1 Introduction

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Microbes produce an extraordinary array of microbial defense systems. These include broad-spectrum classical antibiotics, metabolic byproducts, such as the lactic acids produced by lactobacilli, lytic agents such as lysozymes, numerous types of protein exotoxins, and bacteriocins, which are loosely defined as biologically active protein moieties with a bacteriocidal mode of action. This biological arsenal is striking not only in its diversity, but also in its natural abundance. Bacteriocins are found in almost every bacterial species examined to date, and within a species tens or even hundreds of different kinds of bacteriocins are present. Halobacteria universally produce their own version of bacteriocins, the halocins. Streptomycetes are characterized by broad-spectrum antibiotics. This diversity and abundance of antimicrobial weapons clearly suggest an important role for these potent antimicrobials. Less clear is how such diversity arose and what roles these biological weapons serve in microbial communities. One large family of antimicrobials, the protein-based bacteriocins, has served as a model for numerous, detailed explorations regarding their ecological roles and evolutionary histories. Bacteriocins differ from broad-spectrum, classical antibiotics in one critical way – they have a relatively narrow killing spectrum and are toxic only to bacteria closely related to the producing strain. These toxins have been found in all major lineages of Bacteria, and more recently, have been described as universally produced by some members of the Archaea. According to Klaenhammer, 99% of all bacteriocins may make at least one bacteriocin, and the only reason we have not isolated more is that few researchers have looked for them.

The bacteriocin family includes a diversity of proteins in terms of size, microbial targets, modes of action, and immunity mechanisms. The most extensively studied, the colicins produced by *Escherichia coli*, share certain key characteristics. Colicin gene clusters are encoded on plasmids and are composed of a colicin gene, which encodes the toxin; an immunity gene, which encodes a protein conferring specific immunity to the producer cell by binding to and inactivating the toxin protein; and a lysis gene, which encodes

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a protein involved in colicin release through lysis of the producer cell. Colicin production is mediated by the SOS regulon and is therefore principally produced under times of stress. Toxin production is lethal for the producing cell and any neighboring cells recognized by that colicin. A receptor domain in the colicin protein that binds a specific cell surface receptor determines target recognition. This mode of targeting results in the relatively narrow phylogenetic killing range often cited for bacteriocins. The killing functions range from pore formation in the cell membrane to nuclease activity against DNA, rRNA, and tRNA targets. Colicins, indeed all bacteriocins produced by Gram-negative bacteria, are large proteins. Pore-forming colicins range in size from 449 to 629 amino acids. Nuclease bacteriocins have an even broader size range, from 178 to 777 amino acids.

Although colicins are representative of Gram-negative bacteriocins, there are intriguing differences found within this subgroup of the bacteriocin family. *E. coli* encodes its colicins exclusively on plasmid replicons. The nuclease pyocins of *Pseudomonas aeruginosa*, which show sequence similarity to colicins, and other, as yet uncharacterized, bacteriocins are found exclusively on the chromosome. Other close relatives to the colicin family, the bacteriocins of *Serratia marcesens*, are found on both plasmids and chromosomes. In this volume, Chapter 2 further explores this fascinating abundance and diversity of bacteriocin proteins produced by Gram-negative bacteria, while Chapter 3 focuses on signatures of their evolutionary history contained within their DNA sequences.

Bacteriocins of Gram-positive bacteria are as abundant and even more diverse as those found in Gram-negative bacteria. They differ from Gram-negative bacteriocins in two fundamental ways. First, bacteriocin production is not necessarily the lethal event it is for Gram-negative bacteria. This critical difference is due to the transport mechanisms Gram-positive bacteria encode to release bacteriocin toxin. Some have evolved a bacteriocin-specific transport system, whereas others employ the *sec*-dependent export pathway. Second, Gram-positive bacteria have evolved bacteriocin-specific regulation, whereas bacteriocins of Gram-negative bacteria rely solely on host regulatory networks. The conventional wisdom about the killing range of Gram-positive bacteriocins is that they are restricted to killing other Gram-positive bacteria. The range of killing can vary significantly, from relatively narrow as in the case of lactococcins A, B, and M, which have been found to kill only *Lactococcus*, to extraordinarily broad. For instance, some types of lantibiotics, such as nisin and mutacin B-Ny266, have been shown to kill a wide range of organisms including *Actinomyces*, *Bacillus*, *Clostridium*, *Corynebacterium*, *Enterococcus*, *Gardnerella*, *Lactococcus*, *Listeria*, *Micrococcus*, *Mycobacterium*, *Propionibacterium*, *Streptococcus*, and *Staphylococcus*. Contrary to conventional wisdom, these particular bacteriocins are active also against a number of medically important Gram-negative bacteria including *Campylobacter*, *Haemophilus*, *Helicobacter*, and *Neisseria*. Chapter 4 provides a review of the diversity of Gram-positive bacteriocins.

The Archaea have their own distinct family of bacteriocin-like antimicrobials, known as archaeocins. The only characterized member is the halocin family produced by halobacteria, and few halocins have been described in detail. Archaeocins are produced as the cells enter stationary phase. When resources are limited, producing cells lyse sensitive cells and enrich the nutrient content of the local environment. As stable proteins, they may remain in the environment long enough to reduce competition during subsequent phases of nutrient flux. The stability of halocins may help explain why there is so little species diversity in the hypersaline environments frequented by halobacteria. Chapter 5 deals with peptide and protein antibiotics in the domain Archaea, focusing on halocins and sulfolobicins.

As is clear from this brief survey of bacteriocin diversity and distribution, this heterogeneous family of toxins is united only by the shared features of being protein-based toxins that are relatively narrow in killing spectrum, and often extremely hardy and stable. What makes these the weapons of choice in the microbial world remains an intriguing question. Chapters 6 and 7 provide compelling suggestions regarding the ecological role served by bacteriocins in microbial communities. As will be clear from these two reviews, we have only just begun to understand the fundamental roles these potent toxins serve.

Bacteriocins are now being explored for their potential utility in human and animal health applications, and agricultural uses. Before we will succeed in harnessing the vast power of these toxins to serve in human-mediated functions, we require a more complete understanding of how these proteins have evolved and are shared between bacterial lineages, and what roles they serve in natural microbial communities. Their application to meet numerous human challenges is limited only by our imagination and creativity. The microbial world has invested several billion years in selecting and refining the novel functions afforded by this heterogeneous class of proteins. Let's take advantage of this extraordinary evolutionary experiment!