

## Supplementary Data 1

### **Measuring differentiation-state heterogeneity in primary patient samples and patient-derived xenografts.**

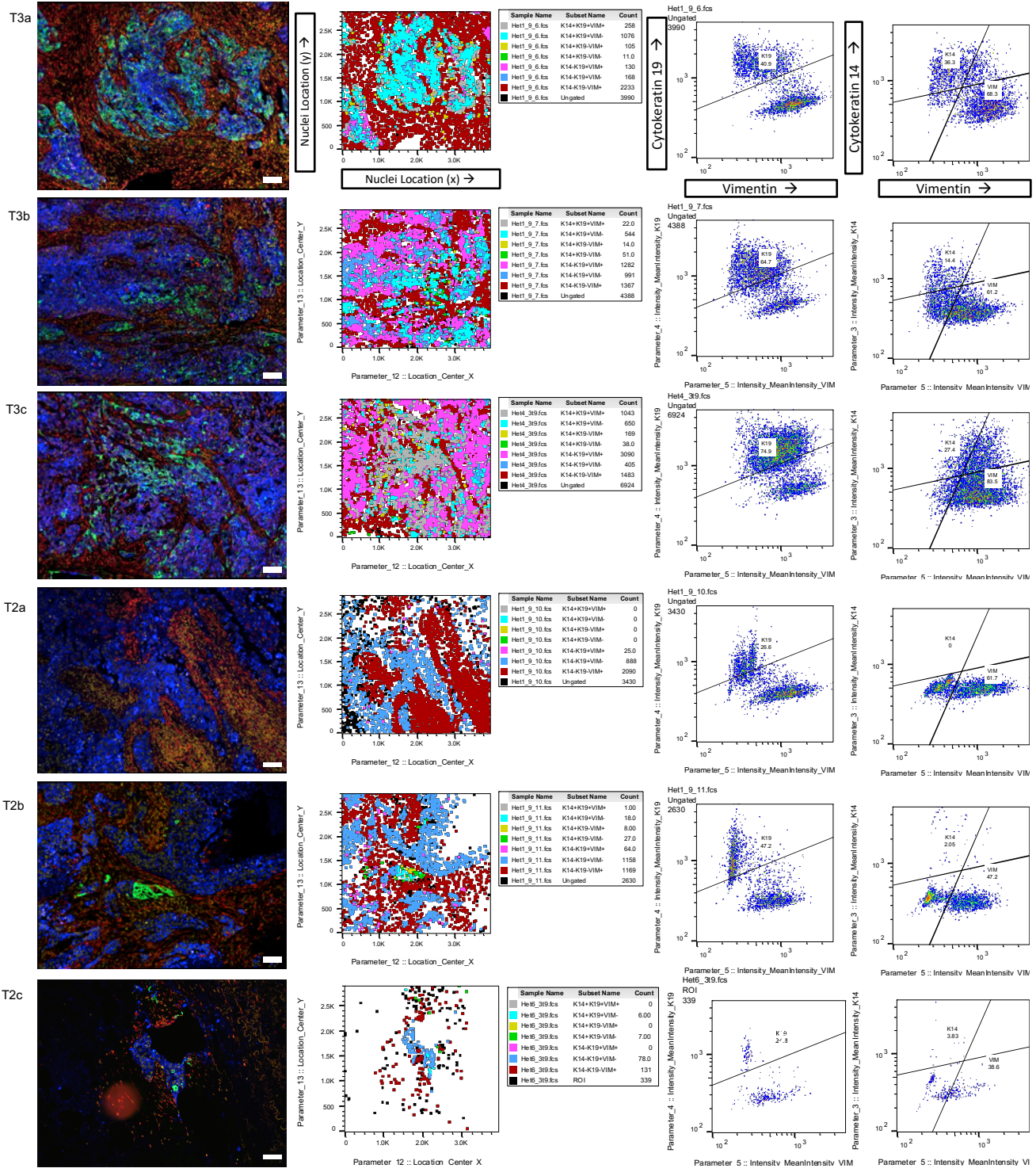
All patient tumors (p.2-7) and patient-derived xenograft tumors (p.8-11) analyzed in this study are presented with the following: 4-color IF images showing DAPI (yellow), K19 (blue), K14 (green) and VIM (red) with normal structures excluded, scale bars = 100 $\mu$ m (left panel). An x/y dotplot showing single cell location and cellular phenotype, with an accompanying legend showing phenotype counts (middle panel). Dot plots comparing single tumor cell intensity of K19 vs. VIM, and K14 vs. VIM, with positivity gates shown for each marker (right panel).

