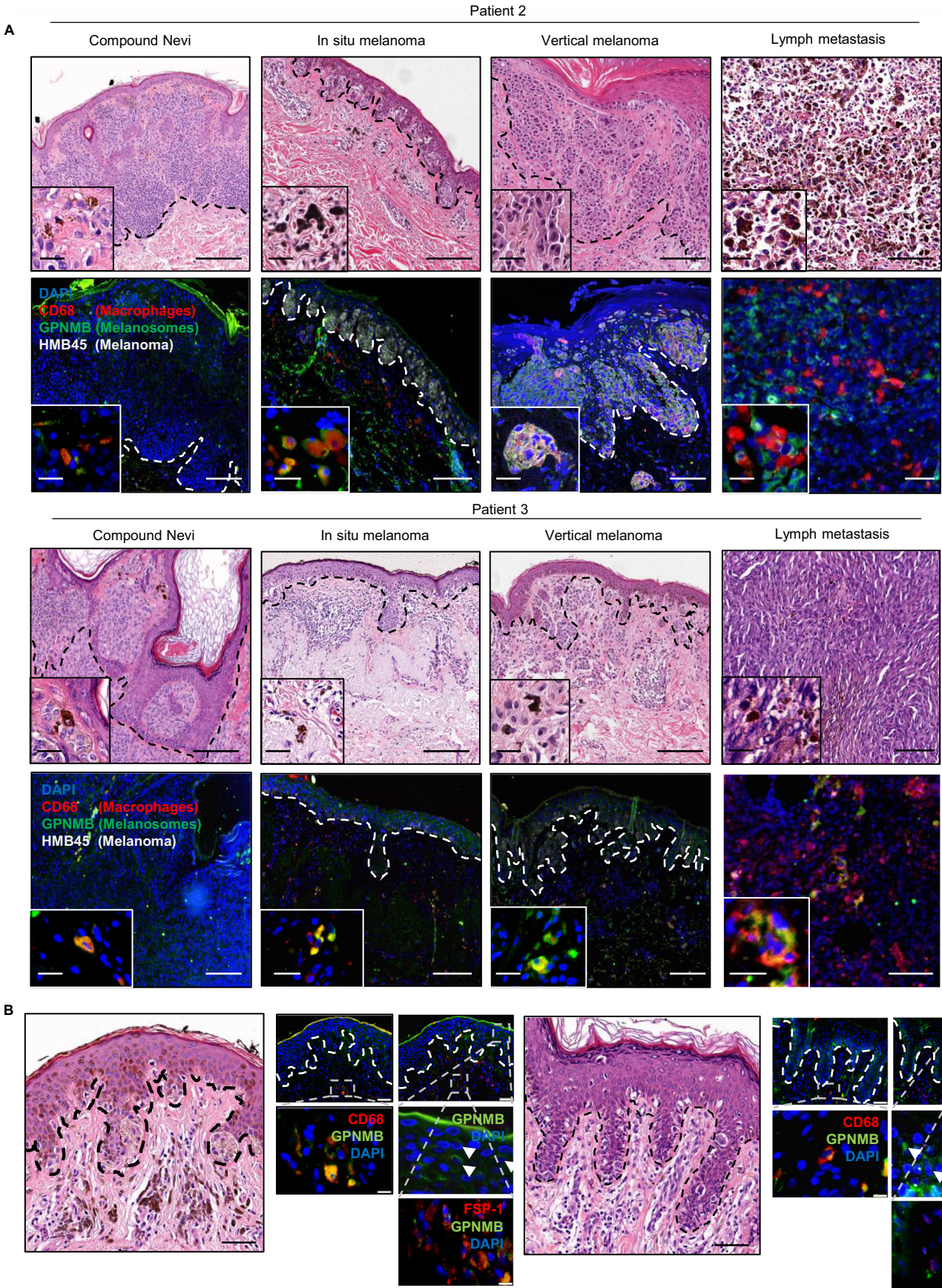
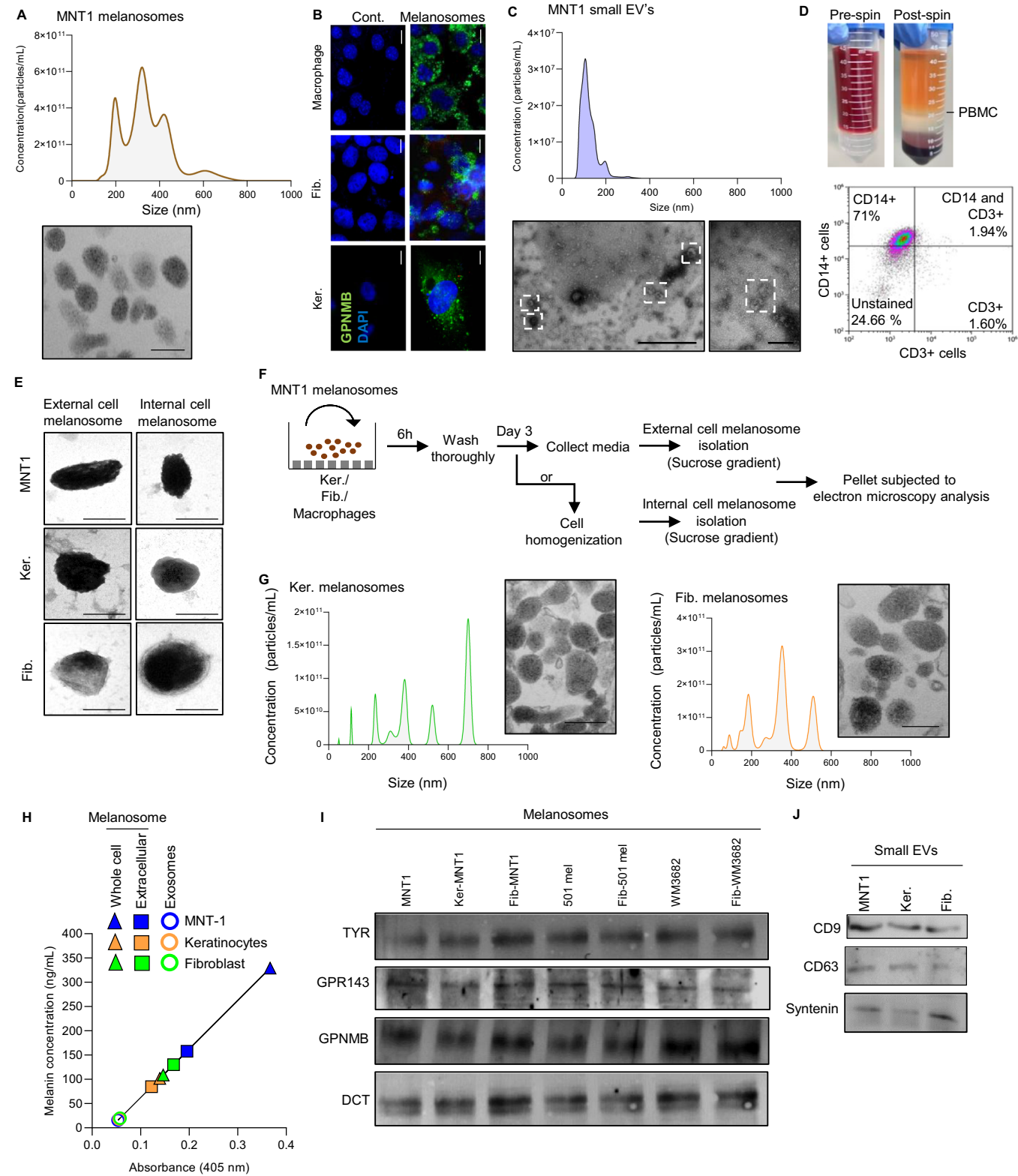


Expanded View Figures

Figure EV1. Associated with Fig. 1: Melanosomes are detected in non-cancerous cells in the tumor microenvironment.

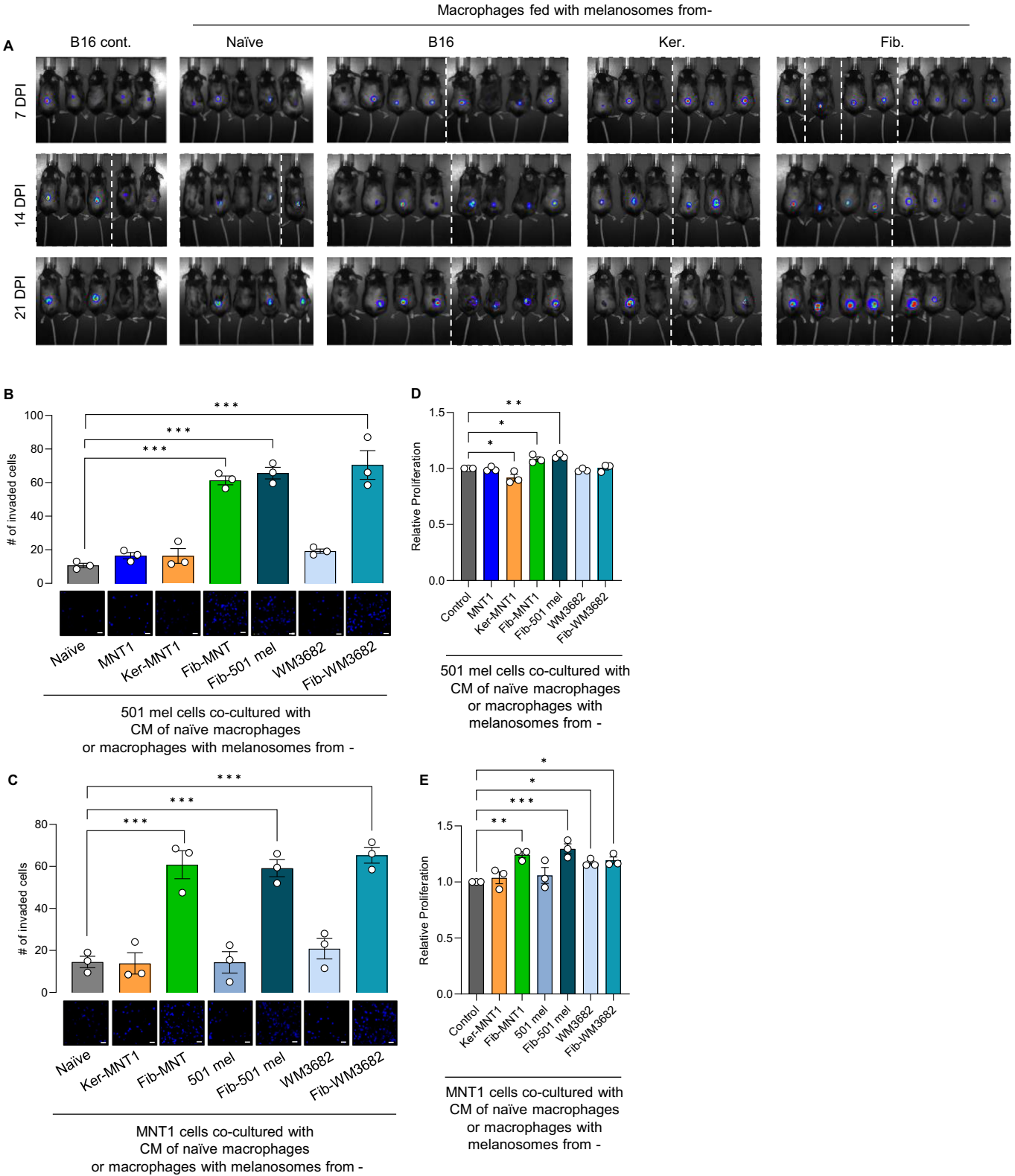
(A) Images of compound nevi, in situ and vertical melanoma and lymph metastasis samples from two patients. Upper panels: Images of H&E-stained sections of compound nevi, in situ, and vertical melanoma and lymph metastasis samples from two patients. Black dashed lines demarcate the epidermal and dermal borders. Scale bars, 200 μ m. Inset images show macrophages containing pigmented vesicles. Scale bars, 50 μ m. Lower panels: Images of consecutive specimens stained for HMB45 (white), melanosomal marker GPNMB (green), and macrophage marker CD68 (red) and with DAPI for nuclei (blue). White dashed lines demarcate the epidermal and dermal borders. Scale bars, 200 μ m. Inset images show the co-localization of macrophages with melanosomes. Scale bars, 50 μ m. (B) Left: Images of H&E-stained sections of in situ melanoma specimens from two patients. Black dashed lines demarcate the epidermal and dermal borders. Scale bars, 200 μ m. Right: Immunofluorescence images of consecutive samples stained for CD68 (red) and GPNMB (green) or FSP1 (red) and GPNMB (green). White dashed lines demarcate the epidermal and dermal borders. Scale bars, 200 μ m. Inset images show keratinocytes within the epidermis containing melanosomes marked with white arrowheads. Scale bars, 50 μ m.





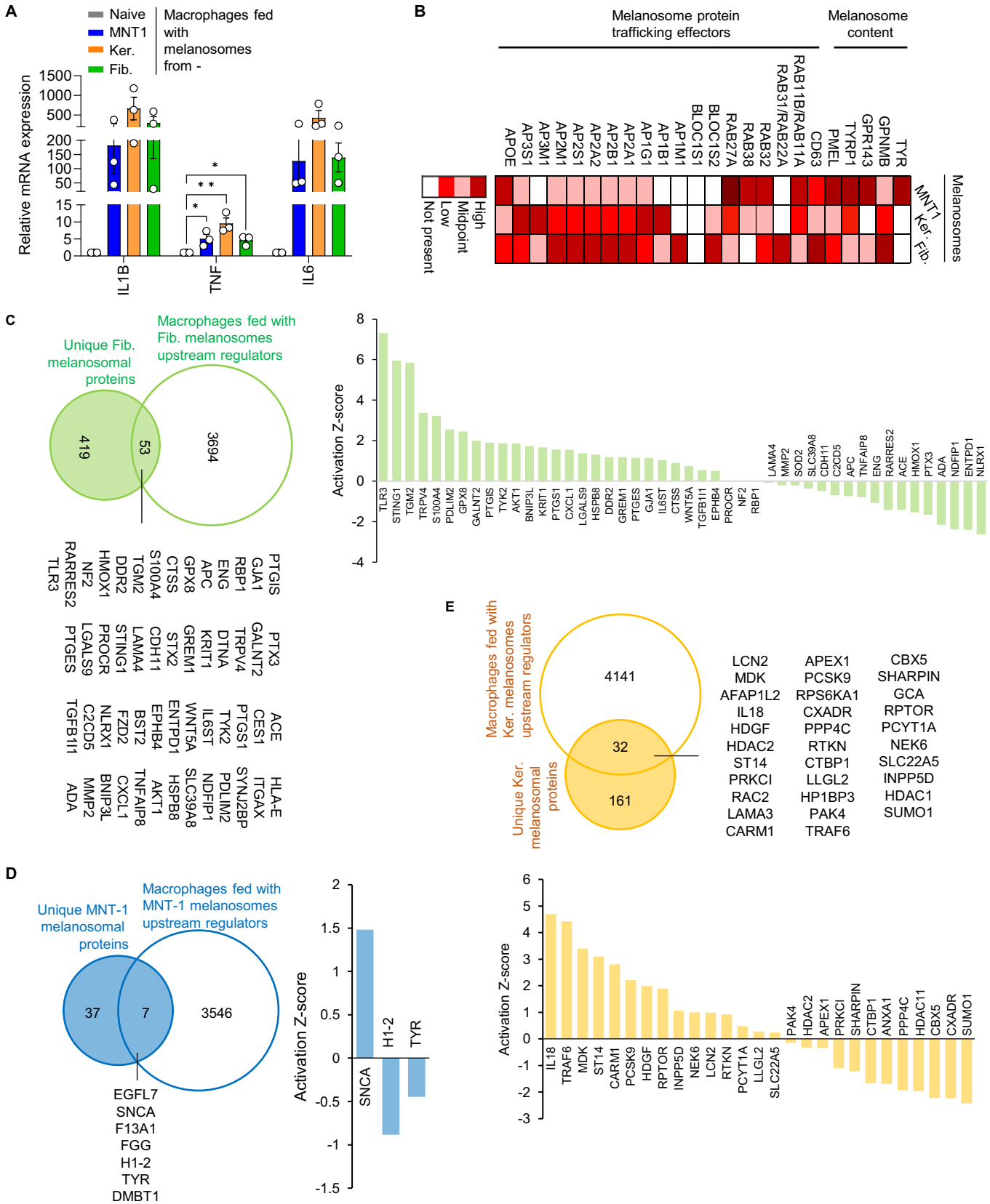
◀ **Figure EV2. Associated with Fig. 2: Cell-to-cell transfer of melanoma cell-derived melanosomes occurs in the tumor microenvironment.**

(A) NanoSight analysis (upper) and TEM analysis (lower) of melanosomes secreted from melanoma cells. Scale bars, 0.5 μm . (B) Immunofluorescence images of control keratinocytes, fibroblasts, and naïve macrophages and of cells cultured with melanosomes for 24 h. DAPI-stained nuclei appear blue. Scale bars, 10 μm . (C) NanoSight analysis (upper) and TEM analysis (lower) of small EVs secreted from melanoma cells. Scale bars, 0.5 μm left image and 0.2 μm for right image. Small EVs are highlighted with white dashed boxes. (D) Top: Photograph of peripheral blood mononuclear cell sample after Ficoll gradient centrifugation. Bottom: Flow cytometry analysis of monocytes isolated using a CD14⁺ cell isolation kit; 71% of isolated cells were CD14⁺. (E) TEM images of melanosomes from inside cells (right) and from conditioned media (left) of indicated samples. Scale bars, 0.2 μm . (F) Workflow of isolation of melanosomes from inside cells and from conditioned media after 3 days of culture. (G) NanoSight analysis (right) and TEM images (left) of melanosomes isolated from the keratinocyte and fibroblast cells cultured with MNT1 melanoma cell melanosomes. Scale bars, 0.5 μm . (H) Quantification of melanin concentration in samples isolated by differential centrifugation, gradient centrifugation, and ultra-centrifugation for secreted melanosomes, internalized melanosomes, and small EVs, respectively, from melanoma cells, keratinocytes, and fibroblasts; $n = 2$ independent experiments. (I) Western blot analysis for signature melanosomal proteins TYR, GPR143, GPNMB, and DCT in the melanosome fraction from indicated cells. (J) Western blot analysis for signature small EV proteins CD9, Syntenin, and CD63 in small EV fractions from indicated cells.



