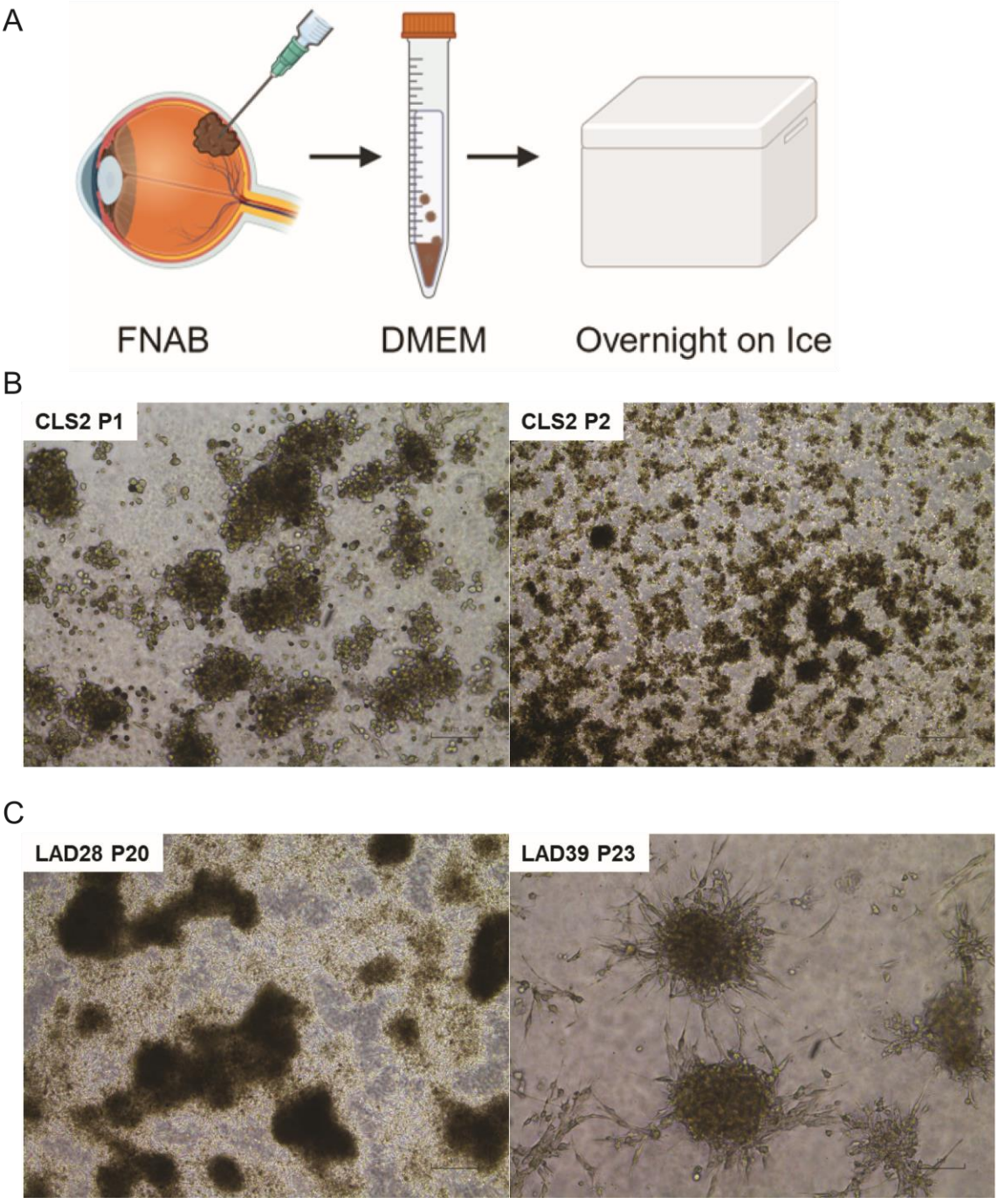
Supplemental Figure 1: Uveal melanoma patient-derived organoids can be generated from externally shipped samples and can be carried for long term passaging.

