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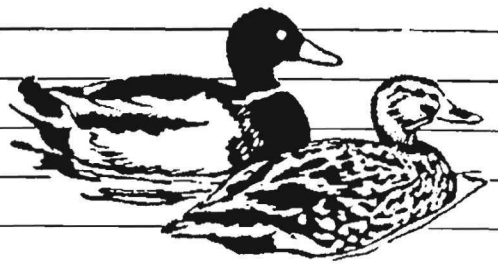

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Exposure of Wild Waterfowl to *Mycoplasma anatis*

Mycoplasmas have been associated with clinical respiratory disease and decreased productivity in domestic fowl. During field studies on the health status and causes of death in ducks, we isolated *Mycoplasma anatis* from wild hens and ducklings. In subsequent laboratory studies, we determined that *M. anatis* decreases the pipping and hatching success of game-farm mallard eggs (RIB 1992, No. 6). *Mycoplasma anatis* also seemed to have an effect on the growth of ducklings inoculated before hatch (RIB 1992, No. 72). The prevalence of mycoplasma infections and their effect on wild waterfowl populations, however, has received limited investigation. We determined previous exposure to *M. anatis* in several species of wild ducks at five study sites in the Central and Mississippi flyways as an indication of its prevalence and distribution.

Measurement of Exposure to *Mycoplasma anatis*

We developed a standard, enzyme-linked immunosorbent assay (ELISA) and used serum antibody titers (measured as percent response compared to standard positive and negative controls) to indicate previous exposure of wild ducks to *M. anatis*. Antisera to *M. anatis* was prepared by hyperimmunization of New Zealand white rabbits with a reference strain culture of the organism. This culture was also used as the antigen in our ELISA test. The ELISA test was standardized to determine positive or negative exposure to *M. anatis* by sampling two groups of domestic Pekin ducks with known mycoplasma infection status. One flock (39 birds sampled) had no previous exposure to *M. anatis*, and the other flock (63 birds sampled) was presently infected. Serum samples were obtained from both groups of ducks, as well as tissue samples (trachea, lung, and reproductive organs) to confirm the status of *M. anatis* infection.

Samples From Wild Ducks

We obtained serum samples from breeding mallards (*Anas platyrhynchos*) and canvasbacks (*Aythya valisineria*) in the Central flyway, and from wintering mallards and American black ducks (*Anas rubripes*) in the Mississippi flyway (Figure). Breeding mallards were captured near Kulm,

North Dakota, and Detroit Lakes, Minnesota, before spring nesting attempts in 1988, 1989, and 1990. Breeding canvasbacks at Agassiz National Wildlife Refuge (NWR) were sampled in spring 1990. In fall and winter 1990-91, 1991-92, and 1992-93, serum samples were collected from juvenile mallards and American black ducks at the Ottawa NWR in Ohio. Adult American black ducks were sampled at the Tennessee NWR during winters 1990-91 and 1991-92; juvenile American black ducks were sampled during 1991-92. All sera were stored at -20°C until testing. Only sera from female birds were used in this study.

A discriminant analysis was conducted to statistically classify the titers of the two reference groups of Pekin ducks. We determined that an ELISA titer cutoff of 31.6% gave a 95% probability of correctly classifying positive titers and increased the specificity of the test so that only birds highly likely to be exposed to *M. anatis* were considered positive. The ELISA values from our wild bird serum samples were classified by using this criteria and analyzed for differences in prevalence by using logistic regression methods.

Prevalence of *M. anatis* Exposure

Because of differences in species and ages of birds sampled, analyses were conducted separately for summer (breeding ducks) and winter data. In summer samples, differences in exposure to *M. anatis* were most pronounced between species; exposure in canvasback ducks was significantly lower (0% vs. 78%, $P < 0.001$) than in mallards (Table 1). This may be because of differences in sensitivity of the test in canvasbacks or differential species exposure to *M. anatis*. No significant differences in prevalence ($P > 0.5$) were seen between locations or years in breeding mallards.

In winters 1990-91 and 1991-92, exposure to *M. anatis* was higher ($P < 0.02$) in adults than in juveniles, but juvenile ducks showed high exposure rates (averaging 50%) during their first winter (Table 2). Juvenile exposure rates also varied annually ($P < 0.005$), primarily because of the increased exposure rate during 1992-93. Juvenile American black ducks did not have significantly different ($P \geq 0.80$) exposure rates than mallards. Adult birds had different ($P = 0.05$) exposure between years, with higher exposure in 1990-91. Other temporal and spatial changes in exposure were difficult to evaluate because adults and juveniles were not consistently sampled at all locations each year.

