

*M. genavense* infections occur only rarely in persons other than AIDS patients (as in the present case), but they always occur in immunocompromised persons (7,8). To date, only 1 case of disseminated infection has been reported in a solid-organ (kidney) transplant recipient; the diagnosis was made by molecular identification in isolates from blood and marrow cultures. That patient died of complications from *M. genavense* infection (9). Because *M. genavense* is a fastidious organism, the infections it causes are difficult to diagnose and their frequency is probably underestimated, which may change with increased use of direct molecular biological methods.

Optimal treatment of *M. genavense* infections has not been established (10). Experience with *M. genavense* infections in AIDS patients and with other nontuberculous mycobacteria infections in solid-organ transplant recipients suggests that at least 2 antimicrobial drugs should be used for a prolonged period; when possible, immunosuppressive drugs should be concurrently reduced (1,3,6,10). Outcome of nontuberculous mycobacteria infections in transplant patients is highly variable (1,5) but was satisfactory in the present patient, who was treated with quintuple antimicrobial-drug therapy and reduced immunosuppressive therapy.

This case of a disseminated infection due to *M. genavense* in a heart transplant recipient was diagnosed early. Universal 16S rRNA gene sequencing after amplification directly from intestinal biopsy specimens enabled fast diagnosis and appropriate management.

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## Isolation of Novel Adenovirus from Fruit Bat (*Pteropus dasymallus yayeyamae*)

**To the Editor:** Bats are thought to be one of the most important reservoirs for viruses such as Nipah virus, severe acute respiratory syndrome (SARS) coronavirus, and Ebola virus (1). These pathogens became known after extensive surveys of bats following outbreaks. As a first step in investigating unidentified pathogens in bats and to help forecast the potential threat of emerging infectious diseases, we tried to isolate and characterize viruses that persistently infect bats. In the process, we isolated a novel adenovirus from a fruit bat in Japan.

*Pteropus dasymallus yayeyamae*, or Ryukyu flying fox, is a fruit bat of Japan. With the permission of the governor of Okinawa, we caught 1 adult male bat of this species and used its spleen and kidneys to establish primary cell cultures. On the 4th passage of the primary adherent cells derived from the spleen, a cytopathic effect (CPE) appeared without any visible

microbe, indicating that the cell culture contained a virus. The virus, tentatively named Ryukyu virus 1 (RV1), caused apparent CPE on primary kidney cells derived from a Ryukyu flying fox and on our established bat kidney T1 (BKT1) cells, which were derived from the kidney of a horseshoe bat (*Rhinolophus ferrumequinum*) and transformed with expression plasmid DNA encoding the large T antigen of replication origin-defective simian virus 40.

To identify the virus, RV1, we applied the rapid determination of viral RNA (RDV) system version 1.0 (2). However, no viral nucleic acid sequence was detected from an RNA sample in the RV1-infected BKT1 cells. For detection of viral DNA, we developed a system for rapid determination of viral DNA sequences (RDV-D) by minor modification to the RDV system for RNA viruses (2–4). The results indicated that 2 of the fragments were homologous to the gene encoding the precursor of terminal protein (pTP) of adenoviruses. Further RDV-D analysis showed that 6 fragments (139 bp, DDBJ/EMBL/GenBank accession no. AB302970) were homologous to the pTP gene and that another

6 fragments (316bp, DDBJ/EMBL/GenBank accession no. AB302971) were homologous to the gene encoding the precursor of protein VI (pVI) of adenoviruses. These results indicated that RV1 must belong to the family *Adenoviridae*.