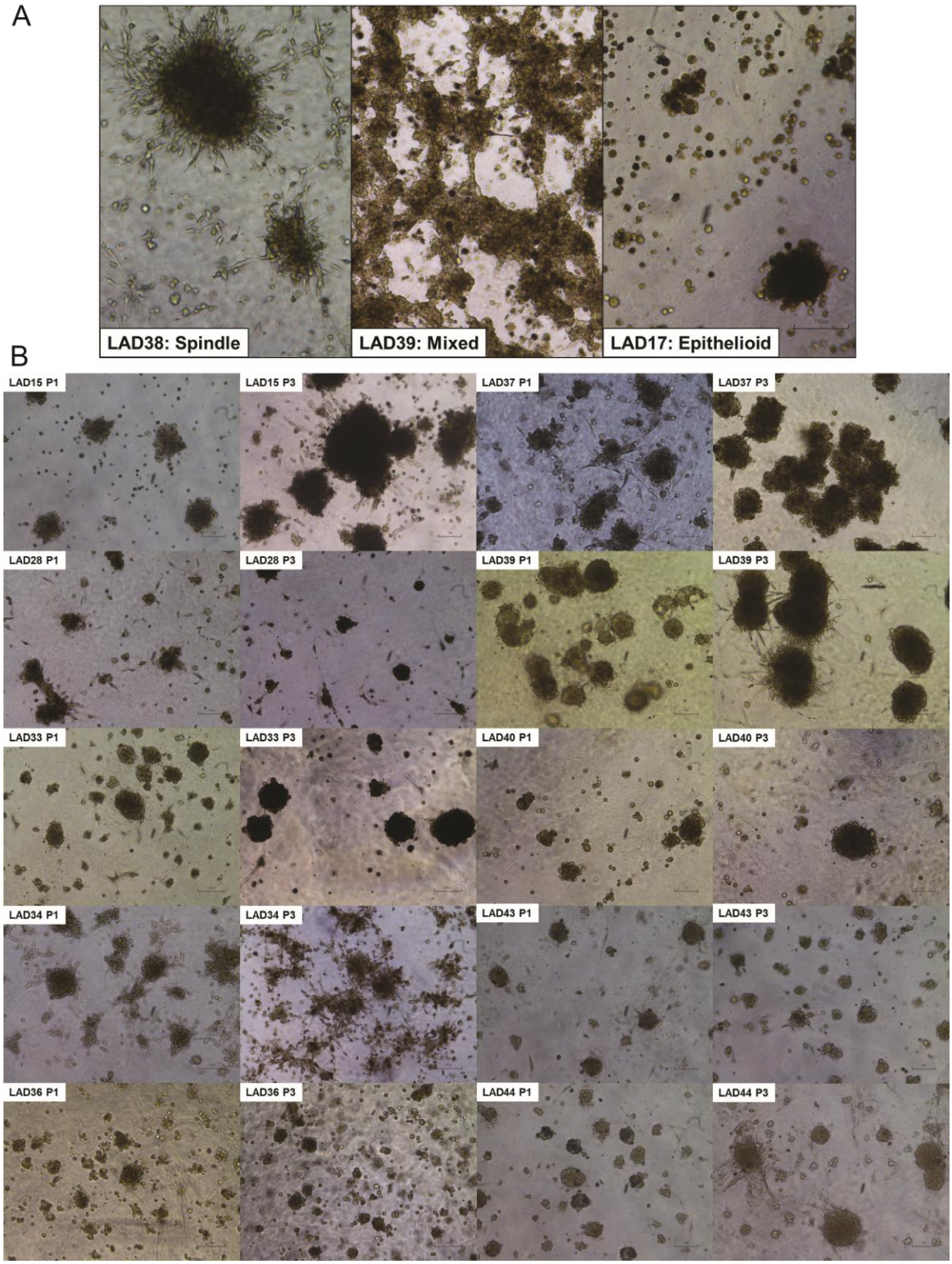


Supplemental Figure 1: Uveal melanoma patient-derived organoids can be generated from externally shipped samples and can be carried for long term passaging.

