



Figure. Water dragon (*Physignathus cocincinus*). Three of the patients with *Salmonella* Kingabwa infections were exposed to this reptile species. Photo credit: Robert Lawton, rklawton@LawtonPhotos.com.

Two thirds of the patients in this outbreak had documented exposure to 1 of 2 lizard species, and half of the patients had pet lizards in their homes. In 2001, the estimated number of households with lizards was 545,000, which represents $\approx 0.5\%$ of all American households (5). Using a standard binomial model, the probability of finding at least 3 of 6 persons chosen at random to be lizard owners is 0.000002. To our knowledge, this is the first investigation to identify a strong association between the rare serotype *S.* Kingabwa and lizards and the first instance of which we are aware that a serotype has been associated with a particular species of lizard dispersed in homes across the United States.

The association between reptile exposure and human salmonellosis is well-established (6–8). CDC has published recommendations for reducing the risk for infection from reptiles (www.cdc.gov/healthypets/animals/reptiles.htm); these include thoroughly washing hands with soap and water after handling reptiles or their cages and keeping reptiles out of food preparation areas. The young age of most patients in this outbreak supports the recommendation that reptiles should not be allowed in households with children <5 years of age.

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Influenza Virus Type A Serosurvey in Cats

To the Editor: Recent reports of cats positive for H5N1 type A influenza virus (*I*) raised the hypothesis that cats might have an epidemiologic role in this disease. Experimental findings seem to support this hypothesis. Experimentally infected cats might act as aberrant hosts (as do humans and other mammals), with symptoms and lesions developing and the virus subsequently spreading to other cats (2,3). The experimental conditions under which this occurs, however, can rarely be observed for domestic or wild cats. No spontaneous cases of transmission from cat to cat or cat to mammal have been reported, and scientifically validated reports about spontaneous disease in cats are rare

(4–6). Reports about cats with circulating influenza virus antibodies are even more rare and occur in unusual epidemiologic situations (7). The true susceptibility of cats to type A influenza viruses in field conditions thus remains to be elucidated.

Based on the assumption that partially susceptible animals should mount an antibody response, we investigated the possible presence of antibodies against the nucleocapsid protein A (NPA), a common antigen of type A influenza viruses, expressed by both avian and human strains (8), in feline serum samples stored at the University of Milan and collected from 1999 to 2005. Only samples for which complete information regarding the cat (owned vs. free-roaming) and its health status were included in the study. Cats were grouped as healthy or sick on the basis of clinical signs; a complete clinicopathologic screening that included routine hematologic tests, clinical biochemical tests, and serum protein electrophoresis; serologic tests for feline immunodeficiency virus and feline leukemia virus infection, which are known to induce immunosuppression; and information regarding the follow-up, including postmortem examination for dead animals. Specifically, 196 serum samples satisfied the inclusion criteria in terms of anamnestic information about the sampled cat and,

according to the above-mentioned diagnostic approach, cats were grouped as reported in the Table. Owned cats were mainly living in the urban area of Milan. By contrast, approximately half of the free-roaming cats included came from rescue shelters from a rural area northwest of Milan. Sixty samples (58.8%) from owned cats and 51 samples (54.2%) from free-roaming cats were collected from September to February, when seasonal human influenza peaks.

Serologic tests for antibodies to type A influenza virus were performed with a competitive ELISA to detect NPA antibodies (9). Negative control serum from specific-pathogen-free chickens and positive control serum specimens from different species (avian, swine, and equine) were included in each plate to provide a full range of controls. Serum samples were considered positive when the absorbance value was reduced to at least 75% compared with 100% for negative control wells.

All cats were negative for type A influenza virus antibodies. The ELISA we used has been validated in several species, including humans (9). Antibodies against NPA are not a major response to influenza infection but likely would have been detected if infections of cat were widespread. Thus, although no positive feline serum samples were used as positive

controls, the negative results are not likely false negatives. Indeed, the negative results of many cats included in the study (the free-roaming ones, especially those affected by severe illness, for which a natural cat/flu virus interaction is unrealistic) might be due to low exposure to the virus because avian influenza outbreaks never occurred in the sampling area included in this study (10). By contrast, many owned cats (those sampled during the winter) likely were exposed to human type A influenza viruses, since approximately half of the viruses responsible for human seasonal influenza isolated in Europe, especially in Italy, are type A (8,10). The close contact between pets and their owners probably exposed cats to these viruses; nevertheless, none of the pet cats seroconverted, even when they had severe systemic diseases or viral induced immunosuppression. Although the number of cats in this study might be statistically insufficient to show low seroprevalences, our results further support the hypothesis that, in field conditions, cats are most probably not susceptible to type A influenza viruses, especially to the human ones (e.g., H3N2, the most diffused among humans, which also did not induced symptoms or lesions in experimental conditions [2]) circulating in the “pre-cat flu era.” In future studies, these results can be used to

