockatoos with necrosis of the distal portion of the beak (an umbrella cockatoo, a citron cockatoo, and a lesser sulfur-crested cockatoo) were euthanatized. The aorta of each bird was identified and catheterized, 100 ml of a warmed mixture of gelatin[k] (80 g) and barium sulfate[l] (2 L) were perfused by hand via a

[a]Coulter Counter ZB16, Coulter Diagnostics, Hialeah, Fla.
[b]Coulter Hemoglobinometer, Coulter Diagnostics, Hialeah, Fla.
[c]Encore, Baker Instruments, Allentown, Pa.
[d]Sepatrex, Gelman Instrument Co, Ann Arbor, Mich.
[e]Cortrosyn, Organon Diagnostics Inc, West Orange, NJ.
[f]Dermathycin, Jensen-Salsbury Laboratories, Kansas City, Mo.
[g]Hummer 1, Technics, Alexandria, Va.
[h]Denton Vacuum Inc, Cherry Hill, NJ.
[i]JEOL JSM 35-C, JEOL, Peabody, Mass.
[j]Phillips EM200, Phillips Electronic Instruments, Mahwah, NJ.
[k]Gelatin Bloom 275, Fisher Scientific, Fair Lawn, NJ.
[l]Solopake, E-Z-EM Co, Westbury, NY.

TABLE 1—Hematologic, serum biochemical, serologic, and endocrinologic findings in healthy cockatoos and in cockatoos with feather and beak dystrophy and necrosis

Evaluation	Determinant	Healthy cockatoos			Cockatoos with FBD		
		No. evaluated	Mean ± SD	Range	No. evaluated	Mean ± SD	Range
Hematology	RBC ($\times 10^6/\mu l$)	5	2.74 ±	2.33 to 3.10	11	2.48 ± 0.52	1.35 to 3.17
	WBC ($\times 10^3/\mu l$)	6	9.6 ± 2.7	6.5 to 13.0	11	7.3 ± 4.5	2.4 to 18.1
	Hb (g/dl)	6	12.9 ± 1.1	10.6 to 17.0	10	11.1 ± 2.8	6.1 to 14.3
	PCV (%)	6	48 ± 3	41 to 53	11	40 ± 8	23 to 50
	Differential						
	Heterophils (%)	6	70 ± 13	63 to 85	11	73 ± 8	58 to 84
	Lymphocytes (%)	6	17 ± 7	15 to 37	11	21 ± 11	4 to 42
	Monocytes (%)	6	0	NA	11	5 ± 4	0 to 13
	Eosinophils (%)	6	0	NA	11	1 ±	0 to 2
	Basophils (%)	6	0	NA	11	1 ± 1	0 to 4
Serum biochemistry	AST (IU/L)	5	151 ± 100	37 to 296	7	176 ± 40	131 to 230
	ALP (IU/L)	5	124 ± 39	75 to 130	7	170 ± 118	52 to 424
	LDH (IU/L)	5	446 ± 174	312 to 782	3	412 ± 72	338 to 510
	Calcium (mg/dl)	5	10.3 ± 0.3	9.4 to 10.8	7	9.5 ± 0.9	8.3 to 10.8
	Phosphorus (mg/dl)	5	4.3 ± 1.6	2.4 to 5.8	7	5.0 ± 1.3	2.7 to 6.6
	Glucose (mg/dl)	5	230 ± 30	170 to 247	7	244 ± 48	189 to 308
	Cholesterol (mg/dl)	5	208 ± 24	172 to 247	4	203 ± 60	133 to 282
	Uric acid (mg/dl)	5	4.0 ± 1.5	2.5 to 6.7	4	5.6 ± 3.6	2.8 to 11.7
Serum electrophoretogram fractions	Prealbumin (g/dl)	6	0.71 ± 0.09	0.55 to 0.81	11	0.47 ± 0.19	0.20 to 0.76
	Albumin (g/dl)	6	2.17 ± 0.23	1.89 to 2.45	11	2.49 ± 0.84	1.48 to 4.10
	β Globulin (g/dl)	6	0.45 ± 0.04	0.37 to 0.49	11	0.33 ± 0.16	0.10 to 0.59
	γ Globulin (g/dl)	6	1.61 ± 0.87	0.93 to 3.12	11	0.70 ± 0.49	0.10 to 1.86
	Total protein (g/dl)	6	4.92 ± 0.73	4.30 to 6.20	11	3.98 ± 0.52	2.90 to 4.70
Endocrinology	Plasma corticosterone						
	Pre-ACTH (ng/ml)	5	14.5 ± 7.2	5.0 to 22.5	10	23.5 ± 12.0	7.5 to 45
	Post-ACTH (ng/ml)						
	2 hours	5	45.5 ± 12.9	37.5 to 67.5	10	61.0 ± 24.7	32.5 to 105
	4 hours	ND	ND	ND	10	46.8 ± 20.6	17.5 to 95
	Plasma thyroxine						
	Prethyrotropin (ng/ml)	NA	NA	NA	9	13.7 ± 6.3	2.3 to 25.0
	Post-thyrotropin (ng/ml)	NA	NA	NA	9	36.0 ± 18.6	4.1 to 73.0

