

# OUTBREAK OF A PAPOVA-LIKE VIRAL INFECTION IN A PSITTACINE NURSERY - A RETROSPECTIVE VIEW.

Susan L. Clubb, DVM  
Pet Farm Inc.  
5400 NW 84 Ave  
Miami, Florida 33166

Richard B. Davis, DVM  
Department of Avian Medicine  
College of Veterinary Medicine  
University of Georgia  
Athens, Georgia 30605

An outbreak of a viral fledgling disease occurred in a nursery of handfeeding psittacine birds in May and June of 1982. The histopathological lesions were similar to those of Budgerigar Fledgling disease (BFD). (Davis et al 1981). The causative agent of BFD is a papovavirus. Surviving birds showed a positive titer to the BFD virus by fluorescent antibody virus neutralization test for BFD virus. The virus was not isolated despite multiple passages in budgerigar embryo fibroblasts and chicken embryo kidney cells. Therefore it will be referred to as a papovavirus like fledgling disease (PVLFD). This paper will describe the events of the outbreak and follow the flock over the next two years.

## HISTORY

On May 18 1982 a 20 day old sun conure died acutely in a psittacine nursery. The bird was ill for only 1 day prior to death and clutch mates appeared to be normal. Two days later a 6 week old blue & gold macaw and another 20 day old sun conure from a different clutch died. These birds had no previous illness and were being handfed and therefore observed closely at least three times daily. Fixed tissues were submitted for histopathology and a preliminary diagnosis of a karyomegalic inclusion body disease similar to papova virus infection in budgerigars was made.\*

At the time of the outbreak 55 psittacine birds of 16 species were at risk. They were divided into two separate nurseries on separate premises but there was contact between the two nurseries. In nursery # 1 the very young conures and the larger species were housed. As the conures reached the age at which they could be fed only three times daily they were transferred to nursery #2. The outbreak occurred almost simultaneously in the two nurseries. In Nursery #1 36 birds of 12 species were at risk of which 15 died (41% mortality). In Nursery #2 19 birds of 5 species were at risk of which 6 died (31% mortality). Figure #1. There were no budgerigars at the farm on in the nurseries.

\* The authors acknowledge the assistance of Dr. David Graham, Cornell University for histopathologic diagnosis.

On the seventh day of the outbreak the histopathological diagnosis was made. At this time 11 birds had died and the birds were spread out in order to stop the spread of the disease. Exposed birds were placed in two new premises which were completely disinfected and were supplied with new housing and feeding utensils. On the 13th and 14th days after they were moved 3 macaws in 3 separate premises died (1 19 day old hyacinth macaw which remained in Nursery #1, a 38 day old scarlet X greenwing macaw in Nursery #3, and a 40 day old scarlet X greenwing macaw in Nursery #4). This indicated that the incubation period may be 14 days or longer.

After June 8 1982 no more acute deaths occurred however many birds in the group were ill. Of 11 birds (4 species) which were chronically affected, 4 died (36%). Figure #2 No losses occurred after June 16, 1982.

#### CLINICAL SIGNS

Clinical signs ranged from peracute death to chronic debilitation and renal failure. The first signs of illness were reduction in daily weight gain and prolonged crop emptying time. This was often followed by vomiting and the observation of reverse peristaltic waves in the crop. Depression followed and most birds had a glassy appearance in the eyes. Anorexia and dehydration were common. When treatment was attempted with subcutaneous or intramuscular injections the birds would hemorrhage severely at the injection site. If a feather was plucked the bird would also hemorrhage excessively and this was used as a preliminary diagnostic test in order to detect and isolate affected birds. One double yellow headed amazon exhibited posterior paresis which progressed to paralysis and death within 18 hours after the appearance of the first symptoms. Most birds died within 24 hours after the appearance of symptoms.

In birds which were chronically affected the most consistent clinical signs were subnormal weight, maldigestion and slow gut transit time, polyuria, secondary candidiasis, abnormal feathering, depression and failure to self feed at a normal age. All birds which recovered appeared clinically normal after reaching adult size except one sun conure which had abnormal feather texture. Two white crown pionus which died 28 and 29 days after the first death had cystic kidneys and died in renal failure.

Serum chemistry profiles revealed extreme elevations of liver enzymes often as much as ten fold. In a 8 week old double yellow headed amazon the LDH was 2129 (normal 100-200), SGOT was 2116 (normal 90-160), and alkaline phosphatase was 734 (normal 40-100). LDH was the most consistently elevated enzyme.



Age at death ranged from 20 days to 56 days.

#### POST MORTEM LESIONS

Hepatomegaly, splenomegaly, petechial and suffusion hemorrhages on the surfaces of the intestines and the myocardium, and ascites were consistently observed.. Skeletal muscle and myocardium were very pale. The liver and spleen were friable and often mottled. Hemorrhage was extensive at injection sites. Swollen kidneys were evident in many birds.

On histological examination slightly basophilic intranuclear inclusion bodies similar to those of BFD were found in several tissues. Multifocal hepatic necrosis was common with occasional karyomegalic inclusions in the necrotic regions. Inclusion bodies were also observed in the spleen, small intestine, kidney, lung, esophagus, proventriculus, and adrenal gland.

