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## Avian Paramyxoviruses from Migrating and Resident Ducks in Coastal Louisiana

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ABSTRACT: Cloacal and tracheal swabs were collected from 1,409 hunter-killed ducks in Cameron Parish, Louisiana, during the 1986 and 1987 waterfowl seasons. Thirty avian paramyxoviruses (PMV's) were isolated from 605 blue-winged teal (Anas discors), 75 mottled ducks (A. fulvigula), 375 gadwalls (A. strepera), 334 green-winged teal (A. crecca), and 20 mallards (A. platyrhynchos). Prevalence of PMV decreased (P = 0.042) from September (4%) through November (2%) to December and January (1%). Juveniles had a higher prevalence of PMV (P < 0.0001) than adults. An isolate from resident mottled ducks documents transmission of PMV's on the coastal wintering areas of Louisiana. The four serotypes isolated, PMV-1, PMV-4, PMV-6, and PMV-8, were typical of PMV's commonly associated with free-living waterfowl.

Key words: Avian paramyxovirus, survey, prevalence, ducks, serotypes, isolates.

Although there are numerous reports of avian paramyxovirus (PMV) (Paramyxoviridae: Paramyxovirus) isolation from wild waterfowl, the epizootiology of these viruses in these diverse populations has received little attention. This is partly because most isolations of PMV from waterfowl have been adjunct to surveillance of these populations for avian influenza viruses (AIV) (Orthomyxoviridae: Influenzavirus) (Alexander, 1986).

The taxonomy of avian PMV's is not clearly established, but nine serotypes designated PMV-1 through PMV-9 are generally recognized (Alexander, 1987). With the exception of PMV-5 and PMV-9, all serotypes have been reported from wild ducks and geese (Ottis and Bachmann, 1983; Nettles et al., 1985; Alexander, 1986; Hinshaw et al., 1985, 1986).

Except for a single report of Newcastle disease (PMV-1) related mortality in common teal (*Anas crecca*) in Iran (Bozorgmehri-Fard and Keyvanfar, 1979), these viruses are not currently associated with either morbidity or mortality in wild waterfowl. Clinical disease or production problems have been reported with PMV-1, PMV-2, PMV-3, and PMV-6 infections in domestic fowl and turkeys (Lipkind et al., 1982; Lang et al., 1975; Alexander, 1986), but a clear connection between PMV in waterfowl and introduction of these viruses to domestic poultry flocks has not been established.

As a result of virus isolation attempts for AIV (Stallknecht et al., 1990), we isolated avian PMV's from resident and migratory wild ducks in coastal Louisiana. Prevalence estimates for PMV in these ducks are compared in relation to annual and seasonal variation, age class, and species.

Ducks were surveyed from lands adjacent to Rockefeller State Wildlife Refuge (Louisiana Department of Wildlife and Fisheries, Cameron Parish, Louisiana, USA; 20°45'N, 92°45'W). Habitat was representative of coastal Louisiana and included a diversity of marsh types ranging from fresh to salt-water. Species selected for this surveillance included blue-winged teal (A. discors), mottled duck (A. fulvigula), green-winged teal (A. crecca), gadwall (A. strepera), and mallards (A. platyrhynchos). Sample collection and virus isolation methods have been described (Stallknecht et al., 1990). Briefly, cloacal and tracheal samples were collected from hunter-killed ducks during the 1986 and 1987 waterfowl hunting seasons. All birds were sampled on the morning they were killed and were aged based on plumage characteristics (Carney, 1964). Samples were transported from the field on wet ice (<12 hr collection and transport time) or liquid nitrogen (>12 hr collection and transport time) and were stored at -70 C until processed.

For virus isolation, samples were thawed, vortexed, and centrifuged at  $1,500 \times g$  for 20 min. Supernatant was inoculated (0.3 ml/egg) via the allantoic route into four 9-day-old specific pathogen free (SPF) embryonated chicken eggs (ECE). Amnio-allantoic fluid (AAF) from eggs was harvested at 72 hr and was tested for hemagglutination. For samples testing negative by hemagglutination, AAF was repassed in four additional ECE. Serotypes of all hemagglutinating viruses were determined with a hemagglutination-inhibition test (Committee on Standard Method for the Hemagglutination and Hemagglutinin-Inhibition Test for Newcastle Disease, 1975) at the National Veterinary Services Laboratories (NVSL; Science and Technology, Animal and Plant Health Inspection Service, United States Department of Agriculture, Ames, Iowa 50010, USA).

Differences in prevalence among years, species, age-class, and month of sampling were tested by chi-square analysis (SAS Institute, Inc., 1985).

During 1986 and 1987, 1,409 samples were collected for virus isolation. Thirty avian PMV's were isolated. Thirteen (43%) of these viruses were recovered on the first ECE passage.

Fifteen PMV's were isolated from 419 blue-winged teal sampled during September 1986 and 1987 for an overall prevalence estimate of 4% (Table 1). All isolates were from juvenile birds.

