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## Carbapenemase-producing *Acinetobacter* spp. in Cattle, France

**To the Editor:** Multidrug resistance in bacteria isolated from animals is an emerging phenomenon, mirroring what is happening among humans. During the past decade, expanded-spectrum β-lactamases in *Enterobacteriaceae* from humans (*1*) and animals (*2*) worldwide have been reported. Among humans, as a consequence of this high rate, use of carbepenems is increasing selection pressure; carbapenem-resistant gram-negative organisms are increasingly reported, including carbapenemase-producing *Enterobacteriaceae* and *Acinetobacter* spp. (*3*).

The most commonly acquired carbapenemases identified in *Acinetobacter* spp. correspond to carbapenem-hydrolyzing class D β-lactamases (*3*). In particular, the worldwide spread of OXA-23–producing *A. baumannii* is considered a serious threat; those strains are frequently involved in nosocomial outbreaks for which therapeutic options are extremely limited (*3*,*4*). Our study objective was to evaluate the possible occurrence of carbapenemase-producing gram-negative bacteria in dairy cattle in France.

In August 2010, at a dairy farm 30 km from Paris, France, rectal swabs were collected from 50 cows. Samples were precultured in buffered peptone water and incubated for 18 h at 37°C. Cultures were inoculated by streaking 100 μL of the suspensions onto Drigalski agar plates (bioMérieux, Balmes-les-Grottes, France) containing 1 μg/mL of imipenem to select for carbapenem-resistant gram-negative isolates. Of the 50 samples, 9 produced growth on imipenem-containing plates. All colonies tested (10 colonies/sample) by using the API 20 NE (bioMérieux) system were first identified as *A. lwoffii*. Molecular techniques based on sequencing of the *gyrA*, *gyrB*, and *rpoB* genes (*5*) enabled more precise identification and indicated that all isolates belonged to the *Acinetobacter* genomospecies (DNA group) 15TU, which is known to be phylogenetically related to *A. lwoffii* and which has been reportedly isolated from sewage, freshwater aquaculture habitats, trout intestines, and frozen shrimp (*6*).

One colony per sample was retained for further investigation (isolates BY1 to BY9). Susceptibility testing and MIC determinations were performed by disk-diffusion assay (Sanofi-Diagnostic Pasteur, Marnes-la-Coquette, France) and Etest (AB bioMérieux, Solna, Sweden) (Table). All isolates except 1 were resistant to penicillins, combinations of penicillins and β-lactamase inhibitors, and carbapenems but susceptible to cefotaxime and of reduced susceptibility to ceftazidime. Isolate BY1 showed higher MICs for carbapenems (Table). In addition, all isolates were resistant to tetracycline, kanamycin, and fosfomycin and remained susceptible to fluoroquinolones, chloramphenicol, gentamicin, amikacin, tobramycin, and sulfonamides. Susceptibility profiles of 3 *Acinetobacter* genomospecies 15TU reference strains showed that they were fully susceptible to penicillins, carbapenems, tetracycline, and kanamycin.

Clonal diversity between the isolates was assessed by pulsed-field gel electrophoresis (*5*), which showed 6 distinct genotypes. Isolate BY1 corresponded to a single clone (data not shown), which indicated that the occurrence of *Acinetobacter* genomospecies 15TU strains among these animals was not the result of dissemination of a single clone.

PCR detection and sequencing of genes that encode carbapenem-hydrolyzing class D β-lactamases (*5*) showed that the 9 *Acinetobacter* genomospecies 15TU isolates harbored a $bla_{\text{OXA-23}}$ gene, whereas the 3 reference strains remained negative. Sequencing confirmed that all isolates expressed β-lactamase OXA-23, which is known to be widespread in *A. baumannii*.

Mating-out assays and plasmid electroporation assays were performed by using $bla_{\text{OXA-23}}$–positive *Acinetobacter* spp. isolates as donors and rifampin-resistant *A. baumannii* BM4547 isolates as a recipient strain (*5*); however, these assays were unsuccessful. Plasmid DNA analysis (*5*) gave uninterpretable results, with DNA degradations.

