Microbicide Activity of Two Reptilian Antimicrobial Peptides on Gram Positive and Gram Negative Bacteria

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Abstract

Previous in-vivo studies have isolated and identified peptides with typical molecular anti-microbial characteristics
in reptiles. In the present study we have tested the putative antimicrobial action of a lizard cathelicidin and of a turtle beta-defensin using the broth microdilution assay on Gram positive and Gram negative bacteria. The addition of the peptides at concentrations indicatively ranging between 0.05-1.9 mg/ml (cathelicidin) and 0.69-4.14 mg/ml (betadefensin) inhibited bacterial growth after 3 hours of incubation as determined by their MIC and IC50 values. Due to the poor solubility and the medium interference the real concentration of the delivered peptides to the bacterial cultures was uncertain. The qualitative evaluation of the anti-microbial damage after treatment with the peptides was done under the electron microscope that showed some alteration and rupture in the plasma membrane, lowering of the ribosomes, swelling and clumping in nucleoid region of Gram negative (E. coli) and Gram positive (S. aureus)bacteria. Immunogold labeling against the two peptides indicated that the peptides were localized not only on the plasma membrane and in cytoplasm of the treated bacteria, but also in the nucleoid region and its protein scaffold.

The present ultrastructural study suggests that these peptides operate a cellular damage initially on the plasma membrane but further also in the ribosomes and on the DNA or its associated proteins. Keywords: Reptiles; Antimicrobial peptides; Bacteria; Antimicrobialtests; Ultrastructure.Protection from potentially pathogenic infections from microbes occurs through di ُerent mechanisms, including the production of antimicrobial peptides. Numerous antimicrobial peptides responsible for a strong innate immunity have been prokaryotes and eukaryotes . Antimicrobial peptides are composed of 8-60 or more amino acids and include several categories among which the best known include the beta-defensins and cathelicidins . He potential utilization of these molecules as Efective new antibiotics is of paramount importance in recent times due the mounting resistance of numerous pathogenic microbes to old and new classes of antibiotics, and therefore e ُorts in discovering e ُective new drugs is very active field of modern infective research.Antimicrobial peptides are not a homogeneous class of compounds, but show a broad diversity in structure and antimicrobial spectrum and interactions .

Previous studies, based on the observation of the high resistance of lizards and turtle to wounds which showed the presence of numerous intercellular and intracellular bacteria in the epidermis ,suggested that potent antimicrobial peptides were possibly involved in the outstanding immunity present in these reptiles. His hypothesis was later confirmed by the isolation of numerous beta-defensins and some cathelicidins from lizard and turtle , and from their prevalent localization in granulocytes and activated keratinocytes.