Incidence of Extended-Spectrum Beta-Lactamase (ESBL) producing multidrug resistant *Escherichia coli* in seafood

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E*scherichia coli* is the most common commensal bacteria and they become opportunistic and obligate pathogens when coexisting with pathogenic strains (Martinez, 2009). The occurrence of *E. coli* in seafood is directly related with either faecal contamination or contamination by the infected handlers. Now-a-days, antibiotics are most commonly used in human therapies, farm animals and aquaculture (Cheong *et al.*, 2014) which increase the incidence of resistant bacterial strains (Sapkota *et al.*, 2008). Beta-lactam antibiotics are the most preferred treatment regimens for many of the infectious diseases (Bush and Jacoby, 2010). The widespread and indiscriminate use of antibiotics coupled with the transmissibility of resistance lead to the emergence of multidrug resistant (MDR) organisms like MDR-Extended-Spectrum Beta-Lactamase (ESBL) producing *E. coli* and become difficult to treat (Kang *et al.*, 2005). In addition, *E. coli* may harbour the ESBL enzymes that confer resistance to most beta-lactam antibiotics mainly of extended-spectrum cephalosporins. ESBLs become easily transferable to other Enterobacterial species when encountered with it (Bradford, 2001). *E. coli* strains producing ESBLs are becoming multidrug resistant (MDR) and are considered to be one of the emerging pathogen world-wide (Ozcakar *et al.*, 2011). ESBLs are a group of enzymes and are encoded by genes on plasmids that hydrolyze all groups of beta-lactam antibiotics, including new generation group of cephalosporins mainly cefotaxime and cefriaxone (Perez *et al.*, 2007). Whereas in the recent years, several studies demonstrated the prevalence of ESBL producing Enterobacteriaceae in food products such as meat, chicken, raw milk and fish (Koo and Woo, 2011; Ryu *et al.*, 2012 and Ojer-Usoz *et al.*, 2013). ESBLs genes enter and disseminate through the food chain *via.* direct contact with humans and animals which could contribute to the spread of these strains (Egea *et al.*, 2012). There is no data available about ESBL-producing bacteria in food and aquaculture products and food of animal origin in Gujarat. Keeping this in view, the present study was carried out during September, 2011 to March, 2015 to monitor the presence of MDR and ESBL producing *E. coli* in seafood in Veraval, Gujarat, India. A total of 82 seafood samples (34.45%) were positive for presumptive *E. coli* on Tergitol - 7 out of 238 samples namely fresh samples (pomfret, horse mackerel, Indian mackerel, tuna, ribbonfish, seer-fish, croaker, ghol, dhoma, sardine, shark, ray fish), dried fishes, from Veraval fish market and frozen samples such as prawns, cuttle fishes, cephalopods and surimi collected from fish processing industries in Veraval region, Gujarat.

The *E. coli* counts ranged from $10^{1 \text{ to } 2}$ x 10^{2}cfu/g. About 140 *E. coli* isolates from 28 samples were confirmed on EMB agar and IMViC test and API 20E bioMerieux. One isolate from each confirmed sample was screened for ESBL producer in comparison with antimicrobial sensitivity. Eleven isolates (39.20%) were multidrug resistant (Fig.1) and four *E. coli* isolates (14.29%) were ESBL producers with triple ESBL detection Ezy MIC Strip (Fig. 2). Antibiotics were screened for determining MDR to cephalosporins (ceftazidime, cefepime, cefuroxime, ceftizoxime, cefoperazone, cefotaxime, ceftriaxone and cefoxitin), beta-lactam and beta-lactam inhibitors (amoxyclav, ticarcillin/ clavulanic acid, piperacillin/ tazobactam and ampicillin/sulbactam ampicillin) monobactam (aztreonam) and cephems and beta-lactam inhibitors (ceftazidime/ clavulanicacid), carbapenems (imipenem and meropenem), fluoroquinolones (ofloxacin, ciprofloxacin, gatifloxacin and levofloxacin), sulphonamides (trimethoprim/ sulfamethoxazole) and aminoglycosides (amikacin and gentamicin). All these ESBL producers and ESBL-Amp C positive were multidrug resistant ≥3 classes of antimicrobials except carbapenem and aminoglycosides. The higher rate of resistance was found with ampicillin (39.29%), trimethoprim/ sulfamethoxazole (32.14%), ciprofloxacin (28.57%) cefepime (17.86%) and cefuroxime (17.86%). Whereas, all the isolates were sensitive to carbapenems (imipenem and meropenem), beta-lactam and beta-lactam inhibitors (piperacillin/ tazobactam and ampicillin/sulbactam) and aminoglycosides (amikacin and gentamicin) and these antimicrobial profiles clearly indicate the presence of ESBL producers. The results of this study indicate the prevalence of ESBLs and multiple antibiotic resistance ESBL producing multidrug resistant *E. coli* in seafood in Gujarat. Their presence revealed that the seafood might be the possible reservoir, transferring to human and posing serious threat to seafood consumers. So, strict hygienic measures are required to reduce the ESBL producing *E. coli* contamination in seafood.

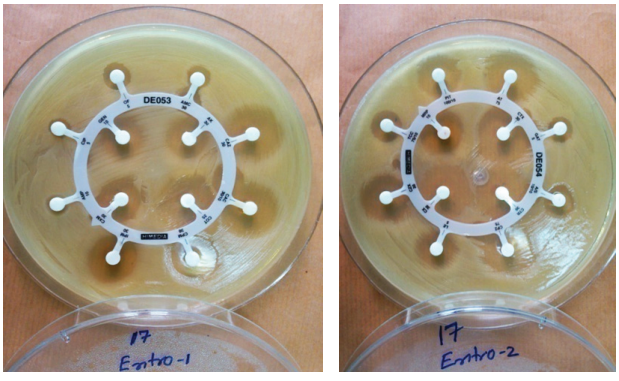

Fig. 1. Antibiogram of *E. coli* isolated from the seafood with 24 antimicrobial agents (HiMedia, Mumbai)

Fig. 2. Detection of ESBL producing *E. coli* with Triple ESBL detection Ezy MICTM Strip (MIX+/MIX) EM 079 (HiMedia, Mumbai)

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