

A MULTIFACTORAL DISEASE SYNDROME IN AFRICAN GREY PARROTS
(*PSITTACUS ERITHACUS*) IMPORTED FROM GHANA

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A multifactoral disease syndrome was observed in four lots of newly imported African Grey Parrots (*Psittacus erithacus*) during 1983. The disease occurred primarily in birds imported from Ghana however African Greys from other countries were susceptible when exposed. Two viruses were associated with the syndrome, a Reovirus and a Paramyxovirus type 3. The reovirus appears to be of primary importance, the role of the paramyxovirus is unknown. The birds were often secondarily infected with fungal and bacterial infections and these may be synergistic with the viral infections. *Aspergillus flavus* and *A. fumigatus* were most commonly isolated followed by *Mucor* Sp. Salmonellosis was also commonly associated with the syndrome. This paper will describe the clinical course of the syndrome.

HISTORY

Four lots of African Grey Parrots from Ghana were quarantined in three different commercial quarantine facilities in Florida (lot #1 and lot #3 were quarantined in the same quarantine station four months apart) in 1983. Two lots contained only African Greys while one lot also contained Jardines Parrots (*Poicephalus gulielmi*) and one contained captive born Cockatiels (*Nymphicus hollandicus*) and Lovebirds (*Agapornis* sp.) from South Africa. The histories of the 4 lots are as follows:

The author acknowledges the assistance of Dr. David Graham, Dept. of Avian and Aquatic Animal Medicine, Cornell University for histopathology and virology, Mr. Jack Crawford, Interamerican Laboratory, Miami Florida for hematology and clinical chemistry, and Dr. Adel Yunis, Dept of Hematology, School of Medicine, University of Miami for bone marrow analysis.

LOT #1

Received 2-21-88

Released 3-24-88

	Received	DOA	Died in quarantine	released	Mortality
African Greys	647	0	15	632	2.3%

Most of the birds in lot #1 were young and 22 were still being hand fed in quarantine. Birds were fed pelleted parrot diet containing 1% chlorotetracycline (CTC). Losses were low in quarantine and the birds were in good condition upon release. No hemagglutinating viruses (HAV) were isolated from samples submitted to the USDA National Veterinary Services Laboratory (NVSL) during quarantine.

Losses began in the flock approximately 10 days after release. Mortality continued at the rate of 1 bird every two days for approximately 2 months (37 birds, 5.8% of those released died post quarantine). Many birds were sold immediately post release and few complaints were received.

Salmonella typhimurium was isolated from the liver of some birds on post mortem exam. Many birds exhibited signs of hepatitis from which *Salmonella* could not be isolated. *E. coli* and *Enterobacter* sp. were often isolated from the gut of affected birds but were not associated with septicemia. *Aspergillus* was also isolated from some birds. The entire flock was vaccinated with a commercially available salmonella bacterin (a). No adverse effects were associated with the vaccine but efficacy could not be determined. The addition of chloramphenicol to the feed (100 mg/bird/day) slowed the death rate but did not stop mortality. (b) Following chloramphenicol therapy however the salmonella was seldom recovered despite the continued mortality due to hepatitis.

Post mortem examination almost uniformly revealed hepatitis. In birds which died in the acute stage of the disease the liver was enlarged, friable and streaked with yellow. In birds which survived the acute stage but died with chronic hepatitis the liver was usually normal in size but was hard and dark colored often appearing dark green or black. Hepatitis was initially suspected to be due to *Salmonella* septicemia.

Histologic examination of birds dying with acute disease revealed multifocal hepatic necrosis, and splenic hyperplasia to be consistent findings in 3 birds which were negative on liver culture.

Suspicion of aflatoxicosis prompted the submission of corn samples to the Florida State Diagnostic Laboratory for analysis. No aflatoxins were found and no other birds in the facility (approx 5,000 birds) fed corn from the same lot developed similar symptoms. It is possible however that the birds were exposed to aflatoxins in the country of origin prior to importation.

