

Supplemental Data

Translational profiles of medullary myofibroblasts during kidney fibrosis

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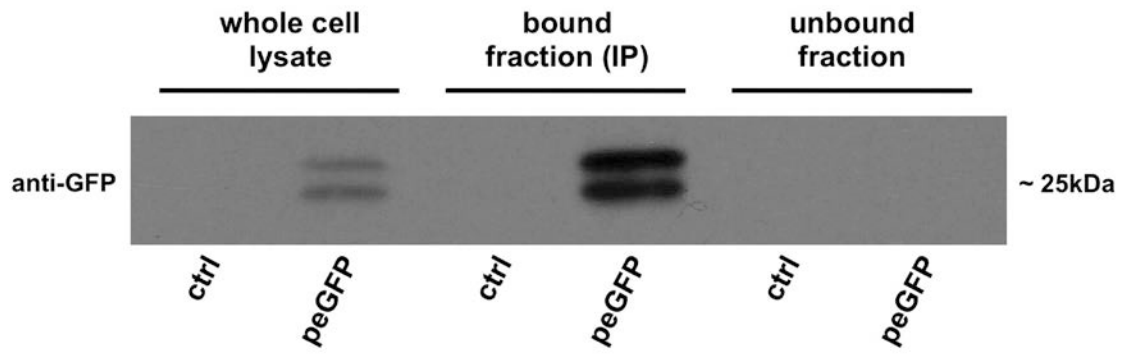


Figure S1. Validation of immunoprecipitation efficiency in eGFP-transfected HEK293 cells. HEK293 cells were transiently transfected with an eGFP expression plasmid and cell lysates immunoprecipitated with anti-GFP monoclonal antibody clones C8 and F7. Immunoblotting documented a strongly enhanced GFP signal in the bound, immunoprecipitated (IP) fraction when compared to the whole cell lysate and the GFP-depleted unbound fraction, respectively, indicating efficient immunoprecipitation of GFP.

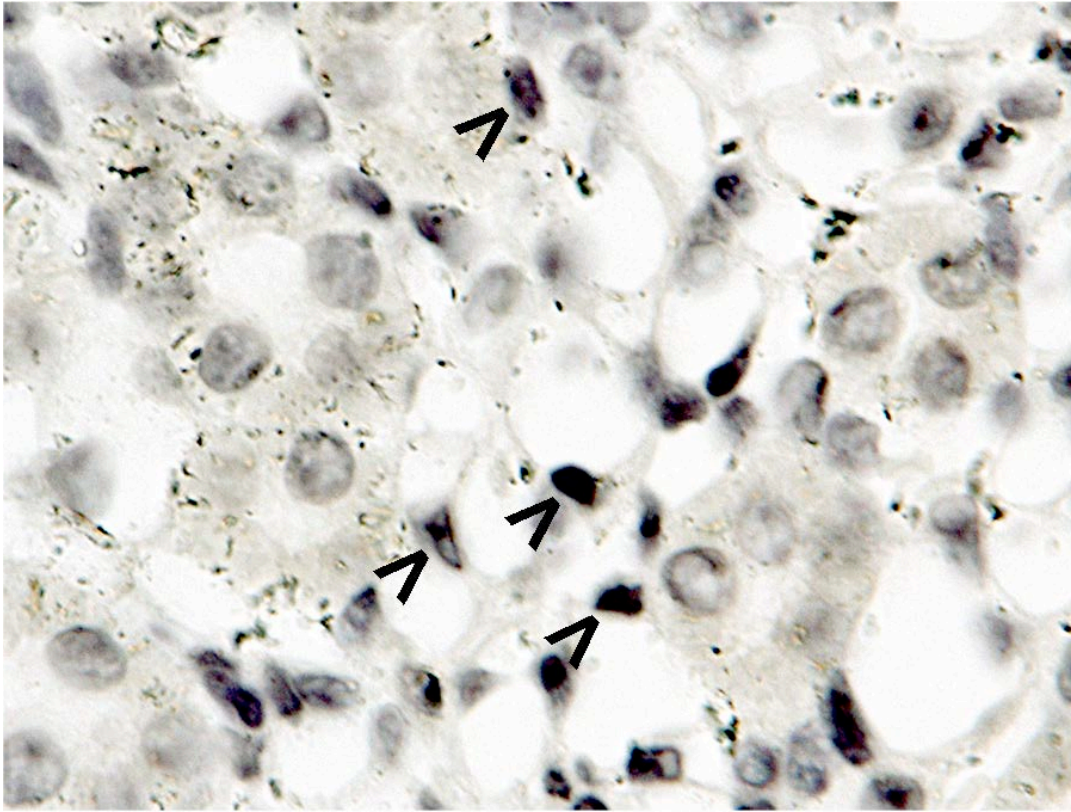


Figure S2. Strong interstitial eGFPL10a expression in kidney medulla from Peri/Fibro^{TRAP} mice. Immunohistochemistry using an anti-GFP antibody reveals strong interstitial reactivity in medulla, consistent with results by epifluorescence.