# Triple Negative Tumors

IF: DAPI K19 K14 VIM

Cell Phenotype (K19 / K14 / VIM):

■ +/+/+ 
 ■ -/+/- 
 ■ -/-/+ 
 ■ +/+/+ 
 ■ +/+/+ 
 ■ -/+/+ 
 ■ +/+/+ 
 ■ -/-/-



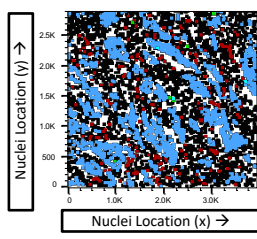
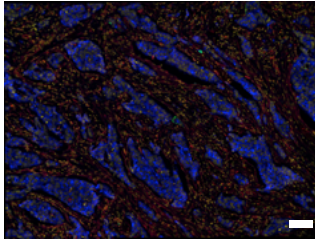
# Triple Negative Tumors

IF: DAPI K19 K14 VIM

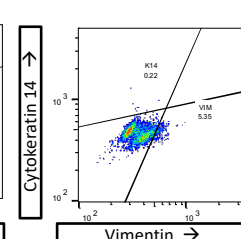
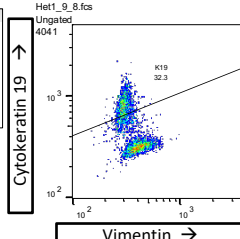
Cell Phenotype

(K19 / K14 / VIM): + / - / - - / + / - - / - / + + / - / + + / + / + + - / - / -

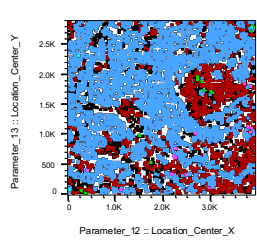
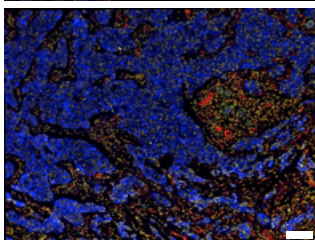
T1a



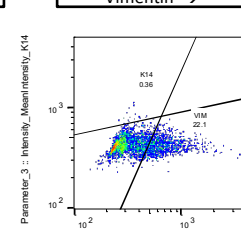
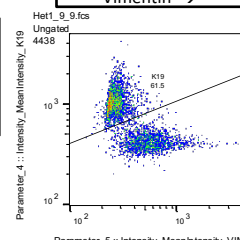
Sample Name	Subset Name	Count
Hett_9_8.fcs	K14+K19+VIM+	0
Hett_9_8.fcs	K14+K19+VIM-	3,000
Hett_9_8.fcs	K14-K19+VIM+	0
Hett_9_8.fcs	K14-K19+VIM-	6,000
Hett_9_8.fcs	K14-K19-VIM+	0
Hett_9_8.fcs	K14-K19-VIM-	1,304
Hett_9_8.fcs	K14-K19-VIM+	216
Hett_9_8.fcs	Ungated	4,041



