Liaqat Ali et al., J Vaccines & Vaccin 2015, 6:6
http://dx.doi.org/10.4172/2157-7560.C1.036

OMICS International
conferenceseries.com

9th Global Summit and Expo on
Vaccines & Vaccination
November 30-December 02, 2015 San Francisco, USA

Characterization of the putative polysaccharide gene cluster in multidrug-resistant *E. faecium* to develop a targeted therapy (vaccine)

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Enterococcus faecium is an emerging multidrug-resistant nosocomial pathogen increased dramatically worldwide and causing bacteremia, endocarditis, urinary tract and surgical site infections in immunocomprised patients. The capsular polysaccharides that contribute to pathogenesis through evasion of the host innate immune system are also involved in hindering leukocyte killing of enterococci. We identified two putative loci with 22 kbp and 19 kbp which contained 11 genes encoding for glycosyltransferases (GTFs); this was confirmed by using genome comparison of already sequenced strains that has no homology to known capsule genes and the EPA (enterococci polysaccharide antigen) locus. The polysaccharide-conjugate vaccines have rapidly emerged as a suitable strategy to combat different pathogenic bacteria, therefore, we investigated a capsular polysaccharide biosynthesis CapD protein in *E. faecium* contains 336 amino acids and had putative function for N-linked glycosylation. The deletion/knock-out *cap*D mutant was constructed and complemented by homologues recombination method and confirmed by using PCR and sequencing. For further characterization and functional analysis, *in-vitro* cell culture and *in-vivo* a mouse infection models were used. Our *ΔcapD* mutant shows a strong hydrophobicity and all strains exhibited biofilm production. Subsequently, the opsonic activity was tested in an opsonophagocytic assay which shows increased in mutant compared to complemented and wild-type strains but more than two fold decreased in colonization and adherence was seen on surface of uroepithelial cells. However, a significant higher bacterial colonialization was observed in *cap*D mutant during animal bacteremia infection. Further experiments will be needed to characterize the putative mega capsular polysaccharide gene cluster to clarify its function, exact mechanism and involvement in pathogenesis of enteroccocal nosocomial infections, eventually to develop a vaccine/ or targeted therapy.

Biography

Liaqat Ali is a PhD student (final year) at University of Freiburg, Germany and undergoing research on human pathogens, (enteroccocal infections, vaccine developement). He has published more then 15 research articles; 5 as a principal author and rest with co-author in reputed peer reviewed journals.

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