The genetic structures surrounding the $bla_{\text{OXA-23}}$ gene were investigated by PCR mapping (*7*), which identified transposon Tn*2008* in isolate BY2 only. Tn*2008* is a major vehicle for the spread of the $bla_{\text{OXA-23}}$ gene in *A. baumannii* in the People's Republic of China (*8*) and the United States (*9*). In the other isolates, the IS*Aba1* element of Tn*2008* had been truncated

Table. Antimicrobial drug MICs for *Acinetobacter* genomospecies 15TU isolates from cows and reference strains, France, August 2010*

| | MIC, μg/mL | | | |
|---|---|---|---|---|
| | *Acinetobacter* genomospecies 15TU | | Reference strain | |
| Drug class | BY1 | BY2–BY9 | NIPH 2171 | NIPH 899 |
| Penicillins and combinations | | | | |
| Amoxicillin | >256 | 128–256 | 4 | 4 |
| Amoxicillin + CLA | >256 | 128–256 | 4 | 4 |
| Cephalosporins | | | | |
| Cefoxitin | 32 | 16–32 | 16 | 16 |
| Cefotaxime | 32 | 16–32 | 8 | 6 |
| Ceftazidime | 32 | 16–32 | 16 | 16 |
| Cefepime | 16 | 4–16 | 4 | 4 |
| Monobactam (aztreonam) | 64 | 32 | 32 | 16 |
| Carbapenems | | | | |
| Meropenem | 16 | 2–4 | 0.5 | 0.5 |
| Imipenem | >32 | 4–6 | 0.25 | 0.25 |
| Doripenem | 8 | 2–4 | 0.5 | 0.5 |
| Cyclines | | | | |
| Tetracycline | >256 | >256 | 0.5 | 0.5 |
| Tigecycline | 0.064 | 0.047–0.064 | 0.047 | 0.125 |
| Quinolones (ciprofloxacin) | 0.5 | 0.5 | 0.25 | 0.25 |
| Aminoglycosides | | | | |
| Gentamicin | 0.5 | 0.25–0.5 | 0.25 | 0.25 |
| Kanamycin | >256 | >256 | 0.5 | 0.5 |
| Sulfonamides | 4 | 4 | 4 | >256 |

*CLA, clavulanic acid (4 μg/mL).

by a novel insertion sequence termed IS*Acsp2* (www-is.biotoul.fr).

The dairy farmer indicated that most animals from which OXA-23 producers had been identified had received antimicrobial drugs in the previous weeks. Although 1 animal had received amoxicillin-clavulanate, most of the others had been given oxytetracycline and neomycin to treat mastitis.

β-lactamase OXA-23 is a common source of carbapenem resistance in *A. baumannii (5)*. Infections with multidrug-resistant OXA-23–producing *A. baumannii* or *A. junii* have been reported from hospitals but not from the community. Our study showed that OXA-23–producers in particular, and carbapenemase producers in general, may be isolated from animals. Among the hypotheses that could explain the selection of this carbapenemase, use of penicillins or penicillin–β-lactamase inhibitor combinations could create selective pressure for β-lactamases because OXA-23 does confer, in addition to decreased susceptibility to carbapenems, a high level of resistance to those compounds. We have previously shown that *A. radioresistens*, an environmental species, was the progenitor of the $bla_{OXA-23}$ gene (*10*). Studies are needed to determine to what extent and at which locations *Acinetobacter* genomospecies 15TU and *A. radioresistens* might co-reside and therefore where the $bla_{OXA-23}$ gene exchange might have occurred.


## Acknowledgments

We thank A. Nemec and L. Dijkshoorn for the *Acinetobacter* genomospecies 15TU reference strains.

