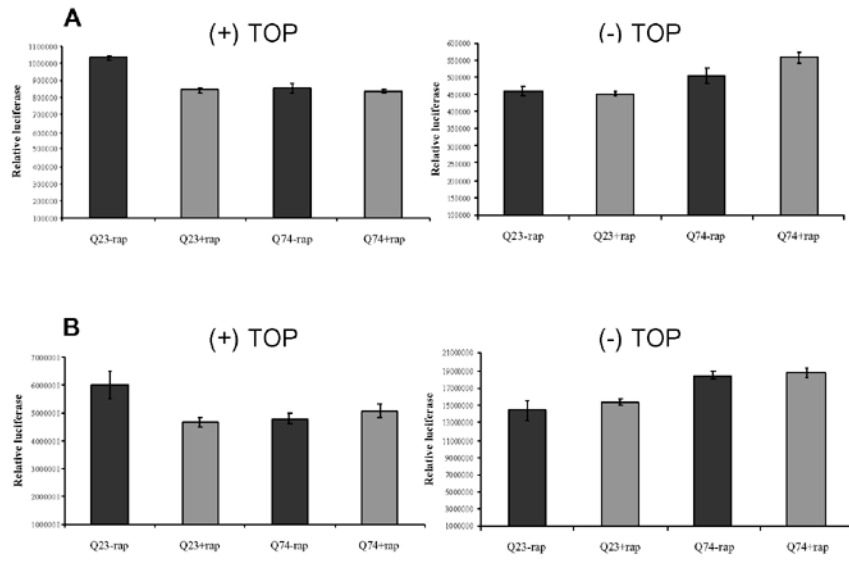


**Figure 4**



## Supplementary figure 4

### Mutant huntingtin dysregulates TOP-dependent translation.

**A.** PC12 cells stably expressing Q23 or Q74 were transfected with a luciferase construct containing a 5'-untranslated region from the eEF2 gene with a terminal oligopyrimidine tract ((+) TOP)<sup>22</sup> or without a TOP sequence ((-) TOP), along with  $\beta$ -galactosidase to control transfection efficiency. Transfected cells were maintained in serum free medium with doxycycline for 36 hours following which the cells were stimulated with 15% serum with or without rapamycin for 3h. Luciferase assays were performed according to a standard protocol<sup>18</sup>. TOP-luciferase activity is significantly reduced with addition of rapamycin in Q23 but unaffected in the mutants. Rapamycin does not reduce control-luciferase. Raw data are shown for one representative experiment in sextuplicate. **B.** COS-7 cells were co-transfected with Q23 or Q74 and a different non-typical TOP-luciferase vector or control-luciferase and  $\beta$ -galactosidase for 48 hours and experiment was performed as in A<sup>18</sup>. The results obtained with this vector were similar to that obtained in A.