AST = aspartate transaminase; ALP = alkaline phosphatase; LDH = lactate dehydrogenase; Hb = hemoglobin; ND = not determined; NA = not applicable.

syringe into the aorta (using slow, steady, manual pressure), and radiography was performed using a cabinet x-ray system.[m]

Results

Clinical disease—The captive-bred lesser sulfur-crested cockatoo first manifested clinical signs of disease at 3 months of age; 2 other clutch mates had the same clinical disease. The parents of these birds were healthy, but in the previous year had produced 2 chicks that developed FBD.

Although initial macroscopic lesions in feathers were found in various feather tracts, the first abnormal feather tracts often were the femoral tracts. Subsequently, abnormal contour feathers were found in all feather tracts. Rectrices often were involved in early stages of the disease. Primary wing feathers generally were the last feathers to develop the most severe changes.

Progression of the disease differed among the birds; however, the time from initial recognition of minimal feather problems to severe involvement generally was 6 months. One triton cockatoo with minimal to moderate FBD at the time of initial evaluation became affected severely during the following 3 months; this bird was alive approximately 2 years after initial evaluation.

Macroscopically, the vane was ragged in appearance, with multiple fractures. Affected feathers would commonly fracture at the proximal rachis. Entire tracts or individual feathers within a tract often did not exsheathe and many of these sheathed feathers were stunted in size. Often, hemorrhage within the pulp was seen. Feathers that did not emerge had thickened sheaths that resulted in midshaft constrictions and terminal clubbing. In severely affected birds, entire tracts were featherless.

Beak abnormalities often were associated with cockatoos with feather disease. Older birds (≥1 year) did not have as high an incidence of beak involvement as compared with juveniles (<1 year). Changes in gross appearance of the beak involved beak overgrowth, fracturing of the distal end, and/or necrosis. Two captive-bred lesser sulfur-crested cockatoos developed FBD between 6 and 10 months of age. Cracking in the hard corneum of the distal portion of the beak was the first lesion noticed. Next, the dark outer corneum was lost, exposing subadjacent lighter-colored horny material. Pathologic beak changes progressed proximally into the palatine area and involved the premaxilla in severely affected birds. Less severe changes were seen in the distal end of the lower beak.

Hematologic and serum biochemical findings—Consistent differences were not found between hematologic values of cockatoos with FBD and healthy moluccan cockatoos (Table 1). White blood cell counts for 3 cockatoos with FBD were lower than values for healthy birds, whereas 2 birds with FBD had values greater than those for healthy cockatoos. A triton cockatoo that was evaluated 3 times during a 4-month period had a decrease in the WBC count one month after the initial evaluation and a slight increase in the WBC count (compared with the second value) 3 months after the second evaluation. However, the second value was still lower than the first.

Serum biochemical values for cockatoos with FBD

[m]Faxitron 43805-N, Hewlett-Packard Corp, McMinnville, Ore.

