

# Soft shelled turtles, food in China, likely spread cholera

June 9 2017

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The pathogen, *Vibrio cholerae* can colonize the surfaces, as well as the intestines of soft shelled turtles. This finding is strong evidence that soft shelled turtles in China, where they are grown for human consumption, are spreading cholera. The research is published in *Applied and Environmental Microbiology*, a journal of the American Society for Microbiology.

Insertion of genes producing bioluminescent proteins into *V. cholerae* enabled the investigators to directly observe the pathogens colonizing the turtles. To infect the turtles, the investigators dipped them in a phosphate buffered saline solution containing the now bioluminescent bacteria, serogroup 0139.

Over the next four days, the researchers checked the turtles at 24 hour intervals. They first detected light signals at 24 hours. At 96 hours, the entire dorsal side of the turtles' shells was emitting bioluminescence. The latter was also easily detected on the dorsal side of the turtles' limbs and necks, and in the calipash, the gelatinous protoplasm, locally regarded as a delicacy, that lies directly beneath the shells' surface.

Determining intestinal colonization was more difficult. The turtles were inoculated intragastrically with the bioluminescent *V. cholerae*. Knowing that digestion takes roughly 34 to 56 hours in 150 gram turtles, the investigators euthanized and dissected the turtles at 72 hours, and checked all their internal organs. Bioluminescence could be detected only in the intestines.

The investigators also identified the different colonization factors—molecular machinery on the surface of *V. cholerae*—that enabled the bacteria to stick to the turtles' dorsal surfaces and intestines.

The motivation for the research was the discovery, through surveillance of the disease in China, that consumption of cholera-carrying soft shelled turtles had caused outbreaks of the disease, said corresponding author Biao Kan, PhD. "Cholera is a life-threatening diarrheal disease," said Kan, who is professor of pathogenic biology and [infectious disease control](#), at the Chinese Center for Disease Control and Prevention, Beijing.

Despite the fact that during the last decade, China has seen less than 200 cases, annually, according to Kan. But he said that of the 39 diseases surveilled under China's Law of Infectious Disease Control and Prevention, cholera is in the most dangerous category, along with plague. He also noted that the O139 serogroup, the major strain spread by the turtles, is an emerging disease in China.

Elsewhere in the world, as of last summer, the Haitian outbreak, which began in 2010, has infected nearly 800,000 people and has caused more than 9,000 deaths, according to National Geographic. In 2014, a total of 190,549 cases worldwide were reported to the World Health Organization, including 2,231 deaths. But the number of reported cases has varied widely year to year, with a post-millennium high of over 600,000 in 2011. But other sources have reported as many as 3 million cases annually this decade.

A side benefit of the study is that the soft shelled turtle could serve as a new animal model for studying how *V. cholerae* interacts with aquatic hosts. Unlike other aquatic models such as zebra fish, soft shelled turtles can be anesthetized and their surfaces sampled out of water for relatively long periods without killing them. Using aquatic models is particularly

beneficial since cholera is a mostly waterborne disease.

Besides soft shelled [turtles](#), aquatic hosts of *V. cholerae* include zooplankton, fish, shellfish, egg masses of midges, waterfowl, and crustaceans. Fish and shellfish are proven to spread this [disease](#).

Provided by American Society for Microbiology

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