DE NOVO DESIGN OF ANTIMICROBIAL PEPTIDE AND THEIR ACTIVITY SIMILAR TO ARD1

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Abstract: A 45 amino acid long peptide has been designed *de novo* with its activity similar to ARD1. Its docking interaction depicts formation of hydrogen bond which alters so many of biological pathways of bacteria (microorganism) thus expressing a strong antimicrobial agent.

Keywords: Antimicrobial peptide, De novo, Modeler.

Introduction:

The continuous exposure to antibiotics with increasing risk of resistance is a great problem to public health. In last few decades AMPs appear as new strategy to combat with bacterial infections and evolved as a hope. More than 2000 antimicrobial peptides have been discovered and stored in Antimicrobial Peptide Database (1).

ARD1 is a naturally occurring antifungal peptide isolated and purified from *Archeoprepona demophon* (Pdb code 1ozz.pdb). ARD1 differs from heliomicin by only two residues but with enhanced activity against the two common human pathogens *A. fumigatus* and *Candida albicans*.

The E.coli FlhDC complex, a prokaryotic heteromeric transcription regulator protein with PDB code 2 AVU is a complex of FlhD and FlhC protein (FlhDC) which regulates the transcription operon in bacteria (2) necessary for their motility in case of flagellar bacteria. The same protein complex is also helpful in lipolytic and hemolytic activity in some insect. About 50 genes are involved in expression of flagellum in bacteria and these all are highly co-ordinated cascade with hierarchy. Among the hierarchy, the class I operon flhdc, whose products are by required for all genes to express the flagella

Material and methods:

Randomly chosen 45 residues has been taken to *de novo* design the AMP as ligand molecule by substituting the amino acid in chain to test their activity against the bacterial target components. The sequence was subjected to modeler9v3 program(3) to model the peptide on homology basis. The requirement of modeler program is one template with known 3D structure and some script file to run the application. In this study to design the 3D structure of unknown sequence the known template molecule is ARD1.

Template sequence (ARD1)
DKLIGSCVWGAVNYTSNCNAECKRRGYKGGHCGSFANVNCW
Query sequence

LLICVVGAAFWYYPPLVCCAFFYIIGGLWPAAAPPYVIICAFFWW

The designed sequences were also subjected to the plug-in *Predator* application (Frishman and Argos, 1997) of the molecular modeling program *Vega ZZ* with return of a predicted of 3D secondary structure for the given sequence. The fig (a) and (b) displays the expected 3D structure of subjected peptide sequence.

