Project 34 : Exploring ribosomal RNA detection for species level identification of bacterial pathogens in a rapid, low cost bioassay format

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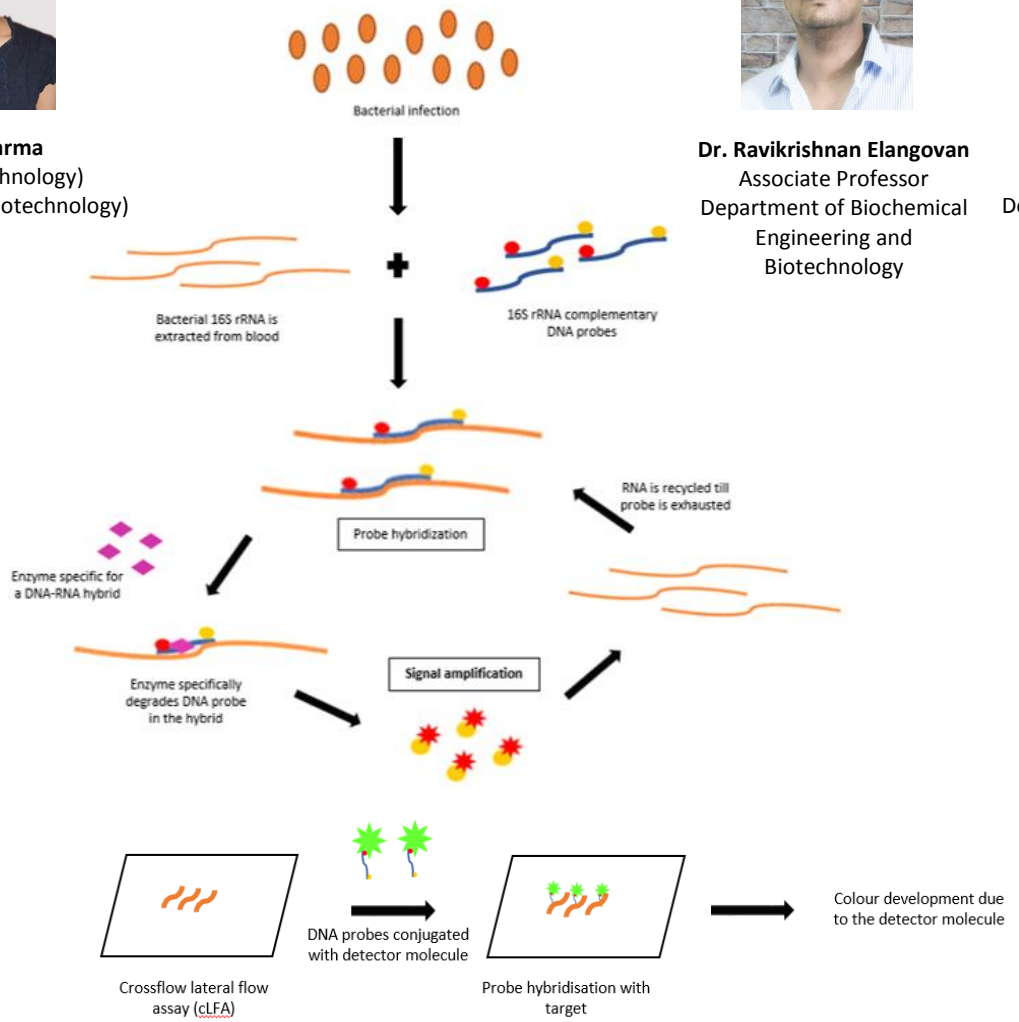

The current gold standard for bacterial diagnosis in a typical biofluid (blood, urine etc.) is culture growth that takes anywhere from 48-72 h or longer to give results. Because of this delay, more often than not, it leads to death in critical conditions such as septicemia. Our hypotheses for this project: (a) the naturally amplified system of house-keeping rRNAs, that remain well conserved across a particular species, can be used for amplification-free & reverse transcription-free, identification of bacterial species in blood/plasma, (b) the rapid detection format along with rRNA-inhibitors can allow rRNA detection in under 30 min without loss in their molecular stability, and (c) a cocktail of anti-sense probes attached to reporter tags (*e.g.* fluorophores, HRPs, nanoparticles *etc.*) can be used to detect a broad range of bacterial types implicated in septicemia.

Based on the above, we would like to develop a point-of-care (POC) assay with a bedside stratagem. It would involve extracting bacterial 16S rRNA from blood and using complementary DNA probes conjugated with gold nanoparticles to detect the RNA. An enzyme which specifically degrades the DNA component in a DNA-RNA duplex would give signal amplification. We would like to replicate our findings from a solution-assay to design an all-exclusive, easy-to-use lateral flow assay (LFA) which will overcome the limitations of detection sensitivity in present assays. If successful, this will be the first POC test of its kind in the world to give species-level information under 30 minutes.