(A) External collaborator uveal melanoma (UM) patient-derived organoid (PDO) pipeline. Fine needle aspiration biopsy of UM is done by collaborating surgeons, and the tissue sample is deposited in cell culture preservation media (DMEM with 10% FBS, PSG, gentamicin, and Fungizone). The samples are shipped overnight on ice and processed in the same fashion as

internally collected samples using gentleMACS dissociation. (B) PDOs have been successfully established and passaged from primary tumor biopsies contributed by external collaborating centers. (C) Some PDOs have been carried beyond passage 20.

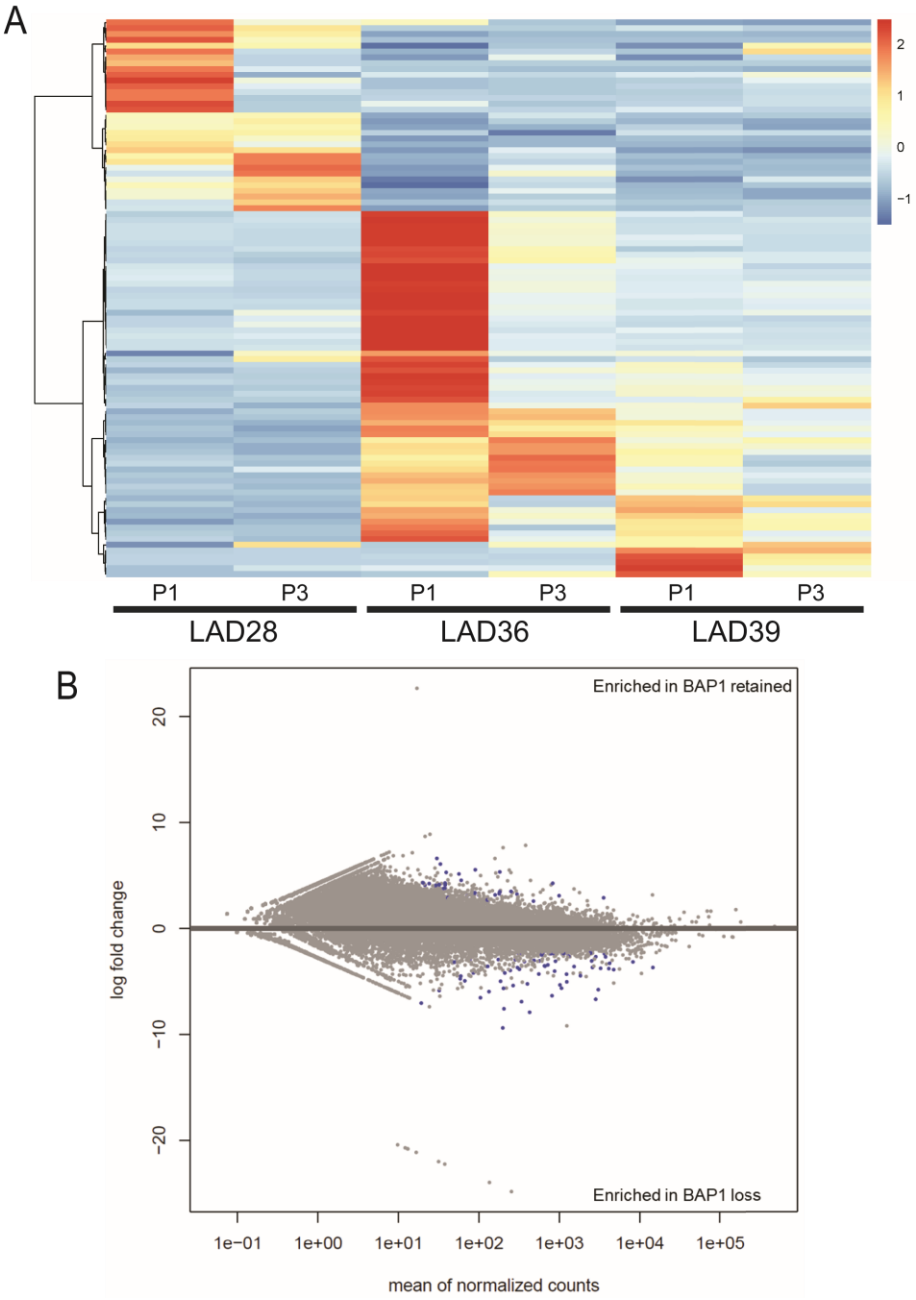
Supplemental Figure 2: Uveal melanoma patient-derived organoid morphology varies between organoids and remains stable through passaging.