One hundred forty-two blue-winged

TABLE 1.Prevalence of avian paramyxoviruses in<br/>early-migrating blue-winged teal, Cameron Parish,<br/>Louisiana, 20 to 27 September 1986 and 19 to 25<br/>September 1987.

Year	Juveniles	Adults	Total	
1986	1/79 (1%)*	0/64 (0)	1/143 (1%)	
1987	14/155 (9%)	0/121 (0)	14/276 (5%)	
Total	15/234 (6%)	0/185 (0)	15/419 (4%)	

\* Number positive/number sampled (% positive).

teal, 266 gadwalls, 222 green-winged teal, 68 mottled ducks, and 20 mallards were sampled during November (Table 2). In 1986, PMV's were isolated from 4 of 242 (<2%) ducks with isolations restricted to blue-winged teal and a mallard. During 1987, PMV's were recovered from all sampled species and were isolated from 8 of 476 (<2%) ducks.

Three PMV's were isolated from 272 (1%) ducks sampled during December and January, which included 145 juveniles and 120 adults (Table 3). All isolations were from green-winged teal.

The prevalence of PMV did not differ by year (P = 0.096) with virus detected in 5 of 430 (1%) ducks sampled during 1986 and from 25 of 979 (3%) ducks sampled in 1987. Prevalence differed by age (P < 0.0001) with virus isolated from 27 of 765 (4%) juveniles and 3 of 574 (<1%) adults.

Species effects were tested only for the November and December and January sample periods (n = 990) to eliminate bias associated with early sampling restricted to blue-winged teal. No difference (P =0.256) between blue-winged teal (2%), gadwall (<1%), green-winged teal (2%), mottled duck (1%), and mallards (5%) was detected.

The prevalence of PMV infection decreased (P = 0.042) progressively with season. Virus was isolated from 15 of 419 (4%) ducks sampled in September, 12 of 718 (2%) ducks sampled in November, and 3 of 272 (1%) ducks sampled in December and January.

Isolated PMV's represented four serotypes including PMV-1 (18 isolates),

Species	Year	Juvenile	Adult	Total
Blue-winged teal	1986	3/56 (5%)*	0/29 (0)	3/85 (4%)
-	1987	1/27 (4%)	0/30 (0)	1/57 (2%)
	Total	4/83 (5%)	0/59 (0)	4/142 (3%)
Gadwall	1986	0/63 (0)	0/39 (0)	0/102 (0)
	1987	2/106(2%)	0/58 (0)	2/164 (1%)
	Total	2/169 (1%)	0/97 (0)	2/266 (1%)
Green-winged teal	1986	0/2 (0)	0/6 (0)	0/8 (0)
	1987	2/108 (2%)	2/106 (2%)	4/214 (2%)
	Total	2/110 (2%)	2/112 (2%)	4/222 (2%)
Mottled duck	1986	b	_	0/27 (0)
	1987	<u> </u>		1/41 (2%)
	Total	-	_	1/68 (2%)
Mallard	1986	1/14 (7%)	0/6 (0)	1/20 (5%)
Total		9/376 (2%)	2/274(1%)	12/718 (2%)

TABLE 2.Prevalence of avian paramyxoviruses in migratory and resident ducks, Cameron Parish, Louisiana,12 to 29 November 1986 and 7 to 28 November 1987.

\* Number positive/number sampled (% positive).

<sup>b</sup> Age not determined.

PMV-4 (seven isolates), PMV-6 (four isolates), and PMV-8 (one isolate). During 1986, isolations were restricted to PMV-1, PMV-4, and PMV-8. During 1987, PMV-1, PMV-4, and PMV-6 were recovered. Viruses recovered from blue-winged teal included PMV-1 (11 isolates), PMV-4 (four isolates), and PMV-6 (three isolates). Serotypes from green-winged teal included PMV-1 (five isolates), PMV-4 (one isolate), and PMV-6 (one isolate). PMV-1 (one isolate) and PMV-4 (two isolates) were isolated from gadwalls. Isolations from mottled ducks and mallards included a single PMV-1 and PMV-8, respectively.

Prevalence and serotype diversity for PMV's observed in this study were consistent with results reported from previous surveys. Ottis and Bachmann (1983) isolated PMV-1, PMV-3, PMV-4, and PMV-6 from 39 of 1,446 (3%) ducks in the Federal Republic of Germany. Isolations representing the PMV-1, PMV-4, and PMV-6 serotypes were reported from 4% of 719

TABLE 3.Prevalence of avian paramyxoviruses in migratory and resident ducks, Cameron Parish, Louisiana,20 December 1985 to 10 January 1986 and 19 December 1986 to 9 January 1987.