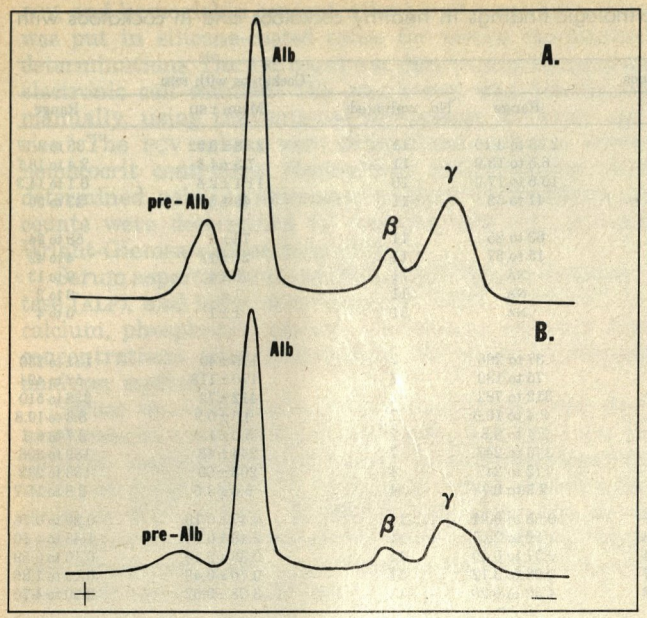

Fig 1—Serum protein electrophoretograms of a healthy moluccan cockatoo (A) and of a moluccan cockatoo with feather and beak dystrophy and necrosis (FBD) (B). Concentrations of the prealbumin (pre-Alb) and γ-globulin fractions of the cockatoo with FBD were lower than those of the healthy cockatoo.

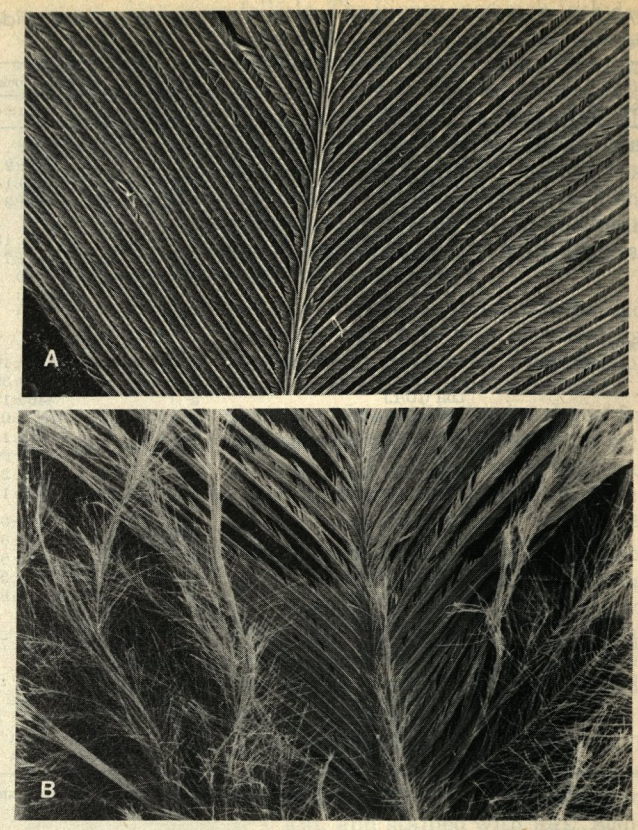

Fig 2—Scanning electron micrographs of a contour feather from a healthy moluccan cockatoo (A) and from a moluccan cockatoo with FBD (B). The affected feather (B) has multiple fractures of the hooklets, barbules, and barbs, and the after feather is poorly developed; ×500.

were not consistently different from those of healthy moluccan cockatoos (Table 1).

Total serum protein concentrations in 6 healthy moluccan cockatoos were significantly greater ($P < 0.01$; Student's t test) than those of 11 cockatoos with FBD (Table 1). Cockatoos with FBD had significantly lower ($P < 0.05$) γ-globulin and prealbumin concentrations than did healthy cockatoos (Table 1; Fig 1).

Serologic findings—Of the 6 CFBD-affected birds evaluated for antibody titers against budgerigar papovavirus, a moluccan and lesser sulfur-crested cockatoo had titers ≥1:20, and 3 Goffin's and an umbrella cockatoo had titers <1:20.

Endocrinologic findings—Compared with pre-ACTH values, all birds developed increased serum levels of corticosterone at 2 and 4 hours after administration of ACTH (Table 1). Significant differences ($P > 0.05$) were not found between corticosterone concentrations of healthy cockatoos and cockatoos with FBD. Compared with prethyrotropin values, 8 of 9 cockatoos with FBD developed increased T_4 concentrations 2 to 4 hours after thyrotropin administration. The cockatoo that did not develop an increased T_4 concentration after thyrotropin administration had a low prethyrotropin concentration and died 40 minutes after the second blood sample was collected.

Pathologic findings—Scanning electron microscopy of affected feathers of birds with FBD indicated that hooklets, barbules, and barbs of the vane were poorly developed and had multifocal fractures (Fig 2). The after feather also was poorly developed.

