

of the prevalence and distribution of the infection in wildlife and livestock populations on the Zimbabwe side of the Great Limpopo Transfrontier Conservation Area is urgently needed. Control options in wildlife are limited (2,10), but chances of success are greater if control measures are initiated at the early stage of disease spread into a new area. Adequate risk-mitigation strategies should be developed and implemented to reduce the risk for bovine TB transmission to livestock and humans living at the periphery of the unfenced Gonarezhou National Park. Failure to promptly assess the situation and adopt appropriate measures would have far-reaching conservation, economic, and public health consequences, not only for Zimbabwe but also for the political and social acceptance of the transfrontier conservation areas in southern Africa.

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References

1. Caron A, Cross PC, du Toit JT. Ecological implications of bovine tuberculosis in African buffalo herds. *Ecol Appl*. 2003;13:1338–45. DOI: 10.1890/02-5266
2. Michel AL, Bengis RG, Keet DF, Hofmeyr M, de Klerk LM, Cross PC, et al. Wildlife tuberculosis in South African conservation areas: implications and challenges. *Vet Microbiol*. 2006;112:91–100. DOI: 10.1016/j.vetmic.2005.11.035
3. Bengis RG, Kriek NP, Keet DF, Raath JP, de Vos V, Huchzermeyer HF. An outbreak of bovine tuberculosis in a free-living African buffalo (*Syncerus caffer*-Sparman) population in the Kruger National Park: a preliminary report. *Onderstepoort J Vet Res*. 1996;63:15–8.
4. Michel AL, Coetsee ML, Keet D, Maré L, Warren R, Cooper D, et al. Molecular epidemiology of *Mycobacterium bovis* isolates from free-ranging wildlife in South African game reserves. *Vet Microbiol*. 2009;133:335–43. DOI: 10.1016/j.vetmic.2008.07.023
5. Grobler DG, Michel AL, De Klerk LM, Bengis RG. The gamma-interferon test: its usefulness in a bovine tuberculosis survey in African buffaloes (*Syncerus caffer*) in the Kruger National Park. *Onderstepoort J Vet Res*. 2002;69:221–7.
6. Frothingham R, Meeker-O'Connell WA. Genetic diversity in the *Mycobacterium tuberculosis* complex based on variable numbers of tandem DNA repeats. *Microbiology*. 1998;144:1189–96. DOI: 10.1099/00221287-144-5-1189
7. Cross PC, Lloyd-Smith JO, Getz WM. Disentangling association patterns in fission–fusion societies using African buffalo as an example. *Anim Behav*. 2005;69:499–506. DOI: 10.1016/j.anbehav.2004.08.006
8. Renwick AR, White PC, Bengis RG. Bovine tuberculosis in southern African wildlife: a multi-species host-pathogen system. *Epidemiol Infect*. 2007;135:529–40. DOI: 10.1017/S0950268806007205
9. Keet DF, Kriek NPJ, RG, Michel AL. Tuberculosis in kudu (*Tragelaphus strepsiceros*) in the Kruger National Park. *Onderstepoort J Vet Res*. 2001;68:225–30.
10. De Lisle GW, Bengis RG, Schmitt SM, O'Brien DJ. Tuberculosis in free-ranging wildlife: detection, diagnosis and management. *Rev Sci Tech*. 2002;21:317–34.

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No Resistance Plasmid in *Yersinia pestis*, North America

To the Editor: Plague, caused by *Yersinia pestis*, is now largely controlled by improved sanitation and the use of antimicrobial drugs. However, before the widespread availability of antimicrobial drugs, an estimated >200 million persons died during pandemics (1). Today, if *Y. pestis* were to acquire antimicrobial drug resistance determinants, plague could again be a deadly disease.

Antimicrobial drug resistance in *Y. pestis* has been documented for

only a few strains. The best available information is for 2 strains isolated in Madagascar in 1995 (2), in which resistance was conferred by plasmids not typically found in *Y. pestis*. Strain 16/95 was resistant to streptomycin only; this resistance was mediated by plasmid pIP1203 (3). Strain 17/95 was resistant to 8 antimicrobial drugs, including some commonly used to treat plague, such as streptomycin, tetracyclines, and sulfonamides (2). Multidrug resistance in 17/95 was mediated by plasmid pIP1202 (4). Both plasmids could be transferred by conjugation from the source *Y. pestis* strains to other *Y. pestis* strains and *Escherichia coli* (3,4). pIP1203 could be transferred from *E. coli* to *Y. pestis* in the midgut of co-infected rat fleas (*Xenopsylla cheopis*), common vectors of plague (5).

