PREVALENCE OF PATHOGENIC ENTERIC BACTERIA IN WILD BIRDS ASSOCIATED WITH AGRICULTURE IN HUMBOLDT COUNTY, CALIFORNIA

By

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PREVALENCE OF PATHOGENIC ENTERIC BACTERIA IN WILD BIRDS

ASSOCIATED WITH AGRICULTURE IN HUMBOLDT COUNTY, CALIFORNIA

by

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ABSTRACT

PREVALENCE OF PATHOGENIC ENTERIC BACTERIA IN WILD BIRDS ASSOCIATED WITH AGRICULTURE IN HUMBOLDT COUNTY, CALIFORNIA

Krysta H. Rogers

Cloacal and fecal samples of 243 wild birds, including 65 house sparrows (*Passer domesticus*), 29 white-crowned sparrows (*Zonotrichia leucophrys*), 16 Brewer's blackbirds (*Euphagus cyanocephalus*), 43 red-winged blackbirds (*Agelaius phoeniceus*), 56 brown-headed cowbirds (*Molothrus ater*), and 34 European starlings (*Sturnus vulgaris*) were sampled for potentially pathogenic bacteria between July 2002 and February 2004, on five dairy farms in coastal Humboldt County, California (USA). Thirty-seven bacterial species in 14 genera were isolated from wild birds. *Escherichia coli* (93/243; 38%), *Pantoea* spp. (41/243; 17%), *Enterobacter* spp. (39/243; 16%), *Yersinia* spp. (29/243; 12%), and *Citrobacter* spp. (27/243; 11%) were the most prevalent bacterial groups recovered from wild birds. Bacterial species composition varied between bird species, farms, and seasons. Fecal samples from 100 dairy cattle on the five farms also were sampled for pathogenic enteric bacteria during November 2002 and April 2004. Twenty-four bacterial species in 13 genera were recovered from cattle. *Escherichia coli* (88/100; 88%), *Citrobacter* spp. (18/100; 18%), *Aeromonas hydrophila* (13/100; 13%), *Proteus vulgaris* (13/100; 13%), and *Citrobacter braakii* (11/100; 11%) were the most prevalent bacteria isolated from cattle. Nineteen bacterial species in 10 genera were recovered from both the 243 wild birds and 100 dairy cattle. Eleven of these 19 bacterial

species had the same API 20E code represented in both birds and cattle. This suggests that there is a potential for bacterial transmission between birds and cattle.

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INTRODUCTION

Agriculture is an important industry in Humboldt County with 26% of the land area devoted to various agricultural practices (California Department of Food and Agriculture 2000). In a 1998 production census livestock and livestock products were valued at over $82,000 (U.S.) (California Department of Food and Agriculture 2000). Locally, private farms raise primarily dairy and beef cattle and to a lesser extent poultry, sheep, and pigs. This environment has created an ideal habitat for gregarious, opportunistically feeding wild bird species. Birds such as Brewer's blackbirds (*Euphagus cyanocephalus*), brown-headed cowbirds (*Molothrus ater*), and European starlings (*Sturnus vulgaris*) benefit from abundant animal feed and the insects attracted to manure. Agricultural practices also contribute to the spread of potentially pathogenic bacteria within the environment through contaminated animal feed, bedding, and waste (Chapman et al. 1993, Cizek et al. 1994, Hancock et al. 1994, Davies and Wray 1997). Of particular concern in the farm environment are the Gram-negative bacteria within the family Enterobacteriaceae, including *Escherichia* spp., *Salmonella* spp., and *Yersinia* spp. Members of this family often are opportunistic pathogens, causing disease in humans, domestic animals, and wildlife under the appropriate conditions (Janda and Abbott 1998). Although many bacteria are a part of the normal intestinal flora of birds, bird species associating with potentially contaminated environments, such as refuse dumps, sewage treatment facilities, agricultural sites, and bird feeders, are more likely to harbor pathogenic bacteria in their intestinal tracts (Quevedo et al. 1973, Fenlon 1985, Cizek et

al. 1994, Casanovas et al. 1995). Coupled with the extensive mobility of birds and migratory habits of some birds, numerous avian species have been implicated as a source of infection for enteric diseases in humans, domestic animals, and other wildlife (Goodchild and Tucker 1968, Altekruse et al. 1994, Kapperud et al. 1998, Craven et al. 2000). For instance, Quevedo et al. (1973) suggested that house sparrows (*Passer domesticus*) captured in horse corrals may transmit *Salmonella* spp. to horses; Craven et al. (2000) implied that house sparrows and European starlings may be responsible for transmitting *Salmonella* spp., *Campylobacter jejuni*, and *Clostridium perfringens* to chickens on poultry farms. However, studies such as these only identify wild bird species as carriers of enteric bacteria through fecal or intestinal analyses. They fail to prove transmission, or in the least, identify similarities in the enteric bacteria carried by the domestic animals in contact with these wild bird species. That is, transmission is suspected, but not proven. Kirk et al. (2002) isolated *Salmonella* spp. from a variety of wild bird species utilizing dairy farms in central California, including brown-headed cowbirds, European starlings, house sparrows, and Brewer's blackbirds. When comparing the *Salmonella* spp. isolated from the birds, with the *Salmonella* spp. previously recovered from cattle in the same geographic area, Kirk et al. (2002) suggested that very few species of *Salmonella* were recovered from both wild birds and cattle. This indicated that wild birds were not an important factor in transmission of *Salmonella* spp. to domestic animals. However, the relationship between domestic animals and wild birds on farms for other bacterial species is relatively unknown.

Birds associated with a contaminated environment, such as a dairy farm, are more likely to carry pathogenic bacteria than birds not inhabiting such an environment (Radwan and Lampky 1972, Cizek et al. 1994, Craven et al. 2000). This suggests the possibility of increased transmission of pathogenic bacteria to domestic animals and humans from birds in these environments. Alternatively, contaminated farm environments may facilitate the passive carrying of bacteria by birds with little likelyhood for transmission, because carrier rates are low (small number of positive birds per total number tested) (Wilson and MacDonald 1967, Plant 1978, Kirk et al. 2002). That is, birds may inadvertently ingest bacteria in their environment, but the bacteria pass through the intestinal tract with no adverse effects to the carrier bird; yet in such cases the birds may aid in dispersal of the bacteria within the environment. Therefore, it is important to assess the prevalence of pathogenic bacteria birds carry, especially in environments that bring them close to domestic animals and humans, to better understand the potential role of wild birds in transmission of pathogenic bacteria.

To date, there has been no systematic assessment of pathogenic bacteria carried by free-living passerines that associate with agricultural practices in Humboldt County. However, bacteria pathogenic to both domestic animals and humans have been isolated from local domestic animals and other wildlife, as well as from environmental sources. For instance, Harber (1988) isolated antibiotic resistant strains of *Escherichia coli* and *Salmonella* spp. from dairy cattle on a farm in Loleta. In a survey of three local large animal veterinary hospitals in Humboldt County, *E. coli* and *Salmonella* spp. were identified as causes of infection in cattle (Carlisle 2002, personal communication, Silver

2002, personal communication). Fecal coliform bacteria also have been routinely isolated from waters of local creeks and urban streams (Cole 2003, Brenneman 2005, personal communication). Although *E. coli* 0157:H7 was not isolated from a sample of approximately 100 resident Canada geese (*Branta canadensis moffitti*) in the Humboldt Bay area, numerous *E. coli* strains have been recovered from area gulls (*Larus* sp.) and wildfowl (Botzler 2002, personal communication). *Yersinia enterocolitica* has been isolated from the feces of a local population of Roosevelt elk (*Cervus elaphus roosevelti*) (Martyny and Botzler 1976) and from the soil in habitats utilized by the elk (Botzler 1979). *Yersinia enterocolitica* and a variety of the enterobacteria were isolated from soil and 6 of 75 captive birds and mammal samples at the Sequoia Park Zoo, Eureka, California (Belltawn et al. 2004). In addition, *Pasteurella multocida* has been isolated regularly from wildfowl in Humboldt County since 1945 (Botzler 2002).

Potentially pathogenic bacteria have been isolated locally from a number of different sources. In addition, there are large populations of wild birds inhabiting local dairy farms. This study was undertaken to determine if wild birds pose a threat to domestic animals through the possible transmission of pathogenic enteric bacteria. As such, the primary objective of this study was to assess the prevalence and diversity of enteric bacteria carried by wild birds associating with agriculture in Humboldt County. Prevalence was then compared between bird species, between birds sampled on different farms, as well as between seasons, males and females, adults and juveniles, and between birds of different body weights. Additionally, the prevalence of enteric bacteria carried by cattle in these areas also was assessed. Prevalence was compared between cattle on

different farms and between seasons. Finally, bacterial species composition and prevalence was compared between birds and cattle.

Since wild bird species can carry pathogenic bacteria in their intestinal tract, I hypothesized that wild birds associating with agriculture are carriers of potentially pathogenic enteric bacteria with the potential to serve as a source of infection for domestic animals. Given that domestic animals, such as cattle, also can carry pathogenic bacteria in their intestinal tract, I hypothesize that dairy cattle will have potentially pathogenic enteric bacteria in their feces. Since the cattle and wild birds are exposed to each other's feces, I predict that the bacteria carried by wild birds and dairy cattle will be similar.

METHODS

Study Area

I tested wild birds and dairy cattle for enteric bacteria on five dairy farms located in McKinleyville (n = 1), Arcata (n = 2), Loleta (n = 1), and Ferndale (n = 1) in coastal Humboldt County, California (Figure 1). Farms ranged in size from 49 to 202 ha (Table 1). Cattle on Farms A, B, C, and D were pastured during the day and confined to a barn at night. Cattle on Farm E were confined to a barn at all times. All cattle were milked twice daily in a milking barn.

