

Supplementary Materials of “Nanoscopic Approach to Study Early Stages of Epithelial to Mesenchymal Transition (EMT) of Human Retinal Pigment Epithelial (RPE) Cells *in vitro*”

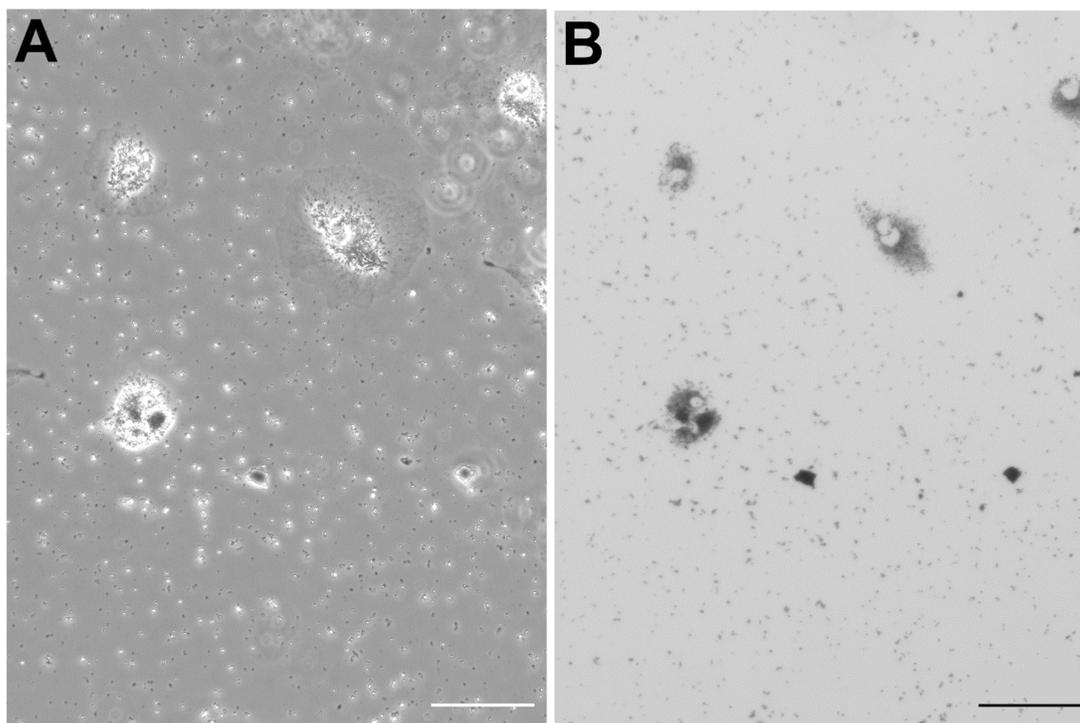


Figure S1. Phase contrast (**A**) and DIC (**B**) images of hRPE cells on day 3 in culture. The attached to the surface RPE cells spread significantly in size by forming lamellipodia (**A**), lose gradually the pigment (melanin granules or melanosomes) (recognized in **B** as dark dots) and expel them into the culture solution (seen in **A** and **B** as dots around the cells). The pigmentation of spread RPE cells is still present/conserved) and accumulated in the central part of cell body (**B**). Scale bars are 50 μm .

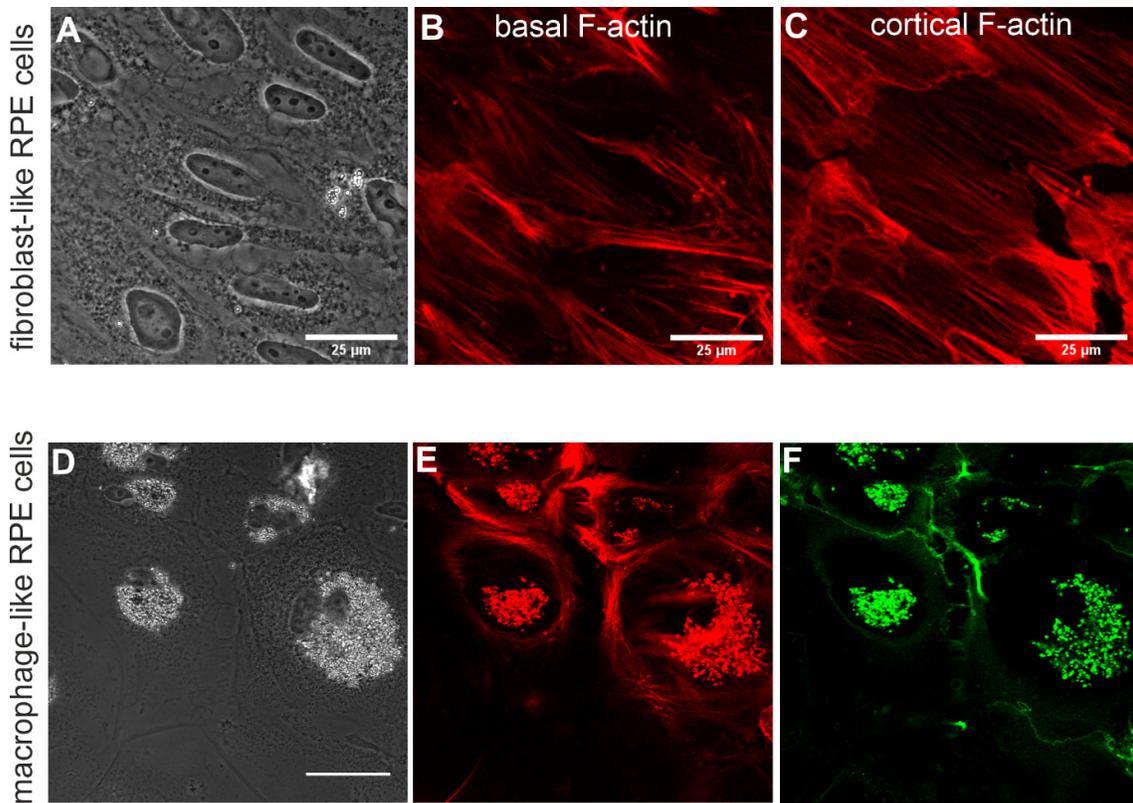


Figure S2. Simultaneously acquired phase contrast (A, D) and immunofluorescence images of fibroblast-like (upper panel) and pigmented macrophage-like RPE cells (lower panel). (B) and (C) show basal and cortical F-actin networks obtained from projection of corresponding z-stack images. (E) and (F) demonstrate melanosome autofluorescence in red-orange (excitation with 540-nm light) and green (excitation with 488-nm light) of macrophage-like RPE cells, which were dual labelled with rhodamine phalloidin (F-actin) and concavalin A (Con A) conjugated with fluorescein isothiocyanate (FITC) dye. Scale bar in (D) is 50 μm .

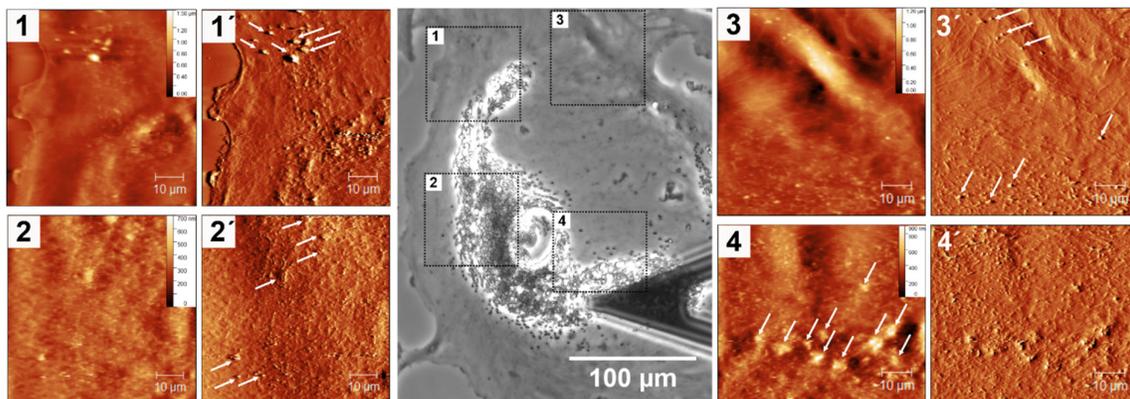


Figure S3. Phase contrast image (in the middle) of one huge macrophage-like cell with AFM (height and deflection) images collected on four different places on cell surface (1–4). Filamentous structure corresponding to the cortical cytoskeleton with stable protrusions can be clearly identified in deflection images (1'–4'). Bound (entering) into the cell membrane (arrows in 1' and 3') and phagocytosed melanosomes (arrows in 2' and 4) can be recognized as well.