

## 국내산 봉독으로부터 분리한 멜리틴의

*Staphylococcus aureus*에 대한 항생제후 효과농촌진흥청, <sup>1</sup>대구가톨릭대학교, <sup>2</sup>The University of Waikato한상미\*, 이광길, 여주홍, 우순옥, 권해용, 장영채<sup>1</sup>, 박관규<sup>1</sup>, Peter Molan<sup>2</sup>**Postantibiotic effect of melittin from honeybee (*Apis mellifera*) venom against *Staphylococcus aureus***Rural Development Administration, <sup>1</sup>Catholic University of Daegu,<sup>2</sup>The University of WaikatoSangmi Han\*, Kwanggil Lee, Joohong Yeo, Soonok Woo, Haeyong Kweon, Youngchae Chang<sup>1</sup>, Kwankyu Park<sup>1</sup>, Peter Molan<sup>2</sup>**Objectives**

Since the ancient times the therapeutic application of honeybee venom (BV) is practised and persisted until the present days. To purify the melittin known as antibacterial peptide, five major peptidergic subfractions were separated, purified and identified from the whole BV. We investigated the antibacterial activity of purified melittin against *Staphylococcus aureus* by the minimum inhibitory concentrations (MIC) and the postantibiotic effect (PAE).

**Materials and Methods**

## ○ Materials

BV was collected by a bee venom collector. *S. aureus* (ATCC 9144) was obtained from the Honey Research Unit, the University of Waikato(Hamilton, New Zealand).

## ○ Methods

The isolation and purification of melittin from bee venom was performed by a Superdex Peptide and a PepRPC column. The PAE was determined by the equation  $T-C$ , where  $T$  is time required for viability counts of an antibiotic exposed culture to increase by 1 log<sub>10</sub> above counts immediately after dilution, and  $C$  is corresponding time for growth control.

**Results**

The peak of melittin was indicated with an arrow in fig 1. The molecular mass and purify of purified melittin were analyzed by a MALDI-TOF mass spectrometry, giving a molecular mass of 2844 (Fig. 2). MIC of the melittin were 0.06  $\mu\text{g ml}^{-1}$  for *S. aureus*. For the *S. aureus*, the mean PAE analysis of melittin was 4.35 h at 1× MIC. *S. aureus* was completely killed by the melittin at 10 times MIC of the melittin. At 5 × MIC and 10 × MIC, no *S. aureus* could be detected within 1 h. Regrowth wasn't observed as early as 18 h.

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The exposure time was 1 h that melittin is enough time to kill for *S. aureus* at concentrations up to  $5 \times \text{MIC}$ .

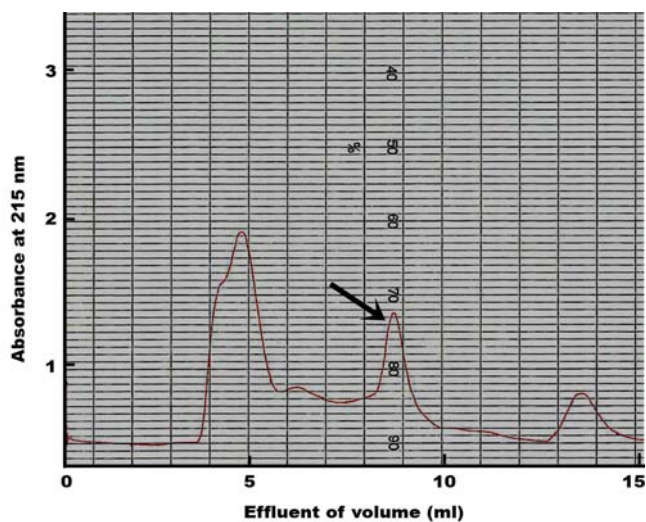


Fig. 1. Reverse phase liquid chromatography. Fraction I from superdex peptide column was loaded onto a PepRPC HR 10/10 column and acetonitrile of 30% was employed for elution. Arrow indicates melittin containing peak

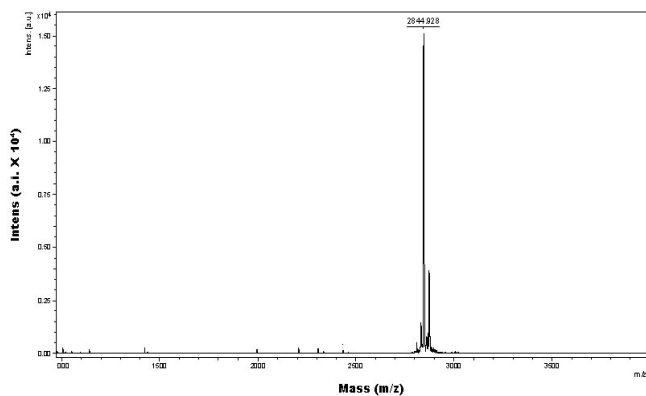


Fig. 2. Mass spectrometry of purified melittin.