#### SEROLOGY

PULFD recovered and exposed birds were tested serologically using a fluorescent antibody virus neutralization test as described by Davis et al (1983). Serum samples were submitted from 7 birds which survived the acute stage of the outbreak and all had a positive reaction. Figure #3.

#### EPIDEMIOLOGY

While most of the PULFD losses in the outbreak occurred in the nurseries some mortality also occurred in the nests of the breeding adults. Nest deaths however occurred only in the nests of a group of sun conures which had been placed into the breeding collection approximately 4 months previously. The sun conures were imported from Guyana and 20 pair were acquired for the farm approximately two weeks after release from quarantine. The birds were quarantined at the farm for an additional 2 months and all appeared normal prior to placing in breeding cages. They were housed in flights or cages and all were in one building. Total time elapsed from leaving Guyana until the outbreak was 7 1/2 months. One adult sun conure from the same group died on June 2 following a three week illness. Karyomegalic inclusion bodies were observed in the spleen of this bird. All other adult sun conures remained clinically normal. Any pairs which had nest mortality were eliminated from the breeding collection.

Sun conures from the same imported lot were subsequently tested serologically and found to have positive BFD titers. These birds had no contact with any birds from the breeding farm. Figure #4.

Sun conures from the same imported lot were associated with a second outbreak of PVLFED on a different farm which occurred in May of 1983.

In September of 1983 a group of newly imported sun conures were tested for papova-virus serology. These birds had just been released from quarantine and had not been in contact with any birds in the United States. Of 13 birds tested 3 showed positive titers and 3 were suspect indicating exposure to papovavirus in Guyana or in quarantine. Figure #5

A subsequent BFD serological survey was undertaken at the breeding farm in order to determine the incidence of seropositive breeding birds in the collection. A total of 106 samples were collected in October of 1983 and January 1984. Of these samples 35 (33%) were positive for BFD (BFD titer  $>10$ ), 22 (20.7%) were suspect (BFD titer of 10) and 49 (46.3%) were negative (BFD titer of  $<10$ ). Many of these seropositive birds have raised normal offspring in the two years since the outbreak. Figure #6

Complete disinfection of the premises was impossible as most of the birds are housed outdoors in suspended cages. Wooden nest boxes are also impossible to disinfect totally. The sun conures were housed in flights with concrete floors and this area was disinfected with a phenolic disinfectant.(A) Sun conures which had dead babies in the nest (two pair only) were eliminated from the breeding collection. Routine cleaning and maintenance was unaltered. The next year however provisions were made for separate nursery facilities for conures and larger birds. No cases of PVLFED have been diagnosed since June 1982.

## DISCUSSION

PVLFED resembles BFD in all respects except ease of isolation of the causative agent. The disease appears to be self limiting. Adult birds exposed to the virus were shown to gain a high titer and subsequently raise normal young. Offspring of adults with titers were shown to be sero-negative.

High mortality occurs in psittacines of many species when exposed to the virus at a young age. In this nursery good hygiene was practiced and the birds were not stressed as they were in the nursery from a very young age, many since hatching. Disinfected feeding utensils were used for each individual bird however time constraints did not permit hand washing between each bird. Birds were kept on corn cob bedding and fecal contaminated dust particles could have resulted in the airborne spread of the disease.

(A) One Stroke Environ - Vestal Products



The fact that no deaths occurred in subsequent years to sero-positive adults is encouraging. Perhaps the virus was naturally eliminated from the environment when the susceptible population (the babies) was no longer available. Seasonal breeding is normal in breeding farms for large psittacines. In contrast, in budgerigar farms, breeding continues year round and the susceptible population that supports the virus is always present. Control of the disease should therefore be only a matter of stopping the breeding cycle and forcing the birds to remain dormant for a period of time. Following this rest the adult population should be immune. This method has already been used successfully in one budgerigar farm (Davis 1983).

Deliberate exposure of adults to the virus followed by a nesting period may be a safeguard against future outbreaks of BFD and PVLF. This practice is currently used to control Avian Encephalomyelitis in poultry. Further controlled research is needed in order to prove the validity of this theoretical means of control. The fact that some sun conures from the suspect lot were associated with a second outbreak of PVLF in June of 1983 (19 months after release from quarantine) indicates the need for investigation of a possible carrier state.

