of bla(OXA-24)-specific primers, the DNA sequence was identical with blaOXA-72(GenBank accession numbers AY739646). A protein of 29kDa decreased in one carbapenem-resistant isolate (isolate 66) and a protein of 24kDa overexpressed in two carbapenem-resistance isolates (isolate 66 and 67). One positive transformant was obtained with PCR, western blot and Nitrocefin test all positive.
Conclusion: These findings indicates that producing class D β-lactamase OXA-23 and OXA-72 was one of the mechanism for carbapenems resistance of *Acinetobacter* spp. in Jinan. The decrease of a 29 kDa out membrane protein and the overexpression of the 24kDa penicillin-binding protein were also related to the carbapenems resistance to in *Acinetobacter* spp. We found carbapenemase OXA-72 which is seldom reported and successfully expressed it in *Pichia pastoris*. Now we are purifying the carbapenemase OXA-72 and do further study on the properties of it.

P1517 Warning! Oxacillinase-mediated carbapenem resistance spreading in Enterobacteriaceae

M. Yilmaz, Y. Aybar Bilir, K. Midilli, R. Ozaras, A. Mert, F. Tabak, R. Ozturk (Istanbul, TR)

Introduction: Resistance to expanded-spectrum cephalosporins in Enterobacteriaceae has been reported to be related to the production of Ambler class A extended-spectrum β-lactamases (ESBLs) and chromosomal and plasmid-encoded AmpC cephalosporinases. However, increasing number of reports regarding β-lactamase mediated resistance to carbapenems in Enterobacteriaceae are being published.
Objectives: Between 2002–2006, we identified 21 Enterobacteriaceae strains that are resistant to any of the carbapenems in the infectious diseases and clinical microbiology laboratory of a 1500 bed university hospital. The aim of the study was to identify the resistance mechanism(s) of carbapenems in these strains and to check for clonal dissemination.
Methods: Bacterial strains and and MIC determinations. All the strains (15 *Klebsiella pneumoniae*, 1 *Escherichia coli*, 2 *Enterobacter* cloaceae and 2 *Enterobacter aerogenes*) were clinical isolates from the Istanbul University, Cerrahpasa Medical Faculty Hospital, Infectious Diseases and Clinical Microbiology Laboratory. The strains were identified with the API 32GN system. Antibiotic susceptibilities and MICs were with disc diffusion assay and E-test.
IEF analysis. Isoelectric focusing (IEF) analysis was performed with an ampholine polyacrylamide gel (pH 3.5 to 9.5).
PCR experiments and DNA sequencing: Using total DNA of each of the strains, PCR amplifications of the blaOXA-48 genes were performed with the primers OXA-48A (5′-TTGGTGGCATCGATTATCGG-3′) and OXA-48B (5′-GAGCACTTCTTTTGTGATGGC-3′), giving rise to a 743-bp fragment.
Molecular typing. Molecular characterisation of the strains was done by macrorestriction analysis of genomic DNA with XbaI (New England BioLabs). DNA fragments were separated by pulsed-field gel electrophoresis (PFGE) in a CHEF-DR II system (Bio-Rad). Electrophoresis conditions were pulse times ranging from 5 to 35 s for 24 h at 6 V/cm and 14°C.
Infection control procedures. Patients from whom carbapenem-resistant Enterobacteriaceae was recovered at any site were visited by a member of the infection control team. Colonisation or infection was determined according to the definition of the Centers for Diseases Control and Prevention for nosocomial infections.
Results and Conclusion: All the carbapenem resistant strains isolated produced OXA-48, a class D oxacillinase significant with carbapenem-hydrolysing activity. No relationship was observed among the seven patients hospitalised in different wards.

P1518 Diversity of antibiotic resistance determinants among the recent population of *Acinetobacter baumannii* strains belonging to European clone II

L. Krizova, M. Maixnerova, L. Dijkshoorn, A. Nemec (Prague, CZ; Leiden, NL)

Objective: The recent increase in *Acinetobacter baumannii* resistance to carbapenems in the Czech Republic has been associated with the spread of strains belonging to European clone II. Although multidrug resistance was a common property of these strains, they differed in resistance to particular agents. The aim of this study was to assess the genetic basis of this variation.
Methods: Sixty-six strains obtained from 37 intensive care units in the Czech Republic in 2005–6 were studied. The strains were previously classified as European clone II by AFLP and were shown to have highly similar or identical PFGE patterns. They were tested for MICs to sulbactam, ceftazidime, meropenem, gentamicin, tobramycin, amikacin, netilmicin and doxycycline, and for the presence of 20 genes associated with resistance to these agents.
Results: All strains were positive for the genes encoding OXA-51, AmpC and the AdeABC efflux system while no strain tested positive for those encoding metallo-β-lactamases, OXA-23 and OXA-24 carbapenemases, or aminoglycoside-modifying enzymes AAC(3)-II, AAC(6′)-I, and ANT(2″)-I. The strains varied with respect to the presence of the genes encoding the following proteins (% positive strains): TEM-1 (80), Tet(B) (92), Tet(A) (5), AAC(3)-I (83), APH(3′)-I (80), APH(3′)-VI (30), OXA-58 (3) and a class 1 integrase (83). ISAba1 was found in 95% strains and three integron variable regions (2.5, 3.0 and 3.5 kb) were identified, differing only in the number of copies of the orfX cassette. The presence of particular genes and the corresponding resistant phenotype were in good agreement. All strains with ISAba1 located upstream of the AmpC gene (n = 33) were resistant to ceftazidime (MIC > 64 mg/l) while those with ISAba1 upstream of the OXA-51 gene (n = 33) showed increased meropenem MICs (2–32 mg/l). Individual strains carried from 4 to 12 resistance genes in 17 combinations. Different combinations were also found in isolates from the same ward and having identical PFGE patterns.
Conclusion: The variation in antibiotic resistance in the studied strains results from the differences in the presence of acquired resistance genes and, possibly from the effect of ISAbaI on the expression of intrinsic genes. The high genetic versatility of European clone II might contribute to its ability to develop resistance to nearly all clinically relevant antibiotics.
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Quinolone and fluoroquinolone resistance – Part 2

P1519 Plasmid-mediated quinolone resistance determinant QepA from ESBL-producing *Escherichia coli* in France

V. Cattoir, L. Poirel, P. Nordmann (Le Kremlin Bicetre, FR)

Objectives: Three different plasmid-mediated quinolone resistance (PMQR) determinants have been described: Qnr, AAC(6′)-Ib-cr and QepA. QepA is an efflux pump responsible for decreased susceptibility to hydrophilic fluoroquinolones, was identified in association with RmtB (responsible for resistance to all aminoglycosides) in two *Escherichia coli* isolates from Belgium and Japan. To date, the single epidemiological survey performed reported a low prevalence of the qepA gene (0.3%) among *E. coli* isolates from Japan. The aim of our study was to determine the presence of qepA gene among ESBL-producing enterobacterial isolates from a French University hospital.
Methods: Screening was performed on 121 ESBL-producing enterobacterial isolates collected in France from January 2007 to October 2007. PCR with primers specific for the qepA gene was used for screening. Detection and identification of ESBL genes was performed using specific