Supplemental Figure 2: Uveal melanoma patient-derived organoid morphology varies between organoids and remains stable through passaging.

Uveal melanoma patient-derived organoids (PDOs) display variable morphology and degrees of pigmentation. (A) PDO morphology often reflects the corresponding primary tumor predominance of spindle, mixed, or epithelioid cell type. (B) Once initial organoid colonies form, PDO morphology can vary between organoids but appears to remain stable within each organoid through early passaging from P1 to P3.

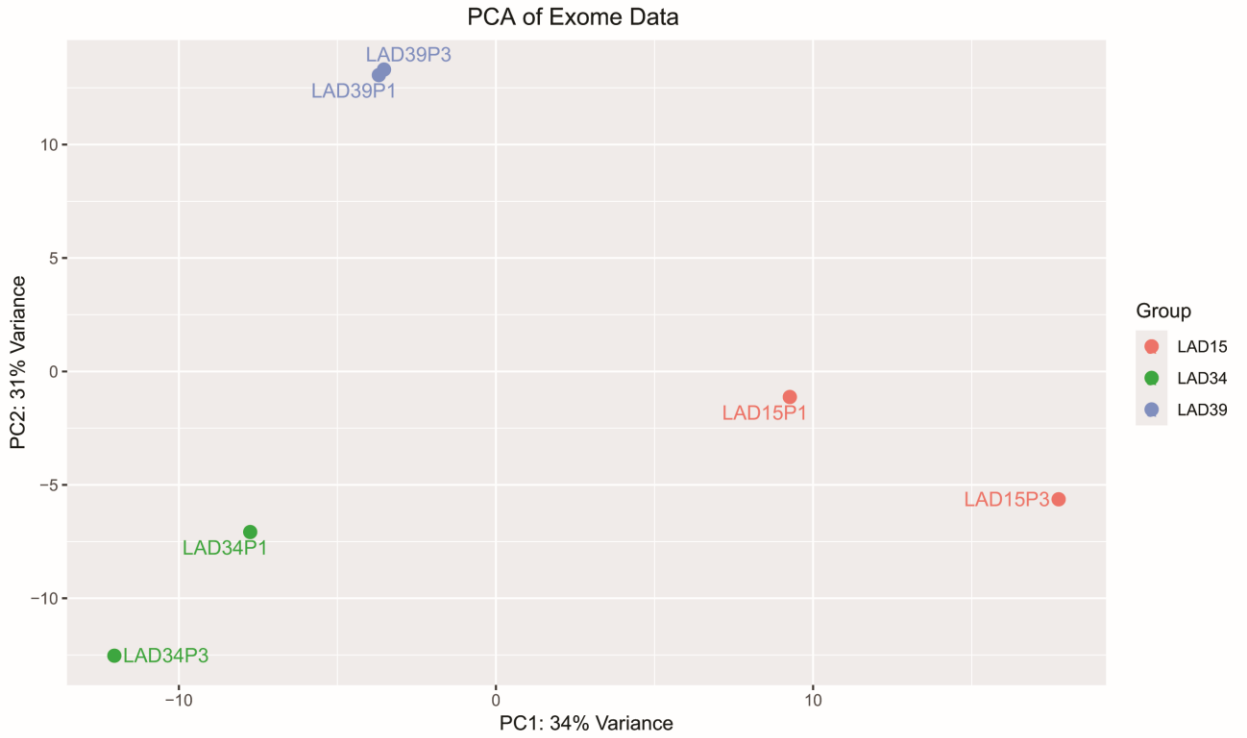
Supplemental Figure 3: Uveal melanoma patient-derived organoid gene expression profile remains stable through passaging and organoids have differential gene expression by BAP1 status.



