INTRODUCTION AND PURPOSE
Recently, emergence of carbapenem resistance was observed among Enterobacteriaceae causing urinary tract infections in Croatia. The aim of the study was to characterize the carbapenem resistance mechanisms and the molecular epidemiology of carbapenem resistance in urine isolates of Enterobacteriaceae from Croatia.

MATERIAL AND METHODS
The antimicrobial susceptibility to a wide range of antibiotics was determined by broth microdilution method. Double-disc synergy test (DDST) was performed to detect ESBLs and modified Hodge test (MHT) was used to screen for production of carbapenemases. MBL-E test was used to screen for production of metallo-β-lactamases. Additionally, the isolates were tested by combined disks tests using four disks of meropenem or imipenem and modified Hodge test (MHT) was used to screen for production of carbapenemases. and modified Hodge test (MHT) was used to test for carbapenemases.

RESULTS
In total 47 strains (31 Enterobacter spp., 12 Klebsiella pneumoniae, 2 Citrobacter spp., and one Escherichia coli and Serratia marcescens, respectively) with reduced susceptibility to carbapenemes, isolated from urine during 2012-2013 from two large hospital centres in Croatia were analyzed. All strains were resistant to amoxicillin alone and combined with clavulanate, cefuroxime, ceftazidime, cefotaxime and ceftriaxone as shown in Table 1. The strains showed variable degree of susceptibility to cephalimimpenem, meropenem, ertapenem, aminoglycosides and ciprofloxacin. Except colistin (all isolates were susceptible), potent activity against carbapenemase producing isolates exhibited also amikacin (50% susceptible isolates). Genes encoding carbapenemases were found in 38 isolates, with VIM-1 metallo-enzyme being the most prevalent and found in 34 strains (twenty five E. cloacae isolates; one strain coharboured VIM-1 and NDM-1), six K. pneumoniae strains, two C. freundii and one S. marcescens isolate, respectively, NDM-1 was found in one Enterobacter cloacae, and KPC-2 in three Klebsiella pneumoniae strains. Nine strains were negative for true carbapenemases and harboured only ESBL belonging to CTX-M family (Table 1). Plasmids encoding VIM MBLS belonged to widespread IncA/C or IncFIB.

Table 1. Minimum inhibitory concentrations of various antibiotics and β-lactamase content of carbapenemase producing isolates of Enterobacteriaceae from urine.

CONCLUSIONS
The study demonstrated emergence of carbapenemase-producing enterobacteriaceae associated with urinary tract infections which could pose a serious therapeutic problem. The predominance of VIM-1 and KPC-2 β-lactamase was observed. In the previous studies Enterobacteriaceae positive for ESBLs belonging to CTX-M family were found to be dominant multiresistant pathogen among urinary isolates but continuing shift to carbapenemase producers is observed.