Histologic examination of feathers from cockatoos with FBD indicated similar changes in all birds. Feathers that did not exsheathe had hyperkeratotic sheaths (Fig 3), resulting in the terminal clubbing and midshaft constrictions that were seen macroscopically. In the epidermal collar of many emerging feathers, ballooning degeneration and necrosis of epithelial cells were seen (Fig 4). The pulp often was infiltrated with macrophages, small mononuclear cells, plasma cells, and heterophils. Occasionally,

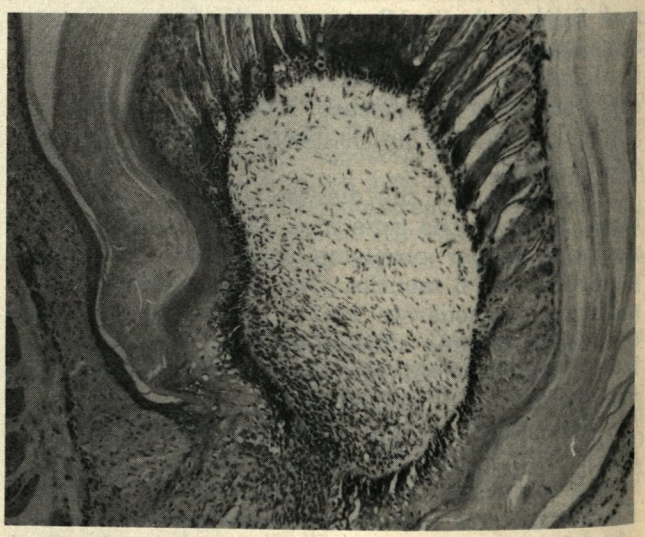

Fig 3—Photomicrograph of a sheathed developing contour feather from a lesser sulfur-crested cockatoo with FBD. The sheath is hyperkeratotic and has resulted in a midshaft constriction of the epidermis. H&E stain; ×112.

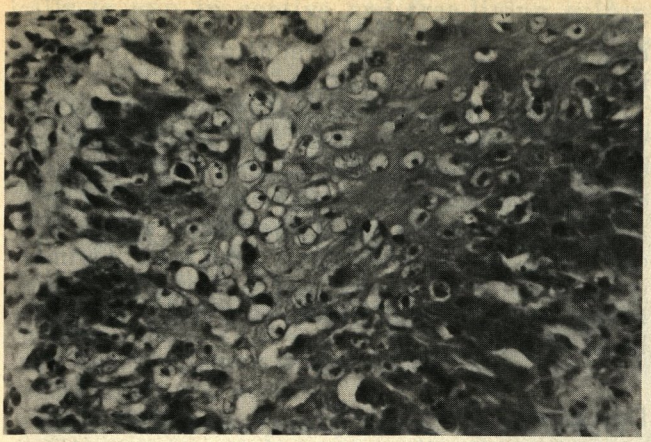

Fig 4—Photomicrograph of the epidermis of an affected feather from a lesser sulfur-crested cockatoo with FBD. Epidermal cells within the collar are undergoing ballooning degeneration and necrosis. H&E stain; ×334.

areas of hemorrhage in the distal pulp were associated with a granulomatous inflammatory response. Mononuclear cells containing basophilic (H&E) spherical granules of uniform size were seen consistently in the pulp and feather epidermis of emerging feathers from cockatoos with FBD. Similar appearing cells, although fewer in number, were seen in the epidermis of recently emerging feathers of healthy cockatoos. The granules stained positive by use of the Giemsa method and were similar histologically to mammalian basophils. However, in feathers of birds with FBD, these cells were more numerous and often contained larger, different sized, magenta-to-basophilic intracytoplasmic inclusions (Fig 5). Single large inclusions were seen within macrophages in the pulp and feather epidermis of cockatoos with FBD. Basophil-like cells and macrophages containing inclusions were seen in cytologic impression smears of pulp stained by the Wright-Giemsa method. Inclusions were not seen in biopsied feathers from healthy

