

SUPPLEMENTARY ONLINE DATA

Proteasomal interaction as a critical activity modulator of the human constitutive androstane receptor

Tao CHEN*, Elizabeth M. LAURENZANA*, Denise M. COSLO*, Fengming CHEN* and Curtis J. OMIECINSKI*¹

*Center for Molecular Toxicology and Carcinogenesis, Department of Veterinary and Biomedical Sciences, 101 Life Sciences Building, Pennsylvania State University, University Park, PA 16802, U.S.A.

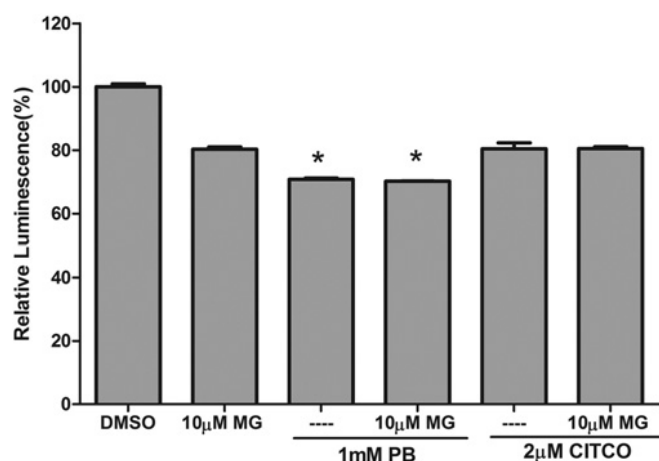


Figure S1 Effect of chemical treatments on the viability of HPHs

HPHs in 24-well plates were treated for 24 h with vehicle control (0.1% DMSO), 1 mM PB or 2 µM CITCO in the presence and absence of 10 µM MG-132 (MG). Cytotoxicity of HPHs was measured with the Cell Titer-Glo Luminescent Cell viability kit (Promega) according to the manufacturer's instructions. Significantly different from DMSO control (*), $P < 0.05$. In addition, the cells were routinely observed with phase contrast microscopy over the course of treatments and no obvious changes were noted in cell morphology or other adverse effects due to any treatments compared with vehicle controls.

¹ To whom correspondence should be addressed (email cjo10@psu.edu).

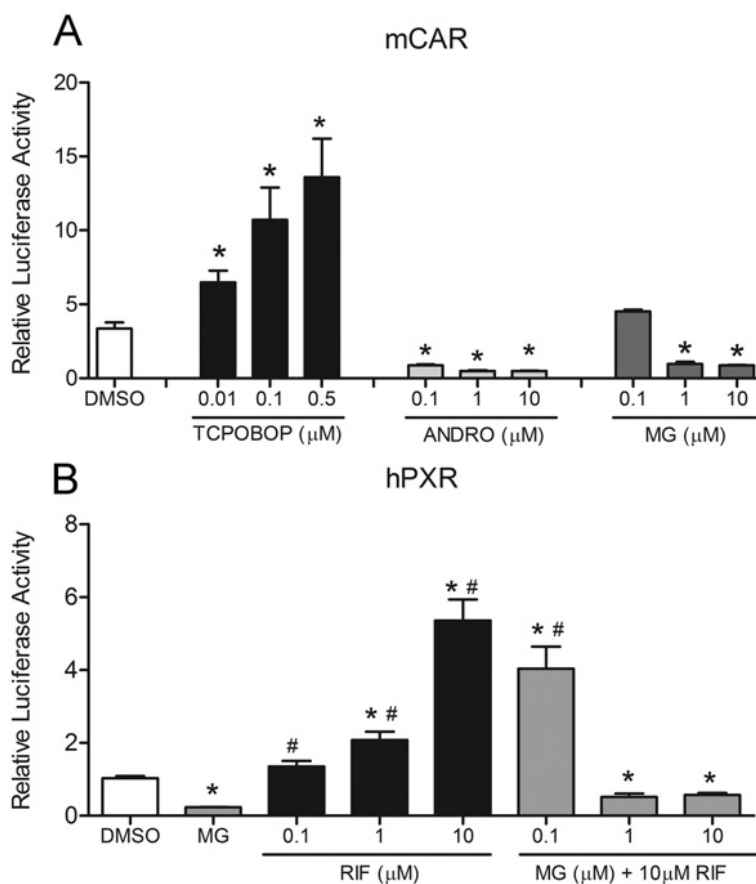


Figure S2 Effect of MG-132 on mCAR and human PXR activation in HepG2-based reporter assays

HepG2 cells were transfected with mCAR or hPXR expression vector along with the CYP2B6-PBREM/XREM reporter and treated for 24 h as described below. Luciferase activities were determined and expressed relative to vehicle control. Data represent the means \pm S.D. ($n = 3$). **(A)** mCAR-transfected cells were treated with vehicle control (0.1% DMSO), mCAR-specific activator (TCPOBOP), mCAR inhibitor (Andro) and MG-132 (MG) at the indicated concentrations. Significantly different from DMSO (*), $P < 0.05$. **(B)** hPXR transfected cells were exposed to vehicle (0.1% DMSO), hPXR-specific activator (Rif), MG-132 alone or co-treated with 10 μ M Rif at concentrations indicated. Significantly different from DMSO (*), MG (#), $P < 0.05$.

Received 20 May 2013/29 October 2013; accepted 14 November 2013

Published as BJ Immediate Publication 14 November 2013, doi:10.1042/BJ20130685