Comparative sequence analysis has indicated that pIP1202 shares an almost identical IncA/C backbone with multidrug-resistant (MDR) plasmids from *Salmonella enterica* serotype Newport SL254 and *Yersinia ruckeri* YR71, suggesting recent acquisition from a common ancestor (6). In this study, this backbone was detected in numerous MDR enterobacterial pathogens (e.g., *E. coli*, *Klebsiella* spp., and multiple *Salmonella* serotypes) isolated from retail meat products. Many of these plasmids transferred at high rates to a plasmid-free *Y. ruckeri* strain, indicating the ability to efficiently transfer among species. Meat products examined in that study originated from 9 US states, including western plague-endemic states such as California, Colorado, New Mexico, and Oregon.

To determine whether the IncA/C plasmid backbone previously found in MDR *Y. pestis* and other species exists in *Y. pestis* isolates from western U.S. states, we screened *Y. pestis* DNA. The 713 isolates were collected from humans, small mammals, and fleas in 14 of the 17 western plague-endemic states (Table), including all states that

reported human cases during 1970–2002 (7). We used Primer Express software (Applied Biosystems, Foster City, CA, USA) to design a TaqMan-MGB single-probe assay to detect *repA*, a plasmid replication gene present in the IncA/C plasmid backbone. We based this assay on the *repA* assay described by Carrattoli et al. (8) and used the same forward primer but a different reverse primer and an additional probe to facilitate screening on a real-time PCR platform.

Real-time PCRs were conducted in 10- μ L reaction mixtures that contained 900 nmol/L of forward (5'-GA GAACCAAAGACAAAGACCTGG A-3') and reverse (5'-TGGCCGAG ATTCAATGATC-3') primers, 200 nmol/L of the *repA*-specific probe (5'-6FAM-AGACTCACCGCA AATG-3'), 1 \times AB TaqMan Universal PCR Master Mix with AmpErase UNG (uracil N-glycosylase) (Applied Biosystems), and 1 μ L of template. Thermal cycling was performed on an Applied Biosystems 7900 HT sequence detection system under the following conditions: 50°C for 2 min, 95°C for 10 min, and 40 cycles of 95°C for 15 s and 60°C for 1 min. DNA extracts from *Y. pestis* strain 17/95 (4) and *Salmonella enterica* serotype Newport strain SL254 (6) were used as positive controls.

Of the 713 *Y. pestis* isolates screened, none was positive for the IncA/C plasmid backbone, indicating that MDR as mediated by pIP1202-like MDR plasmids described by Welch et al. (6) was not in these samples. This finding is encouraging with regard to public health. However, we screened only for the plasmid backbone; MDR genes may have been in some of these samples but not carried by pIP1202-like MDR plasmids, especially considering that plasmids can be readily integrated into the *Y. pestis* chromosome (1).

Could MDR *Y. pestis* arise in North America by acquisition of an MDR plasmid, such as pIP1202, from food-animal production activities in plague-endemic regions? If so, *Salmonella* spp. would be a likely MDR plasmid donor for several reasons. First, *Y. pestis* has several plasmids that are highly similar to those in *Salmonella* spp., indicating active transfer of plasmids between these 2 bacterial groups (6). Second, fleas that are common vectors of plague have been shown to be naturally co-infected with *Salmonella* spp. and *Y. pestis* and capable of transmitting both organisms to rodent hosts (9). Third, MDR plasmids are readily transferred to *Y. pestis* in the flea gut (5). Fourth, transferable MDR plasmids are common among *Salmonella*

Table. *Yersinia pestis* isolates collected from humans, small mammals, and fleas, United States

State	No. isolates	Years collected
Arizona	151	1975, 1977–1984, 1986–1989, 1992–1996, 1998, 2000–2002
California	129	1943, 1962, 1970, 1977, 1979–1980, 1983–1999
Colorado	97	1963, 1968, 1989, 1992, 1995–1997, 1999–2002
Idaho	2	1987, 1997
Kansas	17	1997, 1999
Montana	11	1987, 1992–1993
North Dakota	2	1986, 1993
New Mexico	124	1950, 1976–1977, 1979–1988, 1991–1992, 1994–1995, 1997–2002
Nevada	36	1980–1985, 1987, 1992–1995
Oregon	18	1959, 1970–1971, 1977, 1979, 1981–1984, 1987
Texas	5	Unknown
Utah	55	1965, 1981–1984, 1991–1995, 1999–2001
Washington	2	1984
Wyoming	64	1978, 1980, 1982–1983, 1985–1987, 1989–1990, 1997, 2000–2001

spp. isolates in US food animals (10). Given these linkages, the transfer of an MDR plasmid from *Salmonella* spp. to *Y. pestis* seems possible. However, we emphasize that to date no evidence supports this type of event.