Ophthalmic involvement was evident and was one of the first signs observed in an ill bird. Fixed dilated pupils were commonly observed in individuals in the flock at a distance. On closer examination retinal hemorrhages were observed with these occurring most often around the pecten. Uveitis, hypopyon, and fibrous exudates in the anterior chamber would follow. Posterior and anterior synechia were often observed in birds which recovered from the acute stage of the disease.

Approximately 37 birds were lost post quarantine. Total losses were estimated at 52 birds (8.0%).

LOT #2

Received 5-16-83

Released 6-15-83

	Received	DOA	Died in + quarantine	Released	Mortality
African Greys	597	4	99	494	17.2%

The birds were in poor condition and 70-75% were thin on arrival. Due to the Salmonellosis found in the first lot, these birds were treated with chloramphenicol upon arrival in quarantine. Birds were fed 1% chlorotetracycline pellets and they ate well. Losses were approximately four birds (0.6%) per day initially. Many (approximately 60%) of these had gross lesions of Aspergillosis. The remainder had pneumonia and hepatitis. On the tenth day of quarantine all of the birds were put on flucytosine (100 mg/bird/day) in the feed.(c) After starting the flucytosine the incidence of gross evidence of Aspergillosis decreased, but many birds continued to die with gross lesions of hepatitis. The flock was treated for a total of ten days with flucytosine. Mortality continued with a low percentage showing gross fungal lesions so the birds were treated with Nitrofurazone at the rate of 1 teaspoon per gallon.(d) Parenteral therapy was attempted in affected individuals, with a variety of antibiotics, but this did not significantly alter the course of the disease. Mortality continued at the rate of 3-6 birds per day until release. No hemagglutinating viruses were isolated from samples submitted to NVSL.

Post release, the death rate increased to 10-15 birds per day. All birds were vaccinated for Salmonella but salmonella was not isolated from any birds in the flock. Of 20 birds posted the incidence of gross lesions were as follows; pneumonia - 18(90%),

hepatomegaly, friable liver, yellow mottling - 12(60%), anemia - 4(20%), petechia on viscera - 3(15%), grossly evident lesions of Aspergillosis - 2 (10%), hemorrhagic colitis - 1(5%).

A variety of bacteria were isolated from the intestines of the birds including *E. coli*, *Klebsiella* sp, *Enterobacter* sp. and *Pseudomonas*. In a few cases *Pseudomonas* was isolated from the lungs. Most of these organisms exhibited a wide range of antibiotic susceptibility.

Clinical course was approximately four days from the time clinical signs were first exhibited until death. First signs observed were usually abnormal appearance in the eyes, followed by depression, weakness, and weight loss. Many birds exhibited paralysis which was usually unilateral but often bilateral. After the development of paralysis death usually ensued within several hours to one day. In many birds paralysis was accompanied by edema of the legs and often the head. Skin in these areas was often weeping and cool to the touch. A bloody to dark brown nasal discharge was often evident. Dyspnea was common prior to death. Feces were usually formed and normal in color however urates were often yellow. Ophthalmic lesions were similar to those observed in lot # 1. These clinical signs were also observed in Lot #1 but were less frequent and less severe.

Histopathologic lesions were also similar to those observed in birds from lot #1. Hepatic lesions included congestion with multiple foci of coagulation necrosis, diffuse lipidosis of viable parenchyma. Mild RE cell hyperplasia, lymphoid depletion were observed in the spleens. Diffuse subacute mycotic bronchiolitis and pneumonia was observed with thrombosed large vessels secondary to fungal invasion and penetration of vessel walls.

Cryptosporidium or a similar organism was observed in the intestine of one bird but was non reactive. Fungal cerebritis was found in one bird which was paralyzed prior to death and mucor was isolated from its lung. (19)

Due to the known bone marrow suppression of flucytosine and the anemia and leukopenia observed in the birds therapy was discontinued and the birds were placed on hematinics (d) and lactobacillus supplements (e). Gentian violet (f) was added to the feed to reduce possible fungal infections from moldy feeds. Again the same feeds were fed to a large number of other birds with no corresponding problems.

