# Development of an Antibacterial MAP Homologous to a Loop Region in Human Lactoferrin

**Masachika Azuma** [1]
azuma@translell.eco.tut.ac.jp
**Taiki Kojima** [2]
fwhw8642@mb.infoweb.or.jp
**Masaki Kobayashi** [3]
kobayashimasaki@msg.biglobe.ne.jp
**Carlos Adriel Del Carpio** [1]
carlos@translell.eco.tut.ac.jp

[1] Laboratory for Informatics & AI in Molecular and Biological Sciences, Department of Environmental Life Science, Toyohashi University of Technology, Tempaku, Toyohashi 441-8580, Japan
[2] Department of Surgery II, Nagoya University, School of Medicine, Nagoya 466, Japan
[3] Life Information Analysis Center Inc., Moto, Komaki 485, Japan

## 1 Introduction

We report on the antibacterial activities of an 11 residue peptide (FQWQRNMRKVR) homologous to just over half the loop region of human lactoferricin. The peptide, in the form of a multiple antingen peptide (MAP), exerted significant antibacterial effects against a broad spectrum of bacteria including MRSA.

Lactoferricin is an iron-binding glycoprotein belonging to the transferrin family and found in the specific granules of neutrophils and in secretion fluids such as tears, saliva and even milk. The pepsin digestion of bovine and human lactoferrin promotes the release of antibacterial peptides known as lactoferricin B and H respectively. This may be the key to its antimicrobial activity [1]. Lactoferricin is effective against Gram-positive and negative bacteria as well as yeasts [2], its mechanism of action is however unknown.

Human lactoferricin comprises two chains from residue 1-46 from the N-terminus of lactoferrin and includes an 18 residue loop formed by one of two disulphide bridges [1]. Recently, it has been shown that a much smaller peptide of only 11 residues (FQWQRNMRKVR), homologous to just over half the loop region has potent antibacterial activity and may account for all the activity of the larger peptides.

Here we describe the computer analysis of the structure of such antibacterial MAP, and the experimental results of their antibacterial activities.

## 2 Computer Analysis and Experimental Results

We predicted the eleven amino acid long peptide (FQWQRNMRKVR) three dimensional structure using our 3D structure prediction program (GAX) [3]. The force field implemented in this program corresponds to the MM3 in vacuum. The structure of the MAP with 4 strands of the peptide was also predicted. However for technical reasons it was not possible to obtain the structures for higher branching such as MAP-8 and MAP-16.

The secondary structure of FQWQRNMRKVR, derived from the predicted three dimensional coordinates showed an $\alpha$-helix bordered by two turn like structures at the N and C terminals Fig. 1. The MAP-4 structure conserved the alpha-helical structure in each strand. The structure showed the four strands extended in a symmetrical manner, Fig. 2, although some stable conformers were also obtained where the strands laid crossed with each other.

Experimental results of the tests of antibacterial activities show that MAP-8 and MAP-16 exhibited anti-bacterial activity against Pseudomonas aeruginosa, while the linear MAP-2 and MAP-4 did not, up to a concentration of 200 $\mu$M.

MAP-16, however, showed antibacterial activity against a broad spectra of bacteria, being especially strong against Pseudomonas aeruginosa.

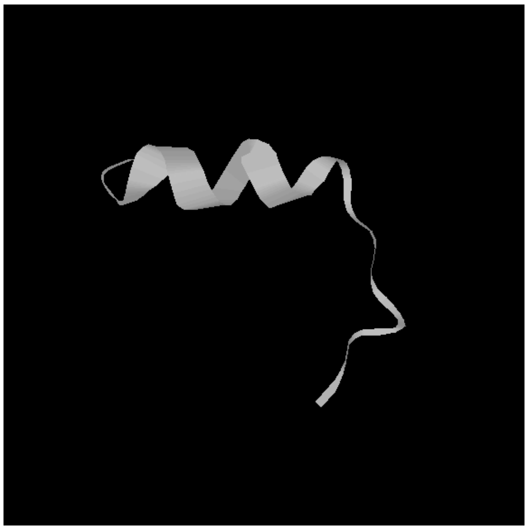

Figure 1: $\alpha$-helix structure.

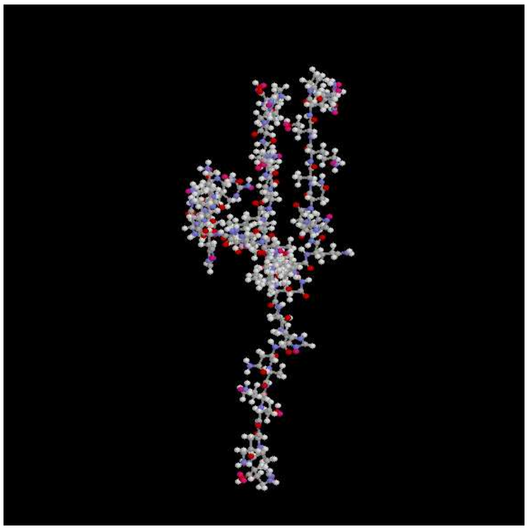

Figure 2: MAP4 structure for the antibacteria.

## 3 Conclusions

Computer modeling of the three dimensional structure of the MAP containing the 11 amino acid long (FQWQRWMRKVR) peptide strongly suggests that the structure (an amphipathic cationic alpha-helix) exerts its antibacterial activity by performing ionic channels or pores in the bacterial membrane. The results are supported by former reports that amphipathic cationic $\alpha$ helices are common patterns to some or the antibacterial peptides or antibacterial domains of peptides such as magainins [4], cecropins [5], and CAP18 [6].

Moreover, MAP structures were originally synthesised as stimulators of antibody production in sensitized animals, and their use included vaccine development. Here we propose a novel possibility for this type of multiple antigen peptides, i.e. the increase in the activity of anti-biotic peptides.

## References


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