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References

1. Perry RD, Fetherston JD. *Yersinia pestis*—etiologic agent of plague. Clin Microbiol Rev. 1997;10:35–66.
2. Galimand M, Carniel E, Courvalin P. Resistance of *Yersinia pestis* to antimicrobial agents. Antimicrob Agents Chemother. 2006;50:3233–6.
3. Guiyoule A, Gerbaud G, Buchrieser C, Galimand M, Rahalison L, Chanteau S, et al. Transferable plasmid-mediated resistance to streptomycin in a clinical isolate of *Yersinia pestis*. Emerg Infect Dis. 2001;7:43–8. DOI: 10.3201/eid0701.010106
4. Galimand M, Guiyoule A, Gerbaud G, Rasoamanana B, Chanteau S, Carniel E, et al. Multidrug resistance in *Yersinia pestis* mediated by a transferable plasmid. N Engl J Med. 1997;337:677–80.
5. Hinnebusch BJ, Rosso M-L, Schwan TG, Carniel E. High-frequency conjugative transfer of antibiotic resistance genes to *Yersinia pestis* in the flea midgut. Mol Microbiol. 2002;46:349–54.
6. Welch TJ, Fricke WF, McDermott PF, White DG, Rosso M-L, Rasko DA, et al. Multiple antimicrobial resistance in plague: an emerging public health risk. PLoS One. 2007;2:e309. DOI: 10.1371/journal.pone.0000309
7. Centers for Disease Control and Prevention. Imported plague—New York City, 2002. MMWR Morb Mortal Wkly Rep. 2003;52:725–8.
8. Carattoli A, Bertini A, Villa L, Falbo V, Hopkins KL, Threlfall EJ. Identification of plasmids by PCR-based replicon typing. J Microbiol Methods. 2005;63:219–28. DOI: 10.1016/j.mimet.2005.03.018
9. Eskey CR, Prince FM, Fuller FB. Double infection of the rat fleas *X. cheopis* and *N. fasciatus* with *Pasteurella* and *Salmonella*. Public Health Rep (1896–1970). 1951;66:1318.
10. Silbergeld EK, Graham J, Price LB. Industrial food animal production, antimicrobial resistance, and human health. Annu Rev Public Health. 2008;29:151–69. DOI: 10.1146/annurev.publhealth.29.020907.090904

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Triatoma infestans Bugs in Southern Patagonia, Argentina

To the Editor: *Triatoma infestans* bugs, the main vector of Chagas disease, historically occupied a large area from northeastern Brazil to Chubut Province in Patagonia, Argentina (1). Large-scale insecticide spraying during the 1980s and 1990s reduced its geographic range and abundance and interrupted transmission of *Trypanosoma cruzi*, mainly in Uruguay, Chile, and Brazil (2). However, *T. infestans* and transmission of *T. cruzi* persist in the Gran Chaco, a large ecoregion in Argentina, Bolivia, and Paraguay (3).

Chubut Province has historically been an area with no risk for vector-mediated transmission of *T. cruzi*; only its extreme northern region was categorized as having a low transmission risk (4,5). However, increased immigration from disease-endemic rural areas in Argentina and Bolivia into Chubut has raised concerns about accidental introduction of *T. infestans* in travelers' luggage (1) and establishment of a transmission cycle.

In January 2007, we conducted a province-wide survey of 21 villages in Chubut Province previously infested with *T. infestans* bugs by using 0.2% tetramethrin as a dislodgeant agent (1 person-hour/house); no *T. infestans* bugs were detected (online Appendix Figure, www.cdc.gov/EID/content/16/4/887-appF.htm). Only *T. patagonica* bugs were found in 11% of peridomestic structures, and none were infested with *T. cruzi*. In June 2007, a *T. infestans*-like bug was found in a primary healthcare center in Comodoro Rivadavia (45°51'S, 67°28'W), a city in southern Chubut Province (online Appendix Figure). Healthcare center staff reported visits by immigrants from Bolivia a few days before this finding and suspected them to be the source. The bug was identi-