**RESULTS (CONTINUED)**

**Gene Thrapy:** Western blot analysis of vector treated retinal tissue lyses using GT-355 antibody (a monovalent antibody specific for glutamylation, Figure 4), detected the full-length (~200Kda) and ~140Kda, suggesting that this functionality critical post-translational modification is detected in both full length (~200Kda), or truncated (~140Kda) RPGRc.

**Protein Interaction:** It has been reported that RPGR mutations impair the interaction between RPGR and RPGRIP1 in vivo and that this in turn disrupts normal RPGR function in photoreceptor cells. We assessed the interaction of RPGR/ORF15 and RPGRIP1 in HEK 293 cells transfected with the respective expression constructs, pTR-SmCBA-RPGRc or C-terminus flag-RPGRIP1. Co-immunoprecipitation studies in transfected HEK293 cells demonstrated that full length RPGR interacts with RPGRIP1 as expected (Figure 5). No interaction was detected when either of the binding partners, RPGRc or RPGRIP1, was co-expressed with a GFP construct.

**Conclusions:**

Codon-optimized RPGR cDNA in the rAAV2tYF-GRK1-hRPGRc vector maintained its integrity during transduction and transcription in vitro and in vivo. Vector-expressed RPGR protein was both glutamylated and able to bind to its partner RPGRIP1, each being required for RPGR to fulfill its functional role in retinal cillum.

**REFERENCES**


