

# Seroprevalence of *Leptospira* serovars in horses in the Walla Walla Valley

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**Objective** – To estimate the seroprevalence of *Leptospira* serovars in horses in the Walla Walla Valley.

**Design** – Observational study.

**Animals** – 172 horses representing 122 clients of the Animal Clinic of Walla Walla.

**Procedures** – From April 2005 through August 2006, serum samples were collected from horses following informed owner consent. The age, breed, sex, and known health problems were recorded for each horse sampled. Serum samples were tested for antibody titers against six *Leptospira* spp. serovars: *bratislava*, *canicola*, *grippotyphosa*, *harjo*, *icterohaemorrhagiae*, and *pomona*. The percent of positive titers were computed as an estimate of the prevalence of exposure to *Leptospira* among horses in the practice area. Age, breed, sex, and specific health problems were examined as associated risk factors.

**Results** – One hundred thirty-three of the 172 horses sampled (77%) had a positive titer to one or more *Leptospira* serovars. Serovar *bratislava* was most common with a seroprevalence of 64%. Elevated titers indicating current exposure were found in 28 horses of 122 owner-locations (23%). Horses 15+ years old were significantly more likely to be seropositive.

**Conclusions** – Seroprevalence of *Leptospira* serovars is high in horses the Walla Walla Valley.

## Introduction

Leptospirosis is a disease syndrome of many animal species and humans caused by the spirochete bacteria *Leptospira* spp. The disease has a worldwide distribution and more than 200 *Leptospira* serovars have been described. Specific *Leptospira* serovars are maintained in the environment by one or more host species that act as reservoirs for disease and transmit infection via their urine. Infection of the maintenance host tends to be chronic and subclinical, whereas infection of an accidental host can result in acute, severe disease.<sup>1</sup>

Leptospirosis in horses manifests as abortions, stillbirth, uveitis, and renal failure.<sup>2,3,4</sup> During the fall of 2002, an associate veterinarian at the Animal Clinic of

Walla Walla (ACWW) investigated an outbreak of fever, lethargy, and uveitis that affected 5 horses in a client herd of 20 horses. Serum antibody titers from the most severely affected horses revealed significant titers to *Leptospira* serovars *bratislava* and *pomona*. In the fall of 2004, significant titers to the same serovars were observed in a recently aborted, unvaccinated heifer and low positive titers to serovars *bratislava*, *canicola*, and *icterohaemorrhagiae* were observed in a horse with chronic uveitis.

The purpose of this study was to conduct a serological survey of antibodies against *Leptospira* serovars in horses in the practice area of ACWW to provide an estimate of the prevalence of exposure to *Leptospira* in the Walla Walla Valley. Knowledge of the prevalence of exposure to *Leptospira* spp. in this area will aid local veterinarians in the diagnosis of disease in companion animals and livestock.

## Materials and Methods

**Sample collection** – From April 1, 2005 through August 31, 2006, serum samples were collected from 172 horses presenting to the ACWW. These horses were owned by 122 clients who agreed to participate in the serological survey. For clients presenting with multiple horses, the two horses that were owned the longest were selected. Data collected at the time of sampling included the age, breed, and sex of the horse as well as any current or past health problems.

**Laboratory analysis** – Serum samples were analyzed for antibody titers against six *Leptospira* spp. serovars: *bratislava*, *canicola*, *grippotyphosa*, *hardjo*, *icterohaemorrhagiae*, and *pomona* by the microscopic agglutination test<sup>2</sup>

**Data analysis** – Descriptive statistics and chi-square tests were computed using SAS® software (SAS Institute, Inc., Cary, NC). Odds ratios (OR) and 95% confidence intervals (CI) were calculated for associated risk factors.

