Gallic Acid Content in Taiwanese Teas at Different Degrees of Fermentation and Its Antioxidant Activity by Inhibiting PKCδ Activation: In Vitro and in Silico Studies

Abstract:

Teas can be classified according to their degree of fermentation, which has been reported to affect both the bioactive components in the teas and their antioxidative activity. In this study, four kinds of commercial Taiwanese tea at different degrees of fermentation, which include green (non-fermented), oolong (semi-fermented), black (fully fermented), and Pu-erh (post-fermented) tea, were profiled for catechin levels by using high performance liquid chromatography (HPLC). The result indicated that the gallic acid content in tea was directly proportional to the degree of fermentation in which the lowest and highest gallic acid content were 1.67 and 21.98 mg/g from green and Pu-erh tea, respectively. The antioxidative mechanism of the gallic acid was further determined by in vitro and in silico analyses. In vitro assays included the use of phorbol ester-induced macrophage RAW264.7 cell model for determining the inhibition of reactive oxygen species (ROS) production, and PKCδ and nicotinamide adenine dinucleotide phosphate (NADPH) oxidase subunit (p47) activations. The results showed that only at a concentration of 5.00 μM could gallic acid significantly ($p < 0.05$) reduce ROS levels in phorbol ester-activated macrophages. Moreover, protein immunoblotting expressed similar results in which activations of PKCδ and p47 were only significantly ($p < 0.05$) attenuated by 5.00 μM treatment. Lastly, in silico experiments further revealed that gallic acid could block PKCδ activation by occupying the phorbol ester binding sites of the protein.

Keywords: Taiwanese tea; degree of fermentation; catechins; gallic acid; antioxidative activity; RAW264.7; protein kinase C; molecular docking