

have significantly shortened the duration of the symptoms is not known.

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## *Trichinella papuae* in Saltwater Crocodiles (*Crocodylus* *porosus*) of Papua New Guinea

**To the Editor:** Until 1995, reptiles

were not known to be hosts of *Trichinella*; however, in that year *Trichinella* was detected in 40% of farm-raised crocodiles (*Crocodylus niloticus*) in Zimbabwe. These crocodiles were infected with a new species, *T. zimbabwensis*, which was experimentally infective in mammals, including primates (1).

The infection of reptiles with *Trichinella* species that are potentially infective for humans has become more important since demand for the meat of crocodiles, caimans, and alligators has increased in many areas of the world. This trend has resulted in the development of national breeding programs in more than 30 countries in North, Central, and South America; Africa; Asia; and Australia (2), which generated an income of approximately \$60 million in 1998 (3).

In 1999 in Papua New Guinea, wild and domestic pigs infected with a new species, *T. papuae*, were found (4,5); this new species was capable of completing its life cycle in reptiles that were infected experimentally (6). *Trichinella* infection has also been found in farm-raised saltwater crocodiles (*C. porosus*) in Papua New Guinea, where a national program for crocodile meat and skin products exists.

Papua New Guinea has one crocodile breeding farm that processes approximately 6,000 animals per year. Following the discovery of *Trichinella*-infected crocodiles in Zimbabwe, the Australian government requested that Papua New Guinea conduct *Trichinella* testing on the crocodile meat exported to Australia. Muscle samples from crocodiles were digested by pepsin and HCl solution according to the standard technique (7). When available, approximately 100 larvae from each infected crocodile were given by mouth to laboratory rats, and 10-20 larvae were stored in 90% ethyl alcohol for molecular identification. Multiplex polymerase chain reaction

(PCR) was used to characterize the larvae, according to a published protocol (8). The primer set oTsr1 and oTsr4 was used to amplify the expansion segment V of the large subunit ribosomal RNA (9). The larvae of all *Trichinella* reference strains were used as controls. PCR products were gel-purified and directly sequenced by using the same primers as those used for PCR amplification. All sequences were aligned by using the Clustal W program from OMIGA 2.0 (Accelrys, San Diego, CA). Final alignment of the expansion segment V sequences was performed manually so microsatellites could be compared.

Muscle samples from 118 saltwater crocodiles (46 farm-born, 71 wild-born and farm-raised, and 1 killed in the wild near the Bensbach River) were tested. All samples from the farm-born crocodiles were negative for *Trichinella*. Of the samples from the 72 wild-born crocodiles (including the 1 killed in the wild), 16 (22.2%) were positive for *Trichinella* larvae, with an average of 7 larvae/g in the biceps. All of the infected crocodiles originated in the Kikori area (Figure). The prevalence of *Trichinella* infection in crocodiles from this area was 32.0% (16/50). Samples from the remaining 21 wild-born and farm-raised crocodiles, and the 1 killed in the wild, were negative for *Trichinella*. These crocodiles originated in nine different locations (Figure).

PCR analysis showed that the parasites belonged to *T. papuae*. However, the crocodile isolates differed from the reference strain of this species by the deletion of a TG dinucleotide and by a single base mutation (G vs. A) in the expansion segment V sequence. Testing for *Trichinella* in crocodile meat has been conducted in Zimbabwe and Papua New Guinea only, and infected crocodiles have been found in both countries. Crocodiles in other parts of the world are also likely to be infected. Since

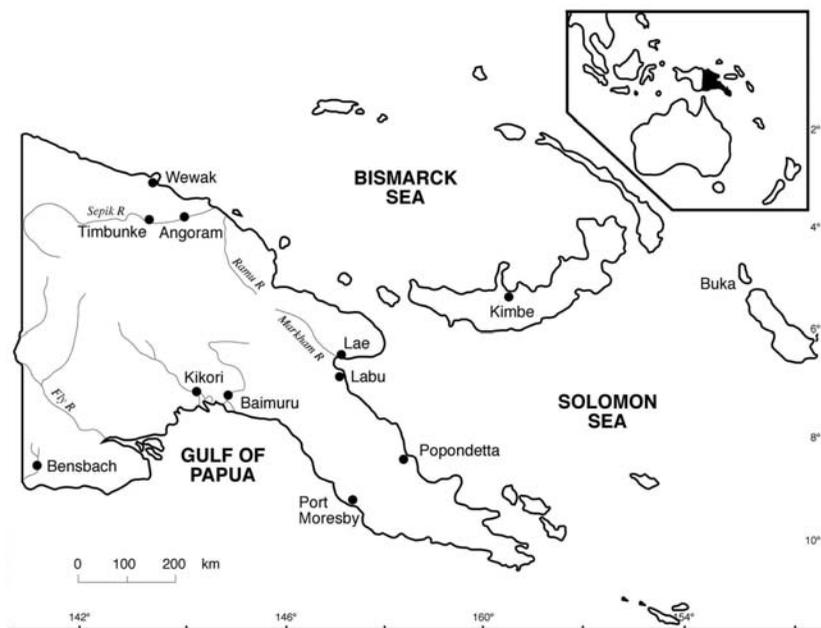


Figure. Papua New Guinea showing the areas of origin of the 72 wild-born saltwater crocodiles (*Crocodylus porosus*): 1 each from Baimuru, Angoram, Timbunke, Kimbe, Bensbach River, and Buka; 2 from Labu; 7 from Wewak; 7 from Popondetta; and 50 from Kikori.

both *T. zimbabwensis* and *T. papuae* infection can develop in reptiles and mammals, eating crocodile meat is a risk. In one region of Papua New Guinea, a high percentage of the local human population had anti-*Trichinella* antibodies (10). Moreover, the risk for human infection may be rising, given the increased marketing of meat from crocodiles, caimans, and alligators in many parts of the world (2). The meat of other carnivorous reptiles, although consumed in very few areas, may also represent a source of infection, as suggested by the large number of larvae of both *T. papuae* and *T. zimbabwensis* in the muscles of experimentally infected monitor lizards (6).

