# Antimicrobial Peptides: ancient conserved molecules as new therapeutics

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*If during the long course of ages and under varying conditions of life, organic beings vary at all in the several parts of their organization, (…) then, considering the infinite complexity of the relations of all organic beings to each other and to their conditions of existence, causing an infinite diversity in structure, constitution, and habits, to be advantageous to them (…) But, if variations useful to any organic being do occur, assuredly individuals thus characterized will have the best chance of being preserved in the struggle for life; and from the strong principle of inheritance they will tend to produce offspring similarly characterized. This principle of preservation, I have called, for the sake of brevity,* ***Natural Selection.*** *"*
*Charles Darwin (1859). The Origin of Species.*

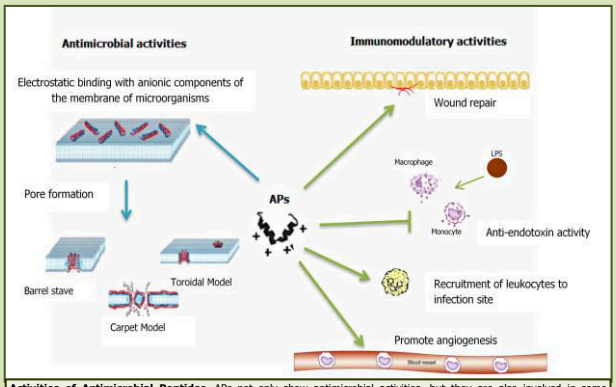
## NEED OF NEW ANTIBIOTICS

The development of antibiotic resistance in bacteria is a good example of natural selection, to the point that these resistance bacteria have been described as ***superbugs.***
It's obvious the need to design new therapeutic strategies to solve this global health problem appeared because of the misuse and overuse of antibiotics.
This work put forward as promising candidates **Antimicrobial peptides** (APs), ancient molecules involved in many defense mechanisms conserved during a lot of generations in a wide range of organisms, because nature can always teach us.

## WHY APS?

APs are small cationic peptides, effector molecules of the innate immune system. They are present in the first barriers of all living organisms: skin, mucosa, epithelia... and that's why these tissues are almost never infected.
They are expressed in a variety of cell types: leucocytes, such as monocytes and neutrophils, epithelia cells, mast cells, so on, and their expression could be constitutive or induced.

Nowadays more than 2000 APs have been reported from different organisms and with different activities. Understanding the relationship between structure and activity may facilitate the rational design of novel antimicrobial agents.




**Activities of Antimicrobial Peptides.** APs not only show antimicrobial activities, but they are also involved in some biological processes related with immunity.
Figure modified from Fabiola Cost, et. al. Elsevier Science Direct, Peptides 36 (2011) and Yuping Lai, et. al. Trends in Immunology (2009)

Due to their **amphipatic properties**, caused by their positive charge and nearly 50% of the amino acids hydrophobic, antimicrobial peptides show **broad antimicrobial activity**: antibacterial, antiviral, antifungal, antiparasitic and so on. APs can bind to anionic components of microorganisms membrane, causing a disruption on it and promoting death. There are several mechanisms proposed for the permeabilization of membrane, e.g. Carpet model, barrel stave or toroidal model.

Furthermore, APs have an impressive variety of additional activities related with some **immunity processes**, such as neutralization of endotoxins released from macrophags and monocytes, chemotaxis , promotion of angiogenesis and wound repair.

## Databases information

| Nº | Activity | Example AP |
|---|---|---|
| 1762 | Antibacterial | Defensins<br>Cathelicidin derived peptides: Protegrin-PG1 |
| 773 | Antifungal | Cathelicidin derived: BMAP-27, Indolicidin. Histatins. Aurein |
| 135 | Antiviral (VIH, VHS, VSV) | α- defensin HNP1, Human β- and synthetic θ-defensin (Retrocyclin 2) |
| 48 | Antiparasitic | BMAP-18, PMAP-23 |
| 22 | Antiinsecticidal | Ponericin |
| 3 | Anti-protist | EcAMP1 |
| 145 | Anticancer | Buforin IIb (derived from histone H2A) |
| 41 | Chemotactic activity | **For neutrophils**: PR-39<br>**For T cells**: HNP-1, LL-37<br>**For monocytes**: ProBac 7, Defensins |

