Carcinoma-associated fibroblast-derived lysyl oxidase-rich extracellular vesicles mediate collagen crosslinking and promote epithelial-mesenchymal transition via p-FAK/p-paxillin/ YAP signaling

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Supplementary methods

CCK-8 assay

Cell viability was measured by CCK-8 assay. Cells were seeded in a 96-well plate (1000 cells/well) and cultured overnight followed by treatment with CAF or NF CM for 72 h. CCK-8 reagent (Biosharp, Shanghai, China) mixed with DMEM/F12 was added in each well and incubated for 1 h. Then, the absorbance at 450 nm was measured using a microplate reader (Bio-Rad, Hercules, CA, USA). Each well was repeated at least 3 times, and the mean absorbance was used to assess cell proliferation.

Wound healing assay

Cells were seeded into a six-well plate (5×10^5 cells per well). Next day, a wound was created in each well using a P1000 pipette tip. Then, cells were treated with CAF and NF CM for 72 h. The wound area at the start and end of each experiment was recorded using an inverted microscopy (Olympus IX71). Each well was repeated at least 3 times, and the mean migration rate was used to assess cell migration ability.

Nanoparticle Tracking analysis (NTA)

Pelleted sEVs were resuspend in PBS. The concentration and size distribution of CAF-S1/S2/S3/S4 sEVs were analyzed using ZetaView (PERTICLE METRIX, German).