Supplemental Figure 3: Uveal melanoma patient-derived organoid gene expression profile remains stable through passaging and organoids have differential gene expression by BAP1 status.

(A) Heatmap showing z scores for gene expression for LAD28, LAD36, and LAD39 at P1 and P3. Same PDOs clustered together at P1 and P3 when examining a subset of genes chosen for differential expression in UM with or without the hallmark feature of BAP1 loss. (B) MA plot of global differential gene expression between a cohort of three BAP1 retained and six BAP1 loss PDOs showing differential expression of 97 genes. PDOs with retained BAP1 had enrichment of 33 genes, while PDOs with BAP1 loss had enrichment of 64 genes.

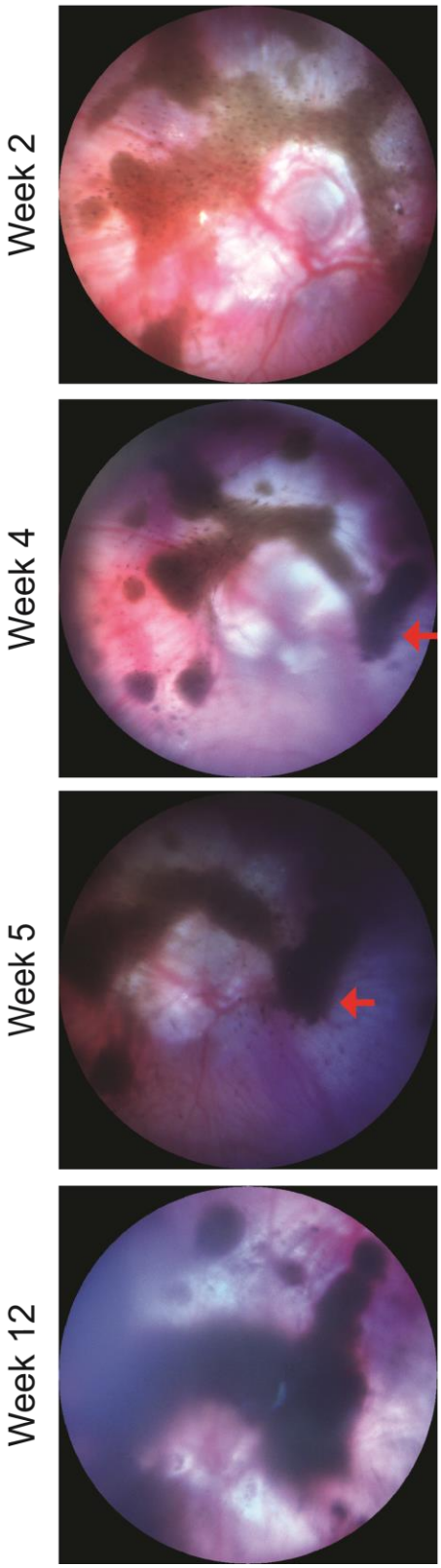
Supplemental Figure 4: Uveal melanoma patient-derived organoids maintain stable whole exome sequencing results through passaging.



Supplemental Figure 4: Uveal melanoma patient-derived organoids maintain stable whole exome sequencing results through passaging.

Principal component analysis (PCA) plot shows comparison of whole exome sequencing results for uveal melanoma patient-derived organoids from paired samples at P1 and P3 for LAD15, LAD34, and LAD39.

