
Abstract
Dietary flavonoids provide various beneficial effects for our health. We investigated the promotive effects of quercetin and myricetin on the intestinal barrier function in human intestinal Caco-2 cell monolayers. Transepithelial electrical resistance (TER) across the monolayers increased rapidly during incubation with quercetin, peaking at 6 h. Lucifer yellow flux, a paracellular marker, was dose-dependently lower after quercetin and myricetin treatments, although quercetin exhibited a more potent effect. Immunoblot analysis of tight junction (TJ) proteins revealed that zonula occludens (ZO)-2, occludin, and claudin-1 were distributed to the actin cytoskeleton fraction by quercetin without increasing their respective whole-cell levels and this distribution was correlated with the increases in TER. The claudin-4 level was elevated by quercetin in both the cytoskeleton fraction and whole cells after 12 h. Confocal microscopy showed the assembly of claudin-1 and -4 at the TJ by quercetin. An inhibitor of protein kinase Cd (PKCd), rottlerin, enhanced the barrier function with changes in the distribution and expression of TJ proteins in a manner very similar to that of quercetin. Phosphorylation of PKCd indicating the enzymatic activity in the cells was decreased by quercetin after 1 h. In the kinase assay, quercetin exhibits direct inhibition of the PKCd isoform. This study demonstrates that quercetin enhances the intestinal barrier function through the assembly of ZO-2, occludin, and claudin-1 by inhibiting PKCd and the increase in claudin-4 expression has an additional role after 12 h.