Species	Year	Juvenile	Adult	Total
Blue-winged teal	1986	0/5 (0)*	0/19 (0)	0/24 (0)
	1987	0/15 (0)	0/5 (0)	0/20 (0)
	Total	0/20 (0)	0/24 (0)	0/44 (0)
Gadwall	1986	0/3 (0)	0/18 (0)	0/21 (0)
	1987	0/60 (0)	0/28 (0)	0/88 (0)
	Total	0/63 (0)	0/46 (0)	0/109 (0)
Green-winged teal	1987	2/62 (3%)	1/50 (2%)	3/112 (3%)
Mottled duck	1987	b		0/7 (0)
Total		2/145 (1%)	1/120 (1%)	3/272 (1%)

\* Number positive/number sampled (% positive).

<sup>b</sup> Age not determined.

ducks in Czechoslovakia (Tumova et al., 1984). In Japan, PMV's were isolated from 2% of 287 ducks (Abenes et al., 1982). Mikami et al. (1987) recovered PMV's from 2% of 166 ducks and reported that isolations from waterfowl in Japan included PMV-1, PMV-4, PMV-6, and PMV-8.

In North America, PMV-1, PMV-3, PMV-4 and PMV-7 were reported from 2% of 1,055 ducks and geese sampled in Pennsylvania and from <1% of 1,511 birds sampled in Maryland (Nettles et al., 1985). In New York, PMV-1, PMV-2, PMV-4, and PMV-6 were isolated from 6% of 1,560 ducks (Deibel et al., 1985), and in Alberta these same serotypes were isolated from 3% of 9,195 ducks (Hinshaw et al., 1985).

In Louisiana, overall prevalence of PMV in ducks was estimated at 2%. Considerable variation, however, was present with prevalence estimates for some species/age groups ranging as high as 9% (juvenile bluewinged teal sampled in September 1987). Four serotypes were found with PMV-1 representing 60% of all isolates. Although differences in prevalence of specific PMV serotypes are apparent among surveys, reported isolations are dominated by the PMV-1 serotype (Ottis and Bachmann, 1983; Nettles et al., 1985; Deibel et al., 1985; Hinshaw et al., 1985). The PMV-4 and PMV-6 serotypes are also very common in waterfowl (Alexander, 1986). Similar results have been reported in sentinel duck studies (Turek et al., 1984; Kelleher et al., 1985).

As most studies have been directed at surveillance for AIV, prevalence estimates for PMV may represent an underestimate of the actual frequency of infection. Protocols for isolation of AIV usually rely on a 48- to 72-hr incubation period as opposed to a 120-hr incubation period recommended for PMV's such as Newcastle disease virus (Slemons and Easterday, 1975; Beard and Hanson, 1984). Detection of 56% of the PMV isolations on second ECE passage as compared to 18% for AIV isolation (Stallknecht et al., 1990) supports this potential bias. Deibel et al. (1985) suggested that intermittent shedding of PMV from ducks may also result in a low estimate of prevalence.

Prevalence of PMV in this study was similar between years. In other surveys of longer duration, annual variation has been observed (Deibel et al., 1985; Hinshaw et al., 1985). Seasonal differences in prevalence of PMV, although statistically significant, decrease only slightly from 4% in September to 1% in December and January. As with AIV (Hinshaw, 1986), the observed seasonal decrease in prevalence of PMV may relate to increased flock immunity. This possibility is supported by observed differences between juveniles (4%) and adults (<1%). Seasonal trends in prevalence of PMV infection in waterfowl are difficult to evaluate since reported prevalence varies little between studies regardless of season or migratory status of the survey population. Likewise, an abrupt peak in prevalence associated with juvenile recruitment, as reported for AIV in duck populations (Hinshaw, 1986), has not been clearly demonstrated with PMV. A seasonal increase in transmission of PMV to domestic sentinel ducks has been reported to occur during late summer and early fall in Minnesota (Kelleher et al., 1985), but this may relate to increased contact associated with peak migration. A winter peak in PMV prevalence also has been reported in domestic ducks (Shortridge, 1980), but this may reflect flock management practices which have little application to wild populations.

Differences in prevalence among species, if present, would be difficult to detect due to the low prevalence in the study population. Prevalence during both years, however, was highest in blue-winged teal and lowest in gadwalls. Similar results are reported for AIV in these samples (Stallknecht et al., 1990) and may relate to differences in behavior, habitat utilization, susceptibility, or duration of viral shedding. As with AIV (Stallknecht et al., 1990), isolations of PMV during the December and January sample period were restricted to green-winged teal. Although this probably relates to increased sample size for this species, similar results from both AIV and PMV have been reported for common teal in Japan (Abenes et al., 1982).

The mode of transmission and the mechanisms contributing to persistence of PMV's within waterfowl populations are unknown. Although prevalence is low, results from the December and January sample period indicate that PMV infections are present in these populations into the winter. In addition, the isolation of PMV-1 from a mottled duck, an indigenous nonmigratory species in Louisiana, documents that transmission of PMV's can occur on these wintering grounds. Whether these viruses persist through a cycle of transmission between susceptible individuals or through persistent infections in individual birds (Vickers and Hanson, 1982), however, cannot be resolved from these results.

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