ABSTRACT

For centuries, *Zingiber officinale* had been used for medicinal purpose, such as painkillers, cancer prevention, anti-inflammatory and anti-oxidant. One of ZO mechanism is to modulate the activity of immune cells, including activity that modulated through Toll receptors signalling pathway. The fruit flies (*Drosophila melanogaster*) DL1 cells have been used to study the activation of ligand-induced Toll receptors. Furthermore, the effect of ZO on mammals toll receptors or toll-like receptors is limited both in vivo and in vitro. Hence, it is important to investigate ZO activity in Toll-pathway.

DL1 cells were cultured for 10 days and inoculated in 384 well plate. Viability assay was observed after 0.05-50 ppm ZO extract and DMSO (solvent control) treatment using CellTiter-Glo® reagent to measure the ATPs. In brief, samples are divided into 3 groups, i.e., 1) DMSO 1% as solvent control, 2) DMSO 25% as toxic control group, 3) ZO extract concentration (0.05-50 ppm) group. The increase in luminescent signal indicates the increase in cell viability. Another batch of DL1 cells were inoculated for Toll receptors activation with Spätzel ligand. The toll receptor activation assay was performed with the same sample grouping. ONE-Glo® reagent was used to measure the luminescent signal from luciferase that produced by activated DL1 cells. The luminescent signal elevation indicates the increase of DL1 cells activation. The data was analysed using ANOVA with confidence level 95% ($p<0.05$) to determine any significant difference between concentrations.

The aforementioned various ZO extract concentrations (0.05-50 ppm) were seen to not affect the viability of DL1 cells. However, ZO extract was able to significantly modulate the activation of Toll receptor signalling pathway. Furthermore, various ZO extract concentrations (0.05-50 ppm) were found to increase the luminescent signal of the cells. Luminescent signal increase with the increase of ZO extract concentration, which means that ZO extract induced the activation of Toll receptor signalling pathway.

Keywords: *Zingiber officinale*, DL1, *Drosophila melanogaster*, Immunomodulator, Toll receptor, Luminescence.