

Analysis of a new signaling pathway involved in thromboxane A₂ receptor-mediated vascular contraction

Kimiaki SUZUKI

Department of Pharmacology, Graduate School of Pharmaceutical Sciences

Thromboxane A₂ (TXA₂), one of the eicosanoids, is an arachidonate product and exhibits its biological activity via G protein-coupled TXA₂ receptors, i.e., TP receptors. Although TXA₂ is well known to be a potent vasoconstrictor, the mechanism underlying the TP receptor-mediated vascular contraction remains to be fully elucidated. The present study was aimed to elucidate the mechanism, particularly focusing on lipid signaling.

U46619, a stable TXA₂ analog, induced vascular contraction consisting of two components in isolated ring preparations of rat aorta. The contraction induced by low-concentration U46619, i.e., at less than 100 nM, was highly sensitive to verapamil, a voltage-dependent Ca²⁺ channel (VDCC) blocker, whereas that induced by higher concentrations of U46619 was less sensitive to verapamil. Moreover, the former was abolished in a 0 Ca²⁺/EGTA extracellular solution, whereas the latter was only partially inhibited under the condition.

We first investigated the mechanism underlying the extracellular Ca²⁺-dependent contraction induced by low-concentration U46619. U46619 at 10 or 20 nM induced a sustained contraction of rat aorta, which was inhibited by ONO-RS-082, a non-selective phospholipase A₂ (PLA₂) inhibitor, and bromoenol lactone, a selective Ca²⁺-independent PLA₂ (iPLA₂) inhibitor, but not by arachidonyl trifluoromethyl ketone, a selective cytosolic PLA₂ (cPLA₂) inhibitor. U46619 was also shown to stimulate arachidonate release in rat aorta, which was detected by liquid chromatography-Mass spectrometry. Moreover, the U46619-induced contraction remaining in the presence of verapamil was largely inhibited by ONO-RS-082 and 2-APB, a cation channel blocker.

Taken together with the results obtained in the present study, we propose the following mechanism: Low-concentration U46619 stimulates Ca²⁺ influx through VDCC and 2-APB-sensitive cation channels, which is primarily mediated by iPLA₂ signaling, thereby eliciting a sustained contraction of rat aorta.