

Supplementary information

Non-canonical chemical feedback self-limits nitric oxide-cyclic GMP signaling in health and disease

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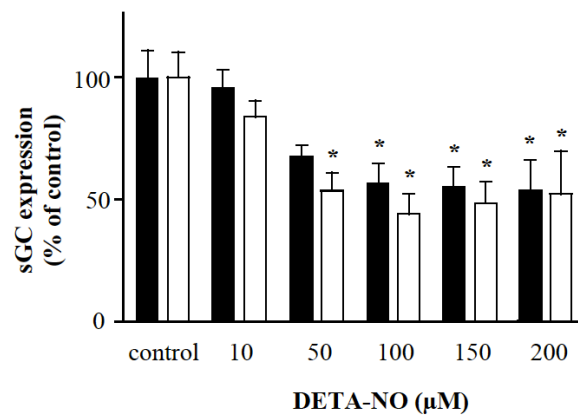
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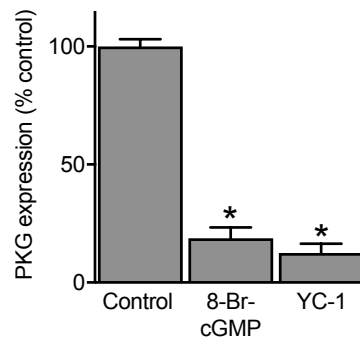
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Key words: endothelial cells; soluble guanylate cyclase; nitric oxide; cyclic guanosine mono phosphate; redox regulation; Bay 58-2667, apo-sGC

