Figure S1. Dox-induced reconstituted TGFBR2 expression and signaling. Western blot analysis confirmed the dox (0.5 $\mu$g/ml)-induced expression of TGFBR2. The TGF-β1 ligand (10 ng/ml) was present in both conditions. Increased phosphorylation levels of Smad2 (pSmad2) were observed in cells cultured in the presence of dox (+dox) compared with those in cells grown in the absence of dox (-dox). Total Smad2 and β-actin proteins served as loading controls. Protein sizes are indicated. Dox, doxycycline; TGFBR2, transforming growth factor β receptor type 2; pSmad, phosphorylated Smad.

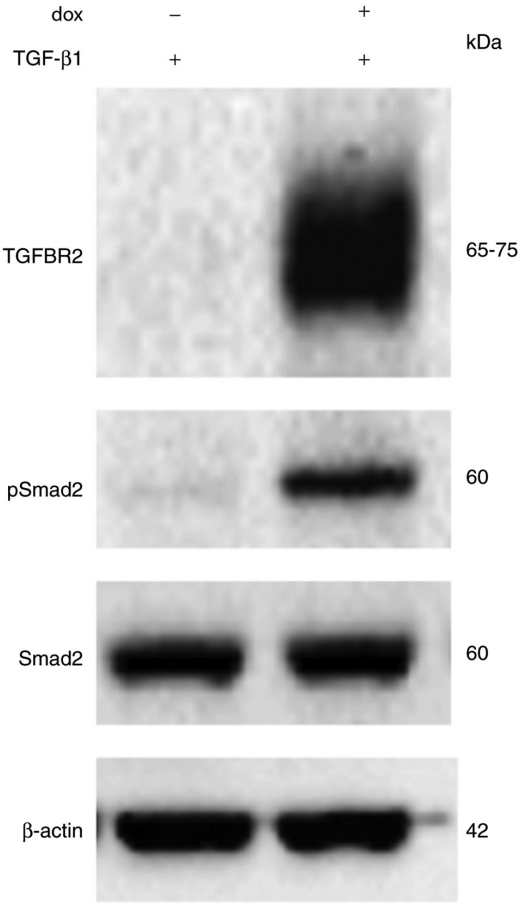

Figure S2. Length distribution of cellular and EV small RNAs based on sequencing data. Sequence lengths and corresponding read count numbers are presented as mean values calculated from four biological replicates per group. TGFBR2, transforming growth factor β receptor type 2; pT, TGFBR2 proficient; dT, TGFBR2 deficient; EVs, extracellular vesicles; nt, nucleotides.

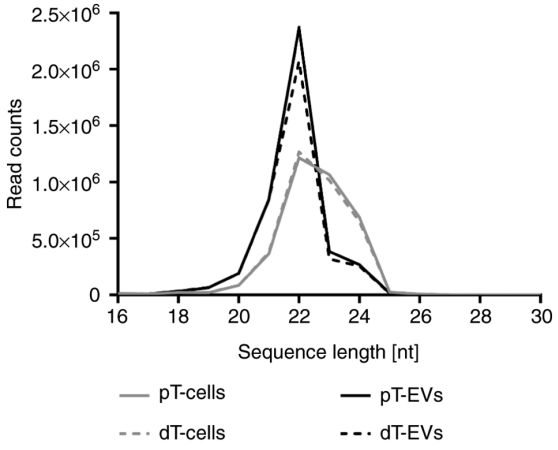

Figure S3. *In silico* network analysis of TGFBR2-regulated miRNAs identified in EVs and predicted target genes. Network analysis was performed *in silico* using miRNet (24). Interactions of 10 TGFBR2-dependent miRNAs with predicted target genes (n=1,022) highlighting candidates (n=18) associated with TGF-β receptor complex signaling (25). TGFBR2, transforming growth factor β receptor type 2; miR/miRNA, microRNA.