Sample Collection

Birds

I tested 243 birds between 23 July 2002 and 14 February 2004. Six bird species were targeted due to their abundance on local dairy farms: Brewer's blackbirds (n = 16), brown-headed cowbirds (n = 56), European starlings (n = 34), house sparrows (*Passer domesticus*; n = 65), redwing blackbirds (*Agelaius phoeniceus*; n = 43), and white-crowned sparrows (*Zonotrichia leucophrys*; n = 29). Birds were live-caught in a modified Australian crow trap (2.44x1.63x2.44m) (Johnson and Glahn 1994) or a mist net (25mm mesh, 12x2.6m and 25mm mesh, 6x5.2m; Avinet, Dryden, New York, USA) located in areas close to the milking barn and cattle enclosures. Scratch grains (Purina Mills, St. Louis, Missouri, USA) and seed (Wild Birds Unlimited, Carmel, Indiana, USA) were offered in trap locations to attract birds 3 to 10 days prior to trapping. The trap and

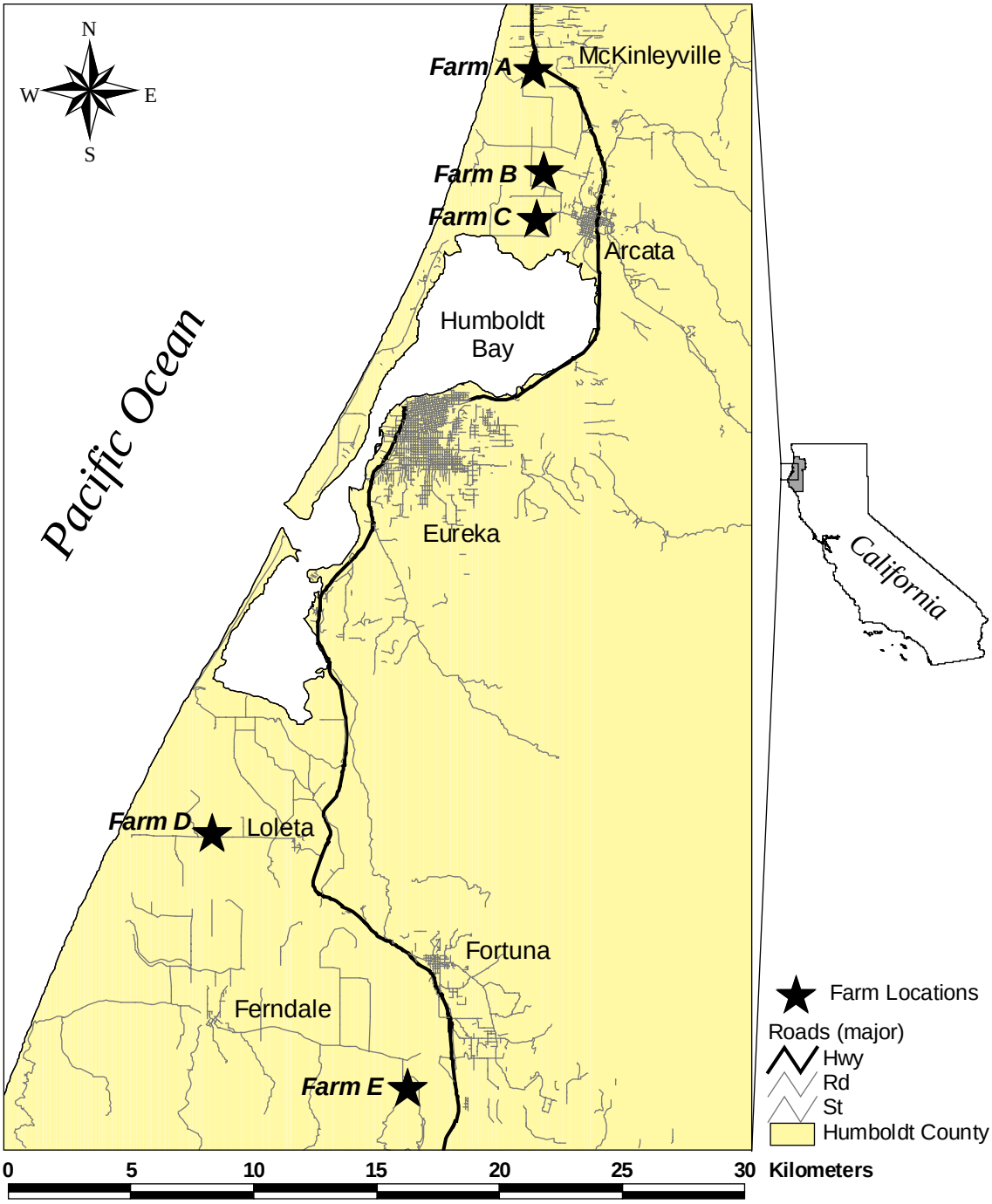

Figure 1. Location of 5 dairy farms on which 243 wild birds and 100 cattle were sampled for potentially pathogenic enteric bacteria between July 2002 and April 2004, in western Humboldt County, California.

Table 1. Attributes of the 5 dairy farms on which wild birds and cattle were sampled for potentially pathogenic enteric bacteria between July 2002 and April 2004 in Humboldt County, California.

Name	Location	UTM[a]	Hectares	Milk Grade[b]	Breed	No. of cattle[c]	Other domestic animals resident on farm	Most common bird species caught[d]
Farm A	McKinleyville	10 405996E 4531445N	65	A	Jersey	120	chickens, dogs	EUST, HOSP, RWBL
Farm B	Arcata N	10 406195E 4526423N	65	B	Jersey	150	chickens, pigs, dogs, cats	BHCO, HOSP, RWBL
Farm C	Arcata S	10 406384E 4525233N	202	A	Holstein	360	dogs, cats, pigeons	BHCO, HOSP, RWBL
Farm D	Loleta	10 393132E 4499950N	162	A	Holstein	325	dogs	BHCO, HOSP, WCSP
Farm E	Ferndale	10 400774E 4489599N	49	A	Holstein	700	dogs, cats, pigeons	BHCO, EUST, WCSP

[a] UTM coordinates in North American Datum 1983

[b] Milk grade according to California Department of Food and Agriculture

[c] Total number of cattle present on farm as reported by farmer at the time of sampling

[d] BHCO = brown-headed cowbird; EUST = European starling; HOSP = house sparrow; RWBL = redwing blackbird; WCSP = white-crowned sparrow

mist nets were routinely checked to remove birds; non-target bird species were released upon capture. Target bird species were held individually in a clean paper bag for up to 30 min until processed. The species of each bird was recorded, as well as its age and sex, when possible. Body mass (g) as well as length (mm) measurements of skull, tarsus and wing were taken. Cloacae and, when applicable, bird feces deposited in the bag, were swabbed with sterile cotton swabs (2 swabs per bird). The swabs were then placed in 2ml sterile saline (0.9% NaCl) and stored on ice in a cooler until transported to the laboratory. All sample collections were performed in accordance with IACUC protocol number 01/02.W.126.A, approved 6 June 2002.

Livestock

Fecal samples also were collected from 100 dairy cattle at each farm (20 samples per farm) between 16 November 2002 and 5 April 2004. Freshly voided fecal samples were swabbed with a sterile cotton swab and then placed in 2ml sterile saline (0.9% NaCl) and stored on ice in a cooler until transported to the laboratory.

Bacterial isolation

In the laboratory, cotton swabs immediately were streaked for isolated colonies onto three different types of media plates: Bacto-MacConkey agar (Difco, Becton-Dickinson, Sparks, Maryland, USA), Levine Eosin Methylene Blue (EMB) agar (BBL, Becton-Dickinson, Cockeysville, Maryland), and Bacto-Trypticase Soy Agar with 0.5% yeast extract (TSA/YE; Difco). Plates were incubated for 24 to 48 hr at 37°C. For the enrichment of *Salmonella* spp., 1 ml of the 2ml saline suspension for the sample was inoculated into 9ml Selenite Broth (Remel, Lenexa, Kansas) and incubated for 18 hr at

37°C. A sample was then streaked onto *Salmonella-Shigella* (SS) Agar (BBL) and incubated at 37°C for 24 hr. If no growth was present after 24 hrs, plates were incubated for an additional 24 hr. For the enrichment of *Yersinia* spp., the remaining 1 ml of saline suspension for the sample was inoculated into Trypticase Soy Broth with 0.5% yeast extract (Difco) and incubated at 4°C for 3 to 12 months. The suspension was then streaked onto *Yersinia* Selective Agar with *Yersinia* Antimicrobic Supplement CN (Difco) and incubated for 24 to 48 hr at 30°C.

At least one representative of each colony type from each type of plate was re-streaked onto TSA/YE plates to obtain pure cultures. Initial biochemical tests, including SIM medium (BBL) and Kligler Iron Agar (Difco), and a Gram stain were used to differentiate colony types. Identification of Gram-negative bacteria was completed with an API 20E system (bioMérieux, Inc., Durham, North Carolina, USA).

Statistical Analysis

Chi-square analyses were used to compare the prevalences of bacteria between species of wild birds, between wild birds sampled on different farms, and between cattle sampled on different farms (Zar 1999). When the expected cell values fell below 5 in a Chi-square analysis, Fisher's exact tests were used to compare differences between two distinct groups (Zar 1999). No statistical tests were possible in some instances involving comparisons between more than two groups that had expected cell values below 5. Fisher's exact tests also were used to compare prevalence of bacteria between birds and

cattle, between seasons, and between birds of different ages, weight, and sex. In addition, a Kruskal-Wallis one-way analysis of variance was used to compare the mean number of isolates (i.e., diversity of bacteria) carried by the same bird species on different farms, by the different bird species on the same farm, by bird species collectively, and by cattle on different farms (Zar 1999). Alpha was equal to 0.05 for each test. Although the potential for Type I error was relatively high due to the number of tests performed, it also was recognized that any attempt to decrease the significance level for any given test, such as applying the Bonferroni adjustment, would increase the occurrence of Type II error. Thus the actual P-values are reported for each statistical test.

RESULTS

Birds

Thirty-seven bacterial species representing 14 genera were isolated from 243 wild birds (Table 2; Appendix A). The most prevalent bacterial groups recorded for birds included *E. coli* (93/243; 38%), *Pantoea* spp. (formally *Enterobacter agglomerans*) (41/243; 17%), *Enterobacter* spp. (39/243; 16%), *Yersinia* spp. (29/243; 12%), and *Citrobacter* spp. (27/243; 11%). No cases were observed in which more than one species of a single genus was isolated from the same bird (Table 2).

