Synopsis Seminar

Seminar Title	: Exploring marine Streptomyces for the development of biopolymer based biomaterials and biofilm mediated degradation of polycyclic aromatic hydrocarbon
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Venue	: Mathematics Seminar Room
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Abstract :

This thesis illustrates the metabolic potential of biofilm-forming marine *Streptomyces* for the biosynthesis of polyhydroxybutyrate (PHB) and extracellular polymeric substances (EPS), while also highlighting their capacity for phenanthrene degradation, showcasing their multifaceted role in sustainable bioremediation and biopolymer production. In this study, sediment samples were collected from four sites of the coastal region of Odisha, India. A total of 24 morphologically distinct*Streptomyces* spp. were isolated, of which 10 isolates showed biofilm-forming and PHB synthesizing potential. The morphological, biochemical, and cultural characterization of the 10 potential isolates were conducted based on the International Streptomyces Project (ISP) protocol. The molecular identification using 16s rRNA and establishment of evolutionary relationship by construction of phylogenetic tree revealed these bacteria as *Streptomyces missionensis* DHS1,*Streptomyces* sp. DHS2, *Streptomyces althioticus* DNS1, *Streptomyces fradiae* DNS4, *Streptomyces nigra* KDS4, *Streptomyces violascens* GPS1, *Streptomyces phaeolivaceus* GPS2, *Streptomyces albidoflavus* GPS3, *Streptomyces drozdowiczii* PPS2, *Streptomyces thermolineatus* PPS3. The quantification of biofilm formation and PHB synthesis revealed that 6 out of these 10 isolates were strong biofilm former and high PHB yielding strains. These strains were further screened for their polycyclic aromatic hydrocarbon (PAH) tolerating potential. Among all these strains *Streptomyces nigra* KDS4 have shown strong biofilm-EPS formation with high PHB yielding (419.3 mg/g of dried biomass) potential. This bacterium bacterium have also shown highest growth and EPS biosynthesis (upto461.33±21.56 mg/L) in presence of different PAHs (naphthalene, phenanthrenen, and pyrene). These findings highlights the integrated capacity of marine *Streptomyces*, particularly *S. nigra* KDS4, for PHB biosynthesis, biofilm-EPS formation, and PAH tolearnce. Further the biofilm-EPS formation in this bacterium was investigated. Confocal laser scanning microscopy (CLSM) and COMSTAT analysis revealed peak biofilm formation at 60 hours of incubation, with subsequent dissociation by 84 hours. The amplification confirmed the presence of *cslA*, encoding cellulose synthase-like proteins involved in EPS and biofilm matrix development. The structural analysis of the EPS using FTIR, ^{1}H NMR, and XRD indicated a heterogeneous biopolymer enriched with polysaccharides, proteins, lipids, and uronic acids. SEM and AFM imaging highlighted a robust fibrillar EPS network with nanoscale roughness aiding surface adhesion. The bacterium form dense biofilm over the polypropylene surface as compared to glass and polystyrene. Thermal stability was demonstrated by differential scanning calorimetry (DSC), with an endothermic peak near 130°C and significant decomposition above 300°C. The EPS exhibited strong antioxidant activity and notable emulsifying capacity, supporting its functional relevance. Furthermore, EPS-based hydrogels displayed excellent porosity and mechanical integrity, emphasizing their suitability for biomedical applications. These findings highlight *S. nigra* KDS4 as a promising candidate for biopolymer production and valorization in environmental and biomedical domains. Further, the PHB synthesizing potential of the bacterium was exploited by optimizing the process parameter and reactor scale production. The optimization of process parameters revealed that slightly alkaline pH, moderate salinity, and a C:N ratio of 20:1 (w/w) using starch and KNO_3 as carbon and nitrogen sources, respectively, significantly enhanced PHB production (2.63 g/L). The PHB synthesis was found to be growth-associated and scalable in bioreactor conditions. Comprehensive characterization using FESEM-EDX, ATR-FTIR, XRD, ^{1}H-NMR, and GC-MS confirmed the identity, purity, and chemical composition of the extracted polymer, which matched the characteristics of standard PHB. The polymer exhibited favorable mechanical properties, including tensile strength and elongation at break, and demonstrated efficient biodegradability in soil. Additionally, cytocompatibility assays confirmed its non-toxic and biocompatible nature, suggesting its suitability for biomedical applications. This is the first report on PHB production from*S. nigra* KDS4, highlighting its potential for sustainable biopolymer production and various industrial applications, including biodegradable packaging, agriculture, and biomedicine. Based on these findings, the coproduction of extracellular polymeric substances (EPS) and polyhydroxybutyrate (PHB) by marine bacterium *S. nigra* KDS4 under phenanthrene stress was investigated, aiming to establish its potential for integrated biopolymer synthesis and bioremediation. phenanthrene-responsive behavior of marine *Streptomyces nigra* KDS4, emphasizing its ability to co-produce extracellular polymeric substances (EPS) and polyhydroxybutyrate (PHB) while facilitating phenanthrene degradation in a biofilm mode. Under phenanthrene stress (100&ndash500 ppm), the bacterium exhibited enhanced biomass and EPS production, with peak EPS yield and structural integrity observed at 500 ppm, as confirmed by CLSM, SEM, and COMSTAT analysis. While PHB biosynthesis was suppressed under stress, EPS composition shifted, displaying increased carbohydrate and protein content. Spectroscopic and microscopic analyses (ATR-FTIR, ^{1}H-NMR, XRD, FESEM-EDX, CD, and 3D EEM) revealed structural and functional remodeling of EPS, including increased crystallinity, aromatic interactions, and altered secondary protein structure. Fluorescence quenching and binding assays demonstrated strong, spontaneous interaction between phenanthrene and EPS fluorophores, confirming a static quenching mechanism. Notably, *S. nigra* KDS4 biofilm achieved 72.56% phenanthrene degradation within 8 days, indicating significant bioremediation potential. These findings collectively demonstrate the

remarkable adaptability of *Streptomyces nigra* KDS4 in modulating EPS and PHB production under phenanthrene stress, supporting its dual functionality in biopolymer synthesis and environmental detoxification. Its robust biofilm architecture, enhanced EPS secretion, and efficient phenanthrene degradation highlight its promise as a sustainable bioresource for integrated bioremediation and industrial biopolymer applications.production and environmental remediation.