This work was partially funded by a grant from the Institut National de la Santé et de la Recherche Médicale (INSERM) (U914), the Ministère de l'Education Nationale et de la Recherche (UPRES-EA3539), Université Paris XI, and mostly by grants from the European Community (TROCAR, HEALTH-F3-2008-223031 and TEMPOtest-QC, HEALTH-2009-241742) and from INSERM (U914). L.P. was funded by a grant-in-aid from the École Nationale Vétérinaire de Maisons-Alfort through an INSERM–École Nationale Vétérinaire de Maisons-Alfort contract.



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DOI: http://dx.doi.org/10.3201/eid1803.111330

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## *Aedes albopictus* Mosquitoes, Yucatan Peninsula, Mexico

**To the Editor:** We collected Asian tiger mosquitoes, *Aedes albopictus* (Skuse), in Cancun in the Yucatan Peninsula of Mexico in September 2011. This mosquito is a nuisance biter of humans and a vector of numerous arboviruses, including those causing dengue, yellow fever, and chikungunya (*1*).

*Ae. albopictus* mosquitoes, which are native to Southeast Asia, emerged in the continental United States in 1985 and thereafter spread rapidly across the southeastern United States and into northern Mexico (*2,3*). These mosquitoes have also been found in the states of Tamaulipas, Coahuila, and Nuevo Leon in northern Mexico, Chiapas in southern Mexico, and south of Mexico in Guatemala and Belize (*3–9*). These findings are now complemented by our collection of *Ae. albopictus* mosquitoes from Cancun in Quintana Roo State, which with Yucatan and Campeche States compose the Yucatan Peninsula. A previous study of the mosquito fauna of Quintana Roo conducted in 2006 did not report any *Ae. albopictus* mosquitoes (*10*).

During September 2011, *Ae. albopictus* mosquitoes were collected from a cemetery in Cancun, which is located in the eastern part of the Yucatan Peninsula (21°8.53′N, 86°52.79′W) (Figure). The collection location was shaded by trees. Water in containers from which larvae were collected had an average temperature of 24.5°C and a pH of 8.5. The larval collection included ≈30 specimens of different developmental stages that were collected from vases and other artificial containers in the cemetery. The containers were examined as part of routine surveillance activities by Servicios Estatales de Salud de Quintana Roo. Larvae suspected to be those of *Ae. albopictus* mosquitoes were reared to adults for identification, and a colony of *Ae. albopictus* mosquitoes from Cancun was established.

$F_0$ or $F_1$ adult specimens were confirmed to be *Ae. albopictus* moszquitoes by species identification at Servicios Estatales de Salud de Quintana Roo (Quintana Roo, Mexico), Universidad Autónoma de Yucatan (Merida, Mexico), and Colorado State University (Fort Collins, CO, USA). The initial mosquito larval collection was composed of 26 *Ae. albopictus*, 3 *Ae. aegypti*, and 1 *Culex* sp. In addition, 6 *Ae. albopictus* female mosquitoes were collected from the cemetery by landing catches.

Finding *Ae. albopictus* mosquitoes in Cancun was not surprising because these mosquitoes have been found in nearby Belize (*9*). Cancun is also a major port for ships carrying tourists and goods that originate in areas to which *Ae. albopictus* mosquitoes are endemic, including Florida and Texas. Nevertheless, the introduction of *Ae. albopictus* mosquitoes into Cancun and the high potential for establishment and spread across the Yucatan Peninsula has major public health implications.

The Yucatan Peninsula is hyperendemic for dengue, with all 4 dengue virus (DENV) serotypes circulating in this region. Should *Ae. albopictus* mosquitoes persist in this region, they may spread and come to play a secondary role to *Ae. aegypti* mosquitoes as local vectors of DENV. *Ae. albopictus* mosquitoes may also change local virus transmission dynamics. For example, DENV transmission may be intensified in rural areas because *Ae. albopictus* mosquitoes are more likely than *Ae. aegypti* mosquitoes to be found in this setting. *Ae. albopictus* and *Ae. aegypti* mosquitoes also may differ in their potential for vertical transmission of DENV, which could