To further confirm that RV1 isolate was an adenovirus, we used PCR and sequencing. We performed the first reaction with the outer primer pair (polFouter and polRouter) of a nested PCR method, targeting the viral DNA polymerase gene with highly degenerate consensus primers that have been described recently (5). A fragment of  $\approx 550$  bp was amplified from RV1 as well as from human adenoviruses-1, -3, -4, and -7 (data not shown). Sequence analysis of the amplified product (DDBJ/EMBL/GenBank accession no. AB303301) showed that RV1 was homologous to tree shrew adenovirus 1 (70.0% amino acid sequence identity), porcine adenovirus 5 (69.2%), canine adenovirus 1 (68.9%), human adenoviruses-3, -16, -21 and -50 (68.9%), and other viruses (>64.8%) in genus *Mastadenovirus*, but less homologous (46.7%–57.8%) to viruses in other genera, *Siadenovi-*

*rus*, *Aviadenovirus*, and *Atadenovirus*. In addition, a phylogenetic tree based on amino acid sequences indicated that RV1 belongs to family *Adenoviridae*, genus *Mastadenovirus* (Figure).

Electron microscopy of RV1-infected BKT1 cells indicated that RV1 accumulated in the nucleus and that the size of capsids was 60–70 nm (data not shown). Restriction endonuclease analysis of the RV1 genome indicated that the genome was  $\approx 20$ –30 kbp (data not shown). These features are consistent with RV1 being an adenovirus.

Until now, a number of RNA viruses have been isolated from bats, but isolation of DNA virus is rare (1). The isolation of the novel adenovirus seems to be possible because of usage of the primary cells originated from the host; DNA viruses might have more restricted host range than RNA viruses and require host-originated cells for the growth. In addition, our success in DNA virus isolation might have resulted from usage of the adult animal latently and persistently infected with DNA viruses such as adenovirus and herpesvirus.

In conclusion, we isolated a novel virus from a fruit bat. This virus was isolated from a healthy bat, which suggests that the virus may persistently infect fruit bats. Although its pathogenicity for humans is still unknown, knowledge of RV1 will be useful in epidemiologic studies of infectious diseases emerging from bats because persistently infecting viruses might be isolated together with primary pathogens. We are planning to establish cell lines from bats and isolate more viruses from persistently infected bats.

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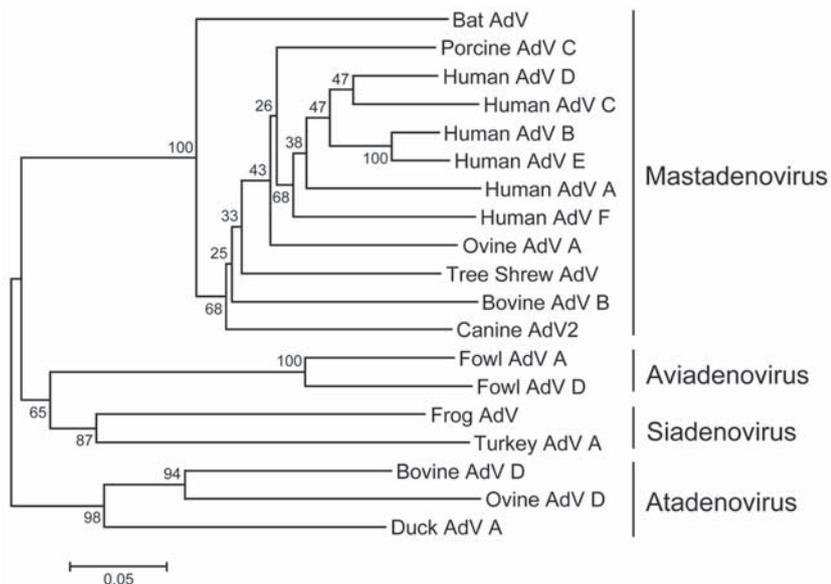


Figure. Phylogeny of adenoviruses based on analysis of partial amino acid sequences of DNA polymerase protein. Trees were estimated by using the neighbor-joining method based on the amino acid pairwise distance and MEGA 4.0 software ([www.megasoftware.net](http://www.megasoftware.net)). Numbers represent percentage bootstrap support (100 replicates).