The prevalence of *E. coli* was higher in female (7/8; 88%) than in male (2/7; 29%) Brewer's blackbirds (Fisher's exact, Z = 2.32; P = 0.041). One Brewer's blackbird was excluded from analysis due to indeterminate sex. The prevalence of *Yersinia* spp. was higher in male (6/25; 24%) than female (0/21; 0%) brown-headed cowbirds (Z = -2.41; P = 0.025). Ten cowbirds were excluded from analysis due to indeterminate sex. For white-crowned sparrows, prevalences of *Citrobacter* (adults 4/19; 21%, juveniles 7/10; 70%; Z = -2.58; P = 0.017) and *Enterobacter* (adults 6/19; 32%, juveniles 8/10; 80%; Z = -2.48; P = 0.021) species each were higher in juveniles than adults. However, because only four of the 52 comparisons in bacterial species composition between birds by age, sex, and weight were significant, the age, sex, and weight were pooled by species in subsequent analyses.

The prevalence of bacterial species varied between farms, with nine bacterial species varying significantly between farms (Table 3). Some bacterial species were

Table 2. Prevalence (number positive/total tested) of enteric bacteria isolated from the 243 wild birds captured on five dairy farms located in western Humboldt County, California between July 2002 and February 2004.

	Birds	
Bacteria	Number positive	Prevalence (%)
Citrobacter braakii	1	0.4
Citrobacter koseri/amalonaticus	6	2.5
Citrobacter koseri/farmeri	5	2.1
Citrobacter youngae	2	0.8
other *Citrobacter* spp.	13	5.3
Enterobacter amnigenus	6	2.5
Enterobacter cancerogenus	2	0.8
Enterobacter cloacae	5	2.1
other *Enterobacter* spp.	26	10.7
Escherichia coli	93	38.3
Escherichia hermannii	15	6.2
Escherichia vulneris	4	1.6
other *Escherichia* spp.	1	0.4
Hafnia alvei	15	6.2
Klebsiella pneumoniae ssp. *ozaenae*	1	0.4
Klebsiella pneumoniae ssp. *pneumoniae*	5	2.1
Klebsiella pneumoniae ssp. *rhinoscleromatis*	5	2.1
Klebsiella terrigena	1	0.4
other *Klebsiella* spp.	1	0.4
Moellerella wisconsensis	6	2.5
Pantoea spp.	41	16.9
Proteus vulgaris	19	7.8
Providencia rettgeri	11	4.5
Providencia stuartii	5	2.1
other *Providencia* spp.	2	0.8
Pseudomonas spp.	24	9.9
Rahnella aquatilis	3	1.2
Serratia fonticola	10	4.1
Serratia liquefaciens	2	0.8
Serratia marcescens	1	0.4
other *Serratia* spp.	4	1.6
Stenotrophomonas maltophila	1	0.4
Yersinia enterocolitica	20	8.2
Yersinia frederiksenii/intermedia	2	0.8
Yersinia kristensenii	2	0.8
Yersinia pseudotuberculosis	1	0.4
other *Yersinia* spp.	4	1.6

Table 2. Prevalence (number positive/total tested) of enteric bacteria isolated from the 243 wild birds captured on five dairy farms located in western Humboldt County, California between July 2002 and February 2004 (continued).

	Birds	
Bacteria	Number positive	Prevalence (%)
all *Citrobacter* spp.[a]	27	11.1
all *Enterobacter* spp.[a]	39	16.0
all *Escherichia* spp.[a]	113	46.5
all *Klebsiella* spp.[a]	13	5.3
all *Providencia* spp.[a]	18	7.4
all *Serratia* spp.[a]	17	7.0
all *Yersinia* spp.[a]	29	11.9

[a] includes all species of a particular genus

Table 3. Selected enteric bacteria isolated from 243 wild birds on each of the five dairy farms, Humboldt County, California, between July 2002 and February 2004 and the significance of the difference in prevalence between farms.

	Farm A	(n = 44)	Farm B	(n = 59)	Farm C	(n = 64)	Farm D	(n = 43)	Farm E	(n = 33)	Significance of difference[a]	
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[b]	P-value
all *Citrobacter* spp.	8	18.2	3	5.1	5	7.8	6	14.0	5	15.2	$X^2 = 2.65$	0.3
Enterobacter cloacae	0	0.0	0	0.0	5	7.8[c]	0	0.0	0	0.0	Z = 3.78	0.001
all *Enterobacter* spp.	10	22.7	5	8.5	7	10.9	8	18.6	9	27.3	$X^2 = 8.50$	0.08
Escherichia coli	18	40.9	26	44.1	25	39.1	12	27.9	12	36.4	$X^2 = 2.99$	0.6
Escherichia hermannii	15	34.1[c]	0	0.0	0	0.0	0	0.0	0	0.0	Z = 8.50	0.0001
Escherichia vulneris	0	0.0	0	0.0	0	0.0	0	0.0	4	12.1[c]	Z = 5.09	0.0003
Hafnia alvei	14	31.8[c]	1	1.7	0	0.0	0	0.0	0	0.0	Z = 7.81	0.0001
Klebsiella pneumoniae ssp. *rhinoscleromatis*	0	0.0	0	0.0	0	0.0	5	11.6[c]	0	0.0	Z = 4.87	0.0001
all *Klebsiella* spp.	2	4.5	3	5.1	1	1.6	7	16.3	0	0.0	NA	NA
Pantoea spp.	18	40.9[c]	5	8.5	6	9.4	8	18.6	4	12.1	$X^2 = 22.87$	0.00004
Proteus vulgaris	0	0.0	4	6.8	8	12.5	2	4.7	5	15.2	NA	NA
Providencia rettgeri	3	6.8	5	8.5	0	0.0	1	2.3	2	6.1	NA	NA
Providencia stuartii	0	0.0	0	0.0	0	0.0	0	0.0	5	15.2[c]	Z = 5.70	0.00004
all *Providencia* spp.	4	9.1	6	10.2	0	0.0	1	2.3	7	21.2	NA	NA
Pseudomonas spp.	5	11.4	5	8.5	4	6.3	2	4.7	8	24.2	NA	NA
Serratia fonticola	0	0.0	4	6.8	3	4.7	1	2.3	2	6.1	NA	NA
all *Serratia* spp.	1	2.3	4	6.8	7	10.9	2	4.7	3	9.1	NA	NA
Yersinia enterocolitica	4	9.1	6	10.2	0	0.0	0	0.0	10	30.3[c]	Z = -2.91	0.01
all *Yersinia* spp.	6	13.6	7	11.9	2	3.1	0	0.0	14	42.4[c]	$X^2 = 14.00$	0.001

[a] NA = unable to perform statistical test due to expected cell value less than 5 for Chi-square analyses or where comparisons between two distinct groups were not possible for Fisher's exact tests

[b] X^2 = test statistic for Chi-square analysis; Z = test statistic for Fisher's exact test

[c] Prevalence significantly different from the other farms for that particular bacterial species

recovered from birds only at a single farm, including *Enterobacter cloacae*, *Escherichia hermannii*, *Escherichia vulneris*, *Klebsiella pneumoniae* ssp. *rhinoscleromatis*, and *Providencia stuartii* (Table 3). Fourteen of the 15 isolates of *Hafnia alvei* occurred at Farm A (Table 3) while, *E. vulneris* was recovered from a single bird species on Farm E (Tables 3, 4). All other bacteria found only on a single farm were recovered from at least two bird species on that farm. Some bacterial species were isolated only once, in one individual bird on one farm, and so were excluded from statistical analyses (Table 2). The diversity of bacteria among birds (mean number of bacterial isolates identified ± SE) was highest on Farm A (2.41±0.26), followed by Farm E (2.18±0.34), Farm B (1.19±0.15), Farm D (1.16±0.15), and Farm C (0.98±0.10) (Kruskal-Wallis one-way ANOVA, H_C = 28.19; df = 4; P = 0.00001). The diversity of bacteria among birds on Farms A and E were significantly higher than on farms B, C, and D (Kruskal-Wallis multiple comparison test).

Most significant variation in prevalence of bacteria among wild bird species was associated with differences between white-crowned sparrows and other birds (Table 4). White-crowned sparrows had higher prevalences of *Enterobacter*, *Pseudomonas*, and *Yersinia* species and a lower prevalence of *E. coli* than any of the other five bird species (Table 4). In addition, white-crowned sparrows were the only bird species from which *E. vulneris* was isolated and the bird species from which four of the five *P. stuartii* isolates were found. However, cowbirds had four of the five *E. cloacae* isolates (Table 4). White-crowned sparrows carried the highest mean (±SE) diversity of bacterial species (2.72±0.35), followed by starlings (2.00±0.29), Brewer's blackbirds (1.50±0.10) redwing

Table 4. Prevalence of selected enteric bacteria isolated from the six wild bird species captured on five dairy farms between July 2002 and February 2004 in Humboldt County, California, and the significance of the difference in prevalence between bird species.

