

Supporting Information

Target-selective cytosolic delivery of cargo proteins using the VHH-presented OLE-ZIP capsules

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(a) Trx-OLE-ZIP

MSDKI IHLTD DSFDT DVLKA DGAIL VDFWA EWCGP CKMIA PILDE IADEY
QGKLT VAKLN IDQNP GTAPK YGIRG IPTLL LFKNG EVAAT KVGAL SKGQL
KEFLD ANLAG SGSGH MHHHH HHSSG LVPRG SAMAA LKKEL QANKK ELAQL
KWELQ ALKKE LAQGG SGGSF GLTGI TLVGT VIGLA LATPL FVIFS PVIVP
AMIAI GLAVT GFLTI GTFGL GGSGG SEQLE KKLQA LEKKL AQLEW KNQAL
EKKLA QKLAA ALEHH HHHH

(b) VHH-OLE-ZIP-EV0

DYKDD DDKGG TQVQL QESGG GLVQA GGSLL LSCAA SGRTF SSYAM GWFRQ
APGKE REFVA AINWS GGSTS YADSV KGRFT ISRDN TKNTV YLQMN SLKPE
DTAAF YCAAT YNPYS RDHYF PRMTT EYDYW GQGTQ VTVSS VDGGS GGSGG
SGGSA LKKEL QANKK ELAQL KWELQ ALKKE LAQGG SGGSF GLTGI TLVGT
VIGLA LATPL FVIFS PVIVP AMIAI GLAVT GFLTI GTFGL GGSGG SEQLE
KKLQA LEKKL AQLEW KNQAL EKKLA Q

(c) VHH-OLE-ZIP-EV1

DYKDD DDKGG TQVQL QESGG GLVQA GGSLL LSCAA SGRTF SSYAM GWFRQ
APGKE REFVA AINWS GGSTS YADSV KGRFT ISRDN TKNTV YLQMN SLKPE
DTAAF YCAAT YNPYS RDHYF PRMTT EYDYW GQGTQ VTVSS AGGVE SAGGS
AGGSA GGSAG GSAGG SGSAG GSAGG STSAG GSAGG SAGGS AGGSA GGSAGS
AGGSA GGSTS AGGSA GGSAG GSAGG SAGGS GSAGG SAGGS TSAGG SAGGS
AGGSA GGSAG GVDGG SGGSG GSGGS ALKKE LQANK KELAQ LKWEL QALKK
ELAQG GSGGS FGLTG ITLVG TVIGL ALATP LRVIF SPVIV PAMIA IGLAV
TGFLT IGTFG LGGSG GSEQL EKKLQ ALEKK LAQLE WKNQA LEKKL AQ

(d) VHH-OLE-ZIP-EV2

DYKDD DDKGG TQVQL QESGG GLVQA GGSLL LSCAA SGRTF SSYAM GWFRQ
APGKE REFVA AINWS GGSTS YADSV KGRFT ISRDN TKNTV YLQMN SLKPE
DTAAF YCAAT YNPYS RDHYF PRMTT EYDYW GQGTQ VTVSS AGGVE SAGGS
AGGSA GGSAG GSAGG SGSAG GSAGG STSAG GSAGG SAGGS AGGSA GGSAGS
AGGSA GGSTS AGGSA GGSAG GSAGG SAGGS GSAGG SAGGS TSAGG SAGGS
AGGSA GGSAG GVESA GGSAG GSAGG SAGGS AGGSG SAGGS AGGST SAGGS
AGGSA GGSAG GSAGG SGSAG GSAGG STSAG GSAGG SAGGS AGGSA GGSAGS
AGGSA GGSTS AGGSA GGSAG GSAGG SAGGV DGGSG GSGGS GGSAL KKELQ
ANKKE LAQLK WELQA LKKEL AQGGS GGSFG LTGIT LVGTV IGLAL ATPLF
VIFSP VIVPA MIAIG LAVTG FLTIG TFGLG GSGGS EQLEK KLQAL EKKLA
QLEWK NQALE KKLAQ

Figure S1. Amino acid sequences of the Trx-OLE-ZIP and VHH-OLE-ZIP-EV_n ($n = 0, 1,$ and 2).

