

PREFACE

Although a few groups of bacteria have long been known to communicate via diffusible chemical signals, we are only now learning that this process is enormously widespread. We are now in a position to begin to appreciate the importance of cell-cell communication in areas as fundamental as bacterial physiology, ecology, evolution, and pathogenesis. Approximately one decade ago, ASM Press published the first comprehensive review of the topic of bacterial cell-cell communication, *Cell-Cell Signaling in Bacteria*, with chapters contributed by leaders in the then-nascent field. We hope readers of this new volume will agree that an enormous amount of information on major aspects of signaling has surfaced since that first book was published and that a fresh view of the topic is now appropriate and important for a diverse audience of researchers, educators, and clinicians.

The past decade has witnessed new insights about the chemical composition, synthesis, and turnover of a variety of bacterial signal molecules. First, the enzymes that synthesize signal molecules are far better understood than they were 10 years ago. At the close of the 20th century, no signal synthase had been studied at the structural level. Currently, the structures of three bacterial signal synthases have been solved, two of which produce AHLs (chapter 16) and one of which synthesizes AI-2 (chapter 19). In other developments, the *Streptomyces coelicolor* 15-residue SapB peptide, required for aerial fruiting body formation, is now known to be synthesized by a nonribosomal peptide synthase (chapter 6). We recently learned that at least one class of extremely hydrophobic signal travels as a component of vesicles derived from the cell outer membrane (chapter 20). This signal, designated PQS (*Pseudomonas* quorum signal) also has antimicrobial properties against gram-positive bacteria. Many new types of signal molecules with a variety of novel structures are under study, including polyamines, rhamnolipids, and metabolites such as indole and amino acids (chapters 3, 7, and 17). During the past decade, a variety of enzymes capable of degrading bacterial communication signals have been described, as well as natural and synthetic small molecules that agonize or antagonize signaling (chapters 10, 24, and 25). Future studies may help us understand whether the

substrates for which these enzymes were selected are signaling molecules or whether the destruction of signal molecules is incidental to the activity for which they were selected.

There has been an explosion of new information on signal receptors and mechanisms of signal transduction. Where 10 years ago there was no structural information about quorum sensing receptors, there now exists structural information for seven of these receptors (chapters 21, 13, 19, and 23) (2, 7). It is striking that at least five of these receptors (PrgX, CprB, TraR, LasR, and SdiA) fully or partially engulf their respective ligands, which contribute to the hydrophobic cores of these proteins. In the case of the cytoplasmic TraR, LasR, and SdiA receptors, ligand binding is required for protein folding and resistance to proteolysis, while PrgX and CprB function as apo-proteins, so their folding must occur in the absence of ligand. Quorum sensing structural studies have provided other surprises. For example, it was found that AI-2 bound to LuxP includes a boron atom and, perhaps equally surprising, that AI-2 bound to the homologous Lsr receptor lacks boron (chapter 19). Also surprising is that the LuxPQ structures provide a new mechanism for two-component signal transduction across the bacterial membrane that differs dramatically from that proposed for signal relay in chemotaxis systems. A cocrystal containing TraR and its antiactivator TraM provides insight into the mechanism for how TraM allosterically prevents TraR from binding DNA (3). Thus, studies of the molecular biology of cell-cell signaling are providing unexpected insights into other areas of molecular biology.

New discoveries about signal transduction pathways and the expression of target genes have also been made. For example, a decade ago we could not have guessed that at the heart of the *Vibrio harveyi* and *Vibrio cholerae* quorum sensing cascades would lie several redundant small RNAs. We could not have predicted that two autoinducers and two AI synthases in *Vibrio fischeri* would influence the activity of LuxR. Large sets of new target genes have been identified using global high throughput techniques such as proteomics and DNA microarrays (6) (chapter 8).

The repertoire of phenotypes affected by cell-cell communication has grown considerably. For example, it has long been appreciated that oligopeptides stimulate sporulation and competence for transformation in *Bacillus subtilis*, but only recently has it been reported that peptides also stimulate the conjugation of the integrative and conjugative element ICEBs1 (chapter 2). Expression of these genes is also induced by DNA damage, similar to the corresponding genes of the STX element of *V. cholerae* (1). A decade ago it was clear that communication is required for biofilm formation in *Pseudomonas aeruginosa* (4), but recently it has been discovered that communication has the opposite effect on biofilms in *V. cholerae* (chapter 9). The surprising finding that increases in population densities activate this trait in *P. aeruginosa* and inhibit the same trait in *V. cholerae* most likely defines the persistent versus acute diseases, respectively, caused by these pathogens.

Studies of *P. aeruginosa* have been especially fast-paced in this past decade. It was already known that this organism has two AHL signals, as well as a quinolone signal called PQS, and that there were two AHL synthases and two AHL receptors. We now know that a third AHL receptor exists that detects one of the known AHLs. The *P. aeruginosa* LasR protein has been extensively stud-

ied *in vitro* and binds both to canonical *las* box binding sites as well as to completely different sites (chapter 8). Binding to some sites is cooperative, while at other sites, the protein binds noncooperatively. Microarrays and random fusions have shown that hundreds of genes are controlled by one or more of these systems, including many genes that encode exported proteins. The crystal structure of the LasR N-terminal domain, complexed with the cognate AHL, was recently determined (2) and revealed interesting structural similarities to TraR of *A. tumefaciens* (chapter 13).

What discoveries might we anticipate in the coming decade? We will look for the elucidation of new classes of signaling molecules, such as the one described late in 2007 (5). We expect that the next 10 years will also witness advances in structural and biochemical studies of signal synthases and receptors, including structural determination of peptide signals complexed with their receptors. There will likely be surprises about the mechanisms of diffusion of different signals, perhaps aided by nanofabrication technologies. Genetic approaches and transcriptional profiling will likely lead to new understanding of network design principles that provide noise reduction, signal integration, and signal amplification. We will learn how bacterial regulatory circuits are wired to provide ordered temporal and spatial expression of large sets of target genes. We also will expect further collaborations between biologists and molecular modelers. We can be reasonably confident about progress in all of these areas. On the other hand, the most exciting discoveries will generally be the ones that no one can even begin to anticipate. To paraphrase a former U.S. Secretary of Defense, it is hard to predict the unknown, but much harder still to predict the unknown unknowns. If the rate of progress of the past decade is to continue at the present pace for another 10 years, we will be able to glimpse a body of knowledge about which we could not even have dreamt when the first ASM volume on this topic went to press just 10 years ago.

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