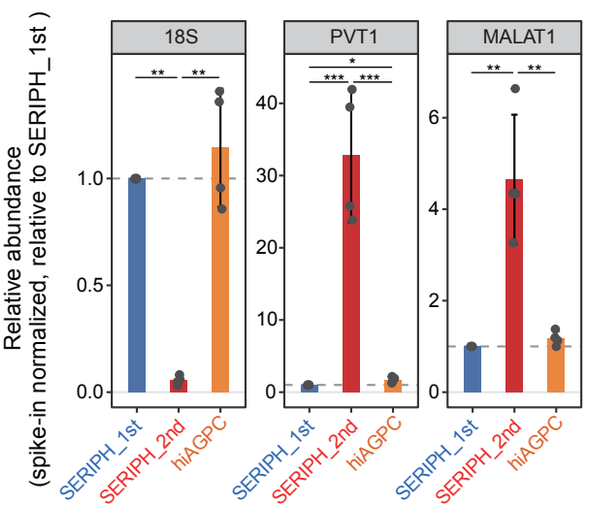
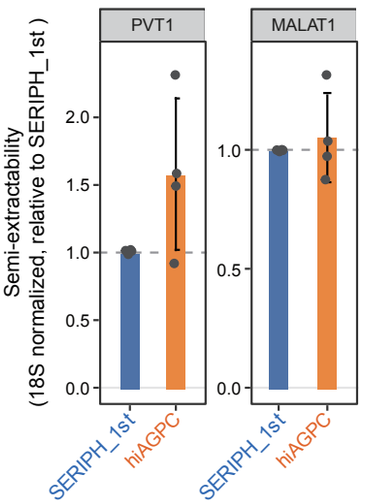
A

Preparation	Mean yield ± SD (μg)	Cell line (n)
SERIPH_1st	119.49 ± 12.74	HEK293 (4)
SERIPH_2nd	3.47 ± 0.85	HEK293 (4)
hiAGPC	137.19 ± 25.14	HEK293 (4)
hiAGPC_leftover	0.74 ± 0.34	HEK293 (4)
SERIPH_1st	123.74 ± 10.18	HeLa (3)
SERIPH_2nd	2.96 ± 0.08	HeLa (3)
hiAGPC	124.66 ± 8.57	HeLa (3)
hiAGPC_leftover	1.19 ± 0.81	HeLa (3)

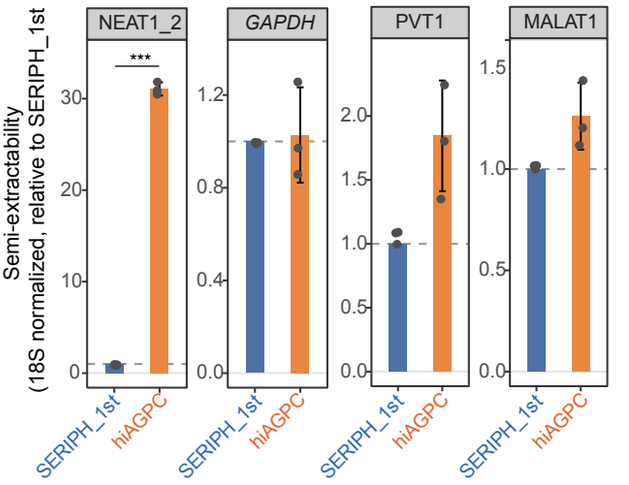
B



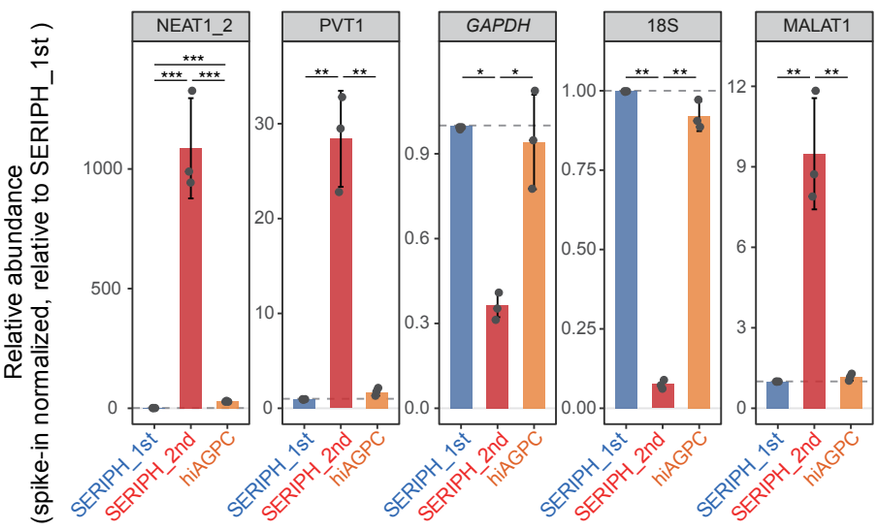
C



D



E



Supplemental Fig. 2

Supplemental Fig S2: RNA yield distribution and validation of selective partitioning in SERIPH

(A) Total RNA recovered from the same homogenate using the conventional method, hiAGPC, or SERIPH (1st-step and 2nd-step fractions) is summarized in the table. RNA was extracted from 1 × 10^6 cells per 1 mL TRI Reagent (8 mL total homogenate). Values represent mean ± SD of 4 technical replicates for HEK293 cells and 3 technical replicates for HeLa cells. For comparison, an additional preparation was performed in which hiAGPC was applied at the first step in place of the conventional extraction.

(B) Semi-extractability assessment of PVT1 and MALAT1 by RT–qPCR in HEK293, analyzed in parallel with Fig. 1B. Four technical replicates were generated from the same HEK293 cell homogenate. Relative RNA abundance was normalized to 18S rRNA, and fold changes were calculated relative to the SERIPH 1st-step fraction for each replicate. Statistical testing was performed as described in Fig. 1B.

(C) Partitioning of MALAT1 lncRNA, 18S rRNA, and PVT1 lncRNA across extraction methods assessed by RT–qPCR. RNA was prepared from the same HEK293 cell homogenates used in Fig. 1C and subjected to SERIPH 1st-step, SERIPH 2nd-step, or hiAGPC extraction. Relative RNA abundance was normalized to an in vitro–transcribed *Cypridina* luciferase spike-in control and expressed relative to the SERIPH 1st-step fraction. Bars represent mean ± SD, and dots indicate individual replicate values. Statistical analysis was performed as described in Fig. 1C. *, **, and *** indicate adjusted $P < 0.05$, < 0.01, and < 0.001, respectively.

(D) RNA Semi-extractability analysis in HeLa cells (three technical replicates). Relative RNA abundance was normalized to 18S rRNA and expressed relative to the SERIPH 1st-step fraction. Statistical testing was performed as described in Fig. 1B.

(E) RNA Partitioning across extraction methods in HeLa cells. RNA prepared from the same homogenates as in (D) was analyzed by RT–qPCR following spike-in normalization. Statistical analysis was performed as described in Fig. 1C.