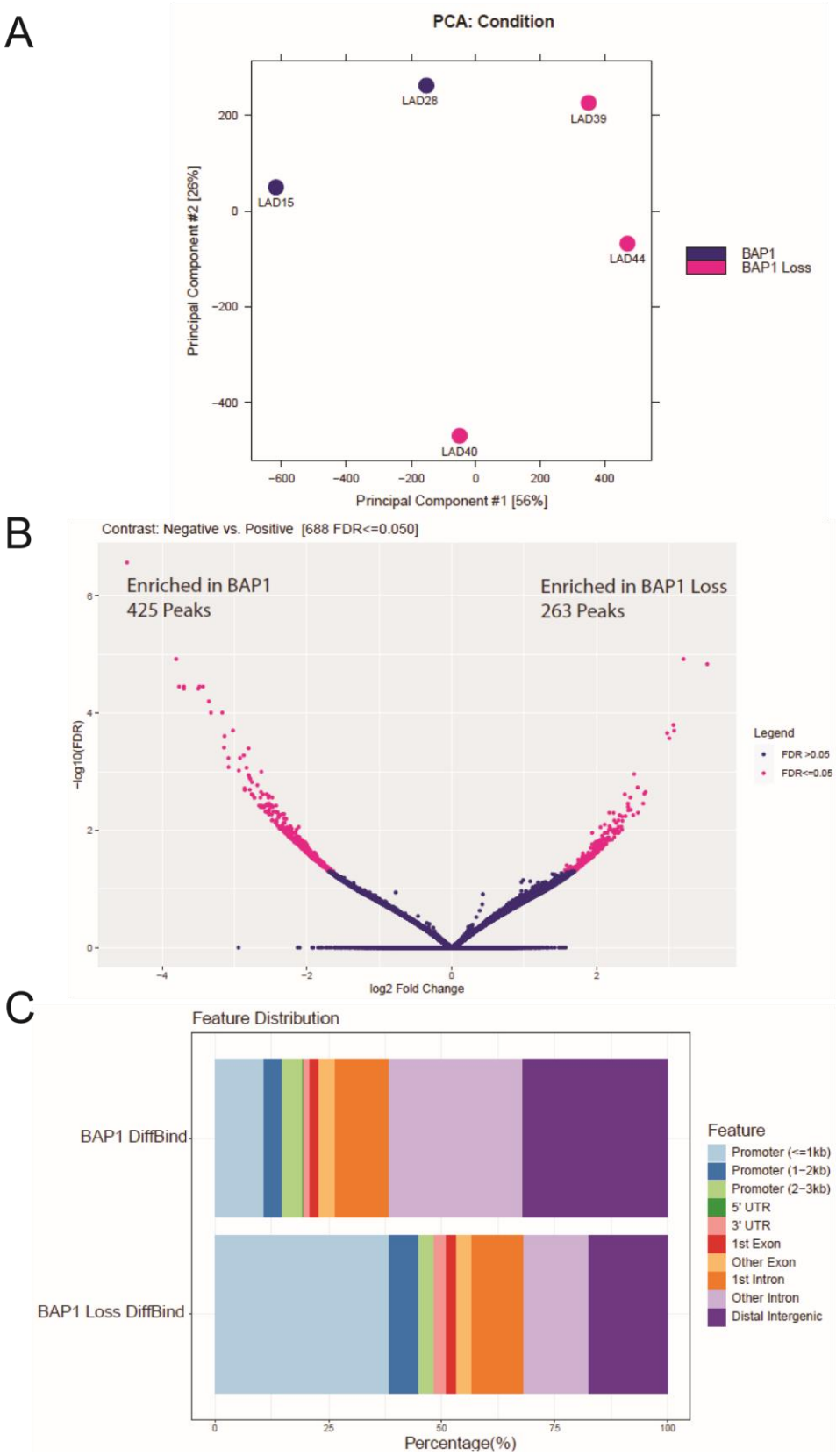
Supplemental Figure 5: Serial monitoring of murine orthotopic xenografts generated from uveal melanoma patient-derived organoids recapitulates *in vivo* human disease.



