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Supplemental Information

**Physical and Molecular Landscapes
of Mouse Glioma Extracellular Vesicles
Define Heterogeneity**

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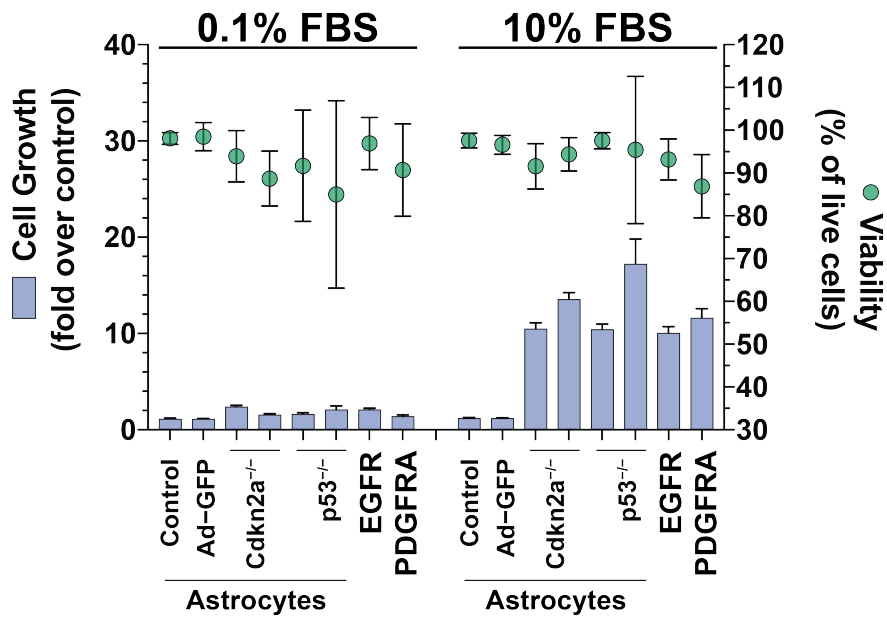


Figure S1. Growth conditions in 0.1% serum, Related to Figure 1.

Cells were grown in conditions (0.1% and 10% fetal bovine serum) for 16 hours and viable cells counted (trypan blue exclusion) in triplicates. Fold over control represent cell numbers relative to cell numbers at time 0 (defined as when cells are switched to 0.1% FBS). Percent viability is the ratio of viable cells by trypan blue staining over total number of cells. Error bars: S.D. n=3 for each cell culture indicated.

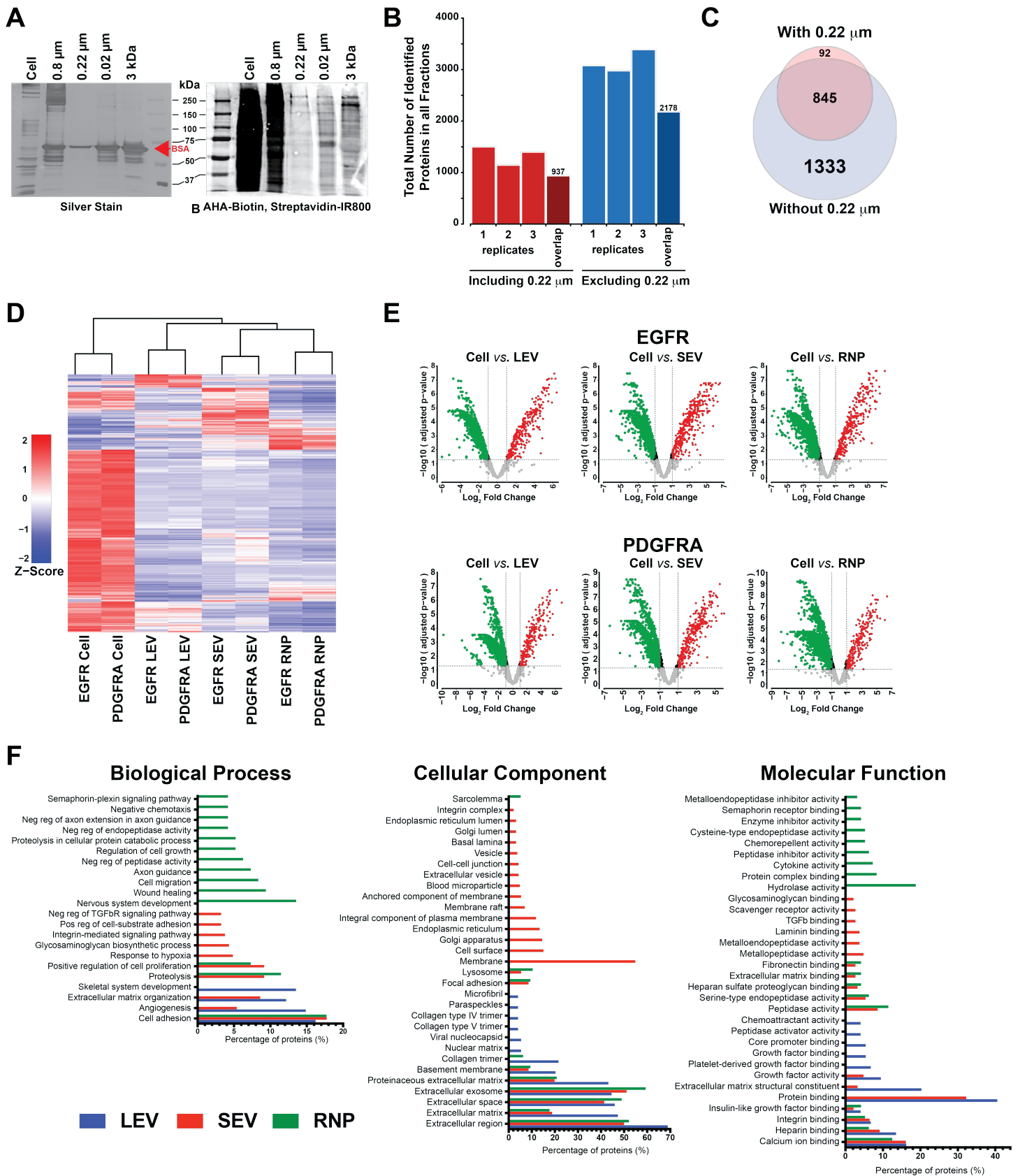


Figure S2. Quantitative proteomics from filtered EV fractions, Related to Figure 2.

(A) Representative photomicrographs of AHA labeled proteins from EGFR-driven mouse GBM primary culture cells separated on 7.5% SDS-page and visualized by silver staining (left) and transferred to PVDF membranes for detection of AHA-labeled proteins (right) from the indicated filter

fractions. The bovine serum albumin (BSA) is undetected in the western blot of AHA labeled proteins. Shown is a representative of at least two independent isolation preparations.

(B) The MEV ≥ 0.22 μm fraction has fewer proteins detectable by MS. Total number of identified proteins in all fractions from two independent experiments. Biological triplicates including the ≥ 0.22 μm fractions consistently identified 937 proteins whereas triplicates of sample excluding the ≥ 0.22 μm fractions identified 2178 proteins.

(C) Venn diagram depicting the overlap in the numbers of identified proteins between the two replicate experiments in (B).

(D) Unsupervised hierarchical clustering analysis of the z-score proportional protein expression values of all 2178 proteins identified in cells, EVs (LEVs and SEVs) and RNP protein fractions. MS spectral count values were z-score normalized prior to cluster analysis.

(E) Volcano plot representations of the differentially expressed proteins in a pair-wise comparison of cells to LEVs, SEVs and RNPs from EGFR and PDGFRA primary culture cells. The significance cut-off was set to a FDR of 0.05 ($-\log(\text{adj.P.val}) \geq 1.3$), the biological cut-off was set to a fold change of ± 2 fold ($-1 \geq \log_2 \text{FC} \geq 1$). Positive fold changes indicate higher protein abundance in the corresponding vesicle type while negative values indicate higher abundance in cells. The four different color codes used represent insignificant proteins (grey), both biologically and statistically significant proteins preferentially enriched in indicated fraction compared to cells (red) and preferentially depleted indicated fraction compared to cells (green), and statistically but not biologically significant proteins enriched and depleted (black).

(F) Functional annotation of vesicle-enriched proteins. Enrichment analysis of EV proteins. EV/RNP-enriched proteins were used in functional and pathway enrichment analysis using the DAVID database (v7.1) with GOTERM Biological Process, Cellular Component and Molecular Function. All terms indicated are significant ($p < 0.05$) following Benjamini correction.