## Results

The horses in this study range in age from 2 to 31 years old. Seventy-two mares, 99 geldings, and one stallion were sampled. Quarter horses and Quarter horse crosses were the most common breed sampled (52%) followed by Arabian and Arabian crosses (12%), Paints (10%) and Appaloosas (6%). Other breeds sampled were Foxtrotter, Haflinger, Miniature Horse, Morgan, Mustang, Norwegian Fjord, Percheron, Saddlebred, Tennessee Walking Horse, Thoroughbred and Welsh Pony.

Of the 172 horses sampled, 133 horses had a positive serum titer to one or more *Leptospira* serovars. The number of reactors per titer to each serovar are shown in Table 1.

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Table 1 - Number of horses seropositive to *Leptospira* spp. by serovar and titer (n = 172)

Serovar	Titer								
	Negative	100	200	400	800	1600	3200	6400	12800
<i>bratislava</i>	62	37	23	19	15	7	5	2	2
<i>canicola</i>	118	29	14	7	4	0	0	0	0
<i>icterohaemorrhagiae</i>	121	22	16	8	3	1	0	0	1
<i>pomona</i>	145	2	6	6	5	3	2	0	3
<i>grippityphosa</i>	162	8	0	0	1	0	1	0	0
<i>hardjo</i>	167	5	0	0	0	0	0	0	0

Antibodies to serovar *bratislava* were most common with 64% of horses yielding a positive or elevated titer, followed by serovars *canicola* (31%), *icterohaemorrhagiae* (30%), *pomona* (16%), *grippityphosa* (6%), and *hardjo* (3%) (Figure 1).

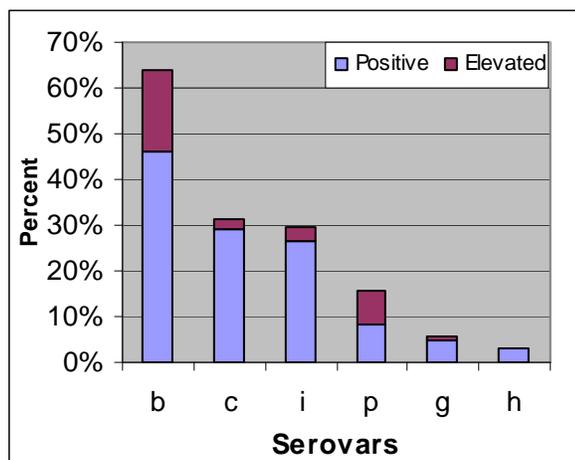


Figure 1 – Percent of horses with positive (1:00, 1:200, 1:400) and elevated (1:800 or greater) serum antibody titers to *Leptospira* serovars *bratislava* (b), *canicola* (c), *icterohaemorrhagiae* (i), *pomona* (p), *grippityphosa* (g), and *hardjo* (h).

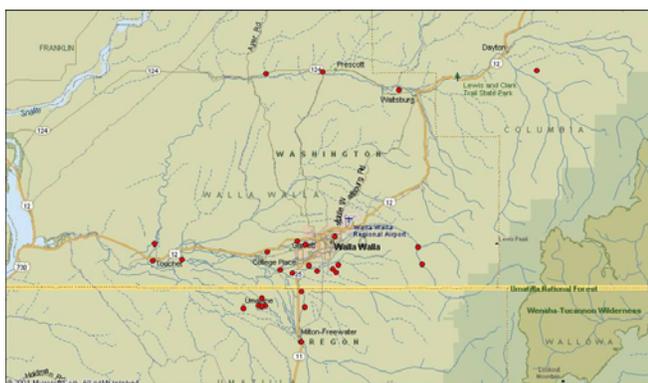


Figure 2 – Locations of 28 horses with one or more elevated (>1:400) serum antibody titers to *Leptospira* spp. indicating > current exposure to the serovar.

Elevated titers to one or more serovars indicating current exposure to that serovar were found in 28 horses out of 122 different owner-locations (23%). These 28 horses were from locations widely distributed throughout the valley (Figure 2).