Supplemental Figure 5: Serial monitoring of murine orthotopic xenografts generated from uveal melanoma patient-derived organoids recapitulates *in vivo* human disease.

Color fundus photography serial monitoring demonstrates successful establishment of intraocular tumors 2 weeks after suprachoroidal injection of uveal melanoma (UM) patient-derived material (LAD39). By week 4, lesions become more dense and deeply pigmented. Slow growth of the intraocular tumors can be seen (red arrows) from weeks 4 to 5. By week 12, the tumor is large and elevated.

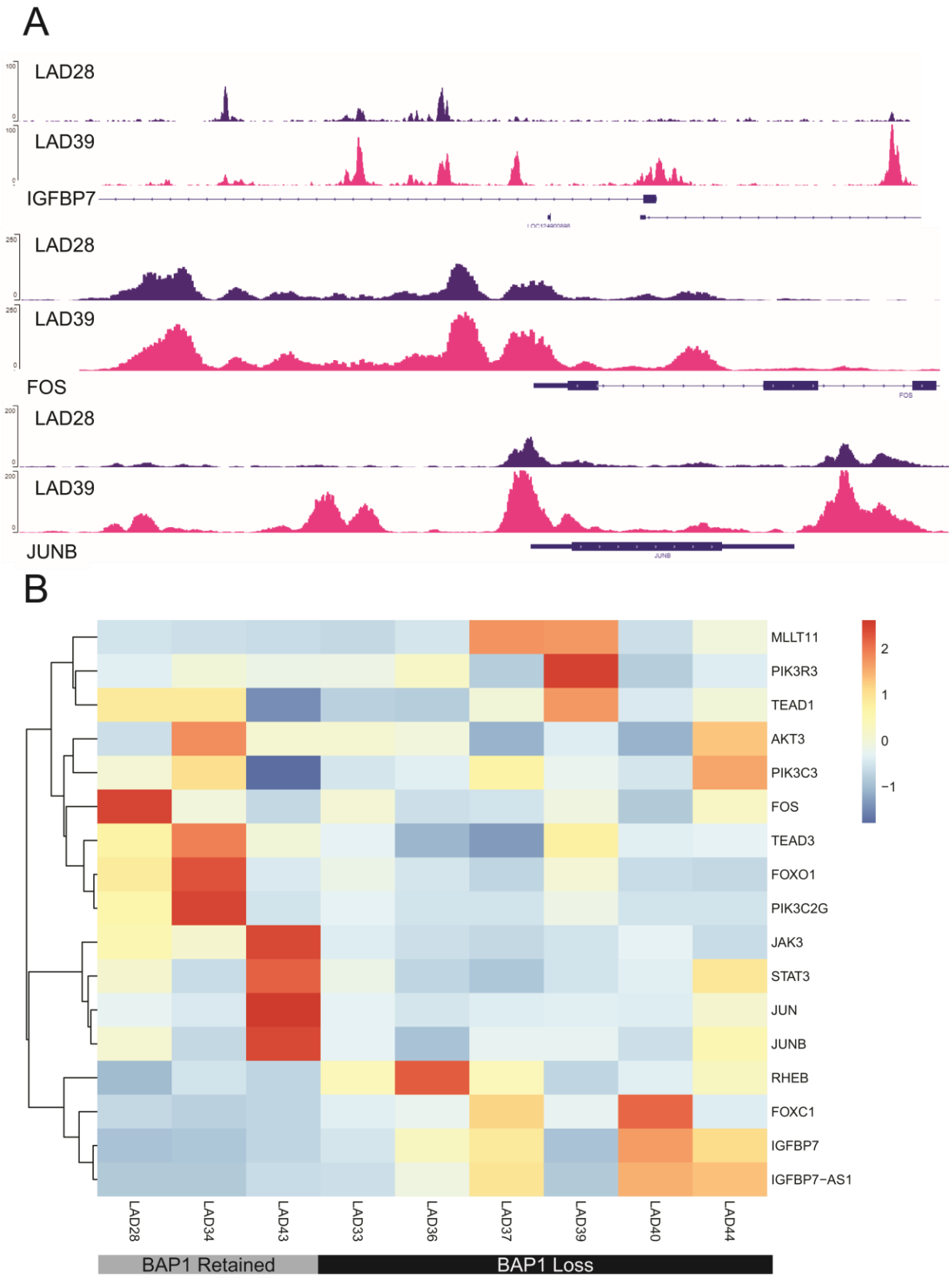
Supplemental Figure 6: Uveal melanoma patient-derived organoids have differential chromatin accessibility based on BAP1 status.



Supplemental Figure 6: Uveal melanoma patient-derived organoids have differential chromatin accessibility based on BAP1 status.

A) PCA plot for ATAC-sequencing using DiffBind for BAP1 retained and BAP1 loss patient-derived organoids (PDOs.) B) Volcano plot showing differentially accessible regions for BAP1 retained vs BAP1 loss (FDR ≤ 0.05). C) Feature distribution of differentially accessible peaks in the genome.

Supplemental Figure 7: Uveal melanoma patient-derived organoids can be used for epigenomics studies to identify drug targets.



Supplemental Figure 7: Uveal melanoma patient-derived organoids can be used for epigenomics studies to identify drug targets.

On ATAC-sequencing analysis, uveal melanoma (UM) patient-derived organoids (PDOs) with retained BAP1 expression had increased chromatin accessibility of promotor/enhancer regions of AKT3, IGFBP7, PIK3C2G, PIK3C3, PIK3R3, and TEAD3. PDOs with BAP1 loss had increased chromatin accessibility for promotor or enhancer regions of FOS, a different site associated with IGFBP7, JUN, JUNB, RHEB, STAT3, and TEAD1. (A) Select tracks for representative BAP1 retained (LAD28, purple) and loss (LAD39, pink) samples are shown, including (top to bottom) IGFBP7, FOS, and JUNB. (B) Integration with RNA-sequencing data was suggestive of increased expression of IGFBP7 (p<0.001, FDR=0.13) and IGFBP7-AS1 (p=0.003, FDR=0.20) in BAP1 loss samples compared to those with retained BAP1 expression.

Supplemental Table 1: Clinical features of patients who donated tumor for uveal melanoma patient-derived organoids (PDOs) characterized by RNA-sequencing

Study Number	Sex	Age	Race	BAP1 Protein Expression	DecisionDx Class	Metastasis	Time to metastasis (months)	Total Follow-up (months)	Status at Last Follow-up
LAD28	M	43	white	retained	1B	No		28.54	Alive
LAD34	F	62	white	retained	1B	No		27.91	Alive
LAD43	M	66	white	retained	1B	No		17.79	Dead, unrelated cause
LAD33	M	68	white	lost	2	No		24.99	Alive
LAD36	M	52	white	lost	2	Yes	5.72	26.66	Alive
LAD37	F	55	white	lost	2	Yes	20.15	22.98	Alive
LAD39	F	62	white	lost	2	Yes	6.64	11.61	Alive
LAD40	M	67	white	lost	2	No		16.57	Alive
LAD44	M	70	white	lost	2	Yes	15.42	17.36	Alive

Supplemental Table 2: Primers used for PCR Resequencing

Primer	Sequence
GNAQ Exon 5 PCR F	GCTCACACATCTGACAGAAGAGC
GNAQ Exon 5 PCR R	TCTTAAAACCATCATGATGTGTTACCCAG
GNAQ Seq Exon 5 F	GTAAGTTCACTCCATTCCCCACA
GNAQ Seq Exon 5 R	TTAATTGAATTGACTTGGATGATCATCGTC
BAP1 Exon 6-7 PCR F	CCACTGGGTACCACATACCAG
BAP1 Exon 6-7 PCR R	TGTTCCTTCCGATTCCTGGAATGC
BAP1 Seq Exon 7 F	AGCTCCCTAGGAGGTAGGC
BAP1 Seq Exon 7 R	TTTGCCTTCCACCCATAGTC