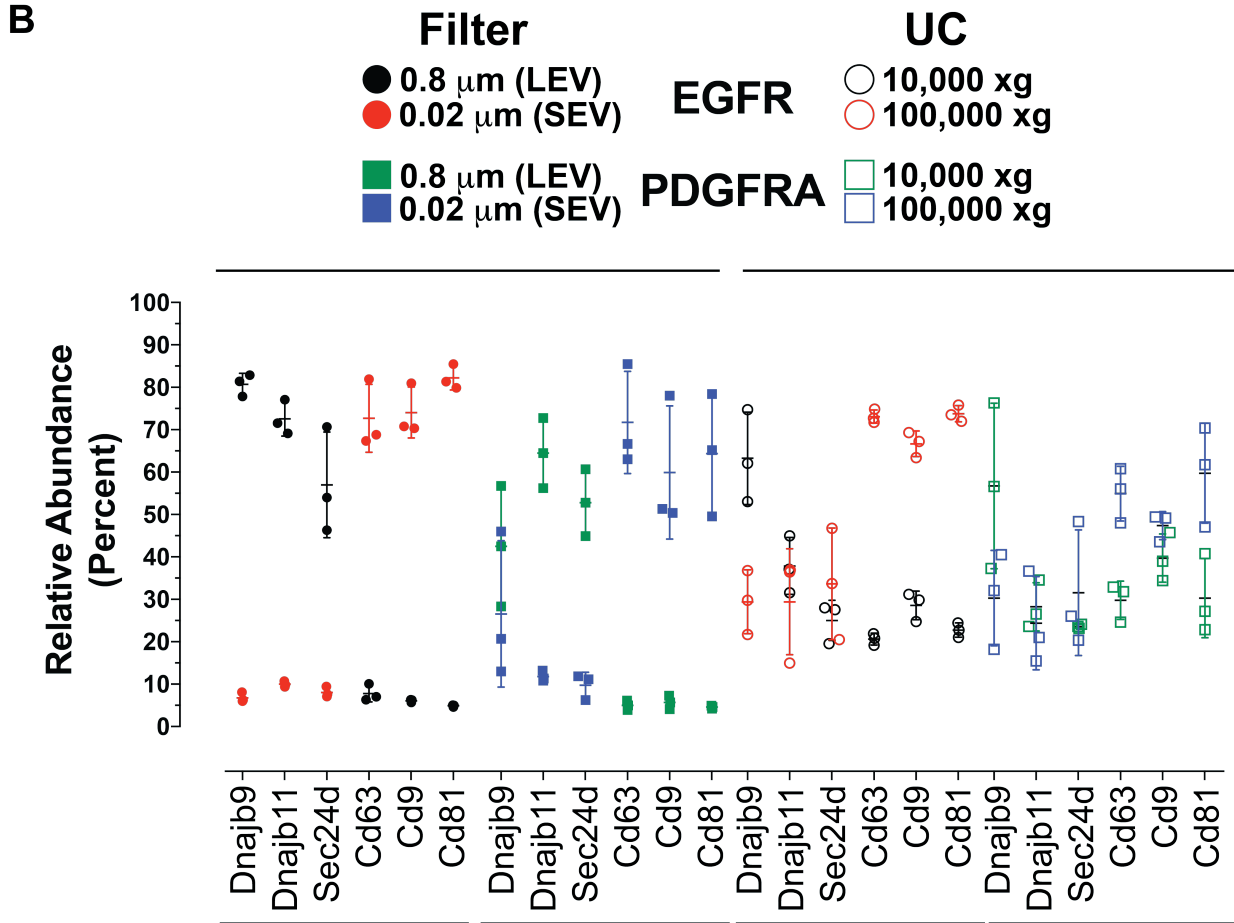
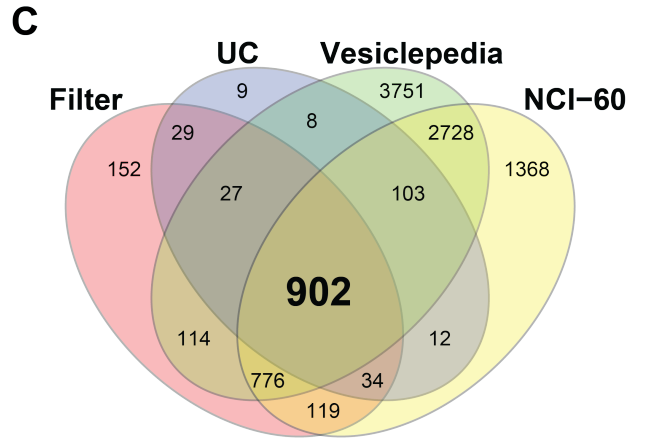
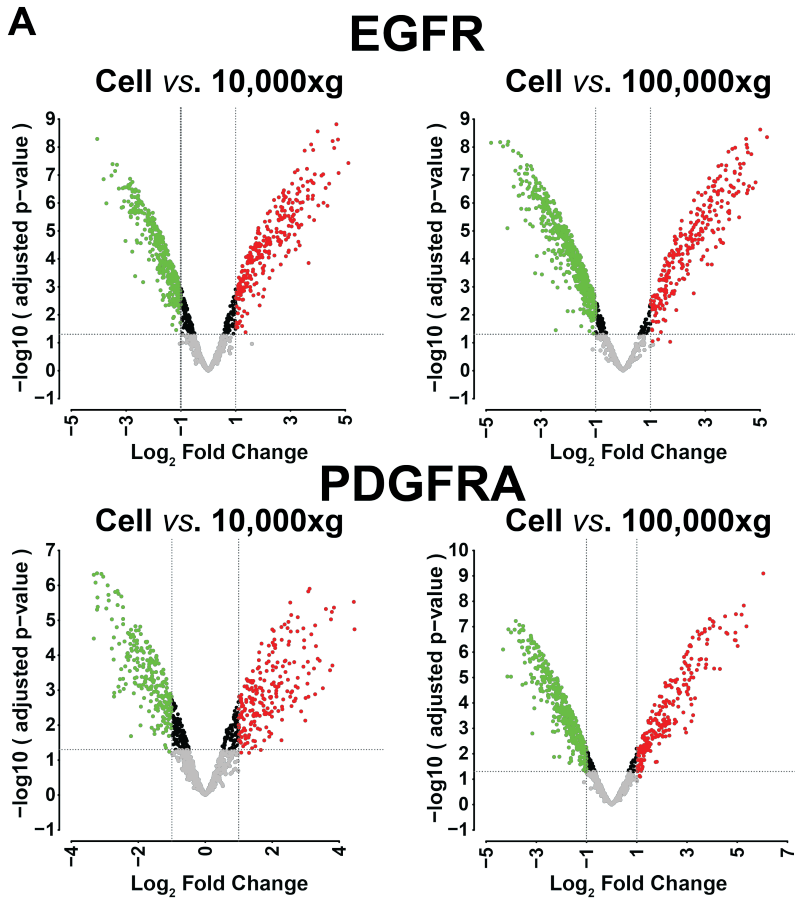


Figure S3. Quantitative proteomics of ultracentrifuged EVs. Related to Figure 3.

