

## 2, 4, 6,-trihydroxy- 3-geranylacetophenone (tHGA) inhibits newly synthesized mediators release in IgE-mediated mast cell degranulation

### ABSTRACT

Mast cells are important effector cells of the innate immune system and participate in allergy reaction. Upon FCERI aggregation, mast cells secrete preformed mediators such as histamine as well as newly synthesized mediators including prostaglandins, leukotrienes and proinflammatory cytokines. Our previous studies demonstrated that tHGA, an active compound isolated from *Melicope ptelefolia*, was able to dose-dependently inhibit the release of histamine and  $\beta$ -hexosaminidase in IgE-mediated mast cell degranulation. However, the inhibitory effects of tHGA on the release of newly synthesized mediators during mast cell degranulation still remain unknown. The current study aims to investigate the in vitro inhibitory effects of tHGA on newly synthesized mediators released in IgE-mediated mast cell degranulation by examining the level of arachidonic acid metabolites such as prostaglandins D2 and leukotriene C4, as well as cytokines such as interleukin 4 and tissue necrosis factor alpha. IgE-sensitized RBL-2H3 cells were pre-treated with tHGA for 20 minutes and later challenged with DNP-BSA for 6 hours. The level of all four mediators released by the degranulated cells was measured by using ELISA kits according to the manufacturer's protocol. Real time-PCR was also performed to examine the effect of tHGA on the gene expression of IL-4 and TNF- $\alpha$ . The results showed that pre-treatment of IgE-sensitized RBL-2H3 cells with non-cytotoxic concentrations of tHGA (1.25, 5 and 20  $\mu$ M) significantly decreased the release and gene expression of all four mediators in a concentration-dependent manner ( $p < 0.005$ ). This preliminary study demonstrated that tHGA does not only inhibit the release of preformed mediators, but also attenuate the release of newly synthesized mediators during mast cell degranulation, via inhibition on their respective gene expression. Further study should look into the effect of tHGA on major signalling pathways to understand the mechanism of action of tHGA that contributes to its inhibitory effects in IgE-mediated mast cell degranulation.

**Keyword:** tHGA; *Melicope ptelefolia*; RBL-2H3 mediators