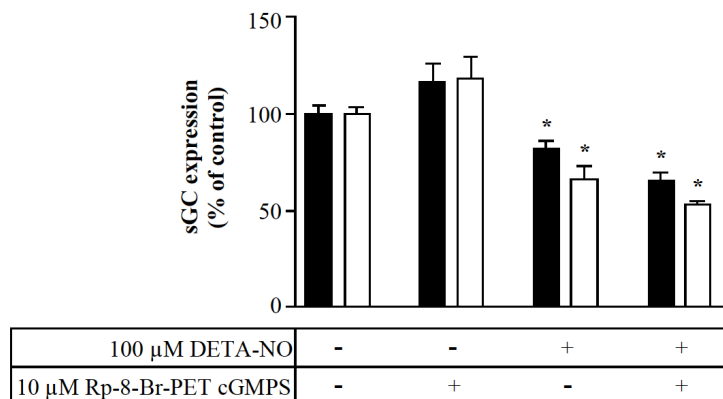
SUPPLEMENTARY FIGURES



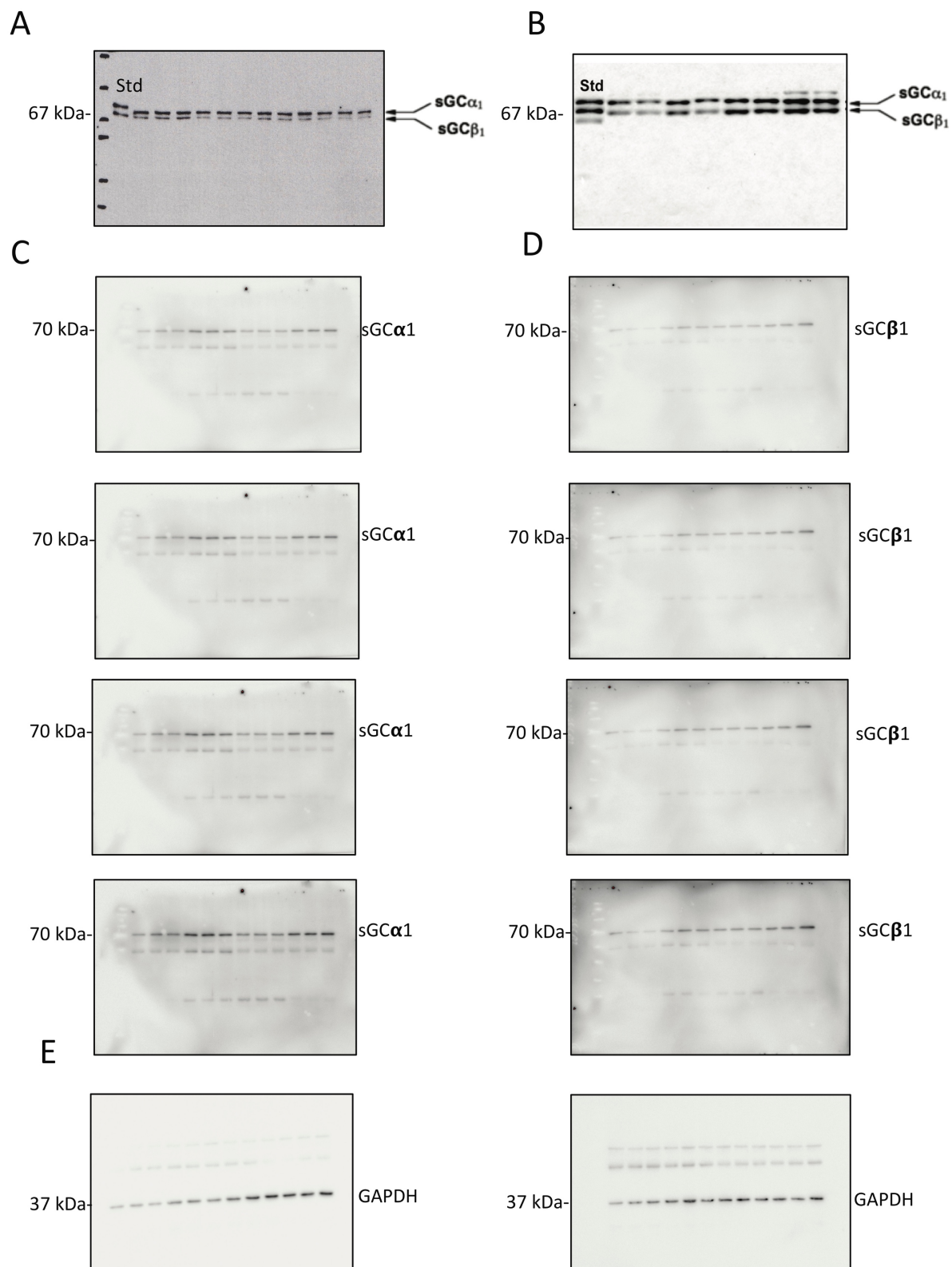
Supplementary Fig. S1. 100 μ M DETA-NO is the maximal concentration to downregulate sGC. Treatment of PPAECs for 72h with different concentrations of DETA-NO showed that 100 μ M is the maximal concentration to downregulate both sGC α_1 (solid bars) and sGC β_1 (open bars) (N=6). *P< 0.05 by one-way analysis of variance (ANOVA), data represent means \pm SEM.



Supplementary Fig. S2. PPAECs display intact NO-cGMP-PKG signaling since treatment with 8-Br-cGMP (500 μ M), or YC-1 for 72hrs reduced PKG expression (N=6-9). *P< 0.05 by one-way analysis of variance (ANOVA), data represent means \pm SEM.



Supplementary Fig. S3. Inhibition of PKG does not prevent DETA-NO induced downregulation of sGC α_1 and sGC β_1 . Treatment of PPAECs for 72h with either 100 μ M DETA-NO, or with the PKG inhibitor Rp-8-Br-PET-cGMPS (10 μ M), or a combination thereof resulted in down regulation of sGC α_1 (solid bars) and sGC β_1 (open bars) level that could not be reversed by Rp-8-Br-PET-cGMPS (N=6). *P< 0.05 by one-way analysis of variance (ANOVA), data represent means \pm SEM.



Supplementary Fig. S4. Representative Immune detection blots of controls, sGC α ₁ and sGC β ₁. A. PPAECs. B. Mouse. C-E. ARDS (Multiple exposures).