Highly Pathogenic Porcine Reproductive and Respiratory Syndrome, China

To the Editor: Since April 2006, a highly pathogenic disease caused by unknown agents and characterized by high fever and a high proportion of deaths in pigs of all ages, emerged in some swine farms in Jiangxi Province, People’s Republic of China. The morbidity rate was 50%–100% and mortality rate was 20%–100%. In the next several months, the disease spread rapidly to most provinces of China. In almost all affected swine herds, the following clinical signs were observed: high and continuous fever, anorexia, red discolorations in the bodies, and blue ears; in the late phase of the disease, diarrhea and other clinical signs might be seen due to the secondary infections. Clinical samples (from lungs, kidneys, liver, and lymph nodes) were collected from animals in different provinces and sent for laboratory diagnosis. DNA and RNA were extracted from the tissue homogenate and PCR or reverse transcription–PCR (RT-PCR) was conducted to detect porcine reproductive and respiratory syndrome virus (PRRSV), classic swine fever virus, porcine circovirus, and pseudorabies virus, respectively (1). In clinical samples, only PRRSV was found to be the dominant virus (48 of 50 samples were PRRSV posi-

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PRRSVs were then isolated successfully on MARC-145 cells with an obvious cytopathologic effect, characterized by cell congregation, contraction, and brushing off at passage 2; immunofluorescence assay using PRRSV NP-, M- and GP5-specific monoclonal antibodies confirmed that the isolated viruses were PRRSV (2,3). Full-length genomic sequencing of 1 of the isolates (HuN4 strain) showed extensive amino acid (aa) mutations in GP5 protein and 2 deletions in Nsp2, 1 aa deletion at 482, and 29 aa deletions at 533–561, compared with the previous Chinese isolates CH-1a and BJ-4.

The newly isolated PRRSV was used to examine the pathogenicity in 60-day-old PRRSV-free piglets, under closed and biosafety (P2) conditions. Each of the piglets (N = 5) received intranasally 10^5.0 50% tissue culture infecting dose of the isolated virus propagated in MARC-145 cells (4,5). The animals were kept in separate rooms throughout the experiment. Clinical observations of respiratory signs, behavior, rectal temperature, and coughing were recorded daily. Blood samples were collected every 2 days and tested for PRRSV-specific antibodies by ELISA (6,7). Tissue samples (from heart, lungs, kidneys, spleen, and lymph nodes) from all animals that died during the experiment were collected and detected by histopathologic examination (8) and virus isolation. Results showed that the clinical manifestations of all pigs were similar to those that appeared in the field investigation (including high and continuous fever, anorexia, red discolorations in the bodies, and blue ears). The specific antibodies to PRRSV were detected at 8 days postinfection, and the high antibody level lasted until the animal’s death, and all infected pigs died at either 7, 8, 12, 16, or 21 days postinoculation, respectively. Furthermore, viruses reisolated from the dead pigs showed an identical homology with the inoculated PRRSV in genes coding for GP5 and partial Nsp2 (2,535–3,307 nt). The results showed that the emerging PRRSV, characterized by deletions in Nsp2, is highly pathogenic to pigs.

To investigate whether the emerging PRRSV was the causative agent of the pandemic diseases on swine farms, an extensive virus survey was conducted. More than 48 samples collected from different swine farms in 12 provinces were found to be PRRSV positive by RT-PCR, based on open reading frame (ORF) 5 and Nsp2 (Figure). Sequence analysis of ORF5 and partial Nsp2 showed that these PRRSVs are highly homologous to each other (98.5%–100% for GP5; 98.2%–100% for Nsp2) and share the same deletions at the same positions of Nsp2 gene with HuN4 strain. Sequence comparison of ORF5 indicated that the HuN4 strain shares 93%, 86%, and 88% nucleotide identities with CH-1a (Chinese isolate), BJ-4 (Chinese isolate), and VR2332 (American isolate), respectively. All the newly isolated PRRSVs belong to the North American type.

Although the cause of the emerging pandemic disease of pigs with a high proportion of deaths in 2006 is unknown, we found high correlation between PRRSV isolation rate and the diseased pigs. The regression test in its natural animal showed that the newly isolated PRRSV was much more virulent than earlier PRRSV isolates. Also, sequence analysis demonstrated a substantial diversity from the PRRSVs isolated during 1996–2005. Further study is needed to answer the question: What role did the newly isolated PRRSV play in the 2006 outbreaks on many of the swine farms in China?

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Recurrence American Cutaneous Leishmaniasis

To the Editor: Leishmaniasis recidivans is an unusual clinical Old World disease primarily associated with Leishmania tropica (1). Recurrence of previously cured cutaneous leishmaniasis (CL) lesions is found in American CL, for which a specific nosologic form known of disease known as leishmaniasis recidiva cutis (LRC) has been identified. Although <30 cases of LRC have been reported from Brazil, Colombia, Peru and Ecuador; these cases were caused mainly by L. braziliensis, L. amazonensis, and L. panamensis (2). We report 7 cases of recurrent American CL caused by L. guyanensis in French Guiana.

Forty-eight military personnel who lived in France spent 3 months in French Guiana in 2004 and took part in a military training program in the rainforest for 15 days. Despite similar exposure conditions, American CL, confirmed by positive direct examination of Giemsa-stained tissue smears, developed in 21 persons. These patients were treated with 1 or 2 courses of either 3 intravenous or 2 intramuscular injections of pentamidine isethionate (4 mg/kg on alternate days). All lesions were cured 1–3 months after treatment had ended. Recurrence of the CL lesion was observed in 7 patients after a disease-free interval of 3–6 months (Table).

New lesions appeared on the edge of a healed scar for each patient, regardless of the location of the primary lesion (Table), and were diagnosed at Rennes University Hospital (positive direct examination or culture) in 2005. For positive cultures, L. guyanensis was identified by genomic and isoenzymatic characterization at the Centre National de Référence des Leishmania, Université de Montpellier, Montpellier, France. Patients were treated with 4 intravenous injections of pentamidine isethionate (4 mg/kg every other day) and were cured without recurrence within 2 years. No differences in age or underlying diseases were noted in patients with recurrent CL.

L. (Viana) guyanensis is highly prevalent in several leishmaniasis-endemic areas of Brazil, Colombia, French Guiana, Guyana, Suriname, Peru, and Ecuador. This organism accounts for >95% of the 5 Leishmania species found in French Guiana, commonly causes localized LCL, and occasionally causes disseminated CL and mucocutaneous leishmaniasis (3). Dedet et al. reported that 6.8% of patients with CL caused by L. guyanensis had a recurrent lesion at the site of a previously cured lesion, which occurred after a mean interval of 7.3 months (4). A total of 33% of our patients had a cured primary infection in <3 months but they had a recurrence after a disease-free interval 3–6 months after treatment.

Additional information on such a recurrent form of CL is needed. Clinical symptoms in our patients were suggestive of LRC as described by Berlin (1), i.e., a recurrence at the site of an original ulcer, generally within 2 years and often on the edge of a scar. LRC may not be uncommon in the New World but rather underreported (2). Few cases of LRC have been reported; these were caused by L. braziliensis, L. amazonensis, and L. panamensis (2, 5, 6). In CL caused by L. guyanensis, borderline clinical symptoms prevent clear distinction of the recurrent form of LRC from early treatment failures or reinnfections. In our patients, the risk for reinfection was excluded because the military personnel lived in France and left French Guiana several months before the recurrence.

Although pentavalent antimony is the recommended treatment for American CL, pentamidine isethionate is widely used in French Guiana. Retrospective analysis showed that 5%–25% of early treatment failures...