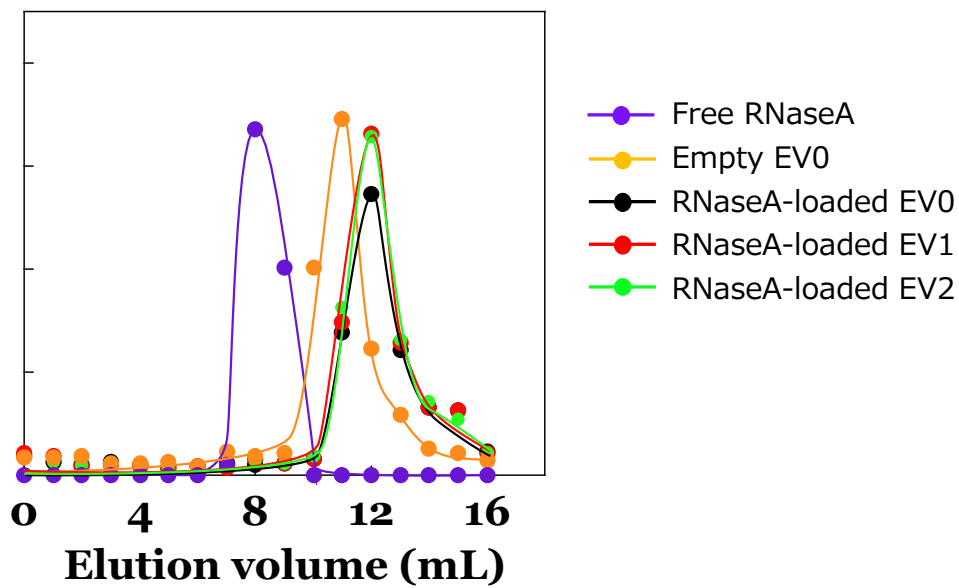


Figure S2. Summary of the GPC chromatograms for RNaseA-loaded EV0, EV1, and EV2, along with empty EV0 and free RNaseA as reference. TOYOPEARL HW-50F served as the gel filtration carrier and was packed into a chromatography column (6 mm in diameter, 800 mm in height), which was then equilibrated. Subsequently, RNaseA-loaded EV0, EV1, and EV2, as well as empty EV0 and free RNaseA, were separately analysed using this column for GPC. FITC-labelled RNaseA was utilized for this experiment.

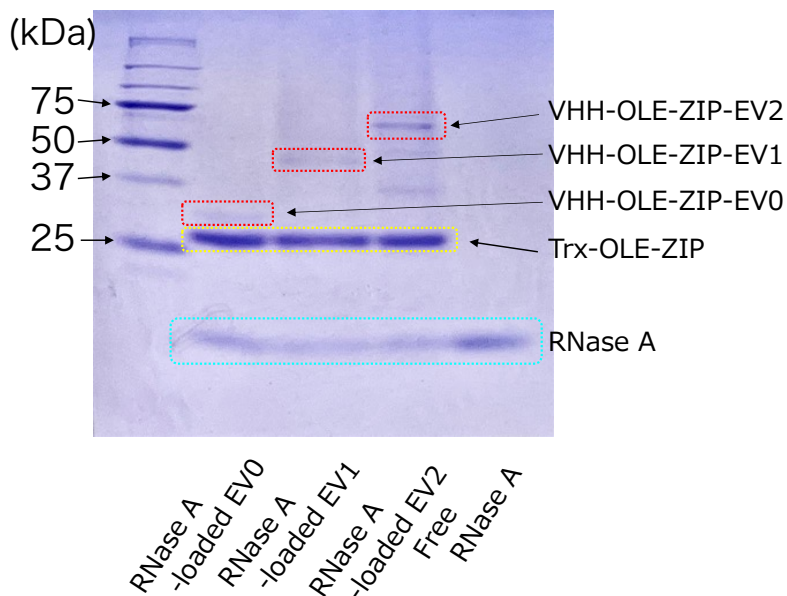
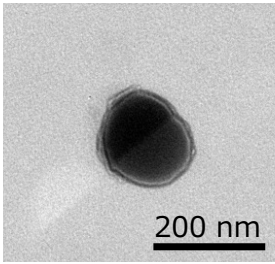
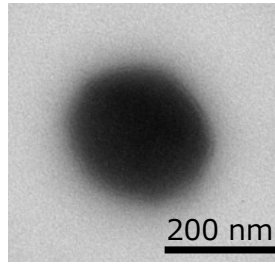


Figure S3. 15% SDS-PAGE analyses of the isolated RNaseA-loaded EV0, EV1, EV2 and free RNaseA.

(a) RNaseA-loaded EV0



(b) RNaseA-loaded EV1



(c) RNaseA-loaded EV2

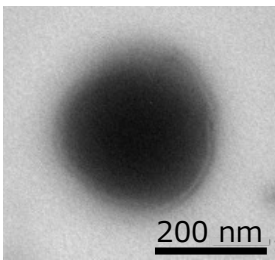


Figure S4. TEM images of the RNaseA-loaded EV0, EV1, and EV2.