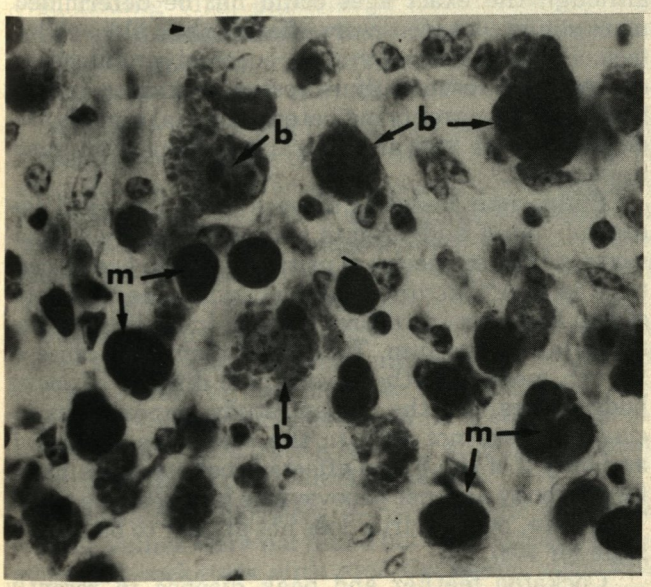

Fig 5—Photomicrograph of macrophages (m) and basophil-like cells (b) containing intracytoplasmic inclusions, in the feather pulp of a lesser sulfur-crested cockatoo with FBD. H&E stain; ×440.

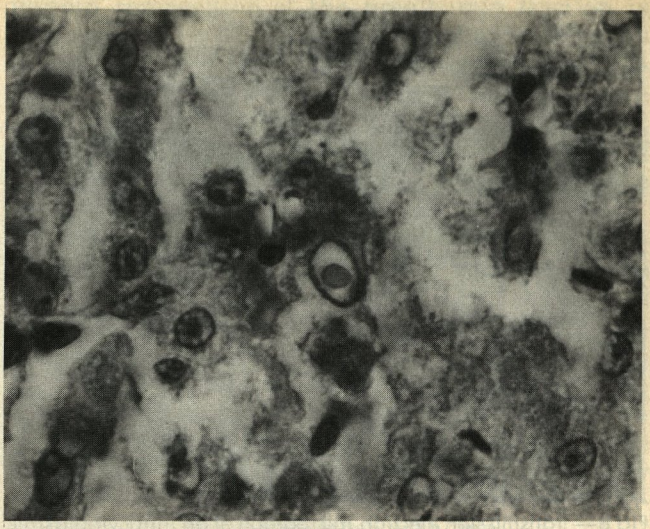

Fig 6—Photomicrograph of a hepatocyte from a lesser sulfur-crested cockatoo with FBD. Notice the intranuclear inclusion. H&E stain; ×295.

cockatoos. The inclusions stained positive for DNA with Feulgen and positive for RNA with methyl green pyronin stains. Amphophilic intranuclear inclusions were seen within follicular epidermal and feather epithelial cells undergoing ballooning degeneration and necrosis. Cells that contained intranuclear inclusions were not as numerous as cells that contained intracytoplasmic inclusions.

Consistent macroscopic or microscopic lesions of internal organs were not seen in birds with FBD. Lesions that were seen were airsacculitis, pericarditis, and hepatitis. In 2 lesser sulfur-crested cockatoos, eosinophilic (H&E) and PAS-negative intranuclear inclusions were found in hepatocytes (Fig 6). Electron microscopically, the intranuclear inclusions were composed of nonviral electron-dense material. The exact nature of this material was not determined.

Electron microscopically, the basophil-like cells contained membrane-bound granules of uniform density (Fig 7). The larger intracytoplasmic inclu-

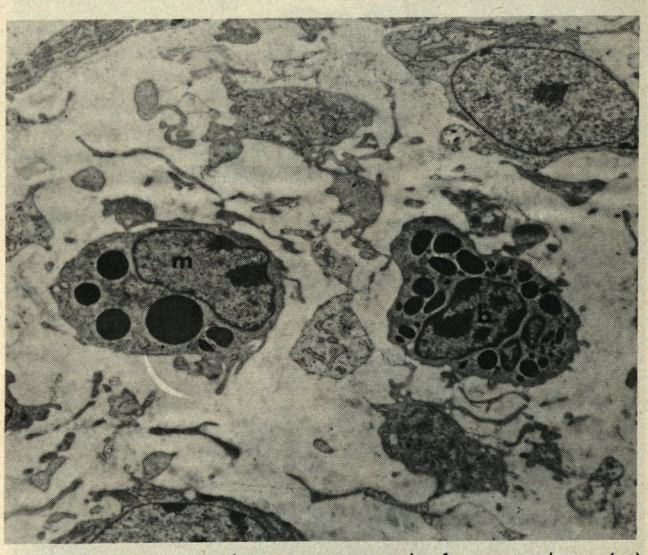

Fig 7—Transmission electron micrograph of a macrophage (m) and a basophil-like cell (b) in the feather pulp of a moluccan cockatoo with FBD. The macrophage contains several large intracytoplasmic inclusions; ×4,000.