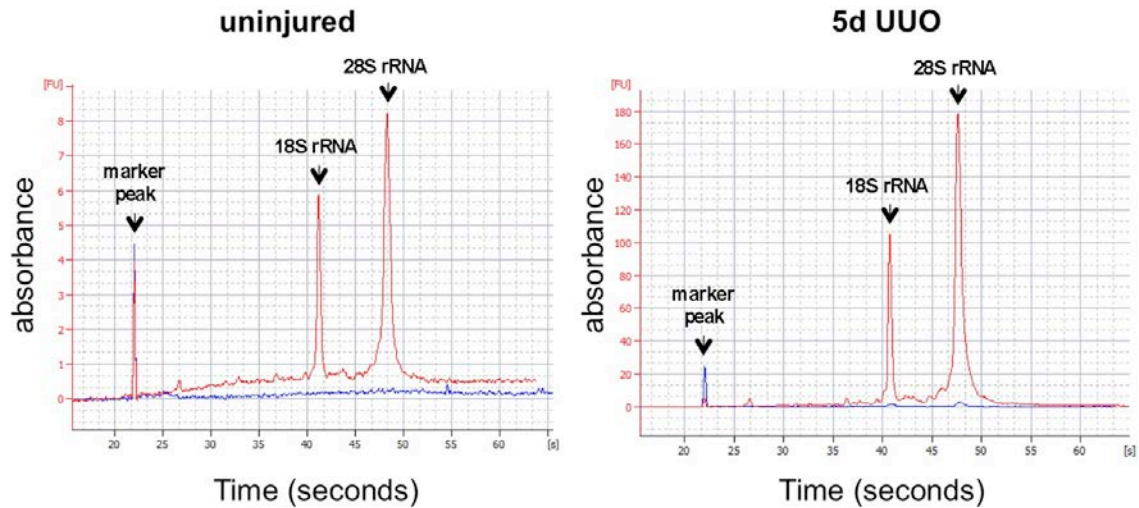


Figure S3. Validation of polysomal RNA isolation using Peri/Fibro^{TRAP} mice and affinity purification procedure. Electropherograms recorded by Bioanalyzer (PicoChip, Agilent Technologies) of TRAP-isolated RNA from uninjured (*left*) and fibrotic (*5d UUO*, *right*) kidney medulla from Peri/Fibro^{TRAP} mice (red traces) and wildtype controls (blue traces). Significant amounts of high quality RNA (RIN > 9.0) are detected only in samples from Peri/Fibro^{TRAP} mice. Note higher RNA yield in fibrotic compared to uninjured kidney indicating substantial induction of renal GFP-L10a expression in Peri/Fibro^{TRAP} animals during fibrogenesis. Y-axis: absorbance in arbitrary fluorescence units (FU); *arrows* mark identity of peaks.

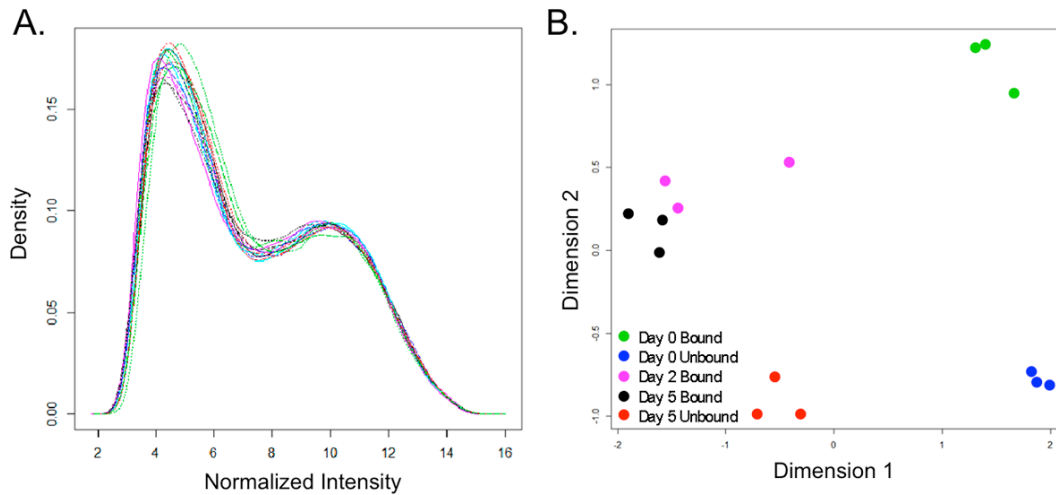


Figure S4. Normalized density curves and multi-dimensional scaling (MDS) plot for all microarrays. **A.** All 15 microarrays were normalized by the Robust Microchip Average and the normalized density curves reveal only small variations between samples, indicating good quality and adequate normalization. **B.** In order to compare the similarity of expression profiles among and between groups, all arrays are represented by MDS plot. As expected, within all groups there is good clustering indicating a high degree of similarity between biological replicates. The exception is the Day 2 bound group which clusters with Day 5 Bound, which may reflect variability in initiation of the fibrotic response. Day 0 and Day 5 groups all show excellent separation from one another and were used for downstream analysis.