Drug Resistance Updates 26 (2016) 43–57

Contents lists available at ScienceDirect

Drug Resistance Updates

journal homepage: www.elsevier.com/locate/drup

Invited review

Mechanisms and consequences of bacterial resistance to antimicrobial peptides

D.I. Andersson*, D. Hughes, J.Z. Kubicek-Sutherland

Uppsala University, Department of Medical Biochemistry and Microbiology, Box 582, SE-751 23 Uppsala, Sweden

ARTICLE INFO

Article history:
Received 14 March 2016
Received in revised form 7 April 2016
Accepted 11 April 2016

Keywords:
Antimicrobial peptides
Anti-bacterial drugs
Resistance
Innate immunity
Selection
Bacterial infections

ABSTRACT

Cationic antimicrobial peptides (AMPs) are an intrinsic part of the human innate immune system. Over 100 different human AMPs are known to exhibit broad-spectrum antibacterial activity. Because of the increased frequency of resistance to conventional antibiotics there is an interest in developing AMPs as an alternative antibacterial therapy. Several cationic peptides that are derivatives of AMPs from the human innate immune system are currently in clinical development. There are also ongoing clinical studies aimed at modulating the expression of AMPs to boost the human innate immune response. In this review we discuss the potential problems associated with these therapeutic approaches. There is considerable experimental data describing mechanisms by which bacteria can develop resistance to AMPs. As for any type of drug resistance, the rate by which AMP resistance would emerge and spread in a population of bacteria in a natural setting will be determined by a complex interplay of several different factors, including the mutation supply rate, the fitness of the resistant mutant at different AMP concentrations, and the strength of the selective pressure. Several studies have already shown that AMP-resistant bacterial mutants display broad cross-resistance to a variety of AMPs with different structures and modes of action. Therefore, routine clinical administration of AMPs to treat bacterial infections may select for resistant bacterial pathogens capable of better evading the innate immune system. The ramifications of therapeutic levels of exposure on the development of AMP resistance and bacterial pathogenesis are not yet understood. This is something that needs to be carefully studied and monitored if AMPs are used in clinical settings.

Contents

* Corresponding author. Tel.: +46 18 4714175.
E-mail address: Dan.Andersson@imbim.uu.se (D.I. Andersson).

http://dx.doi.org/10.1016/j.drup.2016.04.002

1. Introduction to AMPs

1.1. Definition of AMPs and early history

Cationic antimicrobial peptides (AMPs) are relatively small peptides that, by definition, have a net positive charge, and exhibit some, often broad-spectrum, antimicrobial activity. This rather tautological definition encompasses a great diversity of molecules, at both the sequence and the structural level. An online antimicrobial peptide database, APD3 (Wang et al., 2016), accessible at http://aps.unmc.edu/AP/, lists over 2600 examples of AMPs from all kingdoms of life: bacteria, archaea, and eukaryotes (including plants, animals, fungi and protists). AMPs can act directly to protect animals against a wide variety of bacterial, viral, fungal and protozoan infections and because of this are often referred to as host defense peptides. In animals, both vertebrates and invertebrates, AMPs also function as a universal and important part of the innate immune system, modulating activities that promote protection against infections by microorganisms. The APD3 database currently lists 112 different human host defense peptides of which 100 have experimentally determined antibacterial activities. The focus of this review is on the importance of human AMPs in protecting against bacterial infections, on the mechanisms of intrinsic and acquired bacterial resistance to AMPs, and on the opportunities and potential consequences associated with therapeutic use of AMPs.

AMPs are not unique as peptide-based small molecules with antibacterial activity. Many of the antibiotics produced by microorganisms are peptide-based molecules. These are produced by non-ribosomal peptide synthesis (NRPS), a process that involves the expression of large arrays of genes encoding multiple enzymes that work in succession to catalyze the sequence of chemical reactions required to synthetize the antibiotic (Baltz, 2006; Hughes, 2003). Peptide-based antibiotics include β-lactams such as penicillin, cyclic peptide antibiotics like the polymixins and bacitracin, glycopeptides like vancomycin (Yim et al., 2014), and the lipopeptide daptomycin, one of the most recently introduced novel antibiotic classes (Robbel and Marahiel, 2010). The AMPs discussed in this review differ from peptide-based antibiotics in the mechanism of their synthesis and also in their amino acid make-up. AMPs, including those made in human cells, are in contrast to NRPS-antibiotics, produced by the normal process of ribosomal translation on an mRNA template. The primary product is usually a pre-protein that is then processed to the final length of the active AMP. For example, the human cathelicidin AMP, LL-37, is produced as a precursor protein, human cationic antimicrobial protein-18 kDa (hCAP18), and the mature LL-37 peptide is the result of its processing by proteinase 3 (Gudmundsson et al., 1996). This difference in the genetic origins of AMPs and NRPS-antibiotics has consequences for the compositions of the final products. Ribosomally produced AMPs contain only the normal complement of amino acids found in proteins, sometimes with post-translational modifications. NRPS-produced antibiotics, in contrast, are unconstrained by the limitations of ribosomal translation, and usually contain a mixture of normal amino acids together with non-canonical amino acids not found in any proteins (Walsh et al., 2013). In addition, genes for some of the human α-defensins (HNP1 and HNP3) are present in multiple copies and are inherited unequally by different individuals, giving rise to individuals with two or three copies of either or both of these genes, potentially affecting the levels of AMPs produced among individuals (Mars et al., 1995).

Human AMPs vary in length from 5 to 149 amino acids (Bangalore et al., 1990; Cash et al., 2006). The great majority are cationic peptides, but the net charge of different human AMPs ranges from −3 to +20. AMPs have been identified in different human excretions, tissues and cell types, including saliva, tears, sweat and milk, on the skin and tongue, in bone marrow, plasma, kidneys, liver, heart, brain, eyes, intestine, sperm, urinary tract, amniotic fluid, and respiratory tract, and in many different cell types including epithelial/mucosal cells, macrophages, neutrophils, natural killer cells, monocytes, eosinophilic leukocytes, Paneth cells, T-cells and B-cells (http://aps.unmc.edu/AP/). Research into the functions of AMPs goes back at least to the time of Alexander Fleming who discovered lysozyme (Fleming and Allison, 1922). However, it is in the past few decades, especially after the discovery of cationic bacteriocidal peptides in polymorphonuclear leukocytes (Zeya and Spitznagel, 1963), that interest and research has expanded explosively. During the 1990s important new classes of AMPs were discovered in humans, including the β-defensins (Bensch et al., 1995) and the cathelicidins (Gallo et al., 1997) and this stimulated research into understanding the number and variety of human AMPs and, more recently, into their potential applications in antimicrobial therapy to control bacterial infections.

2. Structures of AMPs

Defensins and cathelicidin were recognized early on as important components of the antimicrobial mechanisms of polymorphonuclear leukocytes (PMNs), and were classified as belonging to significantly different structural families (Lehrer and Ganz, 2002; Zanetti et al., 1995). A classification of AMPs into four major families, based on their 3D structures, has been adopted by the APD3 database (Wang et al., 2016).