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Further Western Spread of *Mycoplasma gallisepticum* Infection of House Finches

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ABSTRACT: *Mycoplasma gallisepticum*, an important pathogen of poultry, especially chickens and turkeys, emerged in 1994 as the cause of conjunctivitis in house finches (*Carpodacus mexicanus*) in their eastern range of North America. The resulting epidemic of *M. gallisepticum* conjunctivitis severely decreased house finch abundance and the continuing endemic disease in the eastern range has been associated with repeating seasonal peaks of conjunctivitis and limitation of host populations. *Mycoplasma gallisepticum* conjunctivitis was first confirmed in the western native range of house finches in 2002 in a Missoula, Montana, population. Herein, we report further western expansion of *M. gallisepticum* conjunctivitis in the native range of house finches based on positive polymerase chain reaction results with samples from birds captured in 2004 and 2005 near Portland, Oregon.

Key words: *Carpodacus mexicanus*, conjunctivitis, house finch, *Mycoplasma gallisepticum*.

Mycoplasma gallisepticum conjunctivitis emerged in 1994 as a disease of free-ranging house finches (*Carpodacus mexicanus*) in the mid-Atlantic region of the United States (Ley et al., 1996). An apparently novel strain (Ley et al., 1997) of *M. gallisepticum*, an important poultry pathogen with worldwide distribution, spread rapidly among house finches in their eastern range (Dhondt et al., 1998). Within a year after its emergence mycoplasmal conjunctivitis was found across a region of roughly 800,000 km², and within 3 yr after its emergence the disease had spread over most of the eastern range of the host species, roughly 3,000,000 km² (Dhondt et al., 1998). The resulting epidemic of *M. gallisepticum* conjunctivitis produced an unprecedented decline of eastern house finch populations severely

decreasing host abundance (Hochachka and Dhondt, 2000), and the endemic disease remains associated with repeating seasonal peaks of conjunctivitis and limitation of host populations (Hochachka and Dhondt, 2000; Altizer et al., 2004). Dhondt et al. (1998) were able to document this rapid expansion of the epidemic and the subsequent seasonal variation in prevalence (Altizer et al., 2004) using a volunteer-based monitoring scheme, the *House Finch Disease Survey*. Because conjunctivitis in house finches has been monitored and sampled extensively following initial reports, it is rapidly becoming a model system for understanding emerging infectious diseases in wild avian hosts (Dhondt et al., 1998; Dhondt et al., 2005). *Mycoplasma gallisepticum* was first confirmed in western native house finches in April 2002 when the first cases of mycoplasmal conjunctivitis were observed in a Missoula, Montana, population that had been studied continuously since 1993 (Duckworth et al., 2003). In this communication we describe the further expansion of *M. gallisepticum* conjunctivitis among native western house finches based on three cases yielding positive results by polymerase chain reaction (PCR) from samples taken from birds captured near Portland, Oregon.

Wild-caught house finches submitted to the Audubon Society of Portland Wildlife Care Center were assigned individual identity numbers, and eye lesion scores were recorded (Table 1). For all house finches showing clinical signs of mycoplasmal conjunctivitis, we assigned each eye a 0–3 score based on the severity of conjunctivitis. House finches with eye

TABLE 1. *Mycoplasma gallisepticum* polymerase chain reaction (PCR) results from Oregon^a house finches (*Carpodacus mexicanus*).

Accession no.	House finch id. no.–sex	Eye score ^b	Samples collected ^c	PCR	
				16S rRNA ^d	<i>mgc2</i> ^e
2004.033	257–F	R=0	7 April 2004	–	ND ^f
		L=2	7 April 2004	+	ND
	259–F	R=3	7 April 2004	–	ND
		L=3	7 April 2004	–	ND
2004.044	546–NR ^g	R=3	19 May 2004	+	ND
		L=3	19 May 2004	+	ND
			19 May 2004 ^h	+	ND
2005.012	144–M	R=3	3 August 2005	+	+
		L=1	3 August 2005	+	+

^a House finches with eye lesions typical of *M. gallisepticum* conjunctivitis were submitted to the Audubon Society of Portland, Wildlife Care Center, Portland, Oregon, USA.

^b Eye lesions were scored for right (R) and left (L) eyes on a scale of 0–3 as follows: a zero score represented no signs of conjunctivitis; eyes with minor swelling or redness were assigned a 1; eyes with moderate swelling and discharge received a 2; and severely swollen eyes (those that were nearly or completely swollen shut) were classified as 3.

^c Samples collected were conjunctival swabs inoculated into modified Frey's mycoplasma broth with 15% swine serum.

^d PCR method to detect *M. gallisepticum* 16S rRNA gene (Lauerman, 1998).

^e PCR method to detect *M. gallisepticum* *mgc2* surface protein gene (Garcia et al., 2005).

^f ND = not done.

^g NR = no record.

^h Additional sample was collected by swabbing the choanal cleft.

lesions compatible with mycoplasma conjunctivitis were sampled for mycoplasma culture and *M. gallisepticum* PCR prior to any local or systemic antimicrobial therapy. Briefly, sterile swabs (calcium alginate fiber-tipped ultrafine aluminum applicator swabs, Fisher Scientific, Pittsburgh, Pennsylvania, USA) were used to sample conjunctival sacs of both eyes, and in one case, the choanal cleft. Immediately after sampling from each site, swabs were placed in 2 ml of Frey's mycoplasma broth media with 15% swine serum (Kleven, 1998). Inoculated broths were stored at 4 C for up to 72 hr before overnight shipment on cold packs to North Carolina State University College of Veterinary Medicine (Raleigh, North Carolina). Upon arrival, 1 ml of inoculated broth from each sample was removed for PCR processing. The remaining inoculated broths were incubated at 37 C in humidified air for 4 to 5 wk with 3 passages to additional Frey's broth and agar media to allow for the growth of mycoplasmas (Kleven,

1998). No mycoplasmas were isolated in culture from any of the samples submitted. However, at least one sample from three of four house finches with eye lesions typical of *M. gallisepticum* conjunctivitis showed positive PCR results (Table 1). Conjunctival swabs from right and left eyes of four house finches and a choanal cleft swab from one of these birds were tested using the 16S rRNA gene PCR method described by Lauerman (1998), resulting in six of nine positive reactions. Additionally, right and left conjunctival swabs of the most recently sampled house finch (id. 144-M), which were both positive by the Lauerman method, were also positive using primers to the *mgc2* surface protein gene of *M. gallisepticum*. This method was recently described by Garcia et al. (2005) and reported to have increased specificity and sensitivity compared to the 16S rRNA PCR method.

The increasing reports of house finches with eye lesions typical of *M. gallisepticum*

conjunctivitis (Dhondt, pers. comm.), the observation of eye lesions in captured house finches by veterinary health care professionals, and positive PCR results using two different methods, are compelling evidence for the further western extension of the epidemic in house finches that was first observed and confirmed in the eastern population of house finches in 1994. Culture and isolation of *M. gallisepticum* organisms from West Coast birds has so far not been successful, most likely because of loss of viability during storage and transportation. We have found that culture and isolation success is improved when samples are incubated at 37 C immediately after broths are inoculated. Isolation of *M. gallisepticum* organisms from the western extension of this emerging disease is highly desirable because that would enable further exploration of the molecular epidemiology of the disease and the possible molecular evolution of the pathogen.

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