A phloroglucinol from Melicope ptelefolia attenuates IgE-mediated mast cell degranulation via calcium-dependent signalling pathways

ABSTRACT

Mast cells are important effector cells of the innate immune system that participate in allergic reactions through activation of Lyn, Syk, mitogen activated protein kinase (MAPK), increase of intracellular calcium ion concentration ([Ca2+]i), degranulation and cytokine production. Our previous studies demonstrated that tHGA, a synthetic phloroglucinol originally found in Melicope ptelefolia, was able to inhibit IgE-mediated mast cell degranulation, evidenced by reduced amount of histamine, β-hexosaminidase, PGD2 and LTC4, IL-4 and TNF-α. However, the inhibitory mechanism remains unknown. This current study aims to understand the inhibitory mechanism of tHGA in IgE-mediated mast cell degranulation. IgE-sensitized RBL-2H3 cells were pre-treated with tHGA for 20 minutes and challenged with DNP-BSA for 30-60 min. The effect of tHGA on the influx of calcium ion into cells was studied by using Calcium Detection Kit. The inhibitory pathways of tHGA was determined by examining major signalling molecules involved in IgE-mediated mast cell degranulation using Western blot. The results from calcium influx assay showed that tHGA decreased the influx of calcium into cells. Western blot further confirmed that tHGA does not attenuate the phosphorylation of signalling molecules in calcium-independent NF- -κB signalling pathway but significantly attenuated the phosphorylation of signalling molecules in calcium-dependent pathways including MAPK, at 5 μM and 20 μM ($P < 0.05$). In a conclusion, tHGA attenuates IgE-mediated mast cell degranulation via calcium dependent signalling pathway. The molecular target of tHGA shall be further confirmed to provide an insight into the design of novel pharmacological agents which may be used to regulate the mast cell response.

Keyword: Phloroglucinol; Melicope ptelefolia; Immune system; Mast cell; Allergic reactions