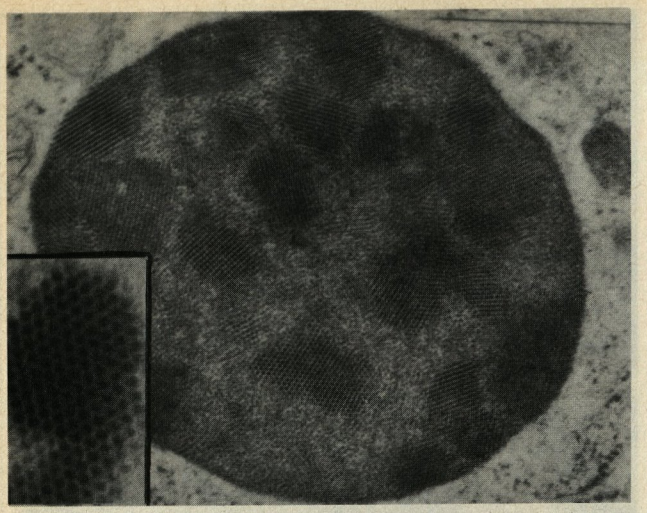

Fig 8—Transmission electron micrograph of an intracytoplasmic inclusion of a macrophage in the feather pulp of a moluccan cockatoo with FBD. The inclusion is not membrane bound and consists of myriads of viral particles; ×18,500. At a higher magnification (inset), particles are organized into a crystalline array; ×70,300.

sions were not membrane bound and consisted of crystalline arrays of nonenveloped viral particles (Fig 8) that were 17 to 22 nm in diameter. Intranuclear inclusions that were seen within follicular epidermal and feather epithelial cells by use of light microscopy were not seen by use of electron microscopy.

Angioradiographic evaluations of beak lesions from cockatoos with FBD indicated a well-developed vascular supply to the area of necrosis (Fig 9). The 3 cockatoos evaluated angioradiographically had osteomyelitis of the premaxilla.

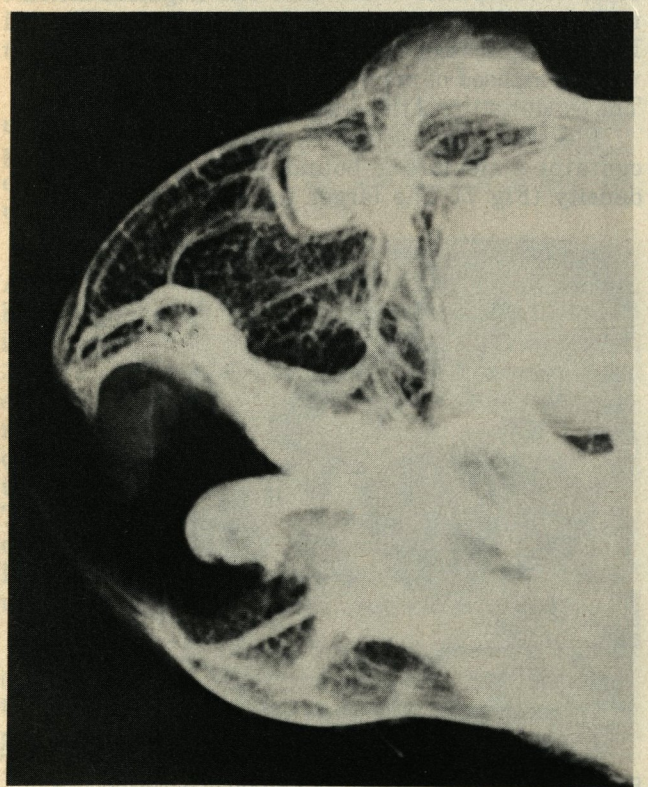

Fig 9—Angioradiograph of a citron cockatoo with beak necrosis. The distal portion of the upper beak has a good vascular supply. Notice the necrosis of the premaxilla.

Microbial isolates from beak lesions of 4 cockatoos with FBD included *Proteus* spp, *Providencia* spp, *Citrobacter diversus, Enterobacter agglomerans, E cloacae, Pseudomonas aeruginosa, Klebsiella pneumoniae, Escherichia coli, Acinetobacter* sp, and *Candida* spp. Cytopathic effects were not seen in primary or subcultures of cockatiel and chicken embryo fibroblasts inoculated with feather pulp suspensions from birds with FBD.