Forty-eight percent of the samples collected were from purebred Quarter horses (QH). Therefore, the breeds represented were grouped into two categories: QH plus QH crossbreds and all other breeds. Eighty-three percent of the QH plus QH crossbreds were seropositive versus 71% of the other breeds, however, this difference was not significant by the chi-square test.

The sex of the horse was also not a significant risk factor for a positive titer. Seventy-six of 99 geldings (77%) were seropositive and 56 of 72 mares (77%) were seropositive. The single stallion in the data set was excluded from the analysis for the association of sex.

The age of the horse was a significant risk factor for a positive titer. Horses aged 15 years and older were more likely seropositive (OR = 2.29, CI = 1.08, 4.8). A higher percentage of older horses had an elevated titer, but this difference was not significant. (Figure 3).

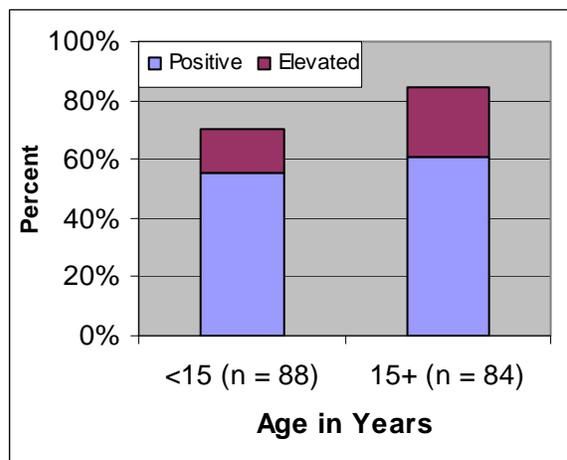


Figure 3 – Percent of horses with positive (1:00, 1:200, 1:400) and elevated (1:800 or greater) serum antibody titers to *Leptospira* spp. by age.

The majority of the horses sampled (n = 157) presented to the clinic for routine vaccinations, anthelmintic administration, dental procedures and health certificates. These horses were in good health at the time of presentation and had no history of health problems. Health problems recorded for the remaining 15 horses were categorized as follows: eye (blindness, uveitis, n = 7), musculoskeletal (arthritis, lameness, laminitis, n = 5), and other (colic, n = 1; nasal discharge, n = 1; melanoma, n = 1). All 15 horses had a positive titer to one or more *Leptospira* serovars. The 7 horses with eye disease all had elevated titers indicating current exposure to multiple serovars with the highest titers to *bratislava* and *pomona*.

## Discussion

Leptospirosis is considered a reemerging disease in human medicine and companion animals.<sup>6,7</sup> Improved diagnostic methods may be partially responsible for this reemergence, but at least in dogs, real estate development in rural areas has been demonstrated to increase the risk of leptospirosis.<sup>8</sup> The population of the Walla Walla valley is growing at a steady average of 1% per year and much of the farmland immediately surrounding the city is being developed for residential housing.<sup>9,10</sup>

Due to the distribution of horse owning clients in the practice area, the ease of sample collection, and the fact there is no vaccine currently in use for leptospirosis in horses, a serological survey of *Leptospira* serovars in horses was conducted to provide an estimate of the prevalence of exposure to the organism in this valley. The results of this study demonstrates the seroprevalence of *Leptospira* serovars in horses in the Walla Walla Valley is remarkably high. Seventy-seven percent of the horses sampled had a positive titer to one or more serovars, with serovar *bratislava* the most common (64% seroprevalence). Also, current exposure to *Leptospira* spp. is not uncommon since 23% of owner-locations had one horse with an elevated titer.

The most recent and geographically nearest seroprevalence study in horses was conducted in Alberta, Canada with samples collected in late 1980s. In this study of 1,923 horses, the seroprevalence rate for serovar *icterohaemorrhagiae* was highest at 94.6%, followed by *bratislava* at 56.6%. The Canadian study also found age significantly associated with a positive titer and calculated a 10% increase in risk of a positive titer with each year of life.<sup>11</sup> The sample size of 172 horses in the present study was too small for more precise analysis of the association of age. The difference in seroprevalence by age was demonstrated best with a cut-point of 15 years.