The presence of a TG dinucleotide in the expansion segment V sequence could be a useful marker for tracing the region of origin of infected meat. The infected crocodiles, all of which were born in the wild, likely acquired infection before they arrived on the farm, since none of the farm-born crocodiles was infected. In Zimbabwe, the source of infection

was the *Trichinella*-infected crocodile meat that had been fed to the other crocodiles; the farm in Papua New Guinea does not engage in this practice, which would explain why none of its farm-born animals was infected.

This study shows the importance of implementing measures to prevent the spread of *Trichinella* infection. For instance, since both *T. papuae* and *T. zimbabwensis* can be easily transmitted from crocodiles to mammals, the discarded parts of crocodiles should be properly destroyed to avoid transmission to synanthropic animals, and the waste products should not be fed to domestic animals, unless the products are frozen or cooked before use. Crocodile-breeding farms should adopt the artificial digestion method used in many countries to screen pigs for *Trichinella* infection (7). Freezing crocodile meat, as practiced in Papua New Guinea, can also prevent infection because freezing destroys *T. papuae* and *T. zimbabwensis* larvae in muscles (1,4). By contrast, salting, drying, smoking, or preserving croc-

dile meat in brine will not destroy trichinellae; these curing methods are not standardized, and the survival of *Trichinella* larvae can depend on factors such as salt concentration, moisture, and temperature (7). Similarly, crocodile meat is frequently vacuum sealed, and the *Trichinella* larvae can retain their infectivity for several months in this environment (7).

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## Panton-Valentine Leukocidin-positive *Staphylococcus aureus*, Singapore

**To the Editor:** Necrotizing community-acquired pneumonia attributable to Panton-Valentine leukocidin-producing strains of *Staphylococcus aureus* has been described as a distinct clinical syndrome with a high death rate in young, immunocompetent patients (1,2). This letter details

the first reported case of necrotizing pneumonia caused by Panton-Valentine leukocidin-positive *S. aureus* in a southeastern Asian country, Singapore.

An 18-year-old girl of Chinese ethnicity with a 4-day history of fever, cough, hemoptysis, and dyspnea sought treatment at Singapore General Hospital in October 2003. This episode had immediately followed an influenza-like prodromal illness for which a general practitioner had prescribed oral erythromycin ethynylsuccinate and medications for symptomatic relief. Her medical history showed an intrauterine *Toxoplasma gondii* infection that had resulted in developmental delay and slight mental retardation. She had never traveled outside Singapore.

On admission, the patient's temperature was 38.4°C, blood pressure was 130/70 mm Hg, and her pulse rate was 108 per min. Bibasal crackles were heard on auscultation of her lung fields, and her respiratory rate was 30 per min despite the use of supplemental oxygen. The results of physical examination were otherwise unremarkable. Initial chest x-ray showed air-space shadowing of the right upper and middle lobes of the lung, as well as blunting of the right costophrenic angle. Blood tests gave the following results: leukocyte count 7.42 x 10<sup>9</sup>/L, neutrophil count 6.53 x 10<sup>9</sup>/L, platelet count 287 x 10<sup>9</sup>/L, hemoglobin level 8.6 g/dL, prothrombin time 15.3 s, and activated partial thromboplastin time 28.7 s. She was experiencing acute renal failure with a serum creatinine level of 783 μmol/L. Liver biochemistry was abnormal with the following values: alkaline phosphatase 513 U/L, alanine aminotransferase 38 U/L, and aspartate aminotransferase 65 U/L. Serum bilirubin level was within the normal range.

The patient was prescribed intravenous ceftriaxone and azithromycin, and hemodialysis was initiated. Within 6 hours of hospitalization, the

patient became hypotensive and hypoxemic and required inotropic support and mechanical ventilation. Intravenous ceftazidime and high-dose cloxacillin were substituted for ceftriaxone at that time. Blood cultures obtained on admission were sterile, but penicillin-resistant *S. aureus* grew from cultures of aspirated endotracheal tube secretions. Results of immunofluorescent tests conducted on bronchial washings for viral antigens of influenza virus A and B, parainfluenza virus, respiratory syncytial virus, and adenovirus were negative. Computed tomographic scan of the thorax on day 3 of hospitalization showed widespread confluent consolidation of the right lung with right pleural effusion and patchy consolidation of the lingular lobe of the left lung. The total leukocyte count increased to 26.3 x 10<sup>9</sup>/L, and disseminated intravascular coagulopathy developed. Results of repeated blood and endotracheal cultures were positive for *S. aureus*, and intravenous gentamicin and rifampicin were added to her antimicrobial cocktail. A transthoracic echocardiogram showed a normal heart with no evidence of endocarditis.

Despite aggressive support, the patient's condition continued to deteriorate. A hemopyopneumothorax developed on the right side on day 4 of hospitalization, which required chest tube insertion. Hemoptysis persisted, and inotropic and ventilatory requirements progressively increased. The patient died on day 20 of hospitalization.

The severity of the patient's infection and the clinical symptoms suggested the presence of Panton-Valentine leukocidin genes in the causative *S. aureus*; tests confirmed the suspicion. *S. aureus* was identified on the basis of colony morphologic characteristics, the coagulation of citrated rabbit plasma (bioMérieux, Marcy l'Etoile, France), and production of a clumping factor (Staphyslide