**Overview of different activities of APs and some examples.** Information source: APD (Antimicrobial peptide database).

## APs AS NEW ANTIBIOTIC DRUGS

| | Conventional antibiotics | Antimicrobial peptides |
|---|---|---|
| Activity | Only Anti-bacteria | Broad spectrum of activity: bacteria, fungi, virus… Immunomodulatory properties: chemotactic, endotoxin neutralization, wound repair, angiogenesis… |
| Antibacterial activity | Single primary target and mode of action. 4-24 h aprox | Differet mode of action in each bacteria Minutes |
| Resistance development | High | Few |
| Pharmacodinamic | Good bioavailibity and biodistribution | Low oral bioavailbilitySusceptibility to salt, pH and serum<br>Poor specific biodistribution<br>hepatic/renal clearance. |
| Pharmacokinetic | Vary | Short half-life<br>Proteolytic degradation |
| Toxicology | Low | High |
| Manufacturing cost | Low | High |

APs seem to figure out the resistance problem because, unlike conventional antibiotics, APs do not have only a single primary target and a single mode of action, each AP could show different modes of action in each microorganism. In addition, they do not need to bind to specific receptors on the membrane surface. Nevertheless, they show some unresolved problems that don't let them go into clinical market.

| Compound/ peptide | Company | Mode of use | Clinical use | Phase |
|---|---|---|---|---|
| **Polysporin** (Gramicidin,polymixin, Bacitracin) | Johnson & Johnson | Topical, ophthalmologic | Triple antibiotic therapy ointment | Approved FDA |
| **Primecrolimus** | Valent Bermuda | Topical | Atopic dermatitis | Approved FDA (2001) |
| **Polymyxin B** | Some | Multiple forms | Gram – infections | Approved FDA |
| **Daptomycin** | Cubist Pharmaceutical | Intravenous | Infections by Gram + / MRSA[1] | Approved FDA (2003/2013) |
| **Micafungin** | Astelles Pharma Inc. | Multiple forms | Antifungical | Approved FDA (2005) |
| **Fuzeon** | Hoffmann-La Roche | Subcutaneous | Against HIV replication | Approved FDA (2003) |
| **Pexiganan MSI-78** | Acces Pharmaceuticals | Topical | Diabetic ulcers | III |
| **MBI-226** | Biowest therapeutic Inc | Topical | Acne | II |
| **PMX 30063** | Polymedix, Inc. | Topical | Acute bacterial skin infections due MSSA[2] | II |
| **Lactoferrin hLFf1-11** | AM-Pharma | Intravenous | Fungal and bacterial infections in HSCT | I/II |
| **Colistidin** | Park Davis s | Oral | Infection due to MSAB | Synergism colistidin+ Rifampicin ( p. III) |

**APs in clinical use and development.** Abbreviations: MSSA- Multiresistant Staphylococcus aureus; HSCT- Haematopoietic stem cells transplant; MRAB- Multidrug-Resistant Acinetobacter Baumannii; MRSA- Methilicidin-resistant Staphylococus aureus

## FUTURE PROSPECTIVE AND CONCLUSIONS

The main hurdless that have impeded the development of such peptides and their clinical use are (1) the toxicity against mammalian cells, (2) high susceptibly to proteases and (3) high manufacturing cost.
Owing to the development in solid phase peptide synthesis is becoming easer and the manufacturing cost cheaper. Several strategies like peptidomimetics, formulation of prodrugs and substitution of L-amino acids to D-aminoacids have been evaluated to enhance the stability and improve toxicity in their administration. Once improved that, maybe in a long term, it would be interesting to address methods of formulation for systemic or oral use.

It is important to note that antimicrobial peptides could enter into therapeutics by various strategies, such as combination of peptides and administration as adjuvants to promote a synergistic effect and enhance the innate immune response.

Needless to say, APs are excellent candidate molecules that must be studied in depth to understand the basis of their limitations. It is important address future studies to the development of safe, stable and effective therapeutic strategies with antimicrobial for bacterial infections and other human diseases.