Discussion

Cockatoo feather and beak disease is a chronic progressive disease. The macroscopic appearance of a bird with FBD depends on the molt cycle stage at which the bird becomes infected actively. Cockatoos appear to loose contour feathers continually throughout the year and do not have a discrete period of molt. As normal feathers are lost and replaced with affected feathers, the bird appears affected more severely. Because primary feather tracts are replaced less often than are other tracts, feathers in primary tracts usually are the last to become affected. Ultimately, affected tracts become totally inactive. Although some birds may die within 6 months after first developing illness, many will survive for several years. Treatment with antibiotics, antifungal agents, and nutrient supplementation does not alter progression of the disease.[1]

In our (SC) experience, approximately 0.5% of imported flocks of white cockatoos and pink cockatoos have FBD; cockatoos most often affected appear to be lesser sulfur-crested, umbrella, citron, and moluccan cockatoos. Only one black cockatoo, a palm cockatoo *(Probosciger aterrimus),* has been seen (SC) with clinical signs of FBD. In the present study, FBD developed in adult and juvenile cockatoos; however, although the exact ages could not be determined, more young cockatoos were affected than older cockatoos. Development of FBD in successive clutches of captive-bred lesser sulfur-crested cockatoos derived from healthy parents indicated potential genetic predisposition for FBD and/or transmission of an infectious agent.

Budgerigar fledgling disease (BFD) is a viral disease of young budgerigars and is caused by a papovavirus.[8] Other species of psittacines also are susceptible to BFD.[9] Intranuclear inclusions characteristic of BFD are seen in various tissues, including feather follicular epithelium. This virus may be a cause of French molt, a feather dystrophic disease of young budgerigars.[10] In the present study, papovavirus inclusions were not seen in the cockatoos evaluated, and only 2 of the 6 FBD-affected cockatoos evaluated for antibody against papovavirus had positive titers. Therefore, papovavirus does not appear to be responsible for FBD.

Cockatoo feather and beak disease in recently imported southeast Asian cockatoos and in captive-bred lesser sulfur-crested cockatoos in the United States was similar macroscopically and microscop-

ically to FBD in captive and free-living sulfur-crested cockatoos in Australia.[2,3] Affected cockatoos in the present study and affected cockatoos in Australia[2,3] had characteristic magenta-to-basophilic intracytoplasmic inclusions in feather epidermis and pulp macrophages. A basophil-like cell was seen in the feather pulp and epidermis of recently emerging feathers of healthy cockatoos and cockatoos with FBD; however, in feathers of birds with FBD, these cells were more numerous and contained intracytoplasmic inclusions. Occasionally, in feather epidermal cells of birds with FBD, amphophilic intranuclear inclusions were seen in areas undergoing degeneration and necrosis.

Electron microscopically, intracytoplasmic inclusions were not membrane bound and consisted of crystalline areas of viral particles that were 17 to 22 nm in diameter. These particles were similar to those found in sulfur-crested cockatoos in Australia.[3] On the basis of conformation and size, the particles resembled parvovirus or picornavirus. Special staining with Feulgen and methyl green pyronin indicated that the inclusions contained DNA and RNA. Although intranuclear inclusions occasionally were seen in the epithelium of affected feathers, they were not seen by use of electron microscopy. The virus may replicate within nuclei of infected epidermal cells. Inclusions may be released when the cells undergo necrosis and may become phagocytized by macrophages and basophil-like cells in the feather pulp and epidermis. If replication is intranuclear, the virus is most consistent with parvovirus; if replication is intracytoplasmic, the virus is most consistent with picornavirus.

In the present study, a causal relationship between the virus and FBD could not be established firmly because Koch's postulates were not fulfilled. The virus could not be isolated in cockatiel and chicken embryo fibroblasts. A cell culture derived from cockatoos may be necessary for isolation. However, finding viral particles in affected feathers of southeast Asian cockatoos that were morphologically similar to those reported in Australian cockatoos is presumptive evidence of a virus-induced disease.

Although intracytoplasmic inclusions were easy to identify in feathers of birds with FBD, all feathers were not suitable for demonstrating inclusions. Therefore, recently emerging feathers that have not exsheathed are recommended for biopsy. Old feathers should be avoided because they are acellular and because cells containing inclusions will not be seen. A biopsy specimen should include a feather and its corresponding follicle, and a minimum of 6 affected feathers should be evaluated per bird. Cells containing characteristic inclusions can be seen in impression smears of pulp from a recently emerging feather stained by the Wright-Giemsa method.