Table. Survey of feline serum samples, collected from 1999 to 2005, for influenza A virus, Italy*

Clinical status	No.	Diagnosis	No.	Virus status	No.	
Pet cats						
Nonsymptomatic	25			FIV	2	
				FeLV	1	
Symptomatic	77	FIP	36			
		Locally extensive inflammation	18	FIV	4	
		Hematologic neoplasia	8	FIV	1	
		Nonhematologic tumors	8			
		Systemic inflammatory or degenerative diseases	7		FIV	2
					FIV + FeLV	1
Free-roaming cats						
Nonsymptomatic	54			FIV	5	
Symptomatic	40	Locally extensive inflammation	27	FIV	7	
		Systemic inflammatory or degenerative diseases	7			
		FIP	6			

*FIV, feline immunodeficiency virus; FIP, feline infectious peritonitis; FeLV, feline leukemia virus.

compare the results of seroepidemiologic investigations among cats living in sites contaminated by avian viruses.

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Pneumonic Plague, Northern India, 2002

To the Editor: A small outbreak of primary pneumonic plague took place in the Shimla District of Himachal Pradesh State in northern India during February 2002. Sixteen cases of plague were reported with a case-fatality rate of 25% (4/16). The infection was confirmed to the molecular level with PCR and gene sequencing (1). A previous outbreak in this region during 1983 was suggestive of pneumonic plague (22 cases, 17 deaths) but was not confirmed. In India, the last laboratory-confirmed case of plague was reported in 1966 from Karnataka State (2).

The index patient for the 2002 outbreak lived in a hamlet in the Himalayas. He went hunting on January 28, 2002, in a nearby forest at a height of ≈500–600 m from his house. There, he killed a sick wild cat and skinned it. He returned home on February 2 and sought treatment for fever, chills, and headache. On February 4, breathlessness, chest pain, and hemoptysis developed; radiologic findings were suggestive of lobar pneumonia, and treatment with augmentin was begun. He died the next day. Subsequently, 13 of his relatives exhibited a similar illness, although 2 additional patients acquired infection in the hospital. The incubation period for those patients was 1–4 days, which is consistent with that of pneumonic plague.

A team of microbiologists, epidemiologists, and entomologists visited the village after 7 more cases were reported until February 12, 2002, followed by a team from the National Institute of Communicable Diseases (NICD), New Delhi. The following case-patient definition was used: a person who sought treatment for fever of rapid onset, chills, chest pain, breathlessness, headache, prostration,

and hemoptysis. A total of 16 cases were reported from 3 hospitals in the area: a local civil hospital, the state medical college, and a regional tertiary care hospital. Clinical material collected from the case-patients and their contacts was initially processed in the laboratories of these hospitals. Wayson staining provided immediate presumptive diagnosis, and confirmatory tests were performed at NICD. Diagnosis of plague was confirmed for 10 (63%) of 16 patients (1).

NICD conducted the following laboratory tests on 2 suspected culture isolates, 2 sputum specimens, 1 lung autopsy material specimen, and 1 lung lavage sample (Table): 1) direct fluorescent antibody test for *Yersinia pestis*; 2) culture and bacteriophage lysis test; and 3) PCR and gene sequencing to detect *Y. pestis*-specific genes (*pla* and *FI*). All these tests confirmed that isolates were *Y. pestis* and met all the World Health Organization's recommended criteria (2).

Antibodies against F1 antigen of *Y. pestis* were detected by passive hemagglutination testing of paired serum samples. Although 5 patients showed a >4-fold rise, 1 patient showed a >4-fold fall in antibody titer. In contrast, samples from 6 patients were negative for *Y. pestis*, and no change was found in the titers from 1 patient. No serum sample was collected from the index patient; for the 2 other patients who died, 1 of the single serum samples became contaminated, and the other was positive for *Y. pestis* (3). Paired serum samples from the case-patients were collected on a single day 4 weeks apart during the visit of the NICD team, regardless of the duration of symptoms.

Antimicrobial drug sensitivity testing was carried out by the Kirby-Bauer disk diffusion method. All isolates were sensitive to doxycycline, tetracycline, chloramphenicol, streptomycin, ciprofloxacin, gentamicin, and amikacin but were resistant to penicillin.