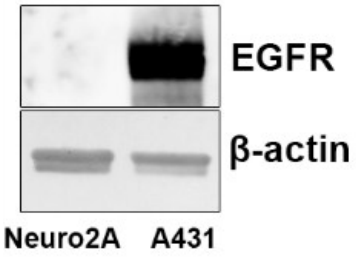
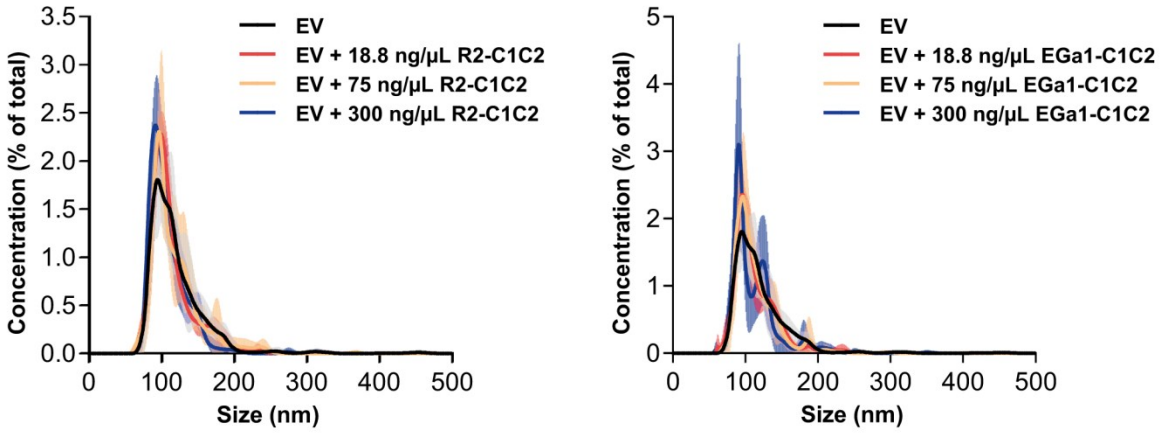
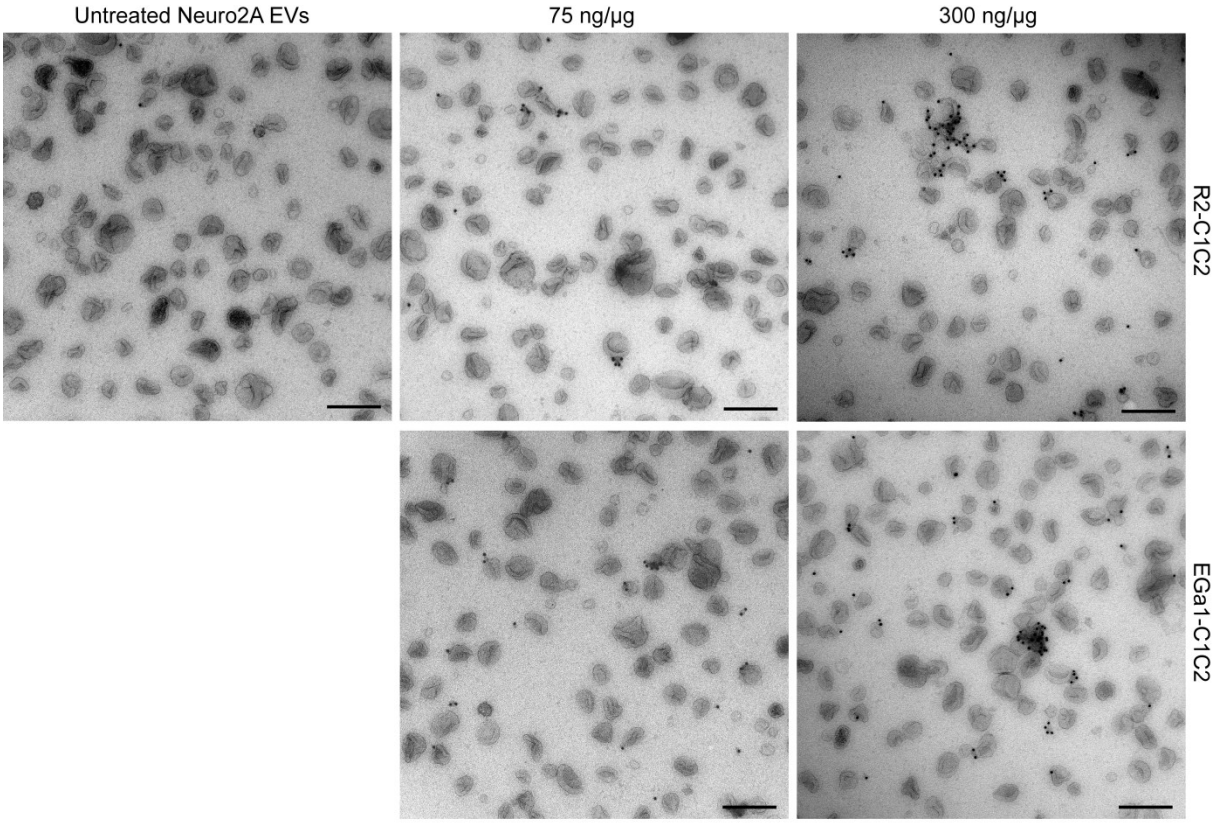
## Supplementary Figures



