

Supplementary Information

Enhanced metastatic potential in the MB49 urothelial carcinoma model

Yu-Ru Liu¹, Peng-Nien Yin¹, Christopher R. Silvers¹, Yi-Fen Lee^{*1,2}

Departments of ¹Urology and ²Pathology, University of Rochester Medical Center, Rochester NY, 14642, USA

Corresponding Author: Yi-Fen Lee, Department of Urology, University of Rochester Medical Center, 601 Elmwood Ave, Box 656, Rochester, NY 14642. Phone: (585) 275-9702. E-mail: yifen_lee@urmc.rochester.edu

⁺The first two authors contributed equally to this work.

Supplementary Table 1. Incidence of lung metastasis in a pilot test of MB49 sub-clones.

| MB49 line | mice with lung metastases |
|------------------|----------------------------------|
| parental | 0/2 |
| F1 | 0/3 |
| F2 | 0/3 |
| F3 | 0/3 |
| S1 | 3/3 |
| S2 | 1/2 |
| S3 | 0/4 |
| S4 | 1/4 |
| S5 | 1/6 |

The lung metastasis incidence of eight candidate MB49 sub-clones was measured 10-21 days following subcutaneous implantation into C57BL/6 mice.

Supplementary Table 2. Primer sequences used in qPCR.

| | Gene name | Forward | Reverse |
|-------------------------------|---------------------------------------|------------------------------|-------------------------------|
| | DCN | TTCCTACTCGGCTGT GAGTC | AAGTTGAATGGCAG AACGC |
| Basal marker genes | CD44 | TTTGCAATGCAGTC AACAGTC | GTTACACCCCAATC TTCATGTCCAC |
| | KRT5 | TCTGCCATCACCCC ATCTGT | CCTCCGCCAGAACT GTAGGA |
| | KRT6B | GCTCAAACATAGGC TCCAGGTT | CCATCAAGAGTCAA ACTAGCCAC |
| | KRT14 | AGCGGCAAGAGTGA GATTTCT | CCTCCAGGTTATTCT CCAGGG |
| CSC marker genes | ALDH1A1 | TGATGTTGTCAGCC CAGTGC | TTGCGACTGTCTTG AGC |
| | CD49 | TCTATTCCTGTTGCC GTCACAA | CGATGACAGCATT CCCGATA |
| | CK14 | CGGCCCACTGAGAT CAAAGAC | AAATGCCAAGCCCC ATTTTAGG |
| | CD90 | CTGCAGCTAGGGGA GTCCAG | GATGACCCCTAATG TCCAGGTC |
| EMT marker genes | ITGA5 | CTTCTCCGTGGAGTT TTACCG | GCTGTCAAATTGAA TGGTGGTG |
| | ITGB1 | ATGCCAAATCTTGC GGAGAAT | TTTGCTGAGATTGG TGACATT |
| | ACTA2 (αSMA) | GTCCAGACATCAG GGAGTAA | TCGGATACTTCAGA GTCAGGA |
| | FN1 | GCTCAGCAAATCGT GCAGC | CTAGGTAGGTCCGT TCCCACT |
| | CDH2 (N-cadherin) | TCGCTGCTTTCATAC TGAAC TTT | AGCGCAGTCTTACC GAAGG |
| | TWIST1 | GGACAAGCTGAGCA AGATTCA | CGGAGAAGGCGTAG CTGAG |
| Internal control genes | GOT1 | GCGCCTCCATCAGT CTTTG | ATTCATCTGTGCGG TACGCTC |
| | HPRT | TCAGTCAACGGGGG ACATAAA | GGGGCTGTACTGCT TAACCAG |
| | GAPDH | AGGTCGGTGTGAAC GGATTTG | TGTAGACCATGTAG TTGAGGTCA |