◀ **Figure EV3. Associated with Fig. 3: Macrophages cultured with melanosomes from different cell sources have heterogenous effects on cancer hallmarks.**

(A) In vivo bioluminescent images of C57BL6 mice injected with B16-F10 mCherry cells expressing firefly luciferase reporter co-inoculated with BMDMs incubated with melanosomes derived from B16-F10 melanoma cells, fibroblasts, or primary keratinocytes. Images were taken on days 7, 14, and 21 post injection. Mice inoculated with B16-F10 cells and naïve macrophages were used as controls. Note: The representative images shown in Fig. 3B have been taken as a representative of each group from the raw images shown here. (B) Upper: Numbers of invaded 501mel melanoma cells treated with conditioned media from macrophages cultured with melanosomes from indicated source cells for 24 h. $n = 3$ independent experiments. Lower: Representative images of invasion assay. Scale bar, 50 μm . (C) The proliferation of 501mel melanoma cells treated with the conditioned media of macrophages that had been cultured with melanosomes from indicated cells for 24 h relative to culture with conditioned media from naïve macrophages (control). $n = 3$ independent experiments. (D) Upper: Numbers of invaded MNT1 melanoma cells treated with conditioned media from macrophages cultured with melanosomes from indicated source cells for 24 h. $n = 3$ independent experiments. Lower: Representative images of invasion assay. Scale bar, 50 μm . (E) The proliferation of MNT1 melanoma cells treated with the conditioned media of macrophages that had been cultured with melanosomes from indicated cells for 24 h relative to culture with conditioned media from naïve macrophages (control). $n = 3$ independent experiments. Data information: In panel (B-E), error bars represent $\pm\text{SEM}$ and one-way ANOVA was performed for statistical analysis, $*P \leq 0.05$, $**P \leq 0.01$, $***P \leq 0.001$ was considered significant.



**Figure EV4. Associated with Fig. 4: Macrophages incubated with melanosomes from different cell types are phenotypically and functionally diverse.