Study on a Novel Analgesic Peptide from the Digestive Juice of Land Leeches

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Haemadipsa guangchuanensis, known as land leeches, is a genus of annelids feeding on blood. Although its biology and reproductive ecology had been studied, there were few reports regarding its analgesic active ingredients. In this project, the digestive tract secretions of Haemadipsa was subjected to the biological assays, such as gel filtration, ultrafiltration membrane-based centrifugal filtration, high performance liquid chromatography separation (HPLC) and purification, mass spectrometry (MS) identification, chromogenic reaction, amino acid sequence determination, computational remodeling, polypeptide synthesis and refolding, cytokine secretions induced by lipopolysaccharide (LPS), and Western blot analysis. The results showed that its digestive tract secretions contained a polypeptide with a relative molecular mass of 3951.8 dalton, which turned violet in the biuret test. This 34-amino acid polypeptide was then named as HGSN1. The amino acid homologous sequence alignment suggested that HGSN1 is a novel active peptide. It contained two pairs of disulfide bonds formed by its four cysteine residues. Synthetic and folding HGSN1 could markedly inhibit secretions of TNF-a, IFN-g, IL-6, and IL-10 that were induced by LPS. Mechanistically, HGSN1 inhibited ERK phosphorylation in a dose-dependent manner, suggesting its role in the analgesic effect. In summary, our current work suggests that HGSN1 is a key ingredient secreted by Haemadipsa guangchuanensis that facilitates its painless blood-sucking process. HGSN1 can potentially be adapted as a precursor compound to develop novel analgesic drugs, and it would be of great importance to the research of protection from land leeches and its treatment.

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