Consistent differences in hematologic or serum enzyme values were not found between healthy cockatoos and cockatoos with FBD. Cockatoos with FBD had significantly lower serum total protein concentrations and mean concentrations of prealbumin and γ-globulin fractions than did healthy cockatoos. The protein composition of the prealbumin fraction is unknown and the importance of this remains unclear. The low concentration of the globulin fraction in cockatoos with FBD probably was indicative of a compromised humoral immunologic system, which may be a partial explanation for the findings of gram-negative bacterial and yeast infections in the birds.

Rosskopf et al[4] reported a possible adrenal insufficiency in a captive sulfur-crested cockatoo in the United States, on the basis of finding low pre- and post-ACTH cortisol concentrations. However, corticosterone is the major circulating glucocorticoid in adult poultry[11] and psittacine birds.[6,12,13] In the present report, pre- and post-ACTH plasma concentrations of corticosterone in cockatoos with FBD were not significantly different from those in healthy cockatoos, indicating a normally functioning pituitary-adrenal axis in cockatoos with FBD.

In 8 of 9 cockatoos with FBD, serum concentrations of T_4 before and after thyrotropin administration were similar to those reported for healthy cockatoos.[7] The prethyrotropin and postthyrotropin T_4 concentrations in the remaining cockatoo (which died 40 minutes after a second blood sample was collected) were consistent with anticipated concentrations in a bird that was near death.

Birds with FBD can survive for several years after onset of clinical signs. Many owners prefer to keep such birds, even though they may be completely featherless. Therefore, clients should maintain these birds at ambient temperatures of 25 C to 30 C because they lack their normal thermoregulatory mechanisms. Birds with beak necrosis must be fed a soft-food diet.

References

1. Harrison GJ. Feather disorders. In: Harrison GJ, ed. *The veterinary clinics of North America, small animal practice.* Vol 14. Philadelphia: WB Saunders Co, 1984;179–199.
2. Pass DA, Perry RA. The pathology of psittacine beak and feather disease. *Aust Vet J* 1984;61:69–79.
3. McOrist S, Black DG, Pass DA, et al. Beak and feather dystrophy in wild sulphur-crested cockatoos *(C galerita). J Wildl Dis* 1984;20:120–124.
4. Rosskopf WJ, Woerpel RW, Howard EB, et al. Chronic endocrine disorder associated with inclusion body hepatitis in a sulfur-crested cockatoo. *J Am Vet Med Assoc* 1981;179:1273–1276.
5. Schermer S. *The blood morphology of laboratory animals.* 2nd ed. Philadelphia: FA Davis, 1967;137–169.
6. Lothrop CD, Olsen JH, Loomis MR, et al. Evaluation of adrenal function in psittacine birds, using the ACTH stimulation test. *J Am Vet Med Assoc* 1985;187:1113–1115.
7. Lothrop CD, Loomis MR, Olsen JH. Thyrotropin stimulation test for evaluation of thyroid function in psittacine birds. *J Am Vet Med Assoc* 1985;186:47–48.
8. Bozeman LH, Davis RB, Gaudry D, et al. Characterization of a papovavirus isolated from fledgling budgerigars. *Avian Dis* 1981; 25:972–980.
9. Jacobson ER, Hines SA, Quesenberry K, et al. Epornitic of papova-like virus-associated disease in a psittacine nursery. *J Am Vet Med Assoc* 1984;185:1337–1341.
10. Bernier G, Morin M, Marsolais G. A generalized inclusion body disease in the budgerigar *(Melopsittacus undulatus)* caused by a papovavirus-like agent. *Avian Dis* 1981;25:1083–1092.
11. Frankel AI, Grabel JW, Nalbandov AV. Adrenal function in cockerels. *Endocrinology* 1967;80:1013–1019.
12. Walsh MT, Beldegreen RA, Clubb SL, et al. Effect of exogenous ACTH on serum corticosterone and cortisol concentrations in the Moluccan cockatoo *(Cacatua moluccensis). Am J Vet Res* 1985;46:1584–1588.
13. Zenoble RD, Kemppainen RJ, Young DW, et al. Endocrine responses of healthy parrots to ACTH and thyroid stimulating hormone. *J Am Vet Med Assoc* 1985;187:1116–1118.