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## Letters

Letters commenting on recent articles as well as letters reporting cases, outbreaks, or original research are welcome. Letters commenting on articles should contain no more than 300 words and 5 references; they are more likely to be published if submitted within 4 weeks of the original article's publication. Letters reporting cases, outbreaks, or original research should contain no more than 800 words and 10 references. They may have one Figure or Table and should not be divided into sections. All letters should contain material not previously published and include a word count.

## Fluoroquinolone-Resistant Group B Streptococci in Acute Exacerbation of Chronic Bronchitis

**To the Editor:** Fluoroquinolones (FQs) that are active against streptococcal species (e.g., levofloxacin and moxifloxacin) have been recommended by numerous national health authorities and international organizations for treating acute exacerbations of chronic bronchitis and pneumonia in adults (1). However, use of these antimicrobial drugs for treating community-acquired infections has led to an increase in FQ-resistant strains in bacteria such as *Streptococcus pneumoniae*. Group B streptococci (GBS, e.g., *S. agalactiae*) are the leading cause of invasive infections (pneumonia, septicemia, and meningitis) in neonates. GBS are also associated with bacteremia, endocarditis, and arthritis, and are responsible for deaths and illness in nonpregnant women with underlying diseases and in elderly adults (2). We describe, to our knowledge, the first GBS clinical isolate in France resistant to FQ; the isolate was from a patient treated with levofloxacin.

GBS CNR0717 strain was isolated as the predominant bacterium in a culture ( $>10^7$  CFU/mL) from 2 purulent sputum samples from an 80-year-old man (leukocytes  $>25$ , epithelial cells  $<10$ ) obtained 8 days apart. This patient was treated for 2 weeks with levofloxacin, 750 mg/day, for acute exacerbation of chronic bronchitis. No other relevant respiratory bacterial pathogens were present in

these samples. GBS CNR0717, a capsular serotype IV strain, was suspected to have reduced susceptibility to FQs because no inhibition zone was observed around disks containing norfloxacin and pefloxacin disks, and reduced diameters were observed around disks containing ciprofloxacin and levofloxacin. Antibiograms were performed according to recommendations of the Clinical and Laboratory Standards Institute (3) on Mueller Hinton agar (Bio-Rad, Marnes la Coquette, France) supplemented with 5% horse blood. This strain was susceptible to all other antimicrobial drugs usually active against GBS (penicillin, erythromycin, clindamycin, tetracycline, rifampicin, vancomycin) and showed low-level resistance against aminoglycosides. MICs for 6 FQs (Table) indicate that GBS CNR0717 was highly resistant to pefloxacin and norfloxacin, with MICs  $>64$  mg/L, and showed increased MICs for ciprofloxacin, sparfloxacin, levofloxacin, and moxifloxacin. No reduction of FQ MICs was observed with reserpine (10 mg/L), which indicated that resistance to FQ was not caused by an active efflux pump system.

Three major mutations have been reported for FQ resistance in streptococci at codon positions 81 in *gyrA* and 79 or 83 in *parC* (4). DNA sequence analysis of these regions showed a mutation in *parC* (Ser 79  $\rightarrow$  Tyr) but not in the wild-type susceptible strain (NEM316). No mutation was detected in the *gyrA* gene. FQ resistance in streptococci is acquired through a stepwise process and has been extensively studied in *S. pneumoniae*. First-step mutants conferring low-level resistance generally result from mutations in either *gyrA* or *parC*. There is also

Table. MICs of fluoroquinolones for strains of group B streptococci (GBS), France

Strain	MIC (mg/L)*					
	Pef	Nor	Cip	Spa	Lev	Mox
GBS CNR07017	>64	>64	4	1	4	1
GBS NEM316	16	8	2	0.5	1	0.25

\*Pef, pefloxacin; Nor, norfloxacin; Cip, ciprofloxacin; Spa, sparfloxacin; Lev, levofloxacin; Mox, moxifloxacin.