	BRBL[a] (n = 16)		BHCO[a] (n = 56)		EUST[a] (n = 34)		HOSP[a] (n = 65)		RWBL[a] (n = 43)		WCSP[a] (n = 29)		Significance of difference[b]	
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[c]	P-value
all *Citrobacter* spp.	3	18.8	2	3.6	4	11.8	3	4.6	4	9.3	11	37.9	NA	NA
Enterobacter cloacae	0	0.0	4	7.1[d]	0	0.0	0	0.0	0	0.0	1	3.4	Z = 3.06	0.01
all *Enterobacter* spp.	4	25.0	6	10.7	7	20.6	4	6.2	4	9.3	14	48.3[d]	X^2 = 15.49	0.0004
Escherichia coli	9	56.3	14	25.0	22	64.7	27	41.5	17	39.5	4	13.8[d]	X^2 = 15.31	0.004
Escherichia hermannii	1	6.3	1	1.8	4	11.8	4	6.2	5	11.6	0	0.0	NA	NA
Escherichia vulneris	0	0.0	0	0.0	0	0.0	0	0.0	0	0.0	4	13.8[d]	Z = 5.48	0.0002
Hafnia alvei	1	6.3	0	0.0	6	17.6	3	4.6	5	11.6	0	0.0	Z = 0.75	0.5
Klebsiella. pneumoniae. ssp. *rhinoscleromatis*	0	0.0	1	1.8	0	0.0	4	6.2	0	0.0	0	0.0	NA	NA
all *Klebsiella* spp.	0	0.0	2	3.6	0	0.0	5	7.7	3	7.0	3	10.3	NA	NA
Pantoea spp.	2	12.5	9	16.1	7	20.6	2	3.1	13	30.2	8	27.6	X^2 = 3.26	0.4
Proteus vulgaris	1	6.3	3	5.4	0	0.0	7	10.8	2	4.7	6	20.7	Z = -1.29	0.2
Providencia rettgeri	1	6.3	4	7.1	2	5.9	0	0.0	3	7.0	1	3.4	NA	NA
Providencia stuartii	0	0.0	1	1.8	0	0.0	0	0.0	0	0.0	4	13.8[d]	Z = 4.74	0.0008
all *Providencia* spp.	1	6.3	6	10.7	3	8.8	0	0.0	3	7.0	5	17.2	Z = -0.85	0.5
Pseudomonas spp.	0	0.0	5	8.9	4	11.8	4	6.2	2	4.7	9	31.0[d]	Z = -2.61	0.01
Serratia fonticola	0	0.0	4	7.1	3	8.8	1	1.5	0	0.0	2	6.9	NA	NA
all *Serratia* spp.	0	0.0	6	10.7	4	11.8	3	4.6	1	2.3	3	10.3	NA	NA
Yersinia enterocolitica	0	0.0	3	5.4	2	5.9	3	4.6	3	7.0	9	31.0	NA	NA
all *Yersinia* spp.	0	0.0	6	10.7	4	11.8	3	4.6	4	9.3	12	41.4[d]	Z = -3.28	0.002

[a] BRBL = Brewer's blackbird; BHCO = brown-headed cowbird; EUST = European starling; HOSP = house sparrow; RWBL = redwing blackbird; WCSP = white-crowned sparrow

[b] NA = unable to perform statistical test due to expected cell value less than 5 for Chi-square analyses or where comparisons between two distinct groups were not possible for Fisher's exact tests

[c] X^2 = test statistic for Chi-square analysis; Z = test statistic for Fisher's exact test

[d] Prevalences significantly different from the other bird species for that particular bacterium

blackbirds (1.49±0.18), cowbirds (1.14±0.15), and house sparrows (1.02±0.14) (H_C = 29.95; df = 5; P = 0.00002). The diversity of bacterial species was significantly higher in white-crowned sparrows and European starlings than in cowbirds and house sparrows (Kruskal-Wallis multiple comparison test). Diversity was also significantly higher in white-crowned sparrows than in Brewer's blackbirds and redwing blackbirds (Kruskal-Wallis multiple comparison test).

Statistical comparisons between prevalence of bacterial species carried by the same bird species on different farms and between different bird species on the same farm were not possible due to low sample sizes. However, the mean number of different bacterial species isolated (i.e., diversity) was compared between the same bird species sampled on different farms. The mean number of bacterial isolates for white-crowned sparrows was higher on Farm E than Farm D (H_C = 9.20; df = 1; P = 0.002). For starlings, diversity was highest on Farm A, followed by farms E and B (H_C = 18.73; df = 2; P = 0.00009). Diversity for redwing blackbirds was highest on Farm A, followed by farms B and C (H_C = 11.44; df = 2; P = 0.003). Although not significantly different between farms, bacterial species diversity for cowbirds was highest on Farm B, followed by farms E, D, and C (H_C = 5.44; df = 3; P = 0.1). Similarly for house sparrows, diversity was not significantly different between farms, but was highest on Farm A, followed by farms C, B, and D (H_C = 5.86; df = 3; P = 0.1). Comparisons for Brewer's blackbirds were not possible due to small sample sizes on each farm.

The mean number of bacteria isolated was compared between the different bird species sampled on the same farm. Diversity on Farm A was highest in starlings,

followed by redwing blackbirds and house sparrows (H_C = 11.58; df = 2; P = 0.003). On Farm D, diversity was highest in white-crowned sparrows, followed by cowbirds and house sparrows (H_C = 7.87; df = 2; P = 0.02). Diversity on Farm E was highest in white-crowned sparrows, followed by starlings and cowbirds (H_C = 15.59; df = 2; P = 0.0004). Although not significantly different, diversity on Farm B, was highest in cowbirds, followed by starlings, redwing blackbirds, and house sparrows (H_C = 3.55; df = 3; P = 0.3). Again, while the diversity was not significantly different among bird species on Farm C, bacterial diversity was highest in house sparrows, followed by redwing blackbirds and cowbirds (H_C = 3.90; df = 2; P = 0.1).

Bacterial species prevalences were higher among birds in summer (April through September) for *E. coli*, *E. hermannii*, *H. alvei*, and *Moellerella wisconsensis*, and higher in winter (October through March) for *P. vulgaris*, *Citrobacter* spp., *Enterobacter* spp., and *Yersinia* spp. (Table 5).

By farm, the prevalence of *E. coli* was significantly higher in summer (13/21; 62%) than in winter (12/43; 28%) on Farm C (Z = 2.62; P = 0.01). On Farm D *Citrobacter* spp. was significantly higher (Z = -3.43; P = 0.001) in winter (6/16; 38%) than in summer (0/27; 0%). *Enterobacter* spp. also was significantly higher (Z = -4.07; P = 0.00009) in winter (8/16; 50%) than in summer (0/27; 0%) on Farm D. Among individual bird species, brown-headed cowbirds had higher prevalence (Z = -2.78; P = 0.007) of *Enterobacter* spp. in winter (6/30; 43%) than in summer (0/26; 0%) while European starlings had higher prevalence (Z = 2.69; P = 0.01) of *Enterobacter* spp. in summer (6/14; 43%) than in winter (1/20; 5%). Starlings also had higher prevalence

Table 5. Prevalence of enteric bacteria with significant seasonal differences isolated from the wild birds captured on five dairy farms between July 2002 and February 2004 in Humboldt County, California.

	Birds				Significance of difference	
	Summer[a]	(n = 126)	Winter[b]	(n = 117)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[c]	P-value
all *Citrobacter* spp.	8	6.3	19	16.2	-2.45	0.02
all *Enterobacter* spp.	13	10.3	26	22.2	-2.53	0.01
Escherichia coli	57	45.2	36	30.8	2.39	0.03
Escherichia hermannii	15	11.9	0	0.0	3.85	0.00005
Hafnia alvei	14	11.1	1	0.9	3.32	0.0008
Moellerella wisconsensis	6	4.8	0	0.0	2.39	0.03
Proteus vulgaris	5	4.0	14	12.0	-2.32	0.03
all *Yersinia* spp.	9	7.1	20	17.1	-2.39	0.02

[a] Summer is April through September

[b] Winter is October through March

[c] Fisher's exact Z-value

(Z = 3.23; P = 0.002) of *H. alvei* in summer (6/14; 43%) than in winter (0/20; 0%). The prevalence of *Pantoea* spp. for starlings was higher (Z = 2.69; P = 0.01) in summer (6/14; 43%) than in winter (1/20; 5%). House sparrows had higher prevalence (Z = 2.20; P = 0.04) of *E. coli* in summer (22/43; 51%) than in winter (5/22; 23%).

Cattle

Twenty-four bacterial species among 13 genera were recovered from 100 cattle (Table 6; Appendix B). The most prevalent bacteria isolated from cattle were *E. coli* (88/100; 88%), *Citrobacter* spp. (18/100; 18%), *Aeromonas hydrophila* (13/100; 13%), *Proteus vulgaris* (13/100; 13%), and *Citrobacter braakii* (11/100; 11%).

The prevalences of two bacterial species, *Klebsiella oxytoca* and *K. pneumoniae* ssp. *pneumoniae*, were significantly higher from cattle on Farm C than the other farms (Table 7). Although not statistically significant, the prevalence of *A. hydrophila* was considerably higher for cattle from Farm E (35%) when compared to the other farms (< 15%); while the prevalence of *E. coli* was lower for cattle from Farm A (60%) than on the other farms (95%) (Table 8).

Prevalences also varied significantly for *A. hydrophila* (Z = 2.55; P = 0.01), *E. coli* (Z = 2.64; P = 0.01), and *Klebsiella* spp. (Z = 2.72; P = 0.005) when cattle breeds were considered; although such differences also may reflect differences between farms. That is, the prevalence of *E. coli* was significantly lower among Jersey (31/40; 78%) than Holstein (57/60; 95%) cattle; however, the prevalence of *E. coli* also was lower among

Table 6. Prevalence (number positive/total tested) of enteric bacteria sampled on five dairy farms located in western Humboldt County, California, between November 2002 and April 2004.

	Cattle	
Bacteria	Number positive	Prevalence (%)
Aeromonas hydrophila	13	13.0
Aeromonas spp.	1	1.0
Citrobacter braakii	11	11.0
Citrobacter koseri/amalonaticus	4	4.0
Citrobacter youngae	3	3.0
Enterobacter amnigenus	2	2.0
other *Enterobacter* spp.	2	2.0
Escherichia coli	88	88.0
Hafnia alvei	5	5.0
Klebsiella oxytoca	3	3.0
Klebsiella pneumoniae ssp. *pneumoniae*	5	5.0
Klebsiella terrigena	2	2.0
Pantoea spp.	2	2.0
Proteus vulgaris	13	13.0
Providencia rettgeri	1	1.0
Providencia stuartii	1	1.0
Pseudomonas flourescens/putida	2	2.0
Pseudomonas spp.	1	1.0
Serratia fonticola	6	6.0
Serratia liquefaciens	1	1.0
Shewanella putrefaciens	4	4.0
Yersinia enterocolitica	8	8.0
Yersinia pseudotuberculosis	1	1.0
other *Yersinia* spp.	1	1.0
all *Citrobacter* spp.[a]	18	18.0
all *Enterobacter* spp.[a]	4	4.0
all *Klebsiella* spp.[a]	10	10.0
all *Serratia* spp.[a]	7	7.0
all *Yersinia* spp.[a]	10	10.0

[a] includes all species of a particular genus

Table 7. Prevalence of selected enteric bacteria isolated from 100 dairy cattle sampled on each of the five dairy farms located in western Humboldt County, California, between November 2002 and April 2004 and the significance of the difference in prevalence between farms.