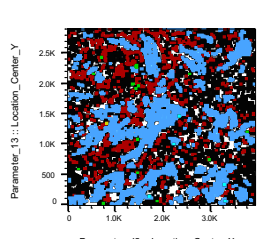
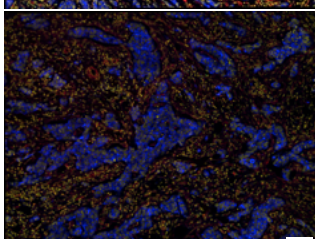
T1b



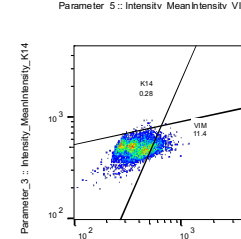
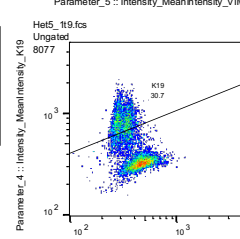
Sample Name	Subset Name	Count
Hett_9_9.fcs	K14+K19+VIM+	0
Hett_9_9.fcs	K14+K19+VIM-	3,000
Hett_9_9.fcs	K14-K19+VIM+	0
Hett_9_9.fcs	K14-K19+VIM-	130
Hett_9_9.fcs	K14-K19-VIM+	140
Hett_9_9.fcs	K14-K19-VIM-	2,713
Hett_9_9.fcs	K14-K19-VIM+	966
Hett_9_9.fcs	Ungated	4,438



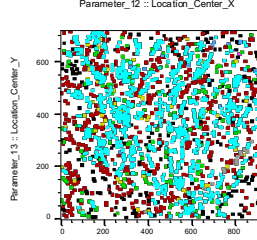
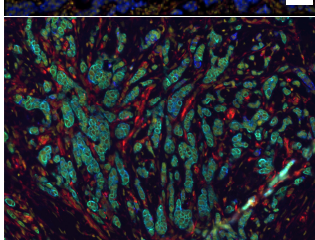
T1c



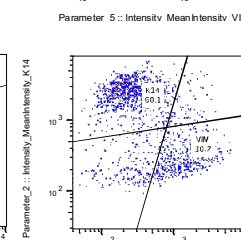
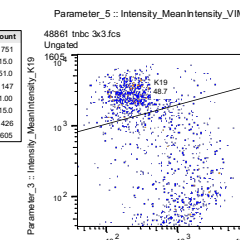
Sample Name	Subset Name	Count
Het5_119.fcs	K14+K19+VIM+	0
Het5_119.fcs	K14+K19+VIM-	5,000
Het5_119.fcs	K14-K19+VIM+	1,000
Het5_119.fcs	K14-K19+VIM-	170
Het5_119.fcs	K14-K19-VIM+	0
Het5_119.fcs	K14-K19-VIM-	2,471
Het5_119.fcs	K14-K19-VIM+	917
Het5_119.fcs	Ungated	8,077



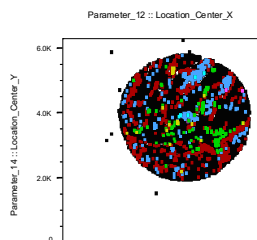
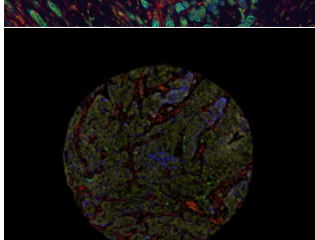
T4



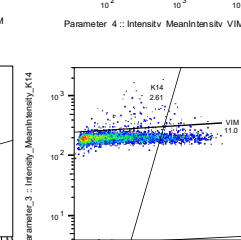
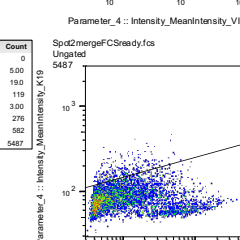
Sample Name	Subset Name	Count
48861 tnc3 3x3.fcs	K14+K19+VIM+	751
48861 tnc3 3x3.fcs	K14+K19+VIM-	150
48861 tnc3 3x3.fcs	K14-K19+VIM+	510
48861 tnc3 3x3.fcs	K14-K19+VIM-	147
48861 tnc3 3x3.fcs	K14-K19-VIM+	1,000
48861 tnc3 3x3.fcs	K14-K19-VIM-	150
48861 tnc3 3x3.fcs	K14-K19-VIM+	426
48861 tnc3 3x3.fcs	Ungated	1,660



