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Amphibian peptides for skin protection and healing.

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BACKGROUND: Amphibians are currently suffering a dramatic decline worldwide, mainly due to chytridiomycosis, a skin infection caused by the pathogenic fungus *Batrachochytrium dendrobatidis* (Bd). An important natural defense of amphibian skin is the production of antimicrobial peptides (AMPs) by granular glands in the dermis. AMPs collected from several species of frogs successfully inhibit the growth of Bd *in vitro*. Besides their anti-microbial and anti-fungal activities, AMPs have been shown to exert other biological effects such as anti-viral, anti-tumor, anti-oxidant, immunomodulating and wound healing.

AIM: We intended to test the efficacy of AMPs as cutaneous defenses in frog species either resistant or susceptible to Bd.

METHODS: 3 frog species (*Gastrotheca nebulanastes* (GN), *G. excubitor* (GE) and *Hypsiboas gladiator* (HG)), were collected in montane scrub, cloud forest and high elevation grassland habitats near Manu National Park in southeastern Peru. AMP secretion was stimulated by injection of norepinephrine into the dorsal lymph sacks. AMPs were then purified by chromatographic techniques. The human endothelial cell line HECV was treated with AMP concentrations ranging from 0.005 to 50 µg/mL. Cell viability was verified by MTT test. Wound healing properties were analyzed by scratch wound assay. AMP inhibition strength against Bd growth was measured *in vitro* by incubating Bd zoospores with different concentrations of AMPs.

RESULTS: Treatment with AMPs secreted from GN, GE and HG did not affect HECV cell viability at any concentration tested. No significant differences in cell migration rate were observed in HECV cells scratched and treated with GN and GE AMPs. Only HG peptides showed wound healing properties as well as strong Bd growth inhibiting ability.

CONCLUSIONS: Stimulation of wound healing mechanisms and inhibition of Bd growth by skin AMPs might both contribute to HG resistance to chytridiomycosis. Understanding the role of skin defenses may lead to the development of novel Bd mitigation strategies. Possible applications of amphibian AMPs in skin medicine deserve attention and further studies.

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