	Farm A	(n = 20)	Farm B	(n = 20)	Farm C	(n = 20)	Farm D	(n = 20)	Farm E	(n = 20)	Significance of difference[a]	
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[b]	P-value
Aeromonas hydrophila	1	5.0	0	0.0	3	15.0	2	10.0	7	35.0[c]	NA	NA
Citrobacter braakii	0	0.0	2	10.0	5	25.0	4	20.0	0	0.0	NA	NA
all *Citrobacter* spp.	0	0.0	6	30.0	7	35.0	5	25.0	0	0.0	$X^2 = 0.48$	0.8
Escherichia coli	12	60.0[c]	19	95.0	19	95.0	19	95.0	19	95.0	NA	NA
Klebsiella oxytoca	0	0.0	0	0.0	3	15.0[c]	0	0.0	0	0.0	Z = 3.52	0.007
Klebsiella pneumoniae ssp. *pneumoniae*	0	0.0	0	0.0	4	20.0[c]	1	5.0	0	0.0	Z = 3.44	0.005
Proteus vulgaris	0	0.0	5	25.0	2	10.0	2	10.0	4	20.0	NA	NA
Serratia fonticola	1	5.0	0	0.0	1	5.0	3	15.0	1	5.0	NA	NA
Yersinia enterocolitica	2	10.0	2	10.0	3	15.0	0	0.0	1	5.0	NA	NA

[a] NA = unable to perform statistical test due to expected cell value less than 5 for Chi-square analyses or where comparisons between two distinct groups were not possible for Fisher's exact tests

[b] X^2 = test statistic for Chi-square analysis; Z = test statistic for Fisher's exact test

[c] Prevalence significantly different from the other farms for that particular bacterium

Table 8. Prevalence (number positive/total tested) of selected enteric bacteria isolated from both wild birds and dairy cattle on five dairy farms located in western Humboldt County, California, between July 2002 and April 2004.

	Birds	(n = 243)	Cattle	(n = 100)	Significance of difference	
	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
Citrobacter braakii	1	0.4	11	11.0	-4.85	0.00008
Citrobacter koseri/amalonaticus	6	2.5	4	4.0	-0.77	0.5
Citrobacter youngae	2	0.8	3	3.0	-1.53	0.2
all *Citrobacter* spp.	27	11.1	18	18.0	-1.72	0.1
Enterobacter amnigenus	6	2.5	2	2.0	-0.77	0.5
all *Enterobacter* spp.	39	16.0	4	4.0	3.06	0.002
Escherichia coli	93	38.3	88	88.0	-10.16	0.0001
Hafnia alvei	15	6.2	5	5.0	0.42	0.8
Klebsiella pneumonia spp. *pneumoniae*	5	2.1	5	5.0	-1.48	0.2
Klebsiella terrigena	1	0.4	2	2.0	-1.44	0.2
all *Klebsiella* spp.	13	5.3	10	10.0	-1.56	0.2
Pantoea spp.	41	16.9	2	2.0	3.78	0.0005
Proteus vulgaris	19	7.8	13	13.0	-1.5	0.2
Providencia rettgeri	11	4.5	1	1.0	1.62	0.2
Providencia stuartii	5	2.1	1	1.0	0.68	0.7
all *Providencia* spp.	18	7.4	2	2.0	1.94	0.1
Pseudomonas spp.	24	9.9	3	3.0	2.15	0.05
Serratia fonticola	10	4.1	6	6.0	-0.75	0.6
Serratia liquefaciens	2	0.8	1	1.0	-0.16	1.0
all *Serratia* spp.	17	7.0	7	7.0	-0.001	1.0
Yersinia enterocolitica	20	8.2	8	8.0	0.07	1.0
Yersinia pseudotuberculosis	1	0.4	1	1.0	-0.65	0.5
all *Yersinia* spp.	29	11.9	10	10.0	0.51	0.7

[a] Fisher's exact Z-value

cattle from Farm A (Jersey) than the other four farms (Table 7). The prevalence of *A. hydrophila* was higher in Holstein (12/60; 20%) than Jersey (1/40; 3%); however, the prevalence of *A. hydrophila* also was higher among cattle from Farm E (Holstein) than the other farms (Table 7). The prevalence of *Klebsiella* spp. was higher in Holstein (10/60; 17%) than Jersey (0/40; 0%); however, the prevalence of *Klebsiella* spp. also was higher among cattle from Farm C (Holstein) than the other farms (Table 7).

The mean (±SE) diversity of bacterial species carried by cattle varied significantly between farms (H_C = 31.85; df = 4; P = 0.000002). Diversity was highest on Farm C 2.55±0.28; n= 20), followed by Farm D (2.00±0.25; n = 20), Farm E (1.80±0.21; n = 20), Farm B (1.75±0.23; n = 20), and Farm A (0.90±0.20; n = 20).

Escherichia coli (Z = -2.85; P = 0.005) was more prevalent in winter (53/55; 96%) than in summer (35/45; 78%). *Citrobacter* spp. (Z = -3.19; P = 0.001) also was more prevalent in winter (16/55; 29%) than in summer (2/45; 4%). Prevalence for all other bacterial species was not significantly different between summer and winter.

Birds and Cattle

Nineteen bacterial species representing 10 genera were recovered from both wild birds and dairy cattle (Table 8). Cattle had a significantly higher prevalence of *C. braakii* and *E. coli* than birds, while birds had higher prevalence of *Pantoea* spp., *Pseudomonas* spp., and *Enterobacter* spp. (Table 8). Eleven of these 19 bacterial species had the same API 20E code represented in both birds and cattle (Table 9).

Table 9. Number of identical API codes for the same bacterial species recorded in both birds and cattle out of the total number of API codes recorded for a given bacterial species.

Bacteria	Identical API codes in birds and cattle (n)	API codes compared (n)
Citrobacter braakii	0	4
Citrobacter koseri/amalonaticus	0	3
Citrobacter youngae	0	2
Enterobacter amnigenus	0	2
other *Enterobacter* spp.	1	9
Escherichia coli	3	14
Hafnia alvei	0	4
Klebsiella pneumoniae spp. *pneumoniae*	1	3
Klebsiella terrigena	0	2
Pantoea spp.	2	8
Proteus vulgaris	1	3
Providencia rettgeri	1	6
Providencia stuartii	1	3
Pseudomonas spp.	1	2
Serratia fonticola	2	5
Serratia liquefaciens	0	2
Yersinia enterocolitica	2	5
Yersinia pseudotuberculosis	1	1
other *Yersinia* spp.	0	2
Total	16	80

When comparing differences in prevalence by farm, birds sampled on Farm A had higher prevalence of *Citrobacter* spp., *H. alvei*, and *Pantoea* spp. than cattle on Farm A (Table 10). Cattle from Farm B had higher prevalence of *Citrobacter* spp., *E. coli* and *P. vulgaris* than the birds sampled on Farm B (Table 10). Cattle from Farm C also had higher prevalence of *Citrobacter* spp., *E. coli*, and *Klebsiella* spp. than birds on Farm C (Table 10). Birds sampled on Farm D had higher prevalence of *Enterobacter* spp. and *Pantoea* spp., while cattle had higher prevalence of *E. coli* (Table 10). Farm D also was the only farm in which no *Yersinia* spp. were isolated from either birds or cattle. Birds on Farm E had significantly higher prevalence of *Enterobacter* spp., *Providencia* spp., *Pseudomonas* spp., and *Y. enterocolitica* than the cattle on Farm E, while cattle had a higher prevalence of *E. coli* (Table 10). In addition, the only two isolates of *Y. pseudotuberculosis* were recovered from the feces of one bird and one cow respectively, from Farm E.

Table 10. Prevalence (number positive/total tested) of enteric bacteria with significant differences in prevalence between the wild birds and dairy cattle sampled on each of five dairy farms located in western Humboldt County, California, between July 2002 and April 2004.

	Farm A				Significance of difference	
	Birds	(n = 44)	Cattle	(n = 20)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
all *Citrobacter* spp.	8	18.2	0	0.0	2.04	0.05
Hafnia alvei	14	31.8	0	0.0	2.85	0.003
Pantoea spp.	18	40.9	0	0.0	3.37	0.0006
	Farm B				Significance of difference	
	Birds	(n = 59)	Cattle	(n = 20)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
all *Citrobacter* spp.	3	5.1	6	30.0	-3.03	0.007
Escherichia coli	26	44.1	19	95.0	-3.98	0.00005
Proteus vulgaris	4	6.8	5	25.0	-2.22	0.04
	Farm C				Significance of difference	
	Birds	(n = 64)	Cattle	(n = 20)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
all *Citrobacter* spp.	5	12.5	7	35.0	-3.03	0.006
Escherichia coli	25	39.1	19	95.0	-4.37	0.00001
all *Klebsiella* spp.	1	3.1	7	35.0	-4.45	0.0001
	Farm D				Significance of difference	
	Birds	(n = 43)	Cattle	(n = 20)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
all *Enterobacter* spp.	8	18.6	0	0.0	2.06	0.05
Escherichia coli	12	27.9	19	95.0	-4.96	0.0001
Pantoea spp.	8	18.6	0	0.0	2.06	0.05
	Farm E				Significance of difference	
	Birds	(n = 33)	Cattle	(n = 20)		
Bacteria	Number positive	Prevalence (%)	Number positive	Prevalence (%)	Test statistic[a]	P-value
all *Enterobacter* spp.	9	27.3	0	0.0	2.56	0.01
Escherichia coli	12	36.4	19	95.0	-4.20	0.00003
all *Providencia* spp.	7	21.2	0	0.0	2.21	0.04
Pseudomonas spp.	8	24.2	0	0.0	2.39	0.02
Yersinia enterocolitica	10	30.3	1	5.0	2.20	0.04

[a] Fisher's exact Z-value

DISCUSSION

A variety of potentially pathogenic bacteria were isolated from the cloacae and feces of wild birds on local dairy farms, as well as from the feces of dairy cattle on the farms. The most prevalent bacterial species isolated from both birds and cattle in this study, *Escherichia coli*, is a common inhabitant of the intestinal tract of numerous vertebrate species (Janda and Abbott 1998). In the past *E. coli* also has been reported from bird species, such as European starlings, house sparrows, house finches (*Carpodacus mexicanus*) (Morishita et al. 1999), brown-headed cowbirds (Johnson et al. 1980), and black-capped chickadees (*Parus atrocapillus*) (Brittingham et al. 1988). While most strains of *E. coli* are considered commensal, some strains are responsible for a variety of illnesses, including urinary tract infections, gastroenteritis, and bacteremia (Janda and Abbott 1998). Specifically, the Shiga toxin-producing *E. coli* (STEC) serotype 0157:H7 has been associated with human outbreaks of hemorrhagic colitis and hemolytic uremic syndrome after the consumption of contaminated food, especially beef (Janda and Abbott 1998). In fact, cattle have been identified as important carriers of *E. coli*, including serotype 0157:H7, and may contribute to environmental contamination (Chapman et al. 1993, Hancock et al. 1994, Rahn et al. 1997). Serotyping of the strains isolated in this study would be necessary to determine if any birds or cattle were carrying pathogenic strains of *E. coli*.

