SUMMARY
AND CONCLUSION
CHAPTER VII

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- A total of 228 enterococcal isolates were collected from clinical samples out of which majority of the isolates were obtained from urine sample.

- *Enterococcus faecium* was the predominant species identified followed by *E. faecalis* and other unusual species such as *E. gallinarum, E. casseliflavus, E. avium, E. durans, E. hirae, E. dispar*. Majority of *E. faecium* was obtained from blood and *E. faecalis* from urine samples.

- PCR identification was highly specific (100%) than the conventional biochemical method especially for the speciation of frequently isolated species viz. *E. faecium* and *E. faecalis*.

- The overall prevalence of HLARE in this study was 17.78% (41/228).

- HLAR phenotype was also observed in unusual species of enterococci such as *E. avium, E. hirae, E. durans* and *E. casseliflavus*.

- Among the 9 aminoglycoside resistance genes tested, *aph(3’)-IIIa* was the most predominant.

- Aminoglycoside acetyl transferase [AAC] which encodes for intrinsic resistance in *E. faecium (aac(6’)-IIi* gene) was detected in 20.17% isolates only. This gene was also detected in 8.7% of *E. faecalis* isolates in our study.
0.02% high level gentamicin resistant isolates and 9.3% high level streptomycin resistant isolates did not carry any of the genes tested. This may be due to other mechanism.

1.75% of VRE were recorded in our study isolates with an MIC of 128-256µg/ml and all were E. faecium isolates with vanA genotype. Importantly, all these isolates were teicoplanin susceptible phenotypes and aminoglycosides resistant.

Macrolide resistance was detected in 53.9% with MIC range of >256µg/ml was observed of which 32.4% isolates carried ermB gene.

The isolates with the presence or absence of gelE and cylA virulence encoding genes showed an alteration in the expression of their respective phenotypic activity. The hyl gene (hyaluronidase) was present only among blood isolates (5.2%) of E. faecium but was not detected in any other source of sample and species.

A maximum of sixteen replicon types were detected from 20 types tested in this study. This includes pheromone responsive plasmids, resistance and virulence genes encoded plasmids, cryptic plasmids and small mobilizable plasmids.

We have reported the plasmid pUSA05 (rep15) in E. faecium and E. avium that were previously reported from community outbreak S. aureus mupirocin resistant strains, which were HLR to all the aminoglycosides and erythromycin screened by MIC.
 Majority of the isolates obtained from urine were found to carry plasmid replicons especially the community associated outbreak plasmids. *E. faecium* isolates carried maximum number of replicon families than that of *E. faecalis*.

- Isolates with virulence encoding genes and high level aminoglycoside resistant phenotypes carried the conjugative replicon families.

- To conclude, this study analysed the distribution of resistance and virulence encoding genes and their mechanism among different *Enterococcus* species. All three types of high level aminoglycoside resistance mechanisms were detected. To the best of our knowledge this is the first study to report the presence of *aac(6’)-Ii* gene in *E. faecalis* isolates from India. This study was first to detect the presence of aminoglycoside resistance encoding genes and macrolide resistant *ermB* genes in enterococci from Indian settings. All the virulence factors were distributed among both low level and high level aminoglycoside resistant enterococcal isolates. The distribution of plasmid replicon families in enterococci along with their drug resistant determinants were well correlated upto species level. This is the first study to document the presence of replicon types in *Enterococcus* isolates from India.