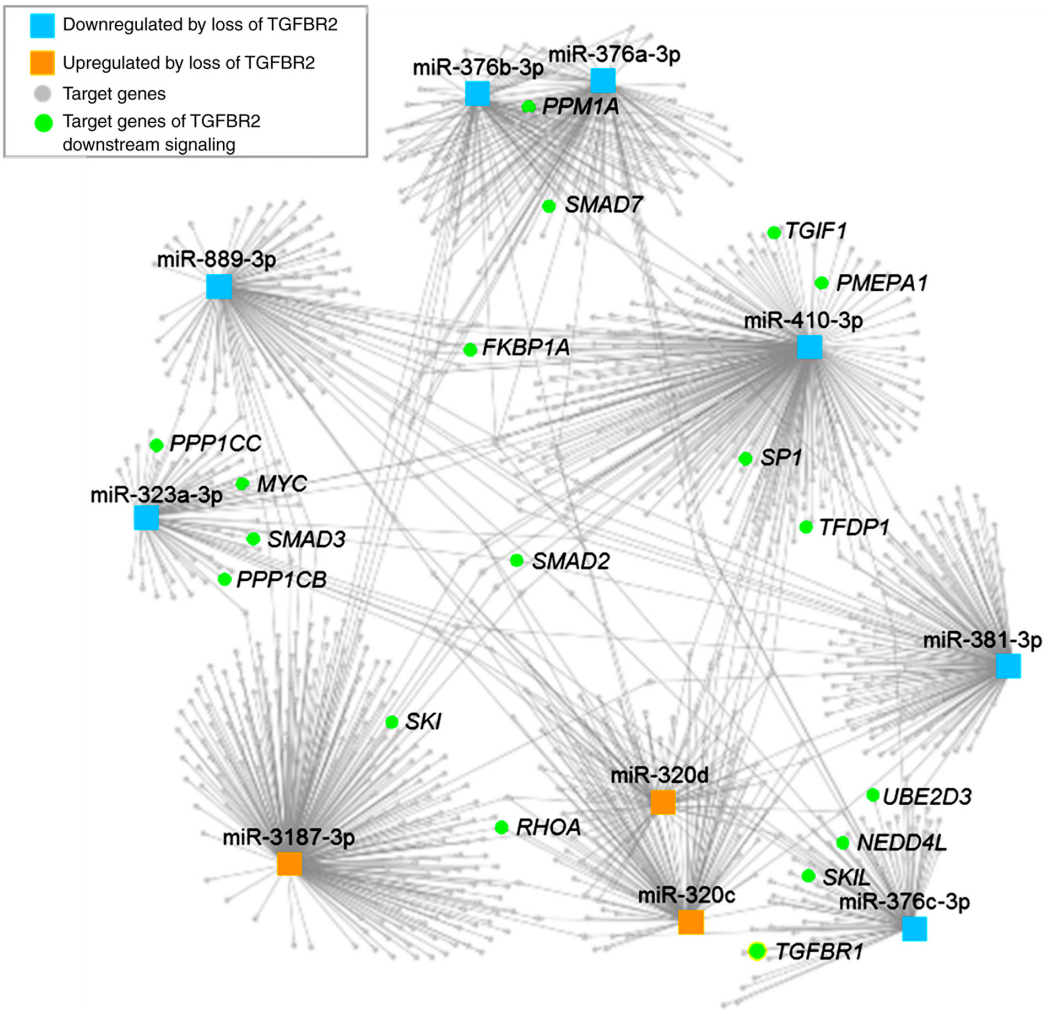

Table SI. Functional enrichment analysis of TGF-β receptor type 2-dependent microRNAs identified in extracellular vesicles.

| Reactome database pathway | P-value |
|---|---|
| Signaling by TGF-β receptor complex | 0.0002 |
| Signaling by NOTCH | 0.0003 |
| Signaling by NGF | 0.0003 |
| NGF signaling via TRKA from the plasma | 0.0003 |
| Cellular response to stress | 0.0004 |
| Transcriptional activity of SMAD2/SMAD3 | 0.0011 |
| Oncogene induced senescence | 0.0011 |
| Pre-NOTCH transcription and translation | 0.0011 |
| Gene expression | 0.0012 |
| Downstream signal transduction | 0.0014 |

Functional enrichment analysis based on hypergeometric testing and reactome database identified signaling by TGF-β receptor complex as the most affected (P=0.0002) pathway (24,25). TGF-β, transforming growth factor-β; NGF, nerve growth factor; TRKA, neurotrophic receptor tyrosine kinase 1.