Bone marrow smears from five birds with anemia and leukopenia were examined by a hematologist familiar with drug induced bone marrow suppression. Bone marrows were cellular and actively producing cells in both the erythrocytic and granulocytic cell lines.

Anemia and leukopenia were believed to be due to intravascular destruction rather than bone marrow suppression.

Twenty birds were checked for blood parasites and all were found free of hemoproteus, microfilaria and trypanosomes.

Hematology and blood chemistry values were usually normal in asymptomatic birds except for leukocytosis observed on estimated WBC count in some birds. All birds were often anemic and leukopenic. Examination of absolute heterophil and lymphocyte number revealed that leukopenic birds were usually heteropenic while absolute lymphocyte counts often remained normal. Anemic birds usually had microcytic, normochromic anemia. SGOT and LDH were elevated in birds which were terminally affected. Figures #1,2 and 3.

Approximately 212 birds were lost post quarantine (42% of released birds). Total losses were estimated at 315 birds (52%).

LOT #3

Received 7-30-83

Release 9-7-83

	Received	DOA	Died in quarantine	Released	Mortality
African Greys	197	1	78	118	40%.

Birds arrived in fair condition and losses were low in the first few days. Heavy losses started after the 15th day and continued for about ten days. Hematinics and Lactobacillus were used in addition to parrot pellets containing CTC. Hemagglutinating viruses were isolated from 3 of 6 submissions by NVSL and typed as Paramyxovirus -3. Clinical and post mortem findings were consistent with those observed in Lot #2. Despite the finding of Aspergillosis in the station flucytosine was not used. Feed samples from Ghana, which were collected from the shipping boxes, were refrigerated until the birds were released, and found negative for Aspergillus post quarantine.

African Greys were quarantined with cockatiels and lovebirds. The losses in these birds were higher than normal for these species from this supplier (cockatiels - 11% mortality, lovebirds - 5% mortality). The cause of this mortality was not determined. No viruses were isolated from cockatiels submitted to the Florida State Diagnostic laboratory post quarantine.

LOT #4

Received 8-13-83

Released 9-21-83

	Received	DOA	Dead in quarantine	Released	Mortality	Total mortality
African Greys	594	122 (20.5%)	83	389	17.5%	43%
Jardines	69	1	6	62		10%

The birds were overheated in transit, resulting in many DOA. Approximately one half of the birds were thin on arrival with many requiring tube feeding. Birds were dying with gross evidence of hepatitis in the first few days. On the 10th day the flock was started on chloramphenicol in the feed. Losses were approximately three to four birds a day. Some birds began to show paralysis, swollen faces and uveitis. Losses continued at the rate of 1 to 3 per day until release.

Although minimal gross lesions of Aspergillosis or Mucor mycosis were observed in these birds, hepatitis and pneumonia were evident. PMV 3 was isolated from 6 of 9 submissions by NVSL. (8)

Approximately 12 days post release losses started in the Jardines from Lot #4. these losses occurred at the same time in birds which were in the facility and those which had been sold. Bacterial and fungal cultures were insignificant. A reovirus was isolated from one Jardine parrot and associated with hepatitis. (9) A Reovirus was also isolated from African Greys in this lot submitted to Cornell University. (3)

Post release mortality figures were not separated for Lots #3 and #4 as they were released almost simultaneously. Of the 507 birds released approximately 81 (15.9%) were lost post release.