Other bacterial species isolated from birds and cattle in this study, such as *Enterobacter* spp., *Citrobacter* spp., *Klebsiella* spp., *Pseudomonas* spp., and *Yersinia*

spp. commonly are isolated from both environmental and clinical sources (Janda and Abbott 1998). *Pseudomonas* spp. and *Klebsiella* spp. frequently are isolated from nosocomial infections involving immunocompromised patients (Janda and Abbott 1998). Although infections involving *Yersinia* spp. occur less frequently than those caused by other Enterobacteriaceae, yersiniosis has been observed in humans, domestic farm animals, and wild birds and mammals (Janda and Abbott 1998). Yersiniae are widely distributed within the environment and across geographic regions (Janda and Abbott 1998). Two species commonly pathogenic for humans and other animals include *Yersinia pseudotuberculosis* and *Y. enterocolitica* (Janda and Abbott 1998). These species have been isolated from a number of asymptomatic carriers, including humans (Fukushima et al. 1984), cattle and pigs (Shayegani et al. 1981, Tsubokura et al. 1989), free-living passerines (Kapperud and Rosef 1983, Kato et al. 1985, Brittingham et al. 1988, Hamasaki et al. 1989), as well as from food, milk, and water (Shayegani et al. 1981, Fukushima et al. 1984, Tsubokura et al. 1989). *Yersinia pseudotuberculosis* and *Y. enterocolitica* each were isolated from both birds and cattle in this study. Without further serotyping, it is unknown whether these were pathogenic strains, although most *Y. enterocolitica* strains isolated from environmental sources tend to be nonpathogenic (Janda and Abbott 1998). No pathogenic strains were observed among the 18 strains of *Yersinia enterocolitica* isolated from 6 captive animals and soil samples at the Sequoia Zoo, Eureka (Belltawn et al. 2004).

Bacterial species composition varied regularly between farms and between bird species. However, it was difficult to distinguish whether these differences were

influenced primarily by differences between farms or between bird species, due to consistent differences between bird species sampled among the farms. Farms varied in the number and species of birds they appeared to attract. In some cases, a bacterial species was found primarily at one farm, but among a variety of bird species. For example, *Escherichia hermannii*, *H. alvei*, and *Pantoea* spp. were isolated from birds on Farm A. All three of these bacterial species are common isolates of soil and water (Janda and Abbott 1998). Other bacterial species were isolated predominantly from one bird species sampled on one farm. For instance, four of five *Enterobacter cloacae* isolates were recovered from house sparrows on Farm C, while *Escherichia vulneris* and *Providencia stuartii* both were isolated primarily from white-crowned sparrows on Farm E. *Enterobacter cloacae* (Janda and Abbott 1998) and *Escherichia vulneris* (Brenner et al. 1982) both are commonly isolated from environmental sources, but have become increasingly common in nosocomial infections in hospitals (Janda and Abbott 1998). *Providencia stuartii* has caused diarrhea in dairy calves (Janda and Abbott 1998) and a few strains have been found to be antibiotic resistant (McHale et al. 1981). Thus bird species utilizing a particular farm may have the potential to influence the bacterial species composition of that farm, and subsequently, increase the availability of a specific bacterium for transmission.

There also was a striking difference between the diversity of bacteria carried by white-crowned sparrows, when compared to the other five bird species sampled, and in particular white-crowned sparrows sampled on Farm E. White-crowned sparrows were caught on farms D and E; however, the white-crowned sparrows sampled on Farm E had

more than twice the mean number of isolates than white-crowned sparrows on Farm D (Farm E, 4.17±0.53, n = 12; Farm D, 1.60±0.37, n = 10). White-crowned sparrows from Farm E had higher prevalence of *Enterobacter* spp. (Z = 0.08; P = 1.0), *Providencia* spp. (Z = -2.32; P = 0.04), *Pseudomonas* spp. (Z = -2.01; P = 0.08), and *Yersinia* spp. (Z = -4.28; P = 0.00003) than white-crowned sparrows from Farm D. However, *Enterobacter* spp. and *Pseudomonas* spp. prevalences between these two farms were not statistically significant. White-crowned sparrows from Farm E also had all four *E. vulneris* and four of the five *P. stuartii* isolates. Interestingly, the prevalence of *E. coli* was significantly lower in white-crowned sparrows than in all other bird species sampled (Table 4). These differences may reflect an interaction between certain farm characteristics, including soil type and number of livestock present, and the life history of white-crowned sparrows, such as diet or migratory tendencies. Diet influences bacterial composition in the intestinal tract among some birds. Glunder (2002) reported chickens being fed a strict seed diet had reduced intestinal colonization of *E. coli*. Brittingham et al. (1988) reported birds with an omnivorous diet had higher prevalence of some bacterial species than birds with a granivorous diet. In addition, white-crowned sparrows are found most abundantly on dairy farms during winter in Humboldt County compared to summer (Harris 1996). I only caught white-crowned sparrows in the winter. In contrast, house sparrows, the only strictly non-migratory species sampled (Harris 1996), had the lowest number of *E. coli* isolates. As such, white-crowned sparrows may be the least adapted to the bacteria found on farms or may ingest bacteria in both their summer and winter habitats; thus they may carry a higher diversity of bacterial species in their intestinal tract. Whether these bacteria

cause disease in white-crowned sparrows is unknown, as is the potential risk of transmission to dairy cattle on farms with large populations of white-crowned sparrows.

Cattle also carried a variety of potentially pathogenic bacteria, some of which varied significantly between farms. For instance, the cattle on Farm C had a high prevalence of *Klebsiella* spp., while no *Yersinia* spp. were isolated from either birds or cattle sampled on Farm D (Table 7). Differences between farms may reflect variation in animal husbandry (Hancock et al. 1994), as well as differences between soil properties, microclimate, or other features on farms (Jamieson et al. 2002). The main distinction in farm operations occurred on farms A and E. Calves on Farm A were kept in individual pens shortly after birth, whereas calves from the other farms were penned together. Although I did not specifically sample calves for bacteria, significantly fewer bacterial species were isolated from the adult cattle sampled on Farm A. In addition, the prevalence of *E. coli* was significantly lower from cattle on Farm A than the other farms. Cobbold and Desmarchelier (2002) found a similar result when studying the transmission of Shiga toxin-producing *E. coli* in dairy calves; the prevalence of Shiga toxin-producing *E. coli* was significantly higher among dairy calves penned together than among those penned individually. Thus, calves penned individually may have lower prevalences of pathogenic bacteria and consequently, the adults on the same farm also may have lower prevalence of the same bacterial species.

Farm E differed from all other farms in that, cattle were not put out to pasture, but confined to a barn 24 hours a day; as such, Farm E had a very high sustained density of cattle. Cattle from Farm E had the highest prevalence of *Aeromonas hydrophila* (Table

7). While *A. hydrophila* is not a member of the Enterobacteriaceae, it is a Gram-negative bacterium within the family Aeromonadaceae. *Aeromonas hydrophila* is commonly found in various water sources (Lightfoot 2003), but also has been recovered from the feces of farm animals and has potential to cause disease (Gray et al. 1990). I propose that the high prevalence of *A. hydrophila* recovered from cattle feces on Farm E most likely has to do with the water flushing system used to clean the cattle pens and that *A. hydrophila* originates as a biofilm in the water used to clean the cattle pens and the cattle inadvertently ingest the bacterium. It is unknown whether the high prevalence of *A. hydrophila* recovered from cattle on Farm E is causing disease in the cattle, but given the potential virulence of *A. hydrophila* (Chopra et al. 2000) it should be considered a significant risk.

Many of the same bacterial species were isolated from birds and cattle (Table 8), indicating there is a potential for transmission of bacteria between birds and cattle on farms. However, only 16 of 80 bacterial strains compared had the same API 20E code (Table 9). This suggests that these bacterial strains are not identical. Transmission, therefore, appears to be infrequent. Also to be considered, however, is that biochemical characteristics of some bacterial species may change when in the intestinal tract of different hosts or under different environmental conditions (Baumler et al. 1998, Gordon and Lee 1999, Buzoleva and Somov 2003). As such, birds and cattle could share more bacteria than is indicated in this study. In addition, without further serotyping of the bacterial species found in this study it is impossible to know the virulence potential of the bacteria and thus the real threat transmission would pose. For instance, *Y.*

psuedotuberculosis was found in one bird and one cow on Farm E. These strains did share the same API code so it is unknown whether these isolates were the same serotype and occurred as a result of transmission.

