Co-transfection of p68/p72 with p53 has no significant effect on levels of p53 protein.

Western blots showing levels of transfected p68, p72 and p53 in H1299 cells. In each case two separate sets of transfections were carried out, in which 10 or 25ng of p53 were transfected in conjunction with increasing amounts of p68/p72 (0-10μg) and the standard 2.5μg of PG13 as used for the co-activation assays. A) Transfection of myc-tagged p68 and p53; B) Transfection of myc-tagged p72 and p53.

For each experiment the western blot probed for p53 was subsequently probed for actin to confirm equal loading of protein. In this experiment, to clearly show levels of transfected p68/p72 and to distinguish these from the endogenous proteins, we used myc-tagged versions of these proteins and probed using an antibody against the myc epitope; this additionally showed that the transfected p68 and p72 were expressed at similar levels. Transfection of untagged p68/p72 similarly had no effect on p53 protein levels (data not shown). *non-specific protein band reacting with the 9E10 (anti-myc) antibody. **residual p53 signal remaining after re-probing blot for actin.