**Revision 1: MCR-15-0130**

**Patient mutation directed shRNA screen uncovers novel bladder tumor growth suppressors**

**SUPPLEMENTAL FIGURE LEGENDS**

Jonathan Hensel1\*, Jason E. Duex1\*, Charles Owens1, Garrett M. Dancik2,

Michael G. Edwards3, Henry F. Frierson4, and Dan Theodorescu1,5

1 Departments of Surgery (Urology) and Pharmacology, University of Colorado, Aurora, Colorado, USA, 80045

2 Department of Mathematics and Computer Science, Eastern Connecticut State University, Willimantic, Connecticut, 06226

3 Division of Pulmonary Sciences and Critical Care Medicine, University of Colorado, Aurora, Colorado, USA, 80045

4 Department of Pathology, University of Virginia, Charlottesville, 22903

5 University of Colorado Comprehensive Cancer Center, Aurora, Colorado, USA, 80045.

\* Equal contribution

**Running Title:** IQGAP1 negatively regulates human bladder tumor growth

**Statement of implication:** This study used gene mutation information from patient-derived bladder tumor samples to inform the development of a screen used to identify novel tumor growth suppressors. This included identification of the protein IQGAP1 as a potent bladder cancer growth suppressor.

**Keywords:** IQ motif containing GTPase activating protein 1 (IQGAP1), bladder neoplasms, Transforming Growth Factor Beta Receptor II, tumor growth suppressor

**Financial Support:** Supported by National Institutes of Health grant CA143971 to DT.

**Corresponding author:** Dan Theodorescu, University of Colorado Comprehensive Cancer Center, Aurora, CO 80045, Tel: 303-724-7135, Fax: 303-724-3162, E-Mail: [dan.theodorescu@ucdenver.edu](mailto:dan.theodorescu@ucdenver.edu).

**Conflict of Interest:** The authors declare no conflict of interest

**Supplemental Fig 1. Knockdown of IQGAP1 has no effect on ERK1/2 activity when cells were grown in monolayer culture.** (A) T24 cells expressing IQGAP1 shRNA were analyzed by western blot for ERK1/2 activity using a phospho-specific antibody as well as an antibody that recognizes total ERK1/2 protein. The blots were also analyzed with α-actinin to confirm equal loading of samples.

**Supplemental Fig 2. Knockdown of TGFBR2 reduces the anchorage independent growth induced by loss of IQGAP1.** (A) Treatment of 253J cells with TGFBR2 siRNA results in a 80% decrease in TGFBR2 expression as assessed by qPCR. (B) Knockdown of TGFBR2 expression in T24 results in abrogation of the previously noted increase in soft agar colony formation that results from loss of IQGAP1. \* p<0.05, \*\* p<0.01.