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Figure #1

BIRDS AT RISK IN NURSERIES AND MORTALITY RATES

Nursery #1

number	species	died
2	Hyacinth macaws	1
3	Blue & gold macaws	1
3	Military macaws	0
1	Buffons X greenwing macaws	0
2	Scarlet X greenwing macaws	2
2	Yellow naped amazons	2
1	Double yellow headed amazon	1
1	Yellow crowned amazon	0
2	Hispanolian amazons	0
3	Slenderbill conures	0
14	Sun conures	8
1	Sulphur crested cockatoo	0
1	African grey parrot	0
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36		15 (41%)

Nursery #2

6	Gold capped conures	0
3	Jenday conures	0
5	Sun conures	3
4	White crowned pionus	2
1	Cockatiel	0
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19		6 (31%)

Figure #2

BIRDS WHICH DEVELOPED CHRONIC ILLNESS.

number	species	died
3	White crown pionus	2
5	Sun conures	2
2	Slenderbill conures	0
1	Hyacinth macaw	0
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11		4

Figure #3

BFD SEROLOGY - BIRDS FROM THE NURSERIES WHICH SURVIVED THE ACUTE STAGE OF THE DISEASE (titer of 16 or greater is positive)

species	BFD titer	
Sun conure	16	
Slenderbill conure	28	
White crown pionus	28	
White crown pionus	256	Bird died of kidney failure
White crown pionus	256	Bird died of kidney failure
Blue & gold macaw	512	
Hyacinth macaw	320	1 year post outbreak

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Figure #4

BFD SEROLOGY - BIRDS FROM SUSPECT LOT WHICH HAD NO EXPOSURE TO BIRDS FROM BREEDING FARM

1.	Sun conure	10	Suspect
2.	Sun conure	20	Positive
3.	Sun conure	20-80	"
4.	Sun conure	80	"
5.	Sun conure	320	"

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Figure #5

BFD SEROLOGY - NEWLY IMPORTED SUN CONURES FROM GUYANA

# tested	BFD titer	
7	Sun conures <5 - 5	Negative
3	Sun conures 10	Suspect
2	Sun conures 20	Positive
1	Sun conure 80	Positive

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Figure #6

SEROPOSITIVE BREEDING PAIRS WHICH HAVE RAISED NORMAL YOUNG SINCE THE OUTBREAK. (including suspects and pairs with only one member positive)

PAIR NUMBER	SPECIES	MALE TITER	FEMALE TITER
3-26	Blue and gold macaws	20	10
3-20	Blue & gold macaws	5	10
3-19	Blue & gold macaw	10	5
3-33	Hyacinth macaws	20	-
3-32	Hyacinth macaws	10	10-20
3-34	Scarlet X greenwing macaws	5	20
3-28	Scarlet X Blue & gold macaws	10	5-10
3-17	Military macaws	10	20
E-9	Red bellied macaw	20	5
	Nobles macaw	5-10	80
C-4	Nobles macaws	10	20-80
2-17	Nobles X hahns macaws	80	20-80
C-8	White crown pionus	-	10
C-1	White crowned pionus	10	10
C-2	Jenday conures	20	80
C-9	Sun conures	10	20
2-1 *	Sun conures	20	20
2-1 *	Sun conures	20	80
A-26	Sun conures	10	10-20
B-1	Sun conures	5	80-320
2-13	Sun conures (Guyana)	10	10
B-2	Green cheeked conures	80	5
B-4	Gold capped conures	10-20	20
2-15	Gold capped conures	<5	10-20
E-14	Queen of bavaria conure	20	20
E-12	Eclectus parrots	20-80	5-10
D-3 *	Eclectus parrots	320	-
	Sulphur crested cockatoo	-	20
C-5	Indian ringnecks	20-80	20-80

\* Off spring of these pairs were tested and found seronegative at weining. (total 8 birds)

titer of 10 = Suspect, Titer >10 = Positive



Figure #7

SCIENTIFIC NAMES OF SPECIES MENTIONED IN PAPER

Hyacinth macaw	( <i>Anodorhynchus hyacinthinus</i> )
Blue and gold macaw	( <i>Ara ararauna</i> )
Scarlet macaw	( <i>Ara macao</i> )
Military macaw	( <i>Ara militaris</i> )
Greenwing macaw	( <i>Ara chloroptera</i> )
Buffons macaw	( <i>Ara ambigua</i> )
Red bellied macaw	( <i>Ara manilata</i> )
Hahns macaw	( <i>Ara nobilis nobilis</i> )
Nobles macaw	( <i>Ara nobilis cumanensis</i> )
Double yellow headed amazon	( <i>Amazona ochrocephala oratrix</i> )
Yellow naped amazon	( <i>Amazona ochrocephala auropalliata</i> )
Yellow crowned amazon	( <i>Amazona ochrocephala ochrocephala</i> )
Hispanolian amazon	( <i>Amazona ventralis</i> )
Eclectus parrot	( <i>Eclectus poratus</i> )
White crowned pionus	( <i>Pionus senilis</i> )
Sulphur crested cockatoo	( <i>Cacatua galerita eleonora</i> )
Sun conure	( <i>Aratinga solstitialis</i> )
Jenday conure	( <i>Aratinga jandaya</i> )
Green cheeked conure	( <i>Pyrrhura molinae</i> )
Gold capped conure	( <i>Aratinga auricapilla</i> )
Gold crowned conure	( <i>Aratinga aurea</i> )
Queen of bavaria conure	( <i>Aratinga guarouba</i> )
Red masked conure	( <i>Aratinga erythrogenys</i> )
Slenderbill conure	( <i>Enicognathus leptorhynchus</i> )
Cockatiel	( <i>Nymphicus hollandicus</i> )