DISCUSSION

The clinical signs, clinical course of disease and post mortem findings in these four lots of birds suggest a common disease syndrome. A single common pathogen which can explain the mortality in all four stations was not evident. Reoviruses have recently been suspected as a cause of hepatitis in other psittacine species as well as African Greys. In a recent review of histopathologic sections from birds in lots #1 and #2 lesions were consistent with those of confirmed cases of hepatitis associated with Reovirus. Reovirus was isolated from birds in lot #4. (6,12)

Reoviruses have been associated with enteric conditions such as cloacal pasting and ulcerative enteritis, acute and chronic respiratory disease, pericarditis and hydropericardium, anemia, inclusion body hepatitis, abnormal feather development, decrease in egg production and death in poultry. Many of these reported syndromes have not been reproduced experimentally. The most common disease associated with Reoviruses in poultry is tenosynovitis. (5)

It is apparent however that a synergism may exist between the Reovirus and other pathogens which increases the severity of the disease syndrome. A synergism between Reoviruses and *Aspergillus* has been observed in poultry. (13)

The role played by PMV-3 is unknown however the frequency of isolation of this virus should prompt further laboratory investigation. In lots #3 and #4 mortality was high with a lower incidence of concurrent Salmonellosis or Aspergillosis and PMV 3 was isolated in both lots by NVSL. This may be an indication of synergism between these two viruses.

In a summary of viral isolations made at NVSL from October 1973 to September 1981, hemagglutinating viruses (HAV) were isolated from 24.5% of the lots imported through commercial stations. Of 1,558 HA isolates which were selected for identification 534 (34%) were typed as PMV-3. Of these 534 PMV-3 isolates, 255(47%) were isolated from Parrots. Parrots were distinguished from Parakeets, Cockatiels and Lovebirds but were otherwise unidentified. In the same eight year period Reoviruses were isolated from 25 lots (4.1%) of psittacines. The incidence of Reovirus may in fact be much higher as they are not HA viruses and therefore may be reported as negative. The primary emphasis of viral isolation in imported birds is to detect the entrance of Velogenic Newcastle Disease and poultry lethal strains of Avian Influenza. As both of these viruses are HA viruses others are often not recognized or identified. (11,16)

Alexander also reported the frequent isolation of PMV-3 in psittacine birds and from African birds entering Great Britain. (1)

The other pathogens involved appear to play a significant role in this syndrome and are probably responsible for the severity of the disease in some lots. Aspergillosis and Salmonellosis have long been incriminated for as causes of mortality in imported African Greys.

The immunosuppressive effect of chlorotetracycline has been demonstrated in turkeys and may also be contributory to the fungal and bacterial superinfections observed in these parrots. In other groups of African Greys, from countries other than Ghana, which are handled identically the results are vastly different emphasizing that the problem is not solely one of environment but one of infection which may be aggravated by environment. The toxins of *Aspergillus flavus* have also been reported as immunosuppressive in poultry. (5,10)

Blood dyscrasias, especially anemia and leukopenia were common findings in this disease syndrome. The specific cause of these dyscrasias is unknown. While birds were treated with both chloramphenicol and flucytosine, which are known to be potentially toxic to bone marrow in man, there was no indication that this occurred in the African Greys. In many leukopenic birds the absolute lymphocyte counts remained relatively normal while the birds had severe heteropenia. The anemia and heteropenia were probably due to overwhelming infection and intravascular destruction.

Ghana African Greys often arrive in quarantine in poor condition. Ghana is a very poor country and food and water supplies are not ideal. In addition the trip is long with stops in Europe. This may contribute to the severity of the disease syndrome.

Prevention of the syndrome will most likely be achieved only by vaccination. Unfortunately African Greys commonly fail to seroconvert following exposure to antigens such as chlamydia and Newcastle vaccines. The development of vaccines and the determination of their efficacy therefore may be a very difficult task. (9,15)

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- a. Salmonella Dublin-Typhimurium bacterin - Bovine isolate: Colorado Serum Company, 4950 York St., Denver Colorado, 80216.
 - b. Chloramphenicol capsules - generic
 - c. Ancobon - Roche, Division of Hoffmann-La Roche Inc., Nutley, New Jersey, 07110
 - d. Nitrofurazone - 9.3% soluble powder, Wholesale Veterinary Supply, Inc., Rockford Illinois 61111
 - e. Hemoplex - Norelco, P.O. Box 1622 SSS, Springfield, Missouri 65805.
 - f. Avi-culture 1 Billion - Wm V Reichert & Son, 1523 Potters road, Park Ridge, Illinois, 60068.
 - g. GV-11 - Norelco, P.O. Box 1622 SSS, Springfield, Missouri 65805.
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Figure #1

