**ABSTRACT**

*Pseudomonas aeruginosa* is a most commonly isolated nosocomial pathogen worldwide. Infections due to this organism are usually seen in patients with impaired immunity such as patients suffering from AIDS, cancer, burn wounds, cystic fibrosis, acute leukemia, organ transplants and intravenous-drug addiction. *P. aeruginosa* strains resistant to multiple drugs (MDRPA) have emerged with increasing frequency from different regions of the country. One of the most important mechanism of resistance is the production of enzyme β-lactamase by the resistant strains. The classes of β-lactamases that are implicated in the resistance are: Extended spectrum β-lactamases (ESBLs), metallo β-lactamases (MBLs) and ampicillin class C β-lactamases (AmpCs). The present study aims at determining the frequency of MDRPA strains in this region by *in vitro* cultural sensitivity assay. A frequency of 34.48% isolates was recorded as MDRPA. The comparative analysis of the virulence factors (the production of protease, lipase, hemolysin and gelatinase as well as biofilm formation) revealed comparable effect between the multiple drug resistant and sensitive isolates. The phenotypic expression of metallo β-lactamase & ampC β-lactamase enzymes was determined by screening and confirmatory tests (combined disc test (CDT) and MRP/MRP+EDTA E-strip test for MBL producing strains & disc antagonism test (DAT) and MIX/MIX+E-strip test for AmpC producing strains). The phenotypic expression was correlated to the presence of selective β-lactamase (*bla*) genes: metallo β-lactamase genes (MBL genes - *blaNDM* and *blaGIM*) and ampicillin class C genes (AmpC genes - *blaPDC* and *blaCMY*) by PCR followed by nucleotide sequencing of the amplicons of different genes under study. Isolates of *P. aeruginosa* (22.22%), 40/180 isolates were preliminary screened positive for MBL production on the basis of resistance to imipenem and/or meropenem and of these, 72.5% (29/40) isolates were further confirmed using combined disc test and MRP/MRP-EDTA E-strip test. However, 60% (108/180) isolates of *P. aeruginosa* were screened as AmpC β-lactamase producers by standard disc diffusion breakpoint for cefoxitin and 48.14% (52/108) were confirmed ampC producers by disc antagonism test and Mix/Mix+E-strip test. A total of 14/180 (7.78%) produced both categories of β-lactamases. Of the phenotypically positive isolates, the *blaNDM* gene was amplified in 37.93% i.e. 11/29 isolates. However, the amplification of *blaGIM* gene was not achieved in any of the isolates tested. The *blaPDC* gene was successfully amplified in 11/52 (21.15%)
isolates. However, the amplification of bla\textsubscript{CMY} gene was not achieved in any of the isolates tested. The amplicons of bla\textsubscript{NDM} and bla\textsubscript{PDC} genes were sequenced for their nucleotides and compared using BLAST\textsubscript{a} analysis with previously reported bacterial strains possessing these genes. The co-occurrence of both genes (bla\textsubscript{NDM} + bla\textsubscript{PDC}) was however, observed in 27.27% (6/22) isolates. Both MBL and AmpC as mechanisms of resistance were thus observed among the isolates of \textit{P. aeruginosa} in the state of Himachal Pradesh. Emergence of such strains is of public health concern as such organisms pose therapeutic challenge.

**KEYWORDS:** \textit{P. aeruginosa}, MDRPA, MBL, ESBL, AmpC, \textbeta-lactamases, bla\textsubscript{NDM}, bla\textsubscript{PDC}, carbapenems.