P 045

NOSOCOMIAL OUTBREAK OF COLISTIN-RESISTANT OXA-48-PRODUCING *KLEBSIELLA PNEUMONIAE* FROM BLOODSTREAM INFECTIONS, GREECE

Samanta Sennati[1], Kalliopi Avgoulea[2], Antonio Cannatelli[1], Tommaso Giani[1], Vincenzo Di Pilato[3], Olympia Zarkotou[2], Athanasios Tsakris[4], Spyros Pournaras[4], Gian Maria Rossolini[5]

[1]Department of Medical Biotechnologies, University of Siena, Santa Maria alle Scotte University Hospital, Siena - Italy
[2]Department of Microbiology, Tzaneio Hospital, Piraeus - Greece
[3]Department of Surgery and Translational Medicine, University of Florence, Florence - Italy
[4]Department of Microbiology, Medical School, National and Kapodistrian University of Athens, Athens - Greece
[5]Department of Experimental and Clinical Medicine, University of Florence, Careggi University Hospital; Clinical Microbiology and Virology Unit, Careggi University Hospital; Don Carlo Gnocchi Foundation, Florence - Italy

Introduction: OXA-48 is a class D carbapenemase originally identified in isolates from Turkey and subsequently detected in several European and North African countries. This enzyme is mostly found in isolates of *Klebsiella pneumoniae* and *Escherichia coli* and shows a peculiar spectrum of activity, being able to hydrolyze carbapenems (although weakly), penicillins, and narrow-spectrum cephalosporins but not expanded-spectrum cephalosporins. In Greece a hospital outbreak of OXA-48-producing ST11 *K. pneumoniae* has been described in 2012, even though nosocomial outbreaks caused by KPC- and VIM-producing *K. pneumoniae* are most commonly reported. In the present study, we investigate an ongoing nosocomial outbreak of colistin-resistant OXA-48-producing *K. pneumoniae* isolates from bloodstream infections that started in June 2014.
Materials and Methods: Eighteen isolates of OXA-48-producing *K. pneumoniae* from blood cultures were collected from different patients during the period June 2014-March 2016 at the Tzaneio General Hospital of Piraeus (Greece), the same hospital that experienced the outbreak in 2012. Species identification was performed with the Vitek 2 automated identification system (bioMérieux, Marcy l'Étoile, France) and MICs were determined by broth microdilution according to CLSI guidelines. Fosfomycin susceptibility testing was performed by agar dilution according to CLSI. EUCAST clinical breakpoints were used for the interpretation. MLST was carried out following the Pasteur Institute scheme and a PCR-sequencing approach was used for the analysis of blaOXA-48 and mgrB genes. The clonal relationship was examined by pulsed-field gel electrophoresis (PFGE).
Results: Four of the 18 OXA-48-producing isolates exhibited a pandrug-resistant (PDR) phenotype being non-susceptible to all tested antibiotics, including colistin, while the remaining isolates showed variable susceptibility to gentamicin, fosfomycin and tigecycline (extensive drug-resistant, XDR, phenotype). In addition to OXA-48 all PDR strains produced CTX-M type ESBL. In all isolates, colistin resistance was due to insertional inactivation of the mgrB gene. Results from PFGE showed two different pulsotypes (A and B). MLST assigned all PDR isolates to ST147 and XDR to ST101, pulsotype A and B, respectively.
Conclusions: In the present study, we describe an ongoing nosocomial outbreak of OXA-48-producing *K. pneumoniae* in Greece caused by the spread of two different clones belonging to ST147, isolated in the first six months of the outbreak, and ST101, collected from 2015 to now. Of note, all ST147 strains showed a PDR phenotype, which seriously limits the therapeutic options for the treatment of such severe infections.

P 046

ANTIBACTERIAL PROPERTIES AND CYTOTOXICITY OF *SATUREJA MONTANA L.* AND *CORIANDRUM SATIVUM L.*

Lorenzo Mondì[1], Catia Longhi[1], Antonietta Lucia Conte[1], Paola Goldoni[1], Maria Grazia Ammendolia[2], Federica Rinaldi[3], Carlotta Marianecci[4], Maria Elisa Crestoni[4], Caterina Fraschetti[4], Francesca Scazzocchio[1]