HEMATOLOGY AND BLOOD CHEMISTRY VALUES FOR BIRDS FROM LOT #2 POST RELEASE

HEMATOLOGY

Bird#	1	2	3	4	5	6	7	8	9	10	11
PCV	59%	50%	58%	52%	32%	45%	54%	52%	55%	59%	51%
TP	4.6	4.0	4.0	4.0	3.0	3.0	3.6	3.8	3.8	4.2	4.0
ESTIMATED WBC	>	>	N	>	>	N	N	<	N	N	>
Polychromatophilic Index	1	1	1	2-3	3	2	2	1	2	2	1

Bird#	12	13	14	15	16	17	18	19	20
PCV	48%	49%	35%	32%	30%	28%	30%	30%	19%
TP	4.0	3.4	5.2	4.2	3.0	4.4	2.8	4.0	3.0
WBC	N	N	<	<	<	<	<	<	<
Polychromatophilic Index	1	4	1	1	1	2	1	1	1

BLOOD CHEMISTRY PROFILES

Bird #	9	12	14	15	16	18	19	20
Alb	1.1	1.0	.9	.7	.8	.6	1.0	.7
Alk P	37	21	31	16	26	10	21	11
Ca	6.4	8.3	7.0	7.4	8.0	6.1	8.5	7.8
Chol	366	220	249	280	170	108	224	156
Gluc	204	192	324	80	190	204	223	200
SGOT	85	189	100	1518	983	231	99	841
LDH	171	173	84	1418	749	91	177	956
Phos	3.8	4.0	4.5	7.1	2.5	5.7	5.6	6.9
T.P.	3.3	3.7	4.0	3.6	2.7	2.7	3.6	3.4
U.A.	2.7	4.2	7.5	2.6	3.0	12.0	4.0	25.5
Glob	2.2	2.7	3.1	2.9	1.9	2.1	2.6	2.7

Continued.

Figure #1 continued

All blood samples were collected two days after release from quarantine. Birds had recieved no drugs other than CTC for approximately 10 days prior to sampling.
Clinical signs exhibited by birds sampled.

Birds 1-11 were showing no signs of disease

12. Weight loss but otherwise normal.

13. Young bird which had weight loss, vomiting, a dark colored thick nasal discharge. The bird was very depressed and near death

14. The bird was thin and depressed.

15. The bird was emaciated, depressed, had uveitis, vomiting nasa discharge and was near death.

16. The bird was thin, vomiting and depressed and had fixed pupil

17. The bird was very thin, had a bloody nasal discharge, uveitis and paresis of the legs.

18. The bird was depressed and thin.

19. The bird was dyspenic, thin and depressed.

20. The bird was thin and near death with a bloody nasal dischang and vomiting.

Estimated WBC - N = normal, < = decreased, > = increased.

Polychromatophilic index as described by Dein (3).

Figure #2

HEMATOLOGY AND BLOOD CHEMISTRY VALUES FROM BIRDS IN LOT # 2 - 1 WEEK AFTER RELEASE FROM QUARANTINE.

Bird #	1	2	3	4	5	6
WBC	7.2	3.2	0.7	1.6	1.6	.4est
RBC	2.82	1.94	2.67	1.38	2.70	2.04
HGB	9.3	7.2	8.9	4.7	10.0	8.0
HCT	34%	29%	30%	16%	33%	26%
MCV	120	149	112	116	122	127
MCH	32.9	37.1	33.3	34	37	39.2
MCHC	27.3	24.8	29.6	29.4	30.3	30.8
PLT	ade	ade	dec	ade	ade	ade
HET	4	2	12	2	2	0
Absolute	.288	.064	.084	.032	.032	0
Lym	89	93	85	95	98	100
Absolute	6.4	2.76	.59	1.52	1.56	.4
Mono	4	2	2	2	0	0
Eos	0	1	1	1	0	0
Bas	3	2	0	0	0	0
SGOT	88	459	153	301	175	545
LDH	205	641	158	168	143	489

Birds had received flucytosine for six days at the time they were sampled. Aspirations for bonermarrow cytology were collected from the femur at this time.