LLICVVGAAFWYYPPLVCCAFFYIIGGLWPAAAPPYVIICAFFWW   45 Residue
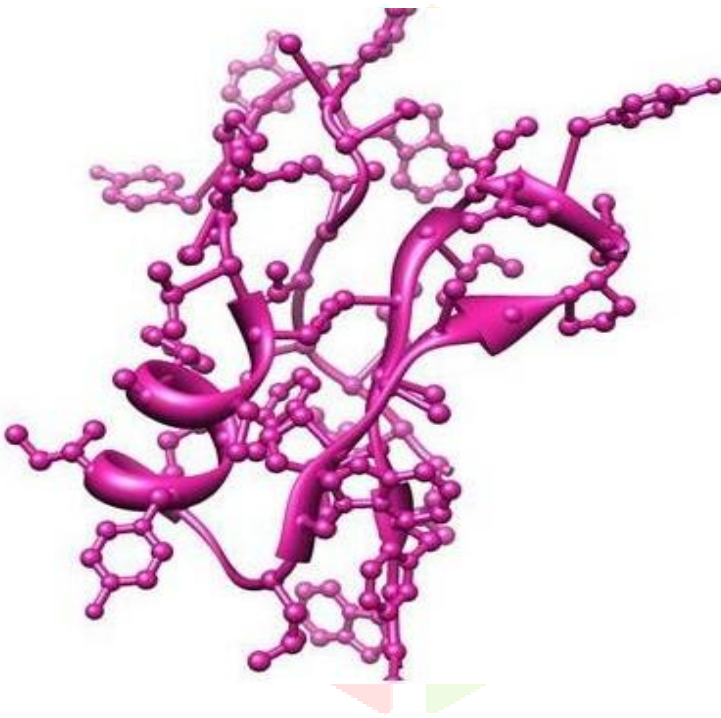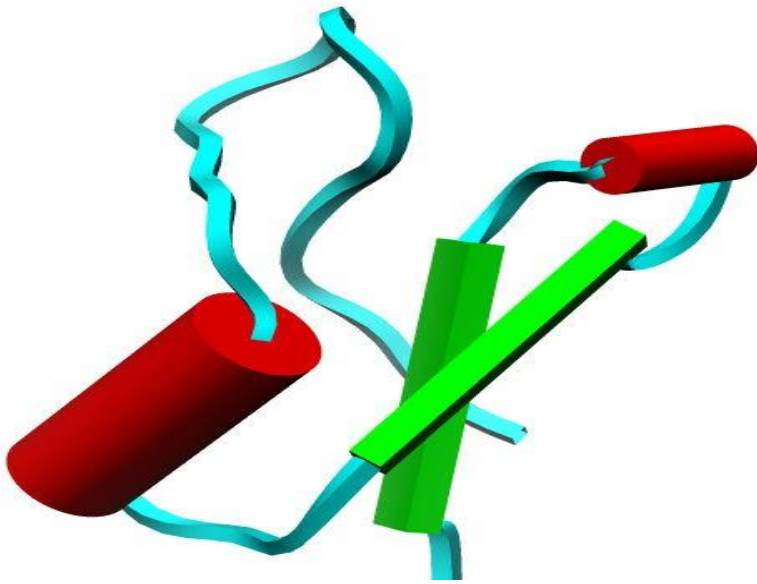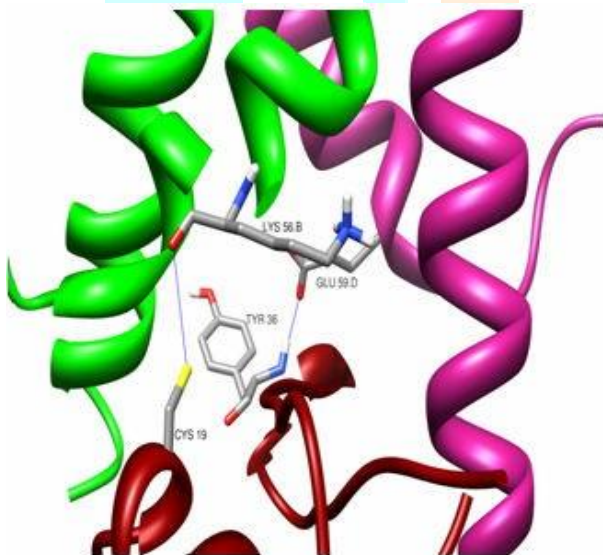__ EEEEEEEEE____EEEEEEEEE_____EEEEEEE___

Fig: (a)

Fig: (b)

Modeler program generated 3D co-ordinate file is viewed by UCSF chimera in ball and stick form fig(a) and in pipes and plank form fig(b).

UCSF Chimera version1 rendered docked image of antimicrobial peptides with oligomeric regulator protein.

Fig (c)

Result and discussion:

Modeler program generated peptide has been docked with bacterial transcription regulator protein flhdc. Docking is a method to predict the orientation of one molecule to another with their possible proximity to form a stable complex(4) by overall minimizing the system free energy. The newly generated peptide binds as a ligand molecule to the regulator protein at two places shown in fig (c).

Regulator protein chain	Peptide as ligand
1. Glu 59D	Tyr 36
2. Lys 56B	Cys 19

Binding with regulator protein with hydrogen bonds and electrostatic interaction suggest that the *de novo* peptide might block the bacterial flagella synthesis which is needed for motility thus inhibiting the growth of bacteria.

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