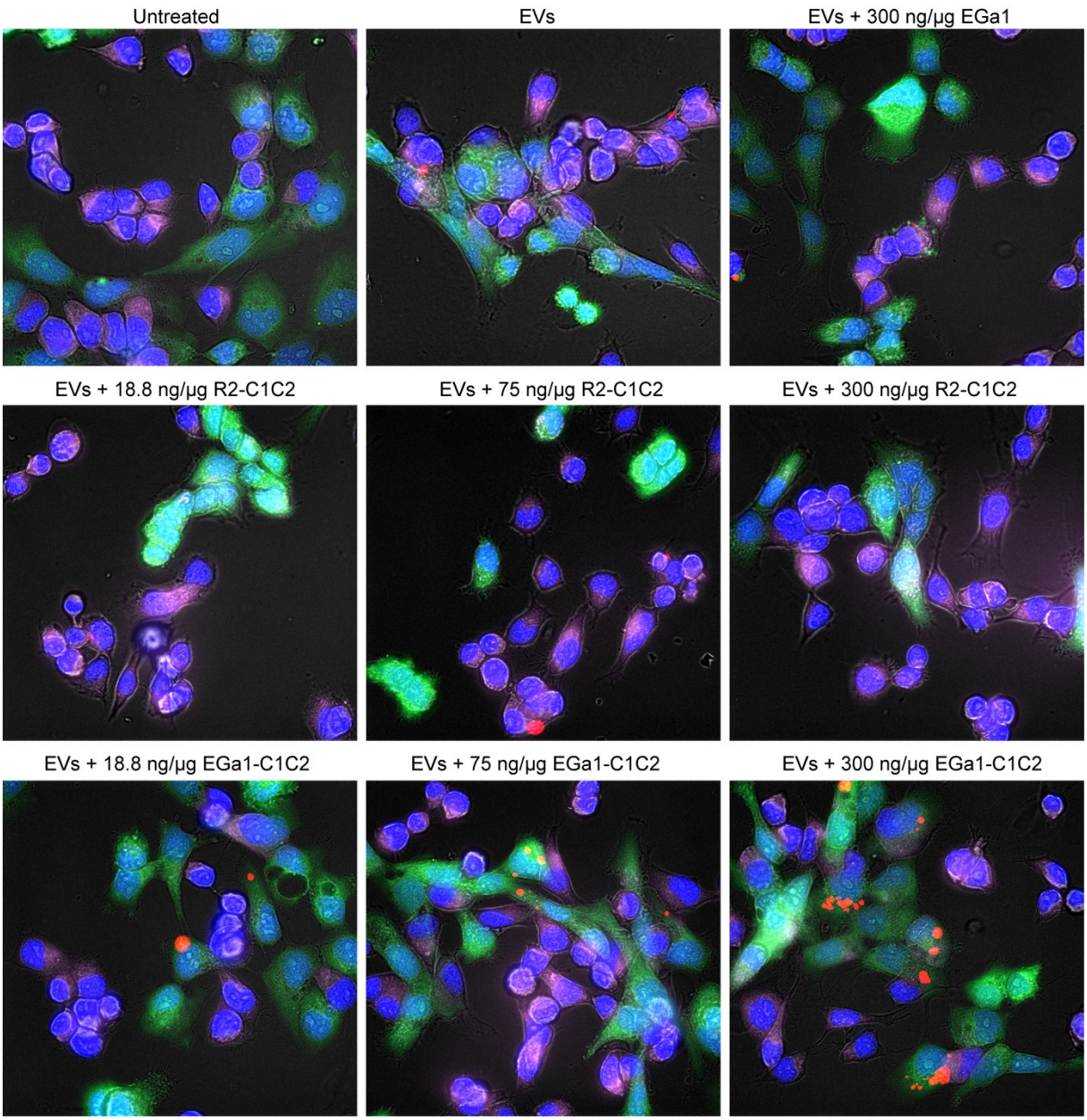

***Supplementary Figure 1: A431 cells express EGFR, whereas Neuro2A cells do not****. Western blot of Neuro2A and A431 cell lysates, probed with anti-EGFR antibodies. Beta-actin was used as a loading control. In each lane 5 µg of protein was loaded.*




***Supplementary Figure 2: Size distribution of Neuro2A EVs does not change when EVs are decorated with C1C2-nanobodies****. Representative size distribution of Neuro2A EVs after decoration with increasing concentrations of R2-C1C2 (left panel) or EGa1-C1C2 (right panel), as determined by Nanoparticle Tracking Analysis. Data is displayed as mean ± SD of 5 measurements.*