[1]Department of Public Health and Infectious Diseases, Microbiology Section, "Sapienza" University of Rome, Rome - Italy
[2]Department of Technology and Health, National Institute of Health, Rome - Italy
[3]Center for Life Nano Science, "Sapienza" University of Rome@ Italian Institute of Technology, Rome - Italy
[4]Department of Drug Chemistry and Technologies, "Sapienza" University of Rome, Rome - Italy

Introduction: Essential oils (EOs) are used in traditional medicine due to recognized therapeutic properties, namely anti-microbial and cytotoxic

activities. Coriander (*Coriandrum sativum L., Apiaceae*) (CDO) and *Satureja montana, L. Lamiaceae* (SGG), are herb widely used as a spice, in food and pharmacy industries and in folk medicine. This study aims to analyze the antibacterial properties of CDO and SGG towards multidrug resistant (MDR) strains. Toxicity in eukaryotic cell lines it has been also valuated.

Materials and Methods: The CDO and SGG (Talia, Rome) compositional analysis was evaluated by gas chromatography and mass spectrometry (GC-MS). MDR uropathogenic *Escherichia coli* ECP19 and ECP32 were isolated at Hospital "Umberto I" Rome. Identification and susceptibility tests were performed by automated VITEK-2 System. *E. coli* ATCC 25922 was used as reference strain. The combined antimicrobial activity of EOs and gentamicin was reported as the fractional inhibitory concentration index (FICI). Bacterial cell modifications was evaluated by propidium iodide staining and electron microscopy. The *in vitro* cytotoxicity of CDO and SGG against human tumor cell lines (Hep-2) was determined by the MTT assay.

Results: The chemical analysis of essential oils showed that the main component of CDO was Linalool (74,7%) whereas in the SGG Carvacrol (51%), Tymol (11%), Terpinene (5,5%) and Cymene (13%). MICs and MBCs of SGG showed a better inhibitory activity respect to CDO values. The FICI values showed a positive interaction between EOs and gentamicin: additive for CDO and synergic for SGG. This effect can be explained by a greater penetration of the antibiotic due to changes of membrane structures as evidenced by propidium iodide staining and electron microscopy studies. Regard Hep-2 cell lines, SGG was able to inhibit cell proliferation more efficiently compared to CDO.

Conclusions: The preliminary results, showing a better biological activity of SGG compared with CDO, encourage further studies to highlight the main active components and their action mechanism on prokaryotic and eukaryotic cells.

P 047

ANTIMICROBIAL ACTIVITY OF NATURAL MOLECULES EXTRACTED FROM *HELICHRYSUM ITALICUM* AGAINST *STAPHYLOCOCCUS EPIDERMIDIS*

Pina Caputo, Vittoria Savio, Elisabetta Buommino, Elena Grimaldi, Brigida D'Abrosca, Antonio Fiorentino, Giovanna Donnarumma

Seconda Università degli Studi di Napoli, Napoli - Italy

Introduction: *Staphylococcus epidermidis* is able to form biofilm and is involved in infectious diseases related to indwelling medical devices, such as peripheral or central intravenous catheters. Bacterial biofilms are highly organized surface-associated communities of bacteria encased within a self produced extracellular matrix. Bacteria within a biofilm exhibit distinct phenotypes from planktonic cells, particularly with respect to growth and gene expression. Biofilm infections are rarely resolved by host defence mechanism and antibiotic therapy generally fails to kill bacteria residing within biofilm. In addition, the increasing emergence of drug-resistant bacteria highlight the need of a continuous screening of new antimicrobials. We reported the antimicrobial activity of new compounds extract of *Helichrysum italicum*, a medicinal plant used in folk medicine as an anti-inflammatory and anti-infective plant.

Materials and Methods: The *H. italicum* compounds dissolved in dimethylsulfoxide and diluted in Luria-Bertani, have been tested for their anti-microbial effect against *S. epidermidis*. The strain has been cultured in LB broth at 37°C. The compounds were added to bacterial cell suspension and the bacterial growth was monitored, measuring the OD in a microtiter plate reader at 600 nm of absorbance. For the rate of killing bacteria the compounds were added to bacterial suspension and incubated at 37 °C for 0, 4, 8, 16 and 24h, after incubations at 37 °C colonies were counted on LB agar by dilution titration method. To assess the ability of molecules to inhibit biofilm formation, compounds were added to microtiter plate at the time of inoculation and the cells were allowed to form biofilms. The biofilm was measured at 570 nm in an microplates reader after Cristal violet (1%) dyes.

Results: *S. epidermidis* growth was differently