Interestingly, the mean diversity of bacterial species recovered from birds was nearly the inverse of the mean diversity of cattle. That is, the mean diversity for birds by farm was highest on Farm A, followed by farms E, B, D, and C; while the mean diversity for cattle by farm was highest on Farm C, followed by farms D, E, B, and A. This seems to support the idea that bacterial transmission between birds and cattle is an unlikely occurrence. The difference in bacterial diversity for birds by farm appears to be related to the bird species sampled on each farm. The bacterial diversity for bird species was highest in white-crowned sparrows, followed by European starlings, and redwing blackbirds and lowest in brown-headed cowbirds and house sparrows. As such, farms with large populations of white-crowned sparrows, European starlings, or redwing blackbirds, Farms A, E, and B, had increased bacterial diversity. Conversely, farms with large populations of brown-headed cowbirds or house sparrows, Farms D and C, had decreased bacterial diversity. The bacterial diversity in cattle by farm seems to be related more to farm characteristics, such as number of cattle, sanitary conditions, soil profile, or other animal husbandry practices. However, when the number of birds sampled on a particular farm increased, more similarities were found between the bacteria carried by birds and cattle. For example, on farms B and C I sampled 59 and 64 birds, respectfully, whereas on the remaining three farms I sampled less than 45 birds on each farm. There were more identical API 20E codes between the birds and cattle sampled on farms B and

C, than the other farms, suggesting that if the sample size was increased more similarities could be found. Therefore, the potential for bacterial transmission between birds and cattle exists, although prevalence appears to be low for most bacterial species. Consequently, transmission between birds and cattle may be more of a concern during an outbreak, when the prevalence of a particular bacterium is unusually high.

MANAGEMENT RECOMMENDATIONS

Based on these results, the transmission of pathogenic bacteria between birds and cattle may occur, though evidence suggests it may be infrequent. It is advisable to continue monitoring both birds and cattle for pathogenic bacteria. However, it also is important to consider the number and species of birds sampled, as well as the number of individual farms investigated. Specific studies on the actual likelihood and rates of transmission of select enteric bacteria between birds and cattle under experimental conditions may be more worthwhile as a first step to quantifying the problem. Determining the virulence of these bacterial species also would be valuable.

LITERATURE CITED

Altekruse, S.F., J.M. Hunt, L.K. Tollefson, and J.M. Madden. 1994. Food and animal sources of human *Campylobacter jejuni* infection. Journal of the American Veterinary Medical Association 204: 57-61.

Baumler, A.J., R.M. Tsolis, T.A. Ficht, and L.G. Adams. 1998. Evolution of host adaptation in *Salmonella enterica*. Infection and Immunity 66:4579-4587.

Belltawn, M., R.G. Botzler, J. Ware, W.K.W. Cheung, S. Abbott, R.N. Brown, J.M. Janda. 2004. Nonplague yersiniae in a zoo. Page 302. In Proceedings of American Association of Zoo Veterinarians, American Association of Wildlife Veterinarians, and Wildlife Disease Association Joint Conference, San Diego, California, August 28-September 3, 2004.

Botzler, R.G. 1979. Yersiniae in the soil of an infected wapiti range. Journal of Wildlife Diseases 15: 529-532.

Botzler, R.G. 2002. Avian cholera on north coast California: Distinctive epizootiological features. Annals of the New York Academy of Sciences 969: 224-228.

Brenner, D.J., A.C. McWhorter, J.K. Leete Knutson, and A.G. Steigerwalt. 1982. *Escherichia vulneris*: A new species of Enterobacteriaceae associated with human wounds. Journal of Clinical Microbiology 15: 1133-1140.

Brittingham, M.C., S.A. Temple, and R.M. Duncan. 1988. A survey of the prevalence of selected bacteria in wild birds. Journal of Wildlife Diseases 24: 299-307.

Buzoleva, L.S. and G.P. Somov. 2003. Adaptation variability of *Yersinia pseudotuberculosis* during long-term persistence in soil. Immunology and Microbiology 135: 536-540.

California Department of Food and Agriculture. 2000 Census of Agriculture. California Agricultural Statistics Service. http://www.cdfa.ca.gov. 1220 N Street, Sacramento, CA, USA, 95814-5607.

Casanovas, L., M. de Simon, M.D. Ferrer, J. Arques, and G. Monzon. 1995. Intestinal carriage of campylobacters, salmonellas, yersinias, and listerias in pigeons in the city of Barcelona. Journal of Applied Bacteriology 78: 11-13.

Chapman, P.A., C.A. Siddons, D.J. Wright, P. Norman, J. Fox, and E. Crick. 1993. Cattle as a possible source of verocytotoxin-producing *Escherichia coli* 0157:H7 infections in man. Epidemiology and Infection 111: 439-447.

Chopra, A.K., X.J. Xu, D. Ribardo, M. Gonzaalez, K. Kuhl, J.W. Peterson, and C.W. Houston. 2000. The cytotoxic enterotoxin of *Aeromonas hydrophila* induces proinflammatory cytokine production and activates arachidonic acid metabolism in macrophages. Infection and Immunity 68: 2808-2818.

Cizek, A., I. Literak, K. Hejlicek, F. Treml, and J. Smola. 1994. *Salmonella* contamination of the environment and its incidence in wild birds. Journal of Veterinary Medicine, Series B 41: 320-327.

Cobbold, R. and P. Desmarchelier. 2002. Horizontal transmission of shiga toxin producing *Escherichia coli* within groups of dairy calves. Applied and Environmental Microbiology 4148-4152.

Cole, J. 2003. Urban streams water quality report for 2003. Wastewater Utilization Program, Environmental Services Department. City of Arcata, California.

Craven, S.E., N.J. Stern, E. Line, J.S. Bailey, N.A. Cox, and P. Fedorka-Cray. 2000. Determination of the incidence of *Salmonella* spp., *Campylobacter jejuni*, and *Clostridium perfringens* in wild birds near broiler chicken houses by sampling intestinal droppings. Avian Diseases 44: 715-720.

Davies, R.H., and C. Wray. 1997. Distribution of *Salmonella* contamination in ten animal feedmills. Veterinary Microbiology 51: 159-169.

Fenlon, D.R. 1985. Wild birds and silage as reservoirs of *Listeria* in the agricultural environment. Journal of Applied Bacteriology 59: 537-543.

Fukushima, H., M. Tsubokura, K. Otsuki, and Y. Kawaoka. 1984. Biochemical heterogeneity of serotype 03 strains of 700 *Yersinia* strains isolated from humans, other mammals, flies, animal feed, and river water. Current Microbiology 11:149-153.

Goodchild, W.M., and J.F. Tucker. 1968. Salmonellae in British wild birds and their transfer to domestic fowl. British Veterinary Journal 124: 95-101.

Gordon, D.M. and J. Lee. 1999. The genetic structure of enteric bacteria from Australian mammals. Microbiology 145:2673-2682.

Glunder, G. 2002. Influence of diet on the occurrence of some bacteria in the intestinal flora of wild and pet birds. Deutsche Tierärtzliche Wochenschrift 109: 253-292.

Gray, S.J., D.J. Stickler and T.N. Bryant. 1990. The incidence of virulence factors in mesophilic *Aeromonas* species isolated from farm animals and their environment. Epidemiology and Infection 105: 277-294.

Hamasaki, S., H. Hayashidani, K. Kaneko, M. Ogawa, and Y. Shigeta. 1989. A survey for *Yersinia pseudotuberculosis* in migratory birds in coastal Japan. Journal of Wildlife Diseases 25: 401-403.

Hancock, D.D., T.E. Besser, M.L. Kinsel, P.I. Tarr, D.H. Rice, and M.G. Paros. 1994. The prevalence of *Escherichia coli* 0157:H7 in dairy and beef cattle in Washington State. Epidemiology and Infection 113: 199-207.

Harber, J. 1988. The identification and characterization of multiple resistant Enterobacteriaceae of one Humboldt County dairy farm. Master's thesis. Department of Biology. Humboldt State University, Arcata, California.

Harris, S. 1996. Northern California birds: A guide to the status, distribution, and habits of the birds of Del Norte, Humboldt, Trinity, northern Mendocino, and western Siskiyou Counties, California. Humboldt State University Press, Arcata, California.

Jamieson, R.C., R.J. Gordon, K.E. Sharples, G.W. Stratton, and A. Madani. 2002. Movement and persistence of fecal bacteria in agricultural soils and subsurface drainage water: A review. Canadian Biosystems Engineering 44: 1-9.

Janda, J.M. and S.L. Abbott. 1998. The Enterobacteria. Lippincott-Raven Publishers, Philadelphia, Pennsylvania.

Johnson, D.M., G.L. Stewart, M. Corley, R. Ghrist, J. Hagner, A. Ketterer, B. McDonnell, W. Newsom, E. Owen, and P. Samuels. 1980. Brown-headed cowbird (*Molothrus ater*) mortality in an urban winter roost. The Auk 97: 299-320.

Johnson, R.J. and J.F. Glahn. 1994. European starlings, damage prevention and control methods. Pages E109-E120. In S.E. Hygnstrom, R.M. Timm, and G.E Larson, editors. Prevention and control of wildlife damage. University of Nebraska-Lincoln.

Kapperud, G., K. Stenwig, and J. Lassen. 1998. Epidemiology of *Salmonella typhimurium* O:4-12 infection in Norway. American Journal of Epidemiology 147: 774-782.

Kato, Y., K. Ito, Y. Kubokura, T. Maruyama, K. I. Kaneko, and M. Ogawa. 1985. Occurrence of *Yersinia enterocolitica* in wild-living birds and Japanese serows. Applied and Environmental Microbiology 49: 198-200.

Kirk, J.H., C.A. Holmberg, and J.S. Jefferey. 2002. Prevalence of *Salmonella* spp. in selected birds captured on California dairies. Journal of American Veterinary Medicine Association 220: 359-362.

Lightfoot, N.F. 2003. Bacteria of potential health concern. Pages 62-79. In J. Bartman, J. Cotruvo, M. Exner, C. Fricker, A. Glasmacher, editors. Heterotrophic plate counts and drinking-water safety. World Health Organization. IWA Publishing, London, UK. http://www.who.int/water_sanitation_health/dwq/en/HPCS.pdf

Martyny, J.W. and R.G. Botzler. 1976. Yersiniae isolated from wapiti (*Cervus canadensis roosevelti*). Journal of Wildlife Diseases 12: 386-389.

McHale, P.J., C.T. Keane, and G. Dougan. 1981. Antibiotic resistance in *Providencia stuartii* isolated in hospitals. Journal of Clinical Microbiology 13: 1099-1104.

