

Fig. I. Silencing of human PP1 catalytic subunits via siRNA. A549 cells were transfected with 100 nM siRNA targeting individual PP1 isozymes. The expression levels of all three PP1 isoforms were analyzed by Western blotting 24, 48, and 72 h post-transfection. The PP1 isozyme targeted by the siRNA is identified by the frame. Loading controls for β-actin have been included for each of the Western blots analyzing the targeted isozyme. The results indicate that each of the siRNAs i) were between 75 and 95% effective in inhibiting their target, and ii) did not downregulate non-targeted PP1 isoforms.

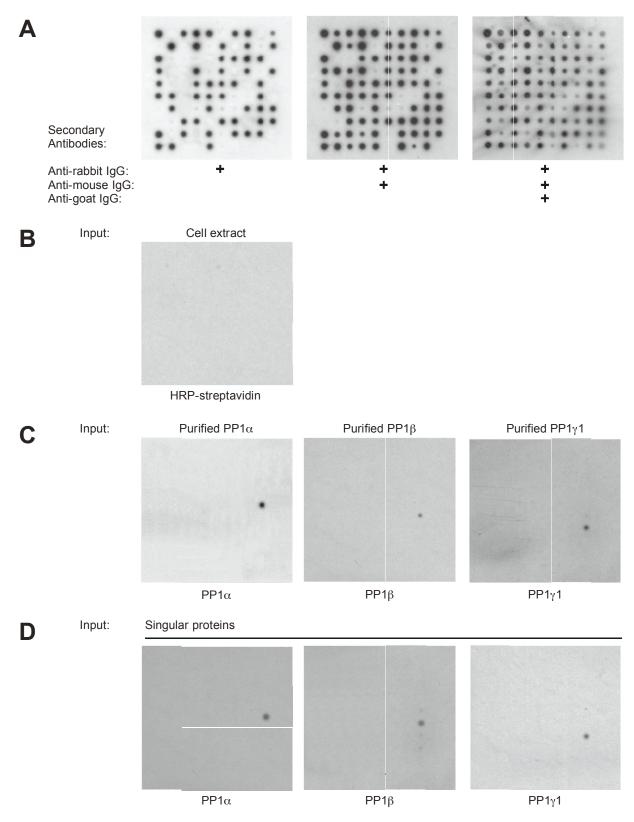


Fig. II. Control incubations to evaluate the antibody arrays. Controls to rule out false negative results: (A) Incubation of exposed membranes to secondary antibodies verify the presence and position of the primary antibodies. Controls to rule out false positive signals: (B) Incubation of one arrray with HRP-streptavidin only leaves a virtually blank membrane. (C) Incubation of one array with purified PP1 isoforms produces one spot in the position (i5, i6, or i7, respectively) of the immobilized PP1 antibodies. (D) After incubation with lysates prepared under conditions that destroy protein complexes, added PP1 antibodies only react with their cognate antigens.