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Campylobacter jejuni in Penguins, Antarctica

To the Editor: The wildlife of Antarctica is highly specialized. Although large animal species are limited primarily to penguins and seals, each species is often abundant. The high degree of isolation potentially protects Antarctic wildlife from diseases distributed in other areas of the world (*1,2*). Despite Antarctica's isolation, however, human- or animal-related pathogens have been found there, or in the sub-Antarctic islands. For instance, serologic evidence of influenza virus A infections in penguins has been found (*3*), and both *Salmonella* spp. and *Mycobacterium tuberculosis* have been isolated from sub-Antarctic and Antarctic animals (*4,5*).

Campylobacter jejuni is a leading cause of bacterial gastroenteritis in humans worldwide; it is usually found in the intestinal tract of various farm and wild animals, particularly birds (*6,7*). We previously reported finding 3 *C.*

jejuni subsp. *jejuni* isolates in macaroni penguins (*Eudyptes chrysolophus*; Figure) from Bird Island (54°00′S, 38°02′W), South Georgia (*1*). Phenotypic tests and 16S rRNA gene sequencing showed that the penguin isolates were identical to each other, and macrorestriction profiling of pulsed-field gel electrophoresis fragments showed that they were very similar to fragments isolated from poultry in Washington in 1984 (*1*). Because the isolates were retrieved from macaroni penguin chicks, we concluded that the animals had acquired the infection locally and that this was likely an instance of introduction of a pathogen to the Antarctic region.

However, restriction fragment pattern resemblance is not identical to genetic relatedness and, given the relevance of the question of origin, this resemblance led us to use a new method for genetic characterization. We reanalyzed the macaroni penguin isolates with multilocus sequence typing (MLST), a method that uses sequence data from 7 unlinked loci for genetic identification (*8*), complemented with *flaA* gene sequencing. A benefit of this method is the increasing availability of epidemiologic databases in which isolates can be compared (e.g., http://pubmlst.org/campylobacter). The isolates were thawed and cultured on conventional blood agar (Columbia agar II containing 8% [vol/vol] whole horse blood) at 42°C in a microaerobic gas environment, with the CampyGen gas-generating system (CN0025A; Oxoid Ltd, Basingstoke, UK) and the BBL GasPak system (BD, Franklin Lakes, NJ, USA). Bacterial DNA was prepared by making a suspension of freshly grown bacterial cells in 200 μL of phosphate-buffered saline (Sigma, St. Louis, MO, USA). Genomic DNA was extracted by use of a Bio Robot M48 (QIAGEN, Hilden, Germany) with a MagAttract DNA mini M48 kit, according to the instructions of the manufacturer. The PCR amplification and nucleotide sequencing followed

the original protocol in principle (*8*). The amplification products were purified and sequenced by using internal separated nested primer pairs.

The 3 isolates from macaroni penguins were all of the same genotype (sequence type [ST]–45) and thus have a common origin. The ST-45 sequence type is the central genotype of the ST-45 clonal complex, a complex often associated with human disease and asymptomatic infection in poultry (*9,10*). Indeed, nearly 42% of the ST-45 samples available in the MLST database have been isolated from humans (31% from poultry), and similar percentages have been observed for the ST-45 clonal complex as a whole. The ST-45 clonal complex is large (composed of 195 individual STs) and has been isolated to date from a variety of environmental sources and different geographic regions, with the exception of the Arctic. The isolates were identical at the *flaA* locus, all having the allele 21/peptide 2 designation (http://pubmlst.org/campylobacter/flaA). This particular peptide is found in 31 records in the database and is thus not unique to the penguin isolates.

Our MLST analysis confirms that the *C. jejuni* isolates from the penguins were of a genotype common among humans with disease and among our food animals. *C. jejuni* is not normally distributed among Antarctic animals (*1,2*), which indicates that this strain may have been imported through human activities. On Bird Island, such activities were carried out by scientists at the British Antarctic Survey base. At the time of the study, toilet wastes from the station were emptied into the surrounding waters, providing a possible transmission route for human-associated *C. jejuni* to reach wildlife, including penguins. Other possible sources of the *C. jejuni* infections include wastes from passing ships or seabirds that pick up the bacteria during offshore feeding excursions (for albatrosses, these can be ≈1,000 km). Once established in a penguin colony, a gastrointestinal pathogen may be transmitted rapidly among individual birds as they are breeding densely and producing a large amount of feces (guano) in the colony. *C. jejuni* infection in birds is normally not associated with overt disease, but other and possibly more devastating pathogens introduced to Antarctic animals could potentially cause outbreaks.