Overall, we found evidence that adult mallards and American black ducks are commonly exposed to *M. anatis*; 68% of the birds we sampled were previously exposed. In addition, samples from juvenile birds indicated that a considerable amount of exposure occurs within the first 8 months after hatch. In contrast, none of the canvasbacks we sampled were exposed, but our sample of canvasbacks was small. In domestic fowl, *Mycoplasma* spp. have caused disease affecting respiratory and reproductive systems, leading to economic loss through decreased productivity and poor growth. The consequence of exposure to *M. anatis* or infection in wild birds, however, is still uncertain. In particular, little is known about the different routes of exposure, how and when clinical infection occurs, and the frequency of vertical (parent to egg) transmission which might effect hatching success.

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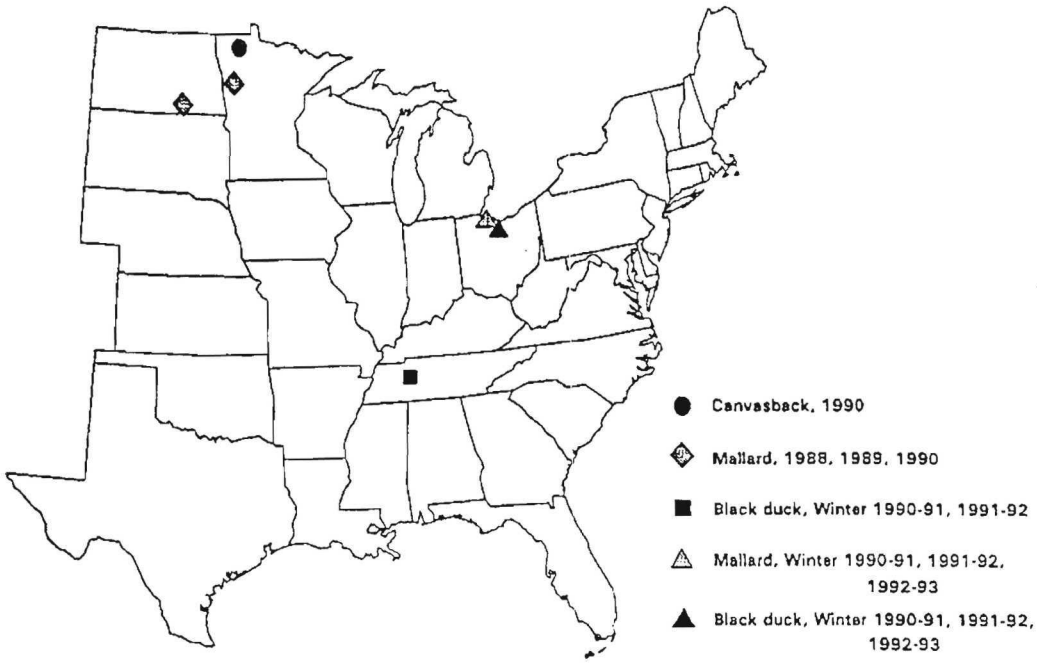

Figure. Study sites in North Dakota, Minnesota, Tennessee, and Ohio where serum samples were obtained to determine the prevalence of exposure to *Mycoplasma anatis*.

Table 1. Prevalence of Mycoplasma anatis exposure in breeding adult hen mallards (*Anas platyrhynchos*) and canvasbacks (*Aythya valisineria*) from the Central flyway.

Location	Species	Year	Prevalence[a]
Minnesota	Mallard	1988	11/16 (69)
Minnesota	Mallard	1989	5/6 (83)
Minnesota	Mallard	1990	16/19 (84)
N. Dakota	Mallard	1988	1/1 (100)
N. Dakota	Mallard	1989	13/18 (72)
N. Dakota	Mallard	1990	10/12 (83)
Minnesota	Canvasback	1990	0/19 (0)

[a] Prevalence: Number positive/Number sampled (%).

Table 2. Prevalence of *Mycoplasma anatis* exposure in wintering mallards (*Anas platyrhynchos*) and American black ducks (*Anas rubripes*) from the Mississippi flyway.

Location	Species	Year	Age	Prevalence[a]
Ohio	Black duck	1990–91	A	2/5 (40)
Ohio	Black duck	1990–91	J	12/33 (36)
Ohio	Black duck	1991–92	J	12/34 (35)
Ohio	Black duck	1992–93	J	31/42 (74)
Ohio	Mallard	1990–91	J	12/28 (43)
Ohio	Mallard	1991–92	J	20/39 (51)
Ohio	Mallard	1992–93	J	26/45 (58)
Tennessee	Black duck	1990–91	A	15/21 (71)
Tennessee	Black duck	1991–92	A	10/24 (42)
Tennessee	Black duck	1991–92	J	4/23 (17)

[a] Prevalence: Number positive/Number sampled (%).