Bird # - Clinical signs

1. Asymptomatic
2. Fat depressed bird with swollen left leg. Post mortem lesions included pneumonia in left lung, liver enlarged with gelatinous appearance and scattered white foci.
3. Bird was vomiting, paralyzed in left leg, thin, and depressed. Post mortem lesions included small liver, normal lungs, congested and mottled kidneys.
4. Bird was paralyzed in left leg and thin but alert.
5. Bird was thin, depressed and vomiting.
6. Bird had bilateral paralysis, was thin, very depressed, and near death.

Figure #3

HEMATOLOGY AND BLOOD CHEMISTRY VALUES FOR CONVALESCING BIRDS - L
#2

Bird #	1	2	3	4
WBC	2.0	5.0	10.0	<1.0
RBC	3.64	3.38	2.58	1.32
HGB	16.7	14.4	12.7	5.6
HCT	49.0	43.0	37.0	15.0
MCV	130	126	145	115
MCH	44	43	48	43
MCHC	34	34	34	38
Heterophils	16	52	63	0
Absolute #	.32	2.6	6.3	0
Lymph	84	48	38	100
Absolute #	1.68	2.4	3.8	1.0
Poly	+2	+1	+2	+3

Bird #	1	2	3	4	5
Alb	1.6	1.3	1.3	.9	1.1
A P	28	46	32	15	8.7
Ca	8.2	9.1	9.5	8.7	8.8
Chol	306	370	319	227	234
Gluc	299	227	254	268	147
SGOT	80	79	63	510	110
LDH	276	434	213	106	602
Phos	3.2	3.4	4.1	5.7	7.8
T.P.	3.7	4.0	5.0	3.4	7.2
U A	11	5.2	9.0	9.4	10.5
Glob	2.1	2.7	3.7	2.5	6.1

Bird # - Clinical signs

1. Uveitis, but otherwise clinically normal. Fecal culture negative for Salmonella.
2. Uveitis, but otherwise clinically normal. Fecal culture negative for Salmonella.
3. Uveitis, but otherwise clinically normal. Fecal culture negative for Salmonella.
4. Bird was thin, pale, weak, ataxic, very weak in left leg.
5. Bird was thin, pale, weak and dyspneic.

Figure #4

NORMAL HEMATOLOGY AND BLOOD CHEMISTRY VALUES FOR AFRICAN GREY PARROTS

	Hawkley (7)	Roskopf (14)	Inter American Lab. (2)*
WBC X 10 ³	3.3-10.0	5-11	
RBC X 10 ⁶	1.0-3.6	2.4-4.5	
HGB g/dl	14.2-17.0		
HCT	48%-51%	43%-55%	
MCV um	137.2-154.1		
MCH pg	41.9-52.8		
MCHC g/dl	28.9-34.0		
Heterophils	56%-76%		
absolute #			
heterophils	1.85-7.3		
Lymphocytes	9%-41%		
Absolute #			
lymphocytes	0.78-2.11		
Albumin g/dl			1.5-2.2
Alkaline phosphatase IU/L			40-100
Calcium mg/dl			9.0-11.0
Cholesterol mg/dl			149-225
Glucose mg/dl		190-350	225-309
SGOT IU/L		100-350	90-160
LDH IU/L		150-450	100-200
Phosphorus mg/dl			2.7-6.8
Total protein g/dl		3.0-5.0	3.8-4.6
Uric Acid mg/dl		4.0-10.0	6.2-10.0
Globulin mg/dl			1.9-3.4

+ Unpublished normals - Not specific to African Grey Parrots

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