Microbial catabolism: from drug targets to biocatalysis

Research Interests

Microbial catabolic activities underpin life on our planet. On the global scale, these enzymes and pathways are essential to the carbon cycle of the biosphere. They also maintain the health of all ecosystems ranging from ocean and forest environments to the different parts of the human body. Microorganisms have evolved an astoundingly versatile armamentarium of enzymes to degrade the vast array of compounds that occur in the biosphere. As a rich source of biocatalysts, microbial catabolic activities are of burgeoning importance for the sustainability of the agricultural, food, textile, pharmaceutical and chemical industries. In pathogens, catabolic activities that play essential roles in the infection process provide potential targets for novel therapeutics, which are urgently needed in this era of increasing antibiotic resistance. The enormous diversity and importance of microbial catabolic activities are being revealed through high-throughput sequencing and related projects. However, our understanding of the molecular basis of these activities lags far behind as reflected by the huge number of poorly-annotated genes in microbial databases. Linking sequence to function is essential for understanding microbial catabolic activities and for exploiting their ability to transform substrates into less toxic compounds or economically useful products that are otherwise difficult to synthesize.

One group of bacteria that catabolises a particularly broad range of organic compounds is the mycolic acid-containing Actinobacteria, including *Mycobacterium* and *Rhodococcus*. Accordingly, these bacteria have considerable biotechnological potential. Indeed, the most commercially successful microbial transformation involves the use of a *Rhodococcus* to produce acrylamide.
These bacteria also include *Mycobacterium tuberculosis* (*Mtb*), responsible for ~1.3 million deaths annually, making it the leading cause of mortality from bacterial infections. Our studies have provided pioneering insights into the catabolism of aromatic compounds and steroids by Actinobacteria, including the organization of these catabolic pathways, the discovery of a cholesterol catabolic pathway in *Mtb*, and the first characterization of a bacterial lignin-degrading enzyme. These findings have important implications for applications ranging from biocatalysts to novel therapeutics.

**Objectives:** The overarching theme of our research is the bacterial catabolism of two classes of organic compounds: lignins and steroids. The fundamental objectives are to understand the logic of catabolic pathways involved and the mechanisms of key catabolic enzymes. The practical objectives include developing novel therapeutics and biocatalysts. The research on steroid catabolism is aimed at elucidating the steps of cholesterol catabolism in *Mtb* and determining its role in virulence, including the influence of steroid metabolites on the host. In elucidating catabolic steps, we are investigating the catalytic mechanism of steroid-ring cleaving enzymes. This research runs in parallel to a collaborative program aimed at characterizing inhibitors of cholesterol catabolism as potential therapeutics. The research on lignin catabolism is aimed at characterizing enzymes involved in lignin depolymerization and catabolism, and developing *Rhodococcus* as a platform for engineering lignin-degrading biocatalysts. Such biocatalysts will be used to improve the carbon efficiency and economics of production of lignocellulose-derived products. The objectives are being pursued using multidisciplinary approaches drawn from biochemistry, molecular genetics, microbiology, chemical biology, structural biology and genomics.

**Significance:** The proposed research provides major new insights into the bacterial degradation of steroids and lignin, two important classes of organic compounds in the biosphere. This includes elucidating the specific functions of genes, their products and entire pathways as well as determining mechanisms of regulation. Our research also helps elucidate the role of cholesterol catabolism in *Mtb* pathogenesis and the catalytic mechanisms of important families of enzymes. The targeted enzymes are important in disease or to the global carbon cycle, particularly in forest ecosystems. Related enzymes are implicated in many other processes. On a more general level, the research provides insights into the function of metals in biological systems, O2-activation, C-C bond cleavage, and the physiology of Actinobacteria. On a practical level, the research contributes to the development of novel therapeutic strategies to combat TB as well as the engineering of enzymes and bacteria for green chemistry applications. Woody biomass has considerable potential as a sustainable alternative to petroleum as a feedstock for high-value products, including resins, carbon fibres and commodity chemicals. Effective lignin-transforming biocatalysts will help develop this potential, reduce dependence on petroleum products, and contribute to revitalizing the forestry industry.