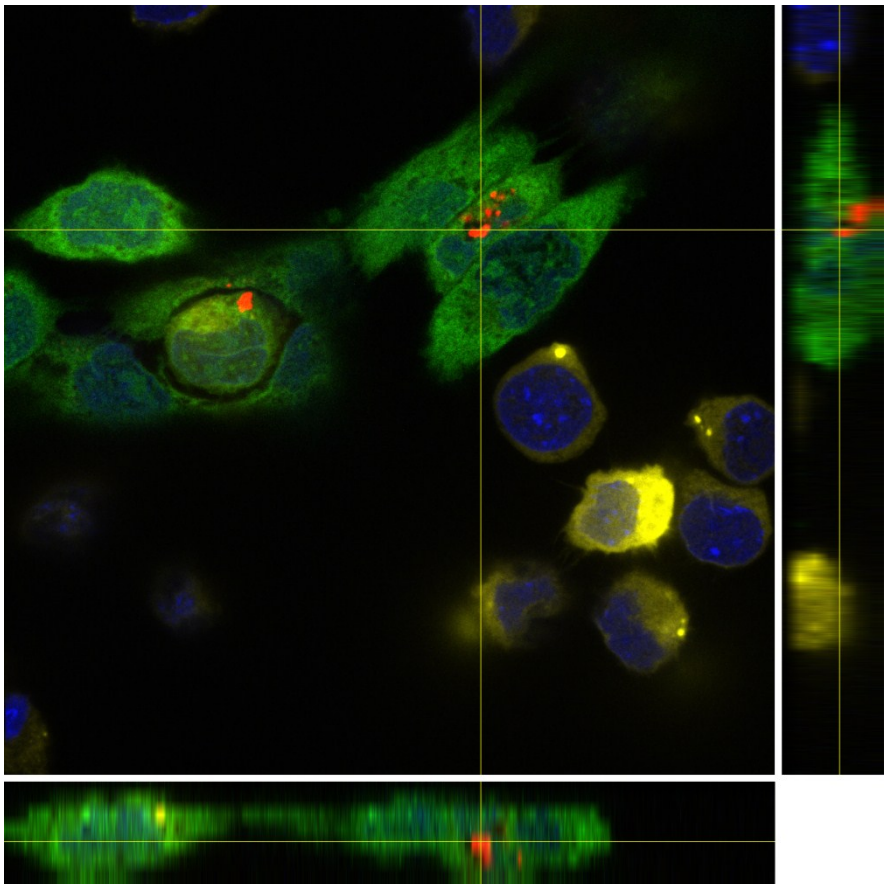



***Supplementary Figure 3: C1C2-nanobodies self-associate with subpopulations of Neuro2A EVs without affecting EV morphology****. Transmission electron microscopy pictures of Neuro2A EVs after decoration with increasing concentrations of R2-C1C2 (top row) and EGa1-C1C2 (bottom row) and after SEC purification. Immunogold labeling was performed with anti-Myc antibodies. Scale bars represent 200 nm.*




***Supplementary Figure 4: Decoration of Neuro2A EVs with EGa1-C1C2 promotes selective uptake by EGFR-overexpressing A431 cells co-cultured with EGFR-negative Neuro2A cells****. Representative fluorescence microscopy pictures of CellTracker Green labeled A431 cells (green) co-cultured with CellTracker Red labeled Neuro2A cells (purple) in a 1:3 ratio, after 4 hours incubation with CellTracker Deep Red labeled SEC-purified Neuro2A EVs (red) decorated with increasing concentrations of R2-C1C2 or EGa1-C1C2 or 300 ng/µg EGa1 without C1C2 domains. Cells were acid washed after incubation to*

*remove surface-bound particles, and stained with DAPI (blue). Differential interference contrast (DIC)*



*pictures are faintly included in overlays to indicate cell boundaries.*

***Supplementary Figure 5: Neuro2A EVs decorated with EGa1-C1C2 are localized inside A431 cells after 4 hours of co-incubation****. Orthogonal presentation of a representative confocal microscopy picture of CellTracker Green labeled A431 cells (green) co-cultured with CellTracker Red labeled Neuro2A cells (yellow) in a 1:3 ratio, after 4 hours incubation with CellTracker Deep Red labeled SEC-purified Neuro2A EVs (red) decorated with 300 ng/µg EGa1-C1C2. Cells were acid washed after incubation to remove surface-bound particles, and stained with DAPI (blue).*

## Supplementary Table

***Supplementary Table 1: Sequences of used primers and oligonucleotides.***

| *Name* | *Sequence (5'-3')* |
|---|---|
| Fw_C1C2 | ATTGCGGCCGCAGGCGGTGGAGGCAGCGGTGGCGGGGGTAGCTGTTCTACACAGCTGGGC |
| Rv_C1C2 | AATCGGCCGAGCCCTGAAAATACAGGTTTTCCTTAAGACAGCCCAGCAGCTC |
| Igκ chain leader sequence | ATGGAGACAGACACACTCCTGCTATGGGTACTGCTGCTCTGGGTTCCAGGTTCCACTGGTGAC |