**

(A) Relative mRNA expression of pro-inflammatory genes *IL1B*, *TNF*, and *IL6* in macrophages cultured with melanosomes from MNT1 melanoma cells, keratinocytes, and fibroblast cells normalized to expression in naïve macrophages; $n = 3$. (B) Heatmap of LFQ intensity of melanosomal proteins and trafficking effector proteins in melanosomes from indicated cell sources. $n = 3$ independent experiments. (C-E) Overlap between the upstream regulators predicted by IPA and unique melanosomal proteins and graphs depicting the activation Z-scores of the upstream regulators found in the overlap from (C) fibroblasts, (D) MNT1 melanoma cells, and (E) keratinocyte as identified in Fig. 4I,J. $n = 3$ individual donors for each melanophage and $n = 3$ independent experiments for melanosome isolation. Data information: In panel (A), error bars represent $\pm$ SEM. One-way ANOVA was performed for statistical analysis, $*P \leq 0.05$, $**P \leq 0.01$ was considered significant.

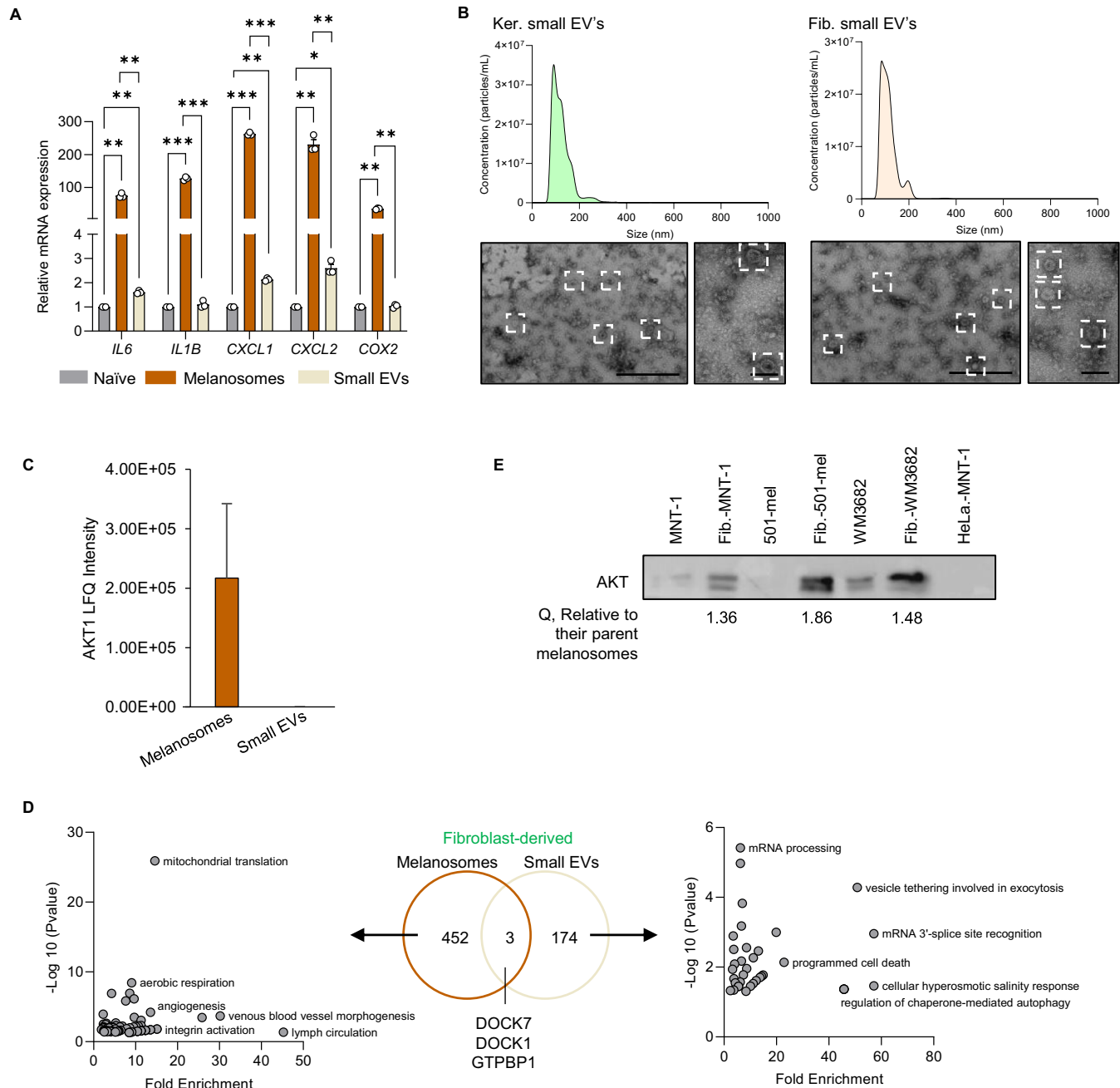


Figure EV5. Associated with Fig. 5: Fibroblasts melanosomes loaded with AKT1 induce angiogenesis in macrophages via the AKT1/mTOR pathway.

(A) mRNA levels of CAF-associated genes in dermal fibroblast cells upon culturing with small EVs or melanosomes. $n = 3$ independent experiments. (B) NanoSight analysis (upper) and TEM analysis (lower) of small EVs secreted from keratinocytes (Ker., left panel) and fibroblasts (Fib., right panel). Scale bars, 0.5 μm and 0.2 μm . Small EVs are highlighted with white dashed boxes. (C) LFQ intensity scores of the AKT1 protein levels in fibroblast-derived small EVs and melanosomes. $n = 2$ independent experiments of isolation of each EVs. (D) Center: Overlap between the uniquely expressed proteins in each fibroblast-derived small EVs or melanosomes compared to the MNT1 or keratinocyte-derived small EVs or melanosomes, by proteomics analysis. MNT1 and keratinocyte EVs were used as controls to identify specific proteins expressed by the EVs of the fibroblast cells. Left and right: Dot plots showing the significantly enriched biological processes of the genes further uniquely expressed by each EV. $n = 2$ independent experiments of isolation for each EVs. (E) Western blot analysis for AKT protein levels in melanosomal fractions isolated from the melanoma cells or fibroblast cells that were cultured with indicated melanoma melanosomes. Data information: In panel (A, C), error bars represent $\pm$ S.E.M. In panel (A), one-way ANOVA was performed for statistical analysis, $*P \leq 0.05$, $**P \leq 0.01$, $***P \leq 0.001$ was considered significant.