Figure S1. Monocyte subset gating strategy by flow cytometry. Left panel, whole population of monocytes from human blood (isolated by elutriation); right panel, subset determination by staining with antibodies anti-CD14 and CD16. P1, whole population; P2, classical CD14⁺⁺/CD16⁻ monocytes; P3, intermediate CD14⁺⁺/CD16⁺ monocytes; P4, non-classical CD14⁺⁺/CD16⁺⁺ monocytes.



 $Figure \ S2. \ Representative \ nanoparticle \ tracking \ analysis \ measurement \ of \ TEVs_{HCT116}. (A \ and \ B) \ Sample \ video \ screen shots \ from \ the \ NTA \ measurement \ obtained \ with \ NanoSight \ LM10HS \ equipped \ with \ the \ LM14 \ 488-nm \ laser \ module \ (Malvern \ Instruments, \ Ltd.).$



Figure S3. Efficiency of $\text{TEVs}_{\text{HCT116}}$ labeling with SYTO RNASelect dye by NTA and flow cytometry. (A) Representative NTA size profile of $\text{TEVs}_{\text{HCT116}}$ without dye. (B) Representative NTA size profile of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$. (C) Representative histogram of $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (D) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (D) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (E) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (D) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (D) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (E) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (E) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (E) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel without labeling. (E) Representative histogram of SYTO RNA-labeled $\text{TEVs}_{\text{HCT116}}$ in FL1 channel. NTA, nanoparticle tracking analysis; TEVs, tumor-derived extracellular vesicles.



Figure S4. Representative CD44 expression on monocytes after 0, 2 and 18 h incubation at 37° C.



Figure S5. Kinetics of $TEVs_{HCT116}$ and $TEVs_{SW1116}$ attachment to monocytes. Data are presented as the mean \pm SEM of three experiments. TEVs, tumor-derived extracellular vesicles; MFI, mean fluorescence intensity.