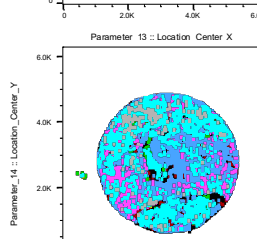
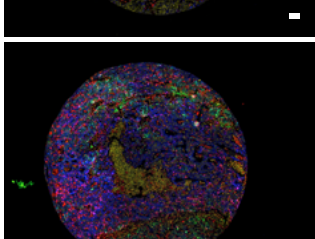
T5



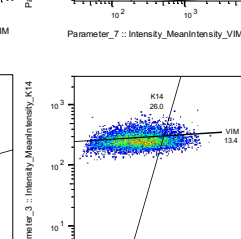
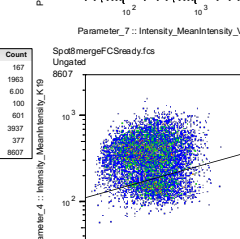
Sample Name	Subset Name	Count
SpotMergeFCsready.fcs	K14+K19+VIM+	0
SpotMergeFCsready.fcs	K14+K19+VIM-	5,000
SpotMergeFCsready.fcs	K14-K19+VIM+	190
SpotMergeFCsready.fcs	K14-K19+VIM-	119
SpotMergeFCsready.fcs	K14-K19-VIM+	3,000
SpotMergeFCsready.fcs	K14-K19-VIM-	276
SpotMergeFCsready.fcs	K14-K19-VIM+	582
SpotMergeFCsready.fcs	Ungated	5,487



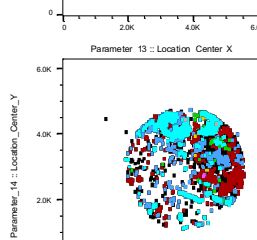
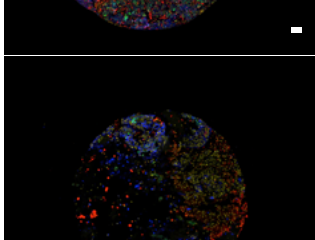
T6



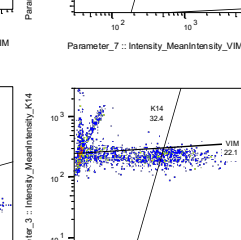
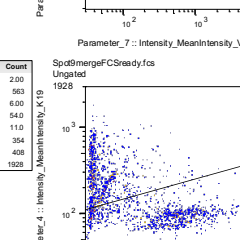
Sample Name	Subset Name	Count
SpotMergeFCsready.fcs	K14+K19+VIM+	167
SpotMergeFCsready.fcs	K14+K19+VIM-	1,993
SpotMergeFCsready.fcs	K14-K19+VIM+	800
SpotMergeFCsready.fcs	K14-K19+VIM-	100
SpotMergeFCsready.fcs	K14-K19-VIM+	601
SpotMergeFCsready.fcs	K14-K19-VIM-	3,937
SpotMergeFCsready.fcs	K14-K19-VIM+	377
SpotMergeFCsready.fcs	Ungated	8,607



T7



Sample Name	Subset Name	Count
SpotMergeFCsready.fcs	K14+K19+VIM+	200
SpotMergeFCsready.fcs	K14+K19+VIM-	563
SpotMergeFCsready.fcs	K14-K19+VIM+	6,000
SpotMergeFCsready.fcs	K14-K19+VIM-	540
SpotMergeFCsready.fcs	K14-K19-VIM+	1,110
SpotMergeFCsready.fcs	K14-K19-VIM-	356
SpotMergeFCsