

Table S1: Comparison between purified WT and mutants of basal and stimulated activities. The values correspond to Fig.3 and include the fold stimulation of each activator compared to basal activity for WT and mutants and percentage of basal activity of the mutants compared to WT. Specific activity is expressed in $\text{nmol}\cdot\text{min}^{-1}\cdot\text{mg}^{-1}\pm\text{SEM}$. Stim.: stimulation. *: The higher basal activity of R40A is explained by a higher amount of sGC in the fraction used, i.e. the fraction eluted with high salt ($\sim 500\text{mM}$) with some absorbance at 431nm as well.

	BASAL	% WT	DEA-NO, 1 μM	fold stim.	YC-1, 100 μM	fold stim.	DEA-NO, 1 μM + YC-1, 10 μM	fold stim.	PPIX, 10 μM	fold stim.
WT	73.1 \pm 2.0	100	4310.0 \pm 298.8	59.0	561.5 \pm 75.2	7.9	6566.9 \pm 208.2	89.8	886.1 \pm 99.6	12.1
T110A	138.0 \pm 18.8	194	4769.2 \pm 327.3	34.5	465.2 \pm 13.9	3.4	5548.9 \pm 803.1	40.2	3843.8 \pm 658.2	27.9
R116A	87.5 \pm 14.8	119	2181.9 \pm 424.6	24.9	299.0 \pm 43.0	3.4	4007.0 \pm 641.0	45.8	2003.0 \pm 126.9	22.9
I41A	103.8 \pm 9.4	140	2187.2 \pm 218.6	21.1	578.5 \pm 59.6	5.6	5973.4 \pm 705.2	57.4	1700.4 \pm 122.7	16.3
R40A *	293.2 \pm 15.4	----	369.5 \pm 70.2	1.9	289.3 \pm 39.9	1	515.3 \pm 43.7	1.7	386.3 \pm 66.4	1.3