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Antibiotic Susceptibility Testing, Molecular Profiling, and 16S rRNA-Based Identification of *Pseudomonas aeruginosa* Isolates from Musculoskeletal Infections

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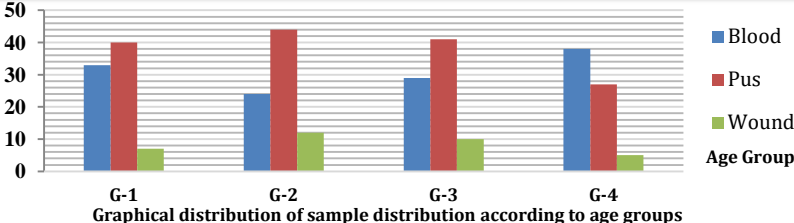
Antibiotic susceptibility testing, molecular profiling, and 16S rRNA-based identification of *Pseudomonas aeruginosa* isolates from musculoskeletal infections

Background

Gram-negative opportunistic pathogen *Pseudomonas aeruginosa* causes musculoskeletal infections in immunocompromised individuals. The primary goal of this research was to describe antibiotic resistance and identification of virulence genes in *Pseudomonas aeruginosa*.

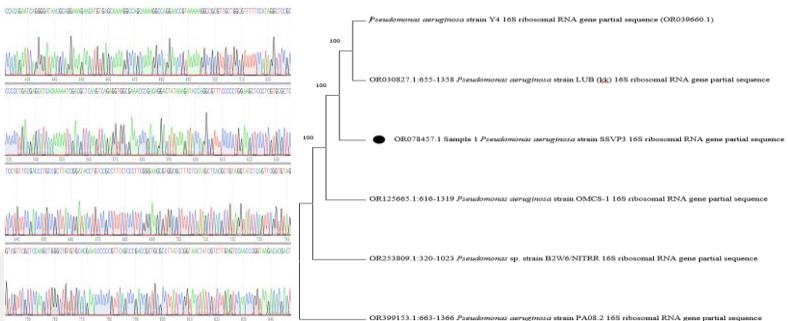
Materials & Methodology

The study was cross-sectional, featuring a randomized selection of samples. Conducted from January to December 2023, the study encompassed a sample size of 320 including blood, pus, and wound swabs [Male:169(52.8%), Female 151(47.1%)]. Age-based categories were ascertained, G1:13-20 years, G2:21-40 years, G3:41-60 years, G4:61-80 years. Identification tests for *Pseudomonas aeruginosa* included API20NE (bioMérieux®, France). Antimicrobial susceptibility testing was following the CLSI,2020 for the Disc Diffusion Test. DNA extraction and purification were carried out through a Genome Jet DNA Purification kit and 16S rRNA primers for the identification of resistance genes *exoA* and *oprL*. Phylogenetic and taxonomic identification was conducted via ribosomal RNA sequencing of PCR-amplified products (BIO-RAD T100TM Thermocycler) and sequencing with an automated sequencer (Illumina MiniSeqTM). Phylogenetic tree was constructed by MEGA11. p-value < 0.05 was statistically significant.



Samples	Quantity	Percentage
Tissue swab	53	19%
Wound swab	35	13.05%
Pus swab	110	41.02%
Blood	70	26.11%

The positive isolates were collected from different specimens.



Molecular characterization of *Pseudomonas aeruginosa*

Results

Prevalence of *Pseudomonas aeruginosa* was 22%. Positivity was recorded from pus (152/320:47.5%), blood (124/320-38.75%) and wound swabs (44/320-13.75%). G4 showed maximum positive isolates (91.25%). Maximum resistance was exhibited against Meropenem (76%) and Imipenem (70%). PCR identified the presence of resistance genes, *exoA* gene (125 bp), and *oprL* (105 bp). The phylogenetic tree was constructed through ribosomal RNA. The gene bank accession number for 16S rRNA gene of *P. aeruginosa* is PQ269824.

Conclusion

Antimicrobial susceptibility testing helps improve the treatment alternatives for stubborn strains. The presence of resistance genes highlights how research at the molecular level enhances treatment approaches for managing multi-drug resistant pathogens.