Mechanism of action of pyrogallol on calcineurin-NFAT signaling

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Calcineurin-NFAT (CN/NFAT) signaling is one of the most well-known signaling pathways involving many biological functions. We have demonstrated that CN/NFAT signaling was responsible for the pathogenesis of allergic rhinitis and identified pyrogallol as an anti-allergic compound. Pyrogallol suppressed NFAT dephosphorylation and CN/NFAT signaling-mediated IL-9 gene up-regulation in RBL-2H3 cells. Pyrogallol improved toluene-2,4-diisocyanate (TDI)-induced nasal symptoms in TDI-sensitized allergy model rats. Here, we investigated the mechanism of action of pyrogallol for CN-NFAT signaling. Pyrogallol inhibited ionomycin-induced dephosphorylation and nuclear translocation of NFAT. Pull-down assay revealed that pyrogallol strengthened interaction between NFATc1 and calcineurin. Further studies demonstrated that calcineurin binding site 2 in NFATc1 was involved in pyrogallol's effect. Poly(U)-binding-splicing factor 60 (PUF60) was identified as NFATc2 binding protein using pyrogallol-immobilized affinity chromatography. Pyrogallol suppressed ionomycin-induced interaction of PUF60 with NFATc2. Knockout of *PUF60* gene suppressed ionomycin-induced IL-9 gene up-regulation in RBL-2H3 cells. These results suggest that pyrogallol suppressed CN/NFAT signaling through the inhibition of NFAT dephosphorylation by the isoform-dependent manner.