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Letters to the Editor

Caz-hi, an Extended-Spectrum TEM β-Lactamase (TEM-61), Is Derived from Caz-lo (TEM-11) by In Vivo Selection

Ceftazidime was introduced into our hospital during the second half of 1987. It was aimed especially for use in the intensive care unit to treat infections with gram-negative bacilli with diminished sensitivity for cefuroxime, at that time the first-line β-lactam antibiotic for serious infections. At that time *Escherichia coli* and *Klebsiella* sp. isolates were susceptible to ceftazidime. An increase in cefuroxime-resistant isolates was noted, resulting in an increase in the use of ceftazidime. A few months later ceftazidime-resistant *E. coli* and *Klebsiella pneumoniae* were isolated. It could be shown that these isolates harbored different plasmid-mediated extended-spectrum β-lactamases (ESBLs), at that time a rare phenomenon (7). The phenotypic characteristics of the isolates and the β-lactamases were reported (10), and the epidemiology is summarized in Fig. 1. From a patient treated with ceftazidime, and in whom the first *K. pneumoniae* producing an ESBL, called "Caz-lo" (Isoelectric focusing point [pl], 5.6), was observed, 10 days later a more resistant *K. pneumoniae*, of the same serotype, with an identical arbitrarily primed PCR profile and with a "Caz-hi" enzyme, was isolated. This was indicative of Caz-hi being an in vivo-selected mutant of Caz-lo. Starting from the Caz-lo *K. pneumoniae*, in vitro selection with ceftazidime resulted in a strain that produced an ESBL with the Caz-hi phenotype. This reinforced the hypothesis of in vivo selection, initially based on epidemiological data only.

The CAZ-lo and CAZ-hi enzymes were detected in other patients, in other *K. pneumoniae* serotypes, and other species but disappeared after a few months and were replaced by other ESBL enzymes (designated FUR-2) (Fig. 1). Here we report newly obtained data on the Caz-hi ESBL enzyme.

The Caz-lo ESBL gene has been sequenced previously and is characterized by a Glu-to-Lys transition at amino acid position 39 (numbering according to Ambler et al. [1]) and an Arg-to-His transition at position 164 (8), with respect to the sequence of TEM-1. According to the standardized nomenclature of β-lactamases, Caz-lo has been named TEM-11 (6).

The sequencing data reported here reveal that Caz-hi (designated TEM-61) differs from TEM-11 by a single transition from Glu to Lys at amino acid position 240 (2). This transition has been shown to occur in other ESBL enzymes (TEM-5, TEM-10, TEM-24, TEM-27, TEM-28, TEM-42, TEM-46, and TEM-49) (3–5) but was never observed in combination with the mutations which are characteristic for TEM-11. It is interesting that a single mutation from an acidic to a basic amino acid at position 240, which also readily explains the shift in pl from 5.6 to 6.5, causes an increase in ceftazidime resistance from a MIC of 8 mg/liter (TEM-11) to a MIC of 256 mg/liter (TEM-61).

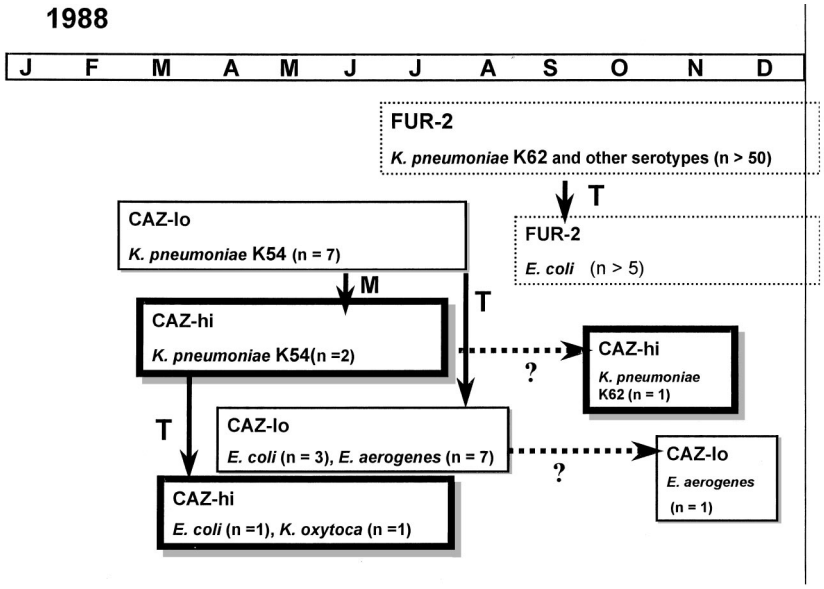

FIG. 1. Epidemiology of ESBLs. M, mutation of enzyme; T, transfer of plasmid to another serotype or another species; ?, period of apparent disappearance of the enzyme. Months are abbreviated at the top.

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Geert Claeys
Thierry De Baere
Mario Vaneechoutte
Gerda Verschraegen
Laboratory for Bacteriology and Virology
University Hospital
De Pintelaan 185
9000 Gent
Belgium