8. Summary

The social networking among prokaryotes helps them to acclimatize in various environmental stresses by undergoing various phenotypic changes like biofilm, secretion of virulence factors, sporulation, bioluminescence, motility, antibiotic resistance, and many more. The prokaryotes possess specific languages (QSSMs) for talking (QS) to the same (*intraspecies*) and different species (*interspecies*) of the group and even with species of another kingdom (*interkingdom*). However, the communication through QS among prokaryotes has been proved harmful to higher eukaryotes like humans, plants, and animals. Among all the phenotypic changes acquire by prokaryotes, biofilm formation is considered to be most vital and harmful to the eukaryotes. Therefore, there is a need to understand the phenomenon of *cell-to-cell* communication and develop various approaches to impede it by designing novel strategies.

QSPs are characterized as the signaling molecule of Gram-positive bacteria. Only one database was available in the literature, which encompasses 231 signaling peptides reported among bacteria. We explored the QSPs, to fetch the unique characteristics by analyzing them, followed by the development of prediction software for identification of the QS potential of unknown peptide(s) and designing novel QSPs (***QSPpred***) (Rajput et al., 2015). The QSPs were analyzed using amino acid composition, amino acid position, QS motifs, and physicochemical properties. However, the models were developed using hybrid of various features like AAC, DPC, N5Bin, C5Bin, and Physico showed accuracy and MCC of 93.00% and 0.86 respectively during 10-fold cross validation using SVM (Rajput and Kumar, 2014). All models constructed using various MLTs like SVM, RF, and *k*-NN are integrated into QSPpred web server. Moreover, this web server incorporates various analyses tools for scanning motif from the unknown sequence(s) (*QSMotifscan*), fragment the protein (*ProtFrag*), generation of mutation in the peptide (*MutGen*), and visualization of important physicochemical properties of the peptide (*PhysicoProp*). The web server is freely available at http://crdd.osdd.net/servers/qsppred/.

Prokaryotes harbors diverse signaling systems namely AHLs, DKPs, DSFs, AI-2, AI-3, and many more. The platform that encompasses complete information of these signaling molecules was lacking. Therefore, we developed a platform named ***SigMol***, which comprehend the information of all major signaling systems of the prokaryotic world (Rajput et al., 2016)). The SigMol contains 1382 signaling molecules from 215 organisms.

Moreover, it contains biological, chemical, and structural information of each molecule. Furthermore, this database is hyperlinked to the external resources like NCBI-taxonomy, PubChem, ChemSpider, PubMed, etc. to make it more informative. Additionally, the standard facilities like browse, search, compare and draw the QSSMs are also integrated into the database. The multilevel communication e.g. intraspecies, interspecies and interkingdom is summarized in form of heatmaps provided on the web server. The SigMol repository can be accessed freely at http://bioinfo.imtech.res.in/manojk/sigmol/.

The phenomenon of multilevel communication through QSSMs is less explored among prokaryotes (Gram positive bacteria, Archaea, and viruses). We performed stepwise bioinformatics survey to unveil the intraspecies, interspecies, and interkingdom communication. The key regulators of QS among Gram-negative bacteria i.e. LuxI and LuxR were explored through similarity, functional characterization, and evolutionary trend in Gram-positive bacteria, Archaea, and viruses. The similarity analyses include AAC, MSA, Motif, and Domain for the comparison with Gram-negative bacteria. The LuxI and LuxR proteins of all the three groups were functionally characterized through GO annotation and ligand binding affinity. Lastly, the evolution of LuxI and LuxR among all the three groups was examined along with their respective BLAST hits, to check the trend for the transfer of QS regulators. Interestingly, all the analyses revealed that QS regulators i.e. LuxI and LuxR among all the three groups were similar and evolved from Gram-negative bacteria and thus possess the potential to involve in multilevel communication (Rajput and Kumar, 2016b, 2017a, b).

The interaction among prokaryotes and with eukaryotes proved harmful to humans. Therefore, various QQ agents especially against biofilm are being designed to impede their communication. We have developed an ***aBiofilm*** (http://bioinfo.imtech.res.in/manojk/abiofilm/), a complete resource for anti-biofilm agents. The resource harbors database, predictor, and data visualization (analyses) modules. The database incorporates 4066 anti-biofilm agents (1400 unique) from 405 articles, and targeting 130 unique organisms. Moreover, the anti-biofilm agents are of varied types like chemicals, phytochemicals, antibodies, enzymes, and many more. Additionally, the repository encompasses biological, chemical, and structural details of each anti-biofilm agent. However, we have developed QSAR based prediction algorithm to identify and design anti-biofilm agents especially chemicals. It has incorporated the

model developed through SVM from the chemical descriptors with accuracy ~80%. Furthermore, the aBiofilm resource contains data visualization module to summarize the overall data for highlighting the relevant information. Moreover, to explore the peptides with anti-biofilm potential, we developed a peptide sequence based prediction algorithm i.e. ***ABPpred*** (http://bioinfo.imtech.res.in/manojk/abppred/) using multiclass approach with an accuracy of 75.00%. This predictor exhibits the capability to categorize the unknown peptide with anti-biofilm potential in 4-classes *viz.* >90%, 50-90%, <50%, and Not active (Rajput and Kumar, 2016a).

Future perspectives

Due to the extensive research undergoing in the field of *cell-to-cell* signaling, various QS and QQ agents have being discovered continuously. Therefore, our database of QS and QQ molecules named *SigMol* and *aBiofilm* should be continuously updated. Consequently, the prediction algorithm like *QSPpred*, *ABPpred*, *aBiofilm predictor* will be redesigned utilizing the updated data and new features. Moreover, we already explored the multilevel communication through LuxI and LuxR regulators, further the cross-communication among microbial world will be explored through interspecies signaling regulator named LuxS. The anti-QS field incorporates various other applications that are quenched through QQ agents like anti-virulence, anti-motility, anti-bioluminescence, and others. Furthermore, the anti-biofilm predictors would be updated from generalized to species specific, to target the specific organisms. The developed tool for targeting the biofilm would be helpful for the researchers to design various anti-biofilm agents, for tackling the problem of antibiotic drug resistance.