

Antimicrobial activities by the novel synthetic antimicrobial peptide against multidrug resistance and biofilm forming strains in a guinea pig model

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Introduction

Membrane-active peptides, including a host of antimicrobials and toxins, have been shown to induce the formation of transmembrane pores. Although the molecular mechanisms by which pore formations occur remain to be thoroughly elucidated, the three mechanisms thus far proposed include the “barrel-stave”, “carpet”, and “toroidal” models. In the barrel-stave model, peptide helices form a bundle with a central lumen within the membrane, appearing similar to a barrel in which the helical peptides function as the staves [1-5].

Stomach mucosa that is infected with *Helicobacter pylori*, the bacterial pathogen associated with gastritis and peptic ulcers, typically shows massive infiltration of inflammatory cells and tissue destruction [6]. Persistence of *H. pylori* in the mucosa has been suggested to be facilitated by *H. pylori*-produced cecropin-like peptides with antibacterial properties. Although *H. pylori* itself is resistant to this peptide, the release of these peptides gives a competitive advantage over other microorganisms [7].

The linear antimicrobial peptide, HP(2-20), is a cationic α -helical peptide that has been isolated from the N-terminal region of the *Helicobacter pylori* ribosomal protein, L1 [8]. This peptide possesses several important functional characteristics; it is bactericidal, it is a neutrophil chemoattractant, and it activates phagocyte NADPH oxidase to produce reactive oxygen species [8].

Results and Discussion

Barely 10 years after ciprofloxacin's introduction into otology, the emergence of ciprofloxacin-resistant *P. aeruginosa* (CRPA) have created new therapeutic challenge in otology. A3 is a synthetic antimicrobial peptide derived from HP (2-20), derived from *Helicobacter pylori*. In a previous study, A3 was shown to possess strong antimicrobial activity in an in vitro test. The peptide A3 showed a strong antimicrobial activity against CRPA in vivo and the cochlear tolerance of A3 was studied in vivo by topical application to the middle ear in guinea pig model. Twenty guinea pigs (5 groups, each group n=4) were each injected by a transtympanic approach with 20 μ l of 8 μ g/ml, 16 μ g/ml, 32 μ g/ml, 128 μ g/ml of A3, and 0.4% gentamicin sulfate was instilled as a control. Auditory brainstem responses (ABR) to click were measured between 1st and 7th days after injection (Table 1). Histologic investigation of cochlea was performed by scanning electron microscope and light microscope. Guinea pigs injected with A3 8 μ g/ml, 16 μ g/ml, 32 μ g/ml

showed no interval changes in response ABR threshold and SEM showed the intact cochlear hair cells. However, 128 μ g/ml showed slight loss of hair cell in the SEM and increased ABR threshold 10 dB more than A3 32 μ g/ml treated group. The control GM treated group showed no detectable ABR threshold, and severe missing of hair cells in the SEM and severe edematous stria vascularis in the LM. Topical application of A3 to the middle ear is well tolerated without cochlear damage and may be a useful as a new otological agent for CRPA otitis media.

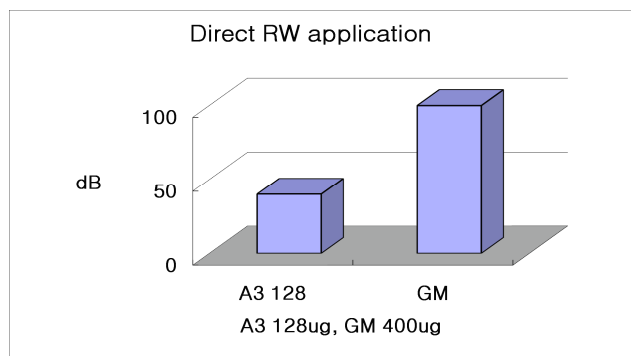


Table 1. ABR : Direct RW approach

- A3 128 μ g and GM 400 μ g (10 μ l) using gelfoam to the RW by postauricular app.
- Avoid trauma to ISJ
- Check C-ABR postop 7th day.

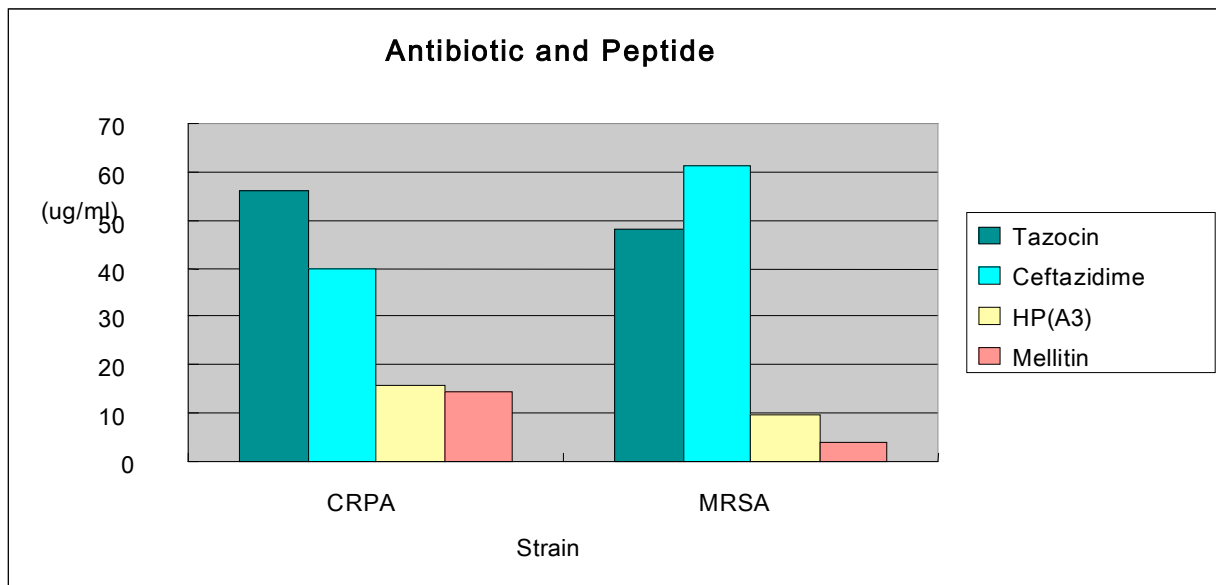
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Antibacterial activity: MIC by ELISA