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Figure. Macaroni penguins (*Eudyptes chrysolophus*).

Photo by Jonas Bonnedahl

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Acute Diarrhea in Children after 2004 Tsunami, Andaman Islands

To the Editor: The Andaman Islands, population ≈350,000, are a territory of India located in the Bay of Bengal, northwest of Indonesia. On December 26, 2004, these islands were struck by an earthquake measuring 9.1 on the Richter scale (*1*) and by the ensuing Great Asian Tsunami (*2*). The fault slip, which caused permanent land subsidence of several meters (*3*) and ingression of sea water, resulted in the displacement of most survivors, many of whom were forced to live in temporary camps on higher ground for periods of more than a year. About 80% of the water supply lines were broken (*4*) and so were most sewage lines, making the situation ideal for transmission of water-borne diseases.

Because an outbreak of cholera had occurred in the Andaman and Nicobar Islands in 2002 (*5*), we were apprehensive about outbreaks of infectious diseases after the tsunami, particularly among children, who are less immune to most infections; therefore, we increased our efforts to identify and contain these possible outbreaks as quickly as possible. However, except for a cluster of cases of rotaviral diarrhea (*6*), no major infectious disease outbreak occurred among residents of the Andaman Islands in the year that followed the tsunami.

Although the incidence of severe cases of diarrhea among children admitted to G.B. Pant Hospital in Port Blair, the only referral hospital in the Andaman Islands, varied greatly from month to month during 2001–2007, the incidence began decreasing after 2005, as indicated by the 12-month moving average (Figure). The mean number of cases per year fell from 361.4 during 2001–2005 to only 255.0 during 2006 and 2007 (p = 0.00025).

The estimated annual incidence of acute diarrhea per 100,000 children in the Andaman and Nicobar Islands was 609 in 2001, 580 in 2002, 595 in 2003, 601 in 2004, 571 in 2005, 370 in 2006, and 420 in 2007. For these incidence estimates, the population at risk during the years 2002–2007 was calculated by extrapolating from the 2001 census population on the basis of an annual population growth rate of 1.53% (the average for 1991–2001) and assuming that children ≤15 years old constituted 36.2% of the total population each year (as they did in 2001). The reduction in the number of acute cases of childhood diarrhea began several months after the tsunami, when the water and sewage systems of the islands had been repaired and renovated in many areas.

According to official reports, the cost of the restoration and renovation of the water and sewage systems after the tsunami was 389.9 million rupees, >2× the projected cost of work on the water and sanitation systems (172.9 million rupees) prior to the tsunami (*4*). In the aftermath of the tsunami, 52 km of new pipelines were laid and 12.5 km of old pipelines were replaced. Water supplies were augmented in 49 areas (*4*). The revamped water and sewage systems eliminated many sources of fecal contamination.

Moreover, by the middle of 2005, post-disaster assistance had been provided by voluntary organizations, missionaries, nongovernmental organizations, and government agencies from mainland India and abroad. This assistance resulted in further improvements in the area's public sanitation infrastructure and hygiene, particularly in the temporary shelters that displaced residents were living in; it also raised awareness among island residents about the threat of water-borne diseases. All of these factors were likely con-
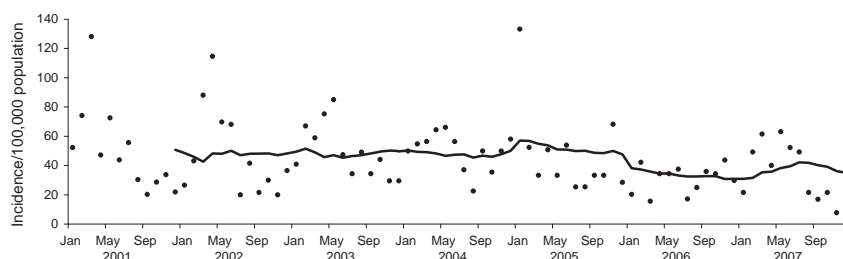

Figure. Estimated monthly incidence (black dots) of acute diarrhea among children ≤15 years of age in the Andaman Islands and 12-month moving average of the monthly incidence (black line), 2001–2007. Data based on cases of disease among children admitted to G.B. Pant Hospital, Port Blair, Andaman and Nicobar Islands, India.