Morishita, T.Y., P.P. Aye, E.C. Ley, and B.S. Harr. 1999. Survey of pathogens and blood parasites in free-living passerines. Avian Diseases 43: 549-552.

Plant, C.W. 1978. Salmonellosis in wild birds feeding at sewage treatment works. Journal of Hygiene 81: 43-48.

Quevedo, F., R.D. Lord, D. Dobosch, I. Granier, and S.C. Michanie. 1973. Isolation of *Salmonella* from sparrows captured in horse corrals. American Journal of Tropical Medicine and Hygiene 22: 672-674.

Radwan, A.I., and J.R. Lampky. 1972. Enterobacteriaceae isolated from cowbirds (*Molothrus ater*) and other species of wild birds in Michigan. Avian Diseases 16:343-350.

Rahn, K., S.A. Renwick, R.P. Johnson, J.B. Wilson, R.C. Clarke, D. Alves, S. McEwen, H. Lior, and J. Spika. 1997. Persistence of *Escherichia coli* 0157:H7 in dairy cattle and the dairy farm environment. Epidemiology and Infection 119: 251-259.

Shayegani, M., I. DeForge, D.M. McGlynn, and T. Root. 1981. Characteristics of *Yersinia enterocolitica* and related species isolated from human, animal, and environmental sources. Journal of Clinical Microbiology 14: 304-312.

Tsubokura, M., K. Otsuki, K. Sato, M. Tanaka, T. Hongo, H. Fukushima, T. Maruyama, and M. Inoue. 1989. Special features of distribution of *Yersinia pseudotuberculosis* in Japan. Journal of Clinical Microbiology 27: 790-791.

Wilson, J.E., and J.W. MacDonald. 1967. *Salmonella* infection in wild birds. British Veterinary Journal 123: 212-219.

Zar, J.H. 1999. Biostatistical analysis, 4th edition. Prentice-Hall Publishers, Upper Saddle River, New Jersey.

PERSONAL COMMUNICATIONS

Botzler, R.G. 2002. Personal Communication. Humboldt State University, Department of Wildlife, CA, 95521

Brenneman, K. 2005. Personal Communication. Humboldt State University, Department of Fisheries, CA, 95521.

Carlisle, B.C. 2002. Personal Communication. 1701 Giuntoli Lane, Arcata, CA, 95521.

Silver, K. 2002. Personal Communication. 1140 Van Ness Ave., Ferndale, CA, 95536.

Appendix A. The following is a list of all the API 20 E codes and the identification confidence for each bacterial isolated observed for 243 wild birds sampled between July 2002 and February 2004 on 5 dairy farms in western Humboldt County, California.

API 20E code	Probable Bacterial Species	ID confidence
3704553	*Citrobacter braakii*	very good
3144533	*Citrobacter koseri/amalonaticus*	very good
3344513	*Citrobacter koseri/amalonaticus*	excellent
3144573	*Citrobacter koseri/farmeri*	very good
1404553	*Citrobacter* spp.	good to genus
1604553	*Citrobacter* spp.	good to genus
1404513	*Citrobacter youngae*	good
1105173	*Enterobacter amnigenus*	good
3104113	*Enterobacter cancerogenus*	good
3305573	*Enterobacter cloacae*	good
1105553	*Enterobacter* spp.	good to genus
1105573	*Enterobacter* spp.	good to genus
1305173	*Enterobacter* spp.	excellent to genus
3105173	*Enterobacter* spp.	excellent to genus
3305173	*Enterobacter* spp.	good to genus
3305363	*Enterobacter* spp.	very good to genus
3305513	*Enterobacter* spp.	very good to genus
3305553	*Enterobacter* spp.	excellent to genus
1144512	*Escherichia coli*	good
4114502	*Escherichia coli*	excellent to species
4144102	*Escherichia coli*	good
4144502	*Escherichia coli*	excellent
5044572	*Escherichia coli*	excellent
5104572	*Escherichia coli*	good
5144502	*Escherichia coli*	excellent to species
5144552	*Escherichia coli*	excellent
5144570	*Escherichia coli*	very good
5144572	*Escherichia coli*	very good
7144552	*Escherichia coli*	good
1144113	*Escherichia hermannii*	good
5144102	*Escherichia* spp.	very good to genus
3004113	*Escherichia vulneris*	acceptable

Appendix A. The following is a list of all the API 20 E codes and the identification confidence for each bacterial isolated observed for 243 wild birds sampled between July 2002 and February 2004 on 5 dairy farms in western Humboldt County, California (continued).

API 20E code	Probable Bacterial Species	ID confidence
4105103	*Hafnia alvei*	very good
4105113	*Hafnia alvei*	excellent
1004713	*Klebsiella pneumoniae ssp. ozaenae*	good
5215773	*Klebsiella pneumoniae* ssp. *pneumoniae*	good
5214773	*Klebsiella pneumoniae* ssp. *pneumoniae*	good
0004321	*Klebsiella pneumoniae* ssp. *rhinoscleromatis*	good
1005773	*Klebsiella* spp.	acceptable to genus
5005773	*Klebsiella terrigena*	good
1244060	*Moellerella wisconsensis*	excellent
1004060	*Moellerella wisconsensis*	excellent
1005333	*Pantoea* spp.	very good
1005373	*Pantoea* spp.	good
1007133	*Pantoea* spp.	very good
1007333	*Pantoea* spp.	very good
1007372	*Pantoea* spp.	very good
1205372	*Pantoea* spp.	good
1245173	*Pantoea* spp.	good
3005162	*Pantoea* spp.	good
0474020	*Proteus vulgaris*	very good
0476021	*Proteus vulgaris*	excellent
0746021	*Proteus vulgaris*	excellent
0234301	*Providencia rettgeri*	excellent
0254310	*Providencia rettgeri*	very good
0264311	*Providencia rettgeri*	very good
0274301	*Providencia rettgeri*	excellent
0274310	*Providencia rettgeri*	excellent
0274311	*Providencia rettgeri*	excellent
0064000	*Providencia* spp.	good to genus
0064200	*Providencia stuartii*	good
0264200	*Providencia stuartii*	good
1264200	*Providencia stuartii*	very good
2004046	*Pseudomonas* spp.	very good to genus
2200004	*Pseudomonas* spp.	good to genus

Appendix A. The following is a list of all the API 20 E codes and the identification confidence for each bacterial isolated observed for 243 wild birds sampled between July 2002 and February 2004 on 5 dairy farms in western Humboldt County, California (continued).

API 20E code	Probable Bacterial Species	ID confidence
1005573	*Rahnella aquatilis*	good
1104753	*Serratia fonticola*	good
5104753	*Serratia fonticola*	very good
5304713	*Serratia fonticola*	very good
5304753	*Serratia fonticola*	very good
7307763	*Serratia liquefaciens*	good
5316721	*Serratia marcescens*	very good
1207763	*Serratia* spp.	good to genus
1207363	*Serratia* spp.	very good to genus
0202000	*Stenotrophomonas maltophila*	acceptable
0154723	*Yersinia enterocolitica*	very good
0154723	*Yersinia enterocolitica*	very good
0154723	*Yersinia enterocolitica*	very good
1155723	*Yersinia enterocolitica*	good
1054533	*Yersinia frederiksenii/intermedia*	very good
0054503	*Yersinia kristensenii*	very good
1014112	*Yersinia pseudotuberculosis*	very good
1054723	*Yersinia* spp.	very good to genus

Appendix B. The following is a list of all the API 20 E codes and the identification confidence for each bacterial isolated observed for 100 dairy cattle sampled between November 2002 and April 2004 on 5 dairy farms in western Humboldt County, California.

API 20E code	Probable Bacterial Species	ID confidence
1006527	*Aeromonas hydrophila*	good
1047527	*Aeromonas hydrophila*	good
2047527	*Aeromonas hydrophila*	good
3047527	*Aeromonas hydrophila*	very good to species
3246127	*Aeromonas hydrophila*	good
3047527	*Aeromonas* spp.	very good to genus
1704753	*Citrobacter braakii*	very good
2704553	*Citrobacter braakii*	very good
3504553	*Citrobacter braakii*	very good
1344513	*Citrobacter koseri/amalonaticus*	very good
3604512	*Citrobacter youngae*	very good
1305553	*Enterobacter amnigenus*	good
3304173	*Enterobacter* spp.	very good to genus
3305173	*Enterobacter* spp.	excellent to genus
1044512	*Escherichia coli*	good to species
5104572	*Escherichia coli*	good
5144542	*Escherichia coli*	very good
5144562	*Escherichia coli*	very good
5144572	*Escherichia coli*	very good
7144552	*Escherichia coli*	good
4105102	*Hafnia alvei*	very good
5104112	*Hafnia alvei*	good
5245773	*Klebsiella oxytoca*	good
5255773	*Klebsiella oxytoca*	good
1215773	*Klebsiella pneumoniae* ssp. *pneumoniae*	good
5215773	*Klebsiella pneumoniae* ssp. *pneumoniae*	good
5204773	*Klebsiella terrigena*	acceptable
1205372	*Pantoea* spp.	good
1007333	*Pantoea* spp.	very good
0476021	*Proteus vulgaris*	excellent

Appendix B. The following is a list of all the API 20 E codes and the identification confidence for each bacterial isolated observed for 100 dairy cattle sampled between November 2002 and April 2004 on 5 dairy farms in western Humboldt County, California (continued).

API 20E code	Probable Bacterial Species	ID confidence
0274310	*Providencia rettgeri*	excellent
0064200	*Providencia stuartii*	good
2201004	*Pseudomonas flourescens/putida*	good
2200004	*Pseudomonas* spp.	good to genus
1104753	*Serratia fonticola*	good
5304753	*Serratia fonticola*	very good
5307723	*Serratia fonticola*	excellent to genus
7307762	*Serratia liquefaciens*	good
0400004	*Shewanella putrefaciens*	acceptable
0400006	*Shewanella putrefaciens*	acceptable
0050723	*Yersinia enterocolitica*	good
0154723	*Yersinia enterocolitica*	very good
1155723	*Yersinia enterocolitica*	good
1014112	*Yersinia pseudotuberculosis*	very good
1055523	*Yersinia* spp.	good to genus