SUPPLEMENTAL INFORMATION:

Interactions of 2-O-Arachidonylglycerol Ether and Ibuprofen with The Allosteric and Catalytic Subunits of Human Cyclooxygenase-2

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Supplemental Fig. S1: Proposed pathway operating to form the major fragment ion from Peak C obtained by MS/MS.
Fig. 2S. Instantaneous inhibition by S-ibuprofen of 2-AG ether oxygenation by huPGHS-2 variants having an Arg-120 substitution in \( E_{\text{allo}} \) or \( E_{\text{cat}} \). Assays of \( \text{O}_2 \) consumption using an \( \text{O}_2 \) electrode were performed as described in Experimental Procedures on the parent publication using 50 \( \mu \text{M} \) 2-AG ether as the substrate. huPGHS-2 heterodimer variants were described previously (15). The designations indicate the mutations in the subunits. For example, Y385F R120A/Native indicates a PGHS-2 molecule having one subunit with both Y385F and R120A mutations and the other subunit as having no mutations (i.e. a Native subunit); R120A/R120A indicates a PGHS-2 molecule with mutations in both subunits. Results are shown for a single experiment involving triplicate determinations. Relative COX-2 activities derived from these data in Fig. 2S are compared in Fig. 6 of the parent publication. Specific activities for the huPGHS-2 variants with 2-AG ether (50 \( \mu \text{M} \)) were as follows: For Native, 25 units/mg; for R120A/R120A, 17 units/mg; for Y385F/Native, 19 units/mg; for Y385F R120A/Native, 24 units/mg and Y385F/R120A, 22 units/mg.