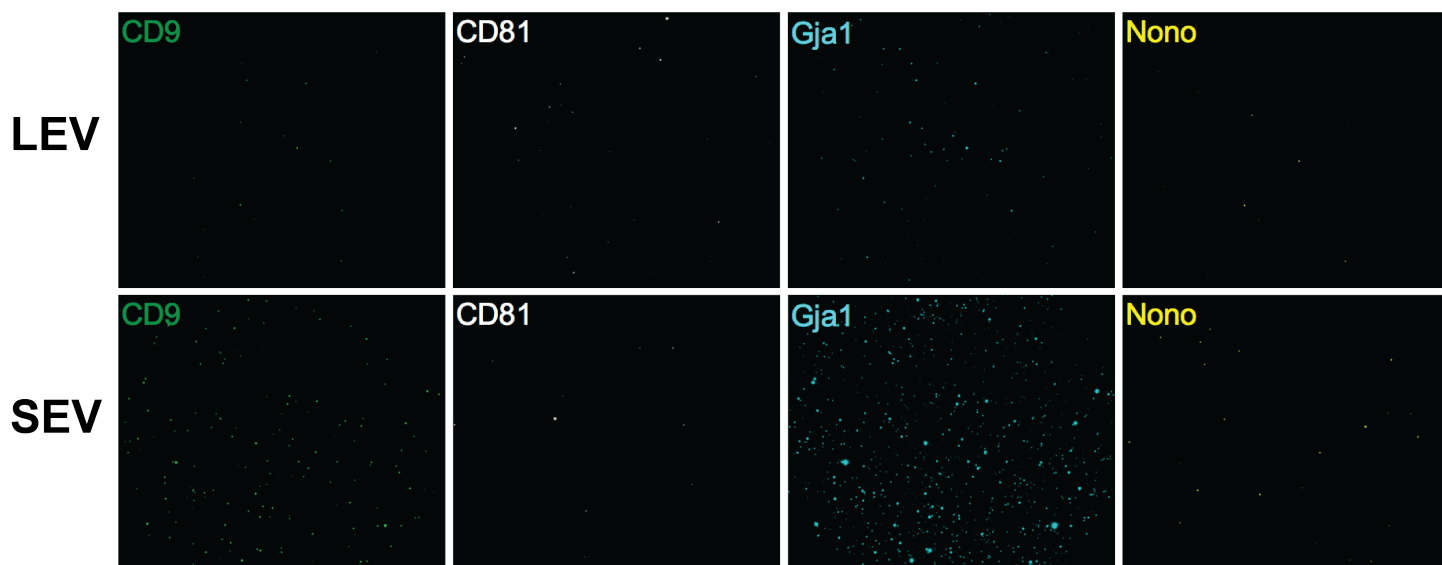
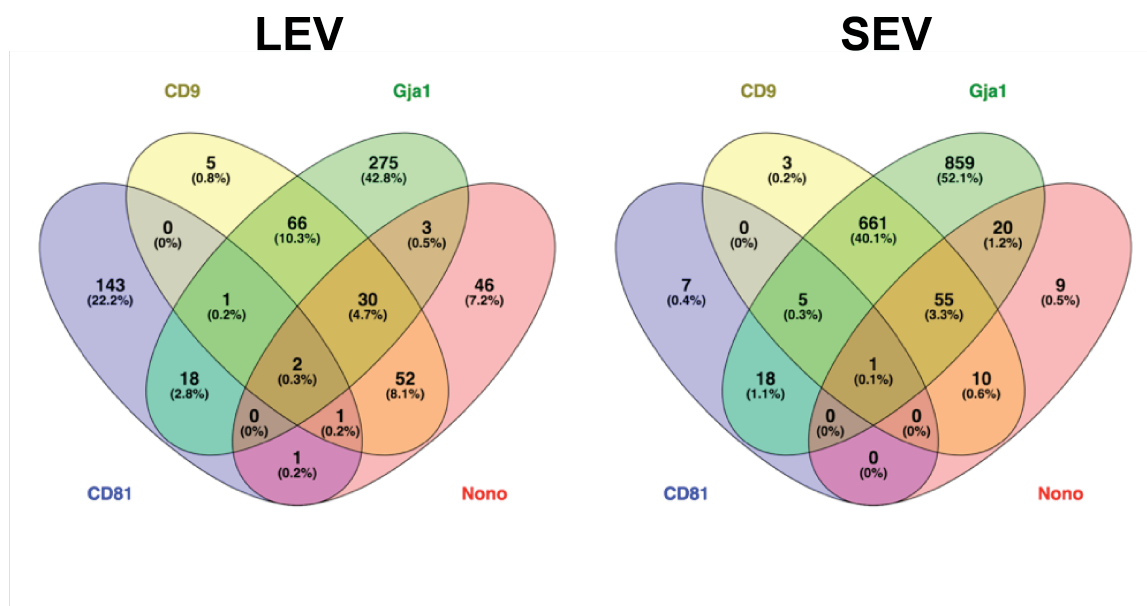
(A) Volcano plot representations of the differentially expressed proteins in a pair-wise comparison of cells to 10,000 xg and 100,000 xg UC from EGFR and PDGFRA primary culture cells. The significance cut-off was set to a FDR of 0.05 ($-\log(\text{adj.P.val}) \geq 1.3$), the biological cut-off was set to a fold change of ± 2 fold ($-1 \geq \log_2 \text{FC} \geq 1$). Positive fold changes indicate higher protein abundance in the corresponding vesicle type while negative values indicate higher abundance in cells. The four different color codes used represent insignificant proteins (grey), both biologically and statistically significant proteins preferentially enriched in indicated fraction compared to cells (red) and preferentially depleted indicated fraction compared to cells (green), and statistically but not biologically significant proteins enriched and depleted (black).

(B) Relative abundance of six markers Dnajb9, Dnajb11, Sec24d, Cd63, Cd9 and Cd81 from EGFR and PDGFRA cultures in LEV, SEV, 10,000 xg and 100,000 xg fractions. The SEV (exosomal) markers Cd63, Cd9 and Cd81 are enriched in the SEV (0.02 μm) and 100,000 xg fractions. For comparative purposes, the proteins Dnajb9, Dnajb11 and Sec24d are enriched in the LEV (0.8 μm) and 10,000 xg fractions. Lines are mean \pm S.D. of biological triplicates. for comparative purposes.

(C) Venn diagram of all proteins identified in our study (2153 from filtered fractions and 1124 from the UC fractions) compared to the Vesiclepedia database of proteins and the NCI-60 EV protein dataset (Hurwitz et al., 2016; Kalra et al., 2012; Pathan et al., 2019). Of the proteins in our datasets, 902 found in both ultracentrifuge and filter matched proteins in Vesiclepedia and NCI-60. A set of 190 proteins from our study represents newly reported vesicle-expressed proteins.

A

Marker	Log2 FC over Cells		
	LEVs	SEVs	RNPs
CD81	-0.12	3.86	-0.94
CD9	-0.75	2.87	-1.66
Nono	2.42	-0.10	-0.20
Gja1	1.90	1.01	-1.20

B**C****Figure S4. Proteomics and single vesicle expression of EV markers. Related to Figure 5.**

(A) Proteomics-derived Log₂ FC expression of the indicated markers from LEV, SEV (exosome) and RNP fractions compared to cells.

(B) Representative photomicrographs of individual immunofluorescence with the indicated markers in LEV and SEV (exosome) fractions.

(C) Co-expression of markers on single EVs. Venn diagram of individual vesicles positive for the indicated markers.