A recent canine leptospira seroprevalence study in Washington state reported an overall 17.1% seropositive rate among healthy dogs. Four of 20 samples (20%) from healthy dogs submitted by veterinary clinics in Walla Walla were positive and none of the positive samples yielded an elevated titer.<sup>12</sup> The difference in seroprevalence between the canine study and the current study may be due to the relatively small sample size in the former study or may

indicate a true difference in the risk of exposure to *Leptospira* spp. between dogs and horses.

Persistent ocular infection with *Leptospira* is strongly associated with recurrent uveitis in horses.<sup>3,13</sup> One field study in New York state found horses seropositive for serovar *pomona* were 13.2 times more likely to have uveitis than seronegative horses.<sup>14</sup> In this study, all seven horses with eye disease had elevated titers to multiple *Leptospira* serovars. This data along with the large numbers of seropositive horses suggests more horses in this practice area may be at risk for uveitis.

## References

1. Radostits OM, Gay CC, Blood DC, Hinchcliff KW. Diseases caused by *Leptospira* spp. In: *Veterinary Medicine, A Textbook of the Diseases of Cattle, Sheep, Pigs, Goats, and Horses.*, 9<sup>th</sup> ed. Philadelphia, WB Saunders Co., 2000;971-996.
2. Donahue JM, Smith BJ, Poonacha KB, et al. Prevalence and serovars of leptospira involved in equine abortions in central Kentucky. *J Vet Diagn Invest* 1995;7:87-91.
3. Wollanke B, Rohrbach BW, Gerhards H. Serum and vitreous humor antibody titers in and isolation of *Leptospira* interrogans from horses with recurrent uveitis. *J Am Vet Med Assoc* 2001;219:795-800.
4. Frazer ML. Acute renal failure from leptospirosis in a foal. *Aust Vet J* 1999;77:499-500.
5. Cole JR, Sulzer CR, Pusell AR. Improved microtechnique for the leptospiral microscopic agglutination test. *App Microbiol* 1973; 25:976-980.
6. Bharadwaj R. Leptospirosis – a reemerging disease? *Indian J Med Res* 2004;120:136-138.
7. Ward MP, Glickman LT, Guptill LF. Prevalence of and risk factors for leptospirosis among dogs in the United States and Canada: 677 cases (1970-1998). *J Am Vet Med Assoc* 2002;220:53-68.
8. Ward MP, Guptill LF, Wu CC. Evaluation of environmental risk factors for leptospirosis in dogs: 36 cases (1997-2002). *J Am Vet Med Assoc* 2004;225:72-77.
9. Hillhouse, V. Newcomers to Walla Walla quickly find it home. In: *Walla Walla Union-Bulletin*, Walla Walla, Washington, Larry J. Duthie, Publisher, August 22, 2003.
10. Hillhouse, V. How does Walla Walla grow? In: *Walla Walla Union-Bulletin*, Walla Walla, Washington, Larry J. Duthie, Publisher, August 26, 2003.
11. Lees VW, Gale SP. Titers to *Leptospira* species in horses in Alberta. *Can Vet J* 1994;35:636-640.
12. Personal communication. December 20, 2006. Margaret A. Davis, DVM, MPH, PhD. Dept. of Veterinary Microbiology and Pathology, Washington State University, Pullman, WA 99164-7040.
13. Brande K, Wollanke B, Niedermaier G, Brem S, Gerhads H. Recurrent uveitis in horses: vitreal examinations with ultrastructural detection of leptospire. *J Vet Med A Physiol Pathol Clin Med* 2007;54:270-275.
14. Dwyer AE, Crockett RS, Kalsow CM. Association of leptospiral seroreactivity and breed with uveitis and blindness in horses: 372 cases (1986-1993). *J Am Vet Med Assoc* 1995;207:1327-1331.