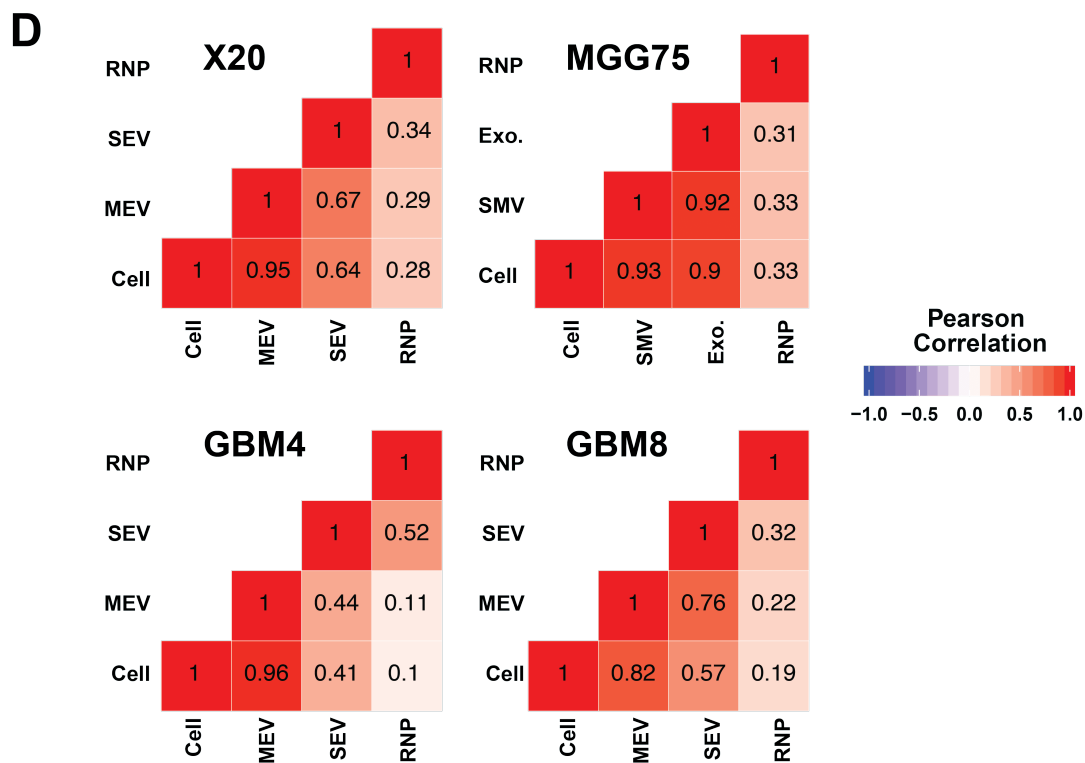
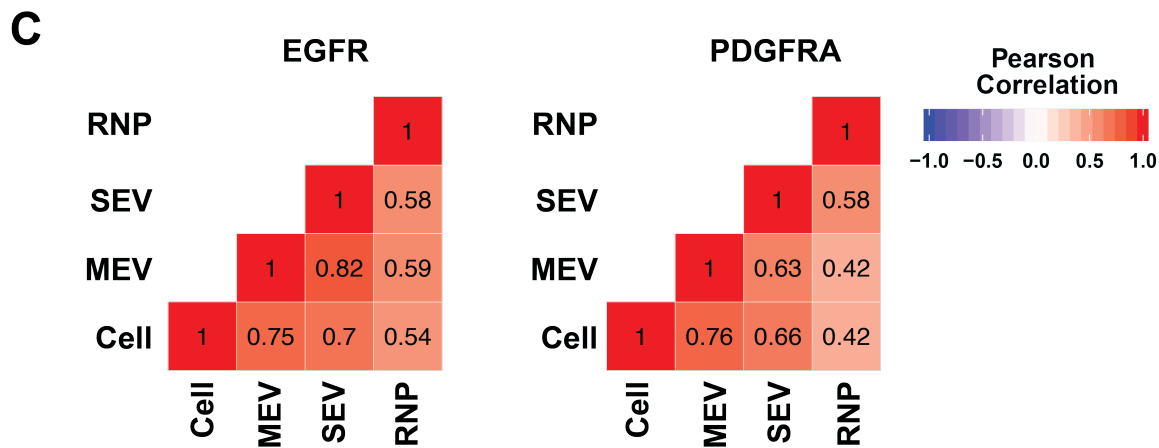
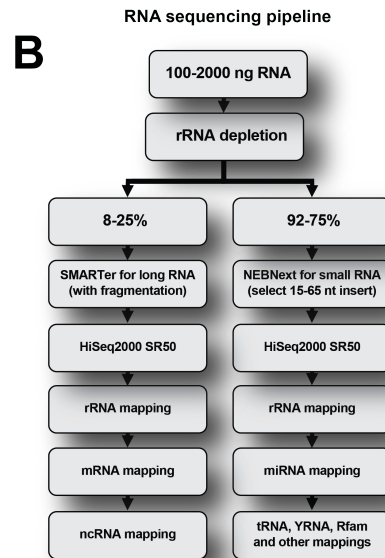
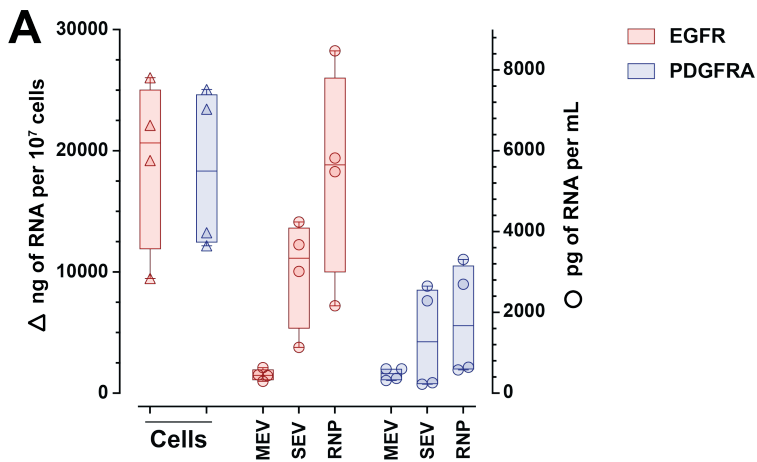


Figure S5 –RNA and Library pipeline. Related to Figure 6.

(A) Cellular RNA yields (ng per 10^7 cells) and EV RNA yields (pg RNA per mL) from conditioned media for EGFR and PDGFRA GBM primary cultures. Biological replicates n=4.

(B) Graphical representation of the optimized pipeline for broad coverage, minimally biased RNA library processing and sequencing. RNA of 15–65 nt was selected for the small RNA libraries.

(C) Pearson correlation of mRNA between mouse GBM EGFR and PDGFRA cell, MEV, SEV (exosome) and RNPs mRNA fractions.

(D) Pearson correlation of mRNA between cell, MEV, SEV (exosome) and RNPs in four human GBM stem cell (GSC) cultures (Wei et al., 2017).

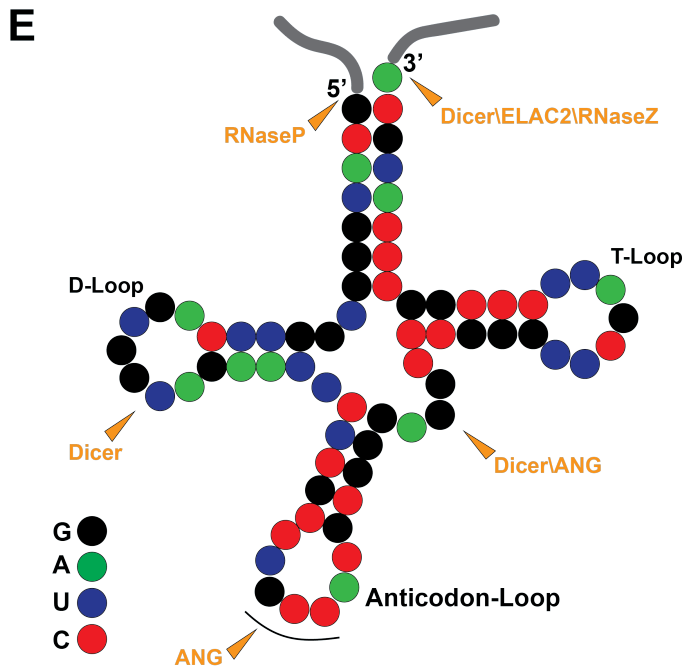
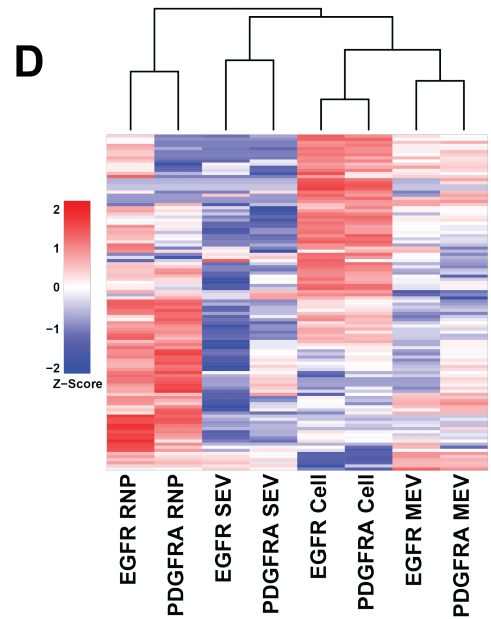
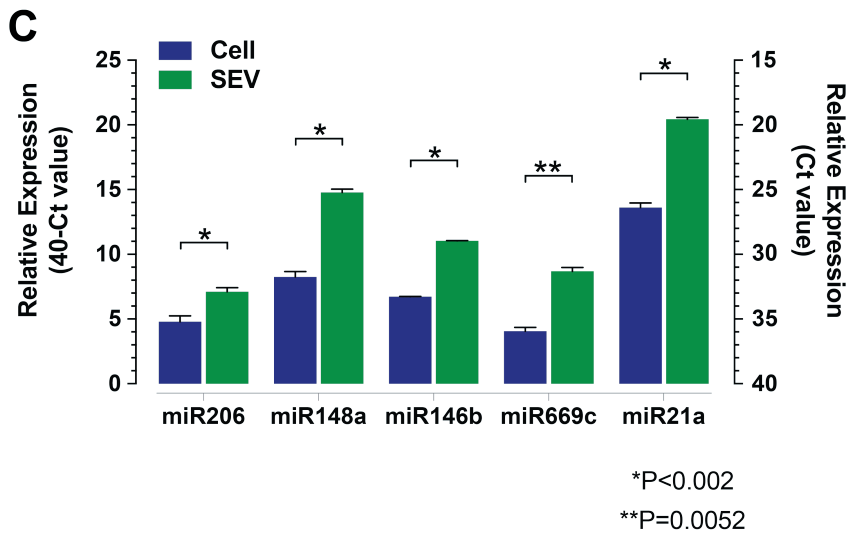
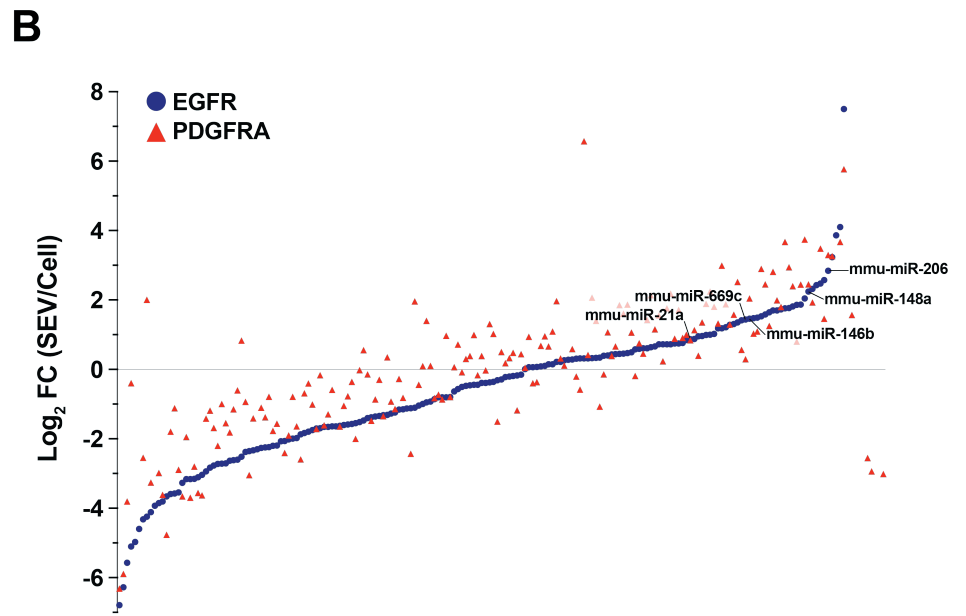
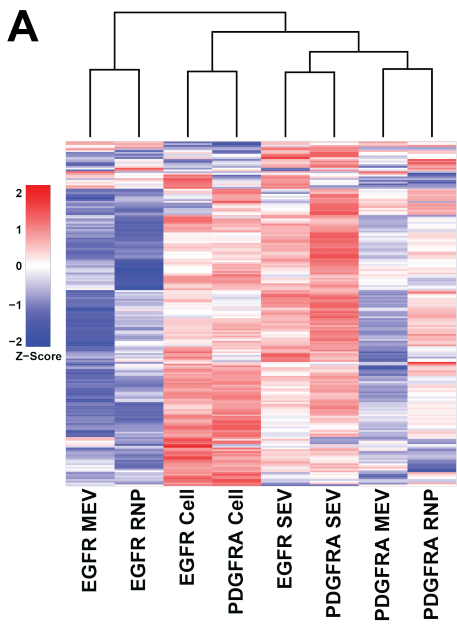


Figure S6. SEV (exosome) enriched miRNAs. Related to Figure 7.

(A) Unsupervised hierarchical clustering analysis of z-score vesicle-enriched miRNA expression values for cells, EV- and RNPs isolated from EGFR and PDGFRA GBM cultures.

(B) Graphical representation of the ratio of expression levels (Log₂ FC exosome/cells) for all mapped miRNAs for both EGFR and PDGFRA GBM derived primary cultures. Selected individual miRNAs used for qRT-PCR validation in (C) are indicated.

(C) Graphical representation of expression levels by qRT-PCR (Taqman probes) of the indicated miRNAs from RNA isolated from EGFR and PDGFRA GBM primary cultures of cells and exosomes. Bar graphs represent an average of three qRT-PCR readings per sample. Averages were analyzed for statistical significance using Student t-test, two tailed. * P<0.002, ** P=0.0052.

(D) Unsupervised hierarchical clustering analysis of z-score vesicle-enriched tRNA genes expression values for cells, SMV, exosomes and RNPs isolated from EGFR and PDGFRA GBM cultures.

(E) Representation of the predicted secondary structure of tRNA GlyGCC and the position of its cleavage (indicated by the arrow) that produces the 5' and 3' fragments which are highly abundant in RNPs. Predicted Gly-GCC tRNA structure was adopted from GtRNAdb.

Table S1. EGFR differentially enriched proteins. Related to Figure 4.

Log2 FC and adjusted p values of EGFR-enriched proteins from Cells, LEVs, SEVs (exosomes) and RNPs. Proteins labeled in Figure 4A are highlighted in red.

Gene Symbol	Cells		LEVs		SEVs		RNPs	
	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val
Ephx1	-3.26	1.0E-03	0.01	9.9E-01	-1.61	2.8E-02	-0.38	8.5E-01
Gsta2	-2.43	4.5E-02	-1.87	3.0E-01	-1.11	1.9E-01	-2.05	5.0E-02
Ahna2	-2.27	5.1E-02	-1.89	2.7E-01	-2.10	3.6E-03	-2.27	2.5E-02
Pvr	-2.17	9.7E-03	-1.19	5.7E-01	-1.84	1.4E-02	-1.90	2.8E-02
Aldh1l2	-1.79	4.5E-02	0.08	9.9E-01	-0.09	9.3E-01	-0.59	8.9E-01
Capg	-1.77	4.6E-02	-0.90	7.9E-01	-0.58	3.8E-01	-1.34	1.1E-01
Bax	-1.69	3.2E-02	-1.40	4.4E-01	-1.60	2.8E-02	-1.56	3.0E-02
Tubb2b	-1.69	4.2E-02	-1.37	7.9E-03	-1.45	4.0E-04	-0.87	1.3E-01
Isg15	-1.44	1.9E-02	-0.63	3.1E-01	-1.19	4.7E-03	-1.41	4.8E-03
Mfn1	0.45	4.0E-01	-7.08	2.1E-03	2.82	1.6E-01	0.62	9.8E-01
Gdf15	0.22	8.9E-01	-5.43	4.9E-03	-2.68	1.4E-03	-3.25	2.5E-02
Col5a3	-0.46	7.3E-01	-4.19	4.6E-06	-1.14	8.8E-02	-2.04	1.1E-03
Col18a1	-0.17	7.8E-01	-3.80	2.1E-02	-1.92	9.1E-03	-2.25	2.1E-02
Igfbp3	0.27	8.3E-01	-3.69	1.5E-06	-3.47	6.5E-10	-3.21	1.5E-06
Pdcl	-1.44	4.6E-02	-3.68	4.7E-01	-1.07	1.0E-01	-1.52	1.3E-01
Sema3b	-0.63	4.8E-01	-2.99	3.3E-02	-1.45	1.1E-01	-1.32	1.3E-01
Wisp2	-0.07	9.6E-01	-2.98	3.8E-02	-1.65	3.6E-02	-2.48	3.0E-02
Tenm3	-1.70	2.1E-02	-1.99	2.1E-01	-1.25	8.7E-02	-1.81	2.3E-02
Scarb2	-1.55	2.2E-02	-1.94	4.7E-01	-0.59	5.8E-01	-0.58	6.4E-01
Moxd1	-2.20	4.2E-02	-2.07	1.4E-02	-4.13	7.8E-09	-2.87	5.7E-07
Lpl	-0.35	7.9E-01	-3.94	6.7E-06	-4.11	9.2E-10	-2.68	2.1E-06
Tcn2	0.70	5.5E-01	-2.20	4.1E-04	-3.86	1.6E-07	-3.71	8.6E-08
Postn	0.04	9.6E-01	-2.94	3.3E-02	-3.82	8.2E-04	-2.81	2.4E-02
Postn-2	-1.26	6.6E-01	-3.08	4.6E-02	-3.81	3.0E-03	-2.77	1.3E-01
Pcdh20	0.37	7.8E-01	-2.00	3.0E-04	-3.81	4.9E-08	-3.45	9.8E-08
Mmp1a	0.06	9.5E-01	-2.81	3.8E-02	-3.36	8.2E-04	-3.27	2.6E-03
Vcam1	0.52	8.2E-01	-0.77	2.8E-01	-3.34	1.5E-06	-2.55	4.8E-05
Pcdhgb6	-1.06	3.9E-01	-0.96	1.6E-01	-3.14	1.5E-07	-3.00	2.8E-06
Igf2r	-0.11	9.1E-01	-0.77	6.8E-02	-3.07	5.8E-08	-2.59	7.8E-07
Gpm6b	-1.01	1.4E-01	-2.75	4.4E-02	-2.97	1.3E-03	-2.06	2.3E-01
Cd151	-0.92	4.3E-01	-0.49	6.3E-01	-2.97	1.1E-07	-1.10	1.4E-01
Ccbe1	0.10	9.4E-01	-2.57	3.2E-03	-2.91	1.4E-06	-2.84	7.2E-07
Serpinh1	-1.82	2.9E-03	-1.80	5.8E-03	-2.53	2.6E-07	-1.68	7.0E-04
Gfra2	1.00	5.6E-01	-3.55	4.6E-03	-3.74	7.3E-05	-5.96	2.4E-05
Klk10	0.55	7.4E-01	-1.63	1.0E-02	-5.11	1.5E-08	-5.36	1.9E-08
Sema6a	0.49	8.4E-01	-1.88	3.0E-01	-2.77	4.7E-03	-4.78	1.8E-03
L1cam	-1.45	6.3E-02	-2.17	3.0E-01	-2.86	7.5E-03	-4.74	1.3E-03
Serpine1	0.63	6.7E-01	-1.67	5.8E-02	-3.42	2.5E-05	-4.71	2.4E-06
Nrcam	-0.34	8.1E-01	-1.01	6.9E-01	-2.95	9.5E-04	-4.44	1.3E-03
Serpine2	0.01	9.9E-01	-2.59	8.2E-04	-4.23	4.2E-09	-4.43	5.9E-09
Spp1	0.18	8.6E-01	-1.59	4.7E-01	-2.45	4.1E-03	-4.36	3.0E-03
Mcam	-1.98	1.5E-02	-1.16	6.7E-01	-3.80	1.1E-03	-4.26	1.3E-03
Pcdh1	-1.20	1.3E-01	-1.01	6.9E-01	-3.19	8.2E-04	-4.22	2.5E-03
Col15a1	-0.30	6.4E-01	-3.40	2.1E-02	-3.26	1.3E-03	-4.00	2.6E-03
Col28a1	1.23	7.3E-01	-1.27	8.4E-02	-2.38	1.5E-02	-3.87	1.3E-03
Thbs1	0.81	5.8E-01	-1.25	1.7E-02	-2.20	2.5E-06	-3.81	1.7E-08
Mrgprf	0.07	9.7E-01	-2.47	5.2E-02	-3.48	1.6E-08	-3.77	3.1E-02
Adamts1	0.75	6.6E-01	-2.50	5.2E-04	-2.87	9.5E-07	-3.76	1.7E-08
Mmp3	0.13	8.6E-01	-2.81	4.6E-02	-2.38	6.8E-03	-3.63	1.1E-02
Pam	-0.87	9.7E-02	-2.36	5.6E-02	-2.87	9.5E-04	-3.63	1.8E-03
Megf10	-0.07	9.5E-01	-2.15	9.0E-02	-2.76	9.5E-04	-3.40	2.6E-03
Prss22	0.18	8.7E-01	-1.83	3.0E-01	-2.87	1.6E-03	-3.31	5.3E-03
Crip2	-1.92	6.6E-02	-2.35	3.1E-04	-2.63	1.4E-06	-3.02	2.4E-06

Table S2. PDGFRA differentially enriched proteins. Related to Figure 4.

Log2 FC and adjusted p values of PDGFRA-enriched proteins from Cells, LEVs, SEVs (exosomes) and RNPs. Proteins labeled in Figure 4A are highlighted in blue.

Gene Symbol	Cells		LEVs		SEVs		RNPs	
	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val
Nnt	5.15	1.7E-02	1.17	8.5E-01	1.14	2.7E-01	0.13	9.9E-01
Ptk7	4.40	3.3E-04	0.79	1.0E-01	0.98	2.9E-02	1.20	2.7E-02
Tagln	4.28	8.6E-05	3.15	3.2E-06	3.03	6.2E-08	3.46	4.3E-07
Crabp2	4.18	2.6E-03	1.45	8.6E-03	2.20	3.6E-06	2.37	6.2E-04
Gsta4	3.09	1.1E-03	0.64	4.2E-01	1.41	5.9E-03	1.83	4.0E-03
Fbxo2	2.94	3.9E-03	1.52	2.2E-02	2.16	4.4E-06	2.06	4.2E-04
Finc	2.89	4.8E-03	1.55	4.9E-01	2.75	9.9E-04	1.13	1.5E-01
Arhgdib	2.83	2.2E-02	0.81	9.9E-01	2.16	8.7E-02	2.83	1.5E-01
Aldh2	2.73	7.7E-05	0.81	1.1E-01	0.74	3.3E-02	-0.12	8.7E-01
Lphn2	2.68	2.9E-03	0.17	7.4E-01	0.29	3.6E-01	-0.13	8.2E-01
Csrp2	2.50	3.1E-02	1.21	9.4E-03	1.92	2.5E-06	1.48	7.3E-04
Tgfb3	2.35	2.1E-02	1.43	3.9E-02	1.11	1.4E-03	0.72	1.2E-01
Gstm2	2.27	9.7E-03	0.97	7.7E-01	1.76	2.6E-02	1.96	3.0E-02
Prkcd	2.23	7.0E-03	1.49	6.2E-01	1.27	3.8E-02	1.12	2.5E-01
Nqo1	2.14	2.9E-03	1.78	5.2E-04	1.79	6.2E-06	1.70	4.2E-04
Mcm4	2.07	2.1E-02	1.43	5.1E-01	1.30	6.0E-02	0.10	9.9E-01
Mcm6	2.06	2.9E-03	1.82	2.3E-03	1.32	1.0E-03	0.93	4.1E-02
Slc29a1	2.05	9.7E-03	1.08	6.7E-01	1.10	8.7E-02	0.11	9.9E-01
S100a13	2.03	7.9E-03	-0.10	8.6E-01	-0.39	3.7E-01	-0.40	6.2E-01
Col2a1	0.78	3.8E-01	3.41	2.1E-02	2.77	9.5E-04	3.30	2.8E-03
Fmod	0.58	5.9E-01	2.74	6.6E-04	2.73	1.1E-06	2.57	9.9E-06
Fbln1	2.72	5.4E-01	4.95	3.0E-04	3.70	1.6E-03	3.48	2.1E-06
Ctgf	1.41	3.8E-01	2.56	1.1E-04	1.62	3.2E-04	1.42	4.4E-03
Bgn	1.01	3.7E-01	2.77	1.2E-05	1.87	3.6E-05	2.73	1.9E-06
Lox	1.14	5.7E-01	3.00	1.1E-05	2.05	6.5E-04	2.12	1.1E-04
Mgp	1.48	1.5E-01	3.88	9.5E-06	1.91	1.1E-03	0.21	7.9E-01
Leprel4	1.16	3.5E-01	2.64	4.6E-06	1.78	1.3E-05	0.87	2.3E-02
Clu	0.90	3.5E-01	3.24	3.5E-07	3.11	6.1E-08	1.97	1.1E-04
Angpt1	1.35	4.5E-01	4.65	2.4E-07	3.38	3.3E-08	2.14	2.5E-04
Col1a2	1.04	4.5E-01	5.36	3.2E-08	2.86	3.0E-07	2.43	5.2E-04
Igfbp2	0.74	7.2E-01	4.04	2.4E-07	5.26	4.4E-10	3.31	6.1E-07
Efemp1	1.14	1.9E-01	4.31	2.4E-07	4.89	2.9E-11	4.20	1.6E-08
Igfbp6	1.54	5.0E-01	4.07	3.2E-06	4.79	1.3E-09	3.31	8.1E-07
Ptx3	1.01	3.9E-01	3.02	5.9E-05	4.79	2.9E-11	2.84	6.8E-06
Apoe	1.09	4.5E-01	2.51	7.6E-05	4.38	5.2E-10	0.22	7.0E-01
Tfpi	1.32	2.4E-01	1.95	3.1E-01	4.08	5.3E-04	3.63	2.5E-03
Comp	2.61	5.3E-01	2.50	1.4E-04	3.69	1.6E-07	2.33	3.7E-05
Sned1	0.35	6.2E-01	2.52	1.9E-01	3.50	1.0E-03	2.47	1.5E-01
Ccnb1	2.55	4.5E-02	3.09	7.0E-02	3.32	1.3E-03	2.74	4.6E-02
Tpm1	1.08	1.4E-01	0.66	9.7E-01	3.30	9.5E-04	0.00	1.0E+00
Fbln1-2	0.79	5.0E-01	2.96	3.1E-04	3.23	1.7E-05	2.86	4.2E-04
Nid2	0.89	3.5E-01	2.22	1.2E-04	3.12	1.6E-07	2.14	6.9E-05
Hist1h2bf	1.55	1.4E-01	1.27	8.4E-01	3.10	2.8E-02	0.91	7.1E-01
Msln	1.35	3.5E-01	2.74	4.4E-06	3.04	4.6E-09	2.23	9.9E-06
Fkbp10	0.97	2.7E-01	2.09	1.4E-04	3.02	2.0E-09	2.78	1.6E-06
Tpm3	0.34	6.1E-01	0.15	9.9E-01	2.96	1.3E-03	-0.57	5.8E-01
Fgfbp1	1.84	2.9E-01	2.07	5.7E-01	2.92	1.6E-02	1.67	6.0E-01
Bmp6	1.34	3.5E-01	2.36	2.4E-05	2.90	7.3E-09	0.85	6.1E-02
Dkk3	0.75	4.1E-01	2.07	4.1E-04	3.37	3.6E-08	4.58	2.7E-08
Nbl1	1.45	2.9E-01	2.42	5.4E-04	2.46	1.0E-05	3.06	1.1E-06
Cst6	0.75	6.7E-01	1.78	1.7E-02	2.69	2.0E-06	2.68	1.6E-06
Cdh11	2.07	3.4E-02	1.83	3.0E-04	2.18	7.9E-06	2.67	4.9E-06
Cx3cl1	2.21	2.8E-01	1.64	1.2E-03	2.36	1.2E-06	2.52	1.2E-06
Col7a1	0.58	6.0E-01	2.48	6.3E-06	2.49	5.0E-07	2.41	2.7E-05

Table S3. EGFR differentially enriched proteins. Related to Figure 4.

Log2 FC and adjusted p values of EGFR-enriched proteins from Cells, 10,000 xg and 100,000 xg ultracentrifugation. Proteins labeled in Figure 4B are highlighted in red.

Gene Symbol	Cells		10,000 xg		100,000 xg	
	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val
Ephx1	-2.86	1.1E-02	-1.41	2.7E-02	-1.37	1.1E-02
Hspa1a	-2.30	1.7E-02	-0.79	1.9E-01	-0.52	1.7E-02
Serpinh1	-1.99	1.1E-02	-0.89	9.4E-02	-1.52	1.1E-02
Uchl1	-1.67	1.4E-01	-0.85	2.8E-02	0.12	1.4E-01
Fabp5	-1.50	1.7E-02	-1.04	4.7E-02	-1.33	1.7E-02
Moxd1	-0.65	6.2E-01	-3.82	2.8E-03	-3.40	6.2E-01
Prelp	-0.42	5.3E-01	-3.40	3.6E-03	-1.75	5.3E-01
Prnp	0.21	8.3E-01	-3.16	8.8E-03	-2.21	8.3E-01
Gdf15	-1.15	4.9E-01	-3.15	2.5E-03	-2.63	4.9E-01
Pam	-1.25	4.5E-02	-2.89	3.5E-03	-2.40	4.5E-02
Itga2	-0.26	6.1E-01	-2.82	2.5E-03	-2.02	6.1E-01
Dmd	-1.21	4.2E-02	-2.72	1.2E-03	-2.59	4.2E-02
Itga1	-0.17	6.0E-01	-2.71	1.3E-03	-2.06	6.0E-01
Vamp3	-0.15	8.9E-01	-2.34	2.3E-03	-1.70	8.9E-01
Olfml2b	-0.84	5.3E-01	-2.19	5.8E-03	-0.93	5.3E-01
Ldlr	-0.32	6.5E-01	-2.10	2.5E-03	-1.81	6.5E-01
Agfg1	0.02	9.8E-01	-2.07	4.7E-03	-1.48	9.8E-01
Ctsc	-1.79	1.5E-02	-1.90	5.4E-03	-1.19	1.5E-02
Col15a1	-0.96	3.9E-01	-3.49	1.8E-03	-4.06	3.9E-01
Igfbp3	-0.22	6.2E-01	-3.46	1.3E-03	-3.92	6.2E-01
Gpm6b	-0.26	7.3E-01	-3.23	1.5E-03	-3.90	7.3E-01
Itga6	-0.88	1.7E-01	-2.89	1.5E-03	-3.20	1.7E-01
Itga7	-0.85	3.2E-01	-2.38	2.8E-03	-3.18	3.2E-01
Itgb1	-0.71	8.1E-02	-2.01	5.5E-03	-3.17	8.1E-02
Epha2	-1.71	1.1E-02	-1.94	7.8E-03	-3.05	1.1E-02
Pvr	-2.19	1.5E-02	-2.67	2.5E-03	-3.02	1.5E-02
Slc7a5	-0.55	2.9E-01	-2.21	5.8E-03	-2.98	2.9E-01
Dip2c	-0.03	9.4E-01	-0.95	3.3E-02	-2.90	9.4E-01
Cd151	-0.74	1.4E-01	-2.47	1.8E-03	-2.87	1.4E-01
Itga4	-0.55	4.7E-01	-2.38	1.5E-03	-2.86	4.7E-01
Col18a1	-0.03	9.5E-01	-2.18	5.2E-03	-2.85	9.5E-01
Tenm3	-0.38	3.1E-01	-2.28	1.5E-03	-2.85	3.1E-01
Gpc6	-0.45	2.7E-01	-1.60	1.7E-02	-2.85	2.7E-01
Il1rap	-0.28	4.3E-01	-2.70	1.3E-03	-2.83	4.3E-01
Slc3a2	-0.60	2.8E-01	-2.22	3.3E-03	-2.75	2.8E-01
Slc16a13	0.04	9.1E-01	-1.91	5.9E-03	-2.74	9.1E-01
Pcdhgc3	-0.96	9.8E-02	-1.85	2.8E-03	-2.74	9.8E-02
P2rx7	-0.94	3.7E-01	-2.10	2.9E-03	-2.64	3.7E-01
Stam	-0.05	9.5E-01	-1.22	1.5E-02	-2.60	9.5E-01
Cd276	-0.21	8.0E-01	-1.69	4.7E-03	-2.58	8.0E-01
Serpine2	0.51	6.5E-01	-2.32	2.1E-02	-2.57	6.5E-01
Slc7a1	-1.36	1.1E-01	-2.52	4.7E-03	-2.56	1.1E-01
Cd81	-0.72	1.1E-01	-1.83	2.5E-03	-2.52	1.1E-01
Nrp1	0.24	7.3E-01	-1.04	4.7E-02	-2.52	7.3E-01
Hgs	0.24	7.4E-01	-1.14	2.7E-02	-2.52	7.4E-01
Cd44	-0.35	6.3E-01	-2.26	2.5E-03	-2.50	6.3E-01
Tcn2	-0.29	5.8E-01	-2.02	4.7E-03	-2.03	5.8E-01

Table S4. PDGFRA differentially enriched proteins. Related to Figure 4.

Log2 FC and adjusted p values of PDGFRA-enriched proteins from Cells, 10,000 xg and 100,000 xg ultracentrifugation. Proteins labeled in Figure 4B are highlighted in blue.

Gene Symbol	Cells		10,000 xg		100,000 xg	
	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val	Log2 FC	adj.P.Val
Tagln	3.57	1.5E-02	2.85	1.2E-02	1.55	1.5E-02
Ptk7	2.79	1.8E-02	1.45	5.3E-03	0.67	1.8E-02
Gstm2	2.61	1.1E-02	1.51	4.6E-02	0.79	1.1E-02
Finc	2.56	1.1E-02	2.03	1.5E-03	1.52	1.1E-02
Slc29a1	2.32	1.5E-02	1.87	8.8E-03	0.49	1.5E-02
Cdh11	2.12	2.3E-02	1.45	2.6E-02	1.52	2.3E-02
Tmem43	1.92	1.1E-01	0.88	1.7E-01	-0.04	1.1E-01
Dab2	1.90	1.7E-02	0.86	1.6E-01	0.68	1.7E-02
Aldh2	1.76	8.5E-02	0.86	2.4E-02	1.39	8.5E-02
Acat2	1.75	6.3E-02	1.25	3.5E-02	1.07	6.3E-02
Nsdhl	1.70	1.5E-02	1.62	8.8E-03	0.30	1.5E-02
Vcam1	1.65	1.4E-02	1.57	1.3E-02	0.21	1.4E-02
Ndrp1	1.59	1.7E-02	1.01	5.6E-02	0.39	1.7E-02
Pkm	1.50	4.9E-02	0.69	6.8E-02	0.93	4.9E-02
Prkcd	2.58	3.1E-03	3.21	1.3E-03	2.23	3.1E-03
Fgfbp1	1.24	5.7E-01	3.06	9.8E-04	2.48	5.7E-01
Lox	0.12	8.9E-01	2.70	1.3E-03	2.46	8.9E-01
Efemp1	1.19	1.6E-01	2.68	3.3E-03	2.60	1.6E-01
U2af2	0.49	2.4E-01	2.06	1.5E-03	1.79	2.4E-01
Col1a2	-0.04	9.7E-01	3.80	1.2E-03	4.14	9.7E-01
Luc7l	-0.10	9.2E-01	3.31	1.3E-03	3.95	9.2E-01
Igfbp6	0.88	4.7E-01	3.04	1.2E-03	3.55	4.7E-01
Lgals3bp	0.60	1.8E-01	1.15	4.4E-02	3.54	1.8E-01
Hist1h4a	0.23	6.8E-01	1.63	8.8E-03	3.21	6.8E-01
Psmb6	0.54	6.5E-01	1.25	7.8E-03	3.14	6.5E-01
Hist1h3a	0.09	8.6E-01	1.28	7.3E-03	3.11	8.6E-01
Hist1h2bf	0.67	2.6E-01	1.79	3.0E-03	3.03	2.6E-01
Clec16a	0.83	4.3E-01	1.63	3.3E-03	3.03	4.3E-01
Angpt1	-0.01	9.9E-01	0.96	1.2E-01	2.96	9.9E-01
Psm2	0.36	7.9E-01	1.33	1.3E-02	2.91	7.9E-01
Psm4	0.75	4.3E-01	1.17	1.2E-02	2.88	4.3E-01
Psm1	0.37	5.0E-01	1.32	8.7E-03	2.82	5.0E-01
Bgn	0.22	6.8E-01	2.15	1.3E-02	2.81	6.8E-01
Vcan	0.45	3.2E-01	0.89	2.4E-01	2.72	3.2E-01
Cdk1	1.58	1.4E-01	1.88	5.7E-03	2.70	1.4E-01
Thbs3	0.33	5.0E-01	0.71	1.6E-01	2.62	5.0E-01
Ssb	-0.05	9.3E-01	2.12	2.5E-03	2.57	9.3E-01
Dhx15	0.28	5.5E-01	1.62	4.7E-03	2.57	5.5E-01
Psm6	0.57	4.3E-01	0.99	1.9E-02	2.55	4.3E-01
Sf3a2	0.65	4.7E-01	2.01	1.5E-03	2.51	4.7E-01
Gmps	0.92	1.4E-01	2.04	1.3E-03	2.50	1.4E-01
Cp	1.52	8.3E-02	1.72	1.4E-02	2.29	8.3E-02
Hnrnpab	0.40	5.0E-01	2.15	1.5E-03	2.23	5.0E-01
Mcm2	1.72	9.8E-02	1.12	7.4E-03	1.78	9.8E-02

Table S5. Composition of long RNA categories. Related to Figure 6.

Subcategory annotations and composition of the long RNA categories represented in Figure 6A.

Categories	Subcategories
Pseudogene	polymorphic pseudogene processed pseudogene pseudogene transcribed processed pseudogene transcribed unitary pseudogene transcribed unprocessed pseudogene translated processed pseudogene unitary pseudogene unprocessed pseudogene
LncRNA	bidirectional promoter lncRNA lincRNA macro lncRNA
mRNA	processed transcript protein coding
mtRNA	Mt rRNA Mt tRNA
RNA decay	non stop decay nonsense mediated decay
Intronic	retained intron sense intronic sense overlapping
small RNA	miRNA snoRNA snRNA sRNA
Other RNA	3prime overlapping ncRNA antisense IG C gene misc RNA ribozyme rRNA scaRNA TEC TR C gene

Table S6. Composition of small RNA categories in Figure 6A. Related to Figure 6.

Subcategory annotations and composition of the small RNA categories represented in Figure 6A.

Categories	Subcategories
Repeats	Low complexity repeat Simple Repeat Satellite repeat SINE LINE LTR
miRNA	
tRNA	
Y RNA	
snRNA	
snoRNA	
srpRNA	
scRNA	
VaultRNA	
rRNA	
Other ncRNA	ncRNA 7SK RNA Pseudogene lncRNA antisense lncRNA scaRNA mitochondrial tRNA
Unannotated	

Table S7. Composition of the tRNA gene clusters. Related to Figure 7.

tRNA Names	tRNA Genes Bins
Asp-GTC-01	chr10.tRNA866-AspGTC
Gly-TCC	chr3.tRNA746-GlyTCC; chr1.tRNA1003-GlyTCC; chr1.tRNA1012-GlyTCC; chr1.tRNA708-GlyTCC; chr1.tRNA1009-GlyTCC; chr11.tRNA1824-GlyTCC; chr1.tRNA1006-GlyTCC
Gly-GCC-01	tRNA; repeat; chr1.tRNA702-GlyGCC; chr1.tRNA704-GlyGCC; chr1.tRNA706-GlyGCC
Asp-GTC-02	chr13.tRNA84-AspGTC; chr13.tRNA993-AspGTC; chr1.tRNA1002-AspGTC; chr13.tRNA991-AspGTC; chr10.tRNA861-AspGTC; chr11.tRNA397-AspGTC; chr1.tRNA1013-AspGTC; chr1.tRNA707-AspGTC; chr11.tRNA69-AspGTC; chr1.tRNA1008-AspGTC; chr5.tRNA1317-AspGTC; chr5.tRNA1315-AspGTC; chr1.tRNA1005-AspGTC
Val-CAC	chr11.tRNA204-ValCAC; chr13.tRNA91-ValCAC; chr13.tRNA966-ValCAC; chr3.tRNA48-ValAAC; chr3.tRNA284-ValCAC; chr1.tRNA710-ValCAC; chr11.tRNA208-ValAAC
Ala-AGC-01	chr13.tRNA978-AlaAGC; chr13.tRNA94-AlaAGC; chr13.tRNA96-AlaAGC
Leu-AAG	tRNA189-LeuAAG; chr13.tRNA62-LeuAAG
Cys-GCA	chr17.tRNA457-CysGCA; chr11.tRNA1442-CysGCA; chr6.tRNA157-CysGCA; chr11.tRNA791-CysGCA; chr11.tRNA1433-CysGCA; chr11.tRNA1432-CysGCA; chr9.tRNA593-CysGCA
Leu-TAG	chr14.tRNA709-LeuTAG
Glu-TTC-01	chr3.tRNA754-GluTTC; chr3.tRNA286-GluTTC
Glu-TTC-02	chr1.tRNA1555-GluTTC; chr14.tRNA364-GluTTC
Glu-TTC-03	chr13.tRNA105-GluTTC; chr14.tRNA352-GluTTC; chr9.tRNA961-GluTTC; chr7.tRNA339-GluTTC
Ala-CGC	chr4.tRNA622-AlaCGC
Glu-CTC-01	chr17.tRNA719-GluCTC
Glu-CTC-02	chr11.tRNA1912-GluCTC; chr1.tRNA1004-GluCTC; chr3.tRNA745-GluCTC; chr13.tRNA1013-GluCTC; chr3.tRNA303-GluCTC; chr10.tRNA90-GluCTC; chr1.tRNA1010-GluCTC; chr1.tRNA1010-GluCTC; chr1.tRNA709-GluCTC; chr1.tRNA1007-GluCTC
Gly-GCC-02	chr11.tRNA1819-GlyGCC; chr13.tRNA77-GlyGCC; chr8.tRNA562-GlyGCC; chr3.tRNA878-GlyGCC; chr8.tRNA892-GlyGCC; chr8.tRNA891-GlyGCC; chr13.tRNA110-GlyGCC; chr1.tRNA699-GlyGCC; chr1.tRNA1400-GlyGCC; chr2.tRNA1747-GlyGCC; chr3.tRNA282-GlyGCC
Ala-AGC-02	chr13.tRNA102-AlaAGC; chr19.tRNA8-AlaAGC
Phe-GAA	chr5.tRNA1316-PheGAA
Glu-CTC-03	chr3.tRNA622-GluCTC
Glu-CTC-04	chr7.tRNA969-GluCTC
Val-AAC	chr13.tRNA95-ValAAC
Gly-CCC	chr3.tRNA752-GlyCCC; chr4.tRNA67-GlyCCC
His-GTG	chr4.tRNA1691-HisGTG; chr3.tRNA291-HisGTG; chr3.tRNA751-HisGTG; chr3.tRNA295-HisGTG; chr2.tRNA587-HisGTG; chr2.tRNA1432-HisGTG; tRNA; chr2.tRNA1431-HisGTG; chr3.tRNA747-HisGTG
Gln-CTG	chr13.tRNA987-GlnCTG
Gln-TTG	chr11.tRNA630-GlnTTG; chr11.tRNA630-GlnTTG; tRNA; chr11.tRNA1493-GlnTTG; chr13.tRNA114-GlnTTG; chr13.tRNA1007-GlnTTG; chr13.tRNA113-GlnTTG
Ser-GCT	chr13.tRNA988-SerGCT
Arg-TCT	chr19.tRNA639-ArgTCT; chr11.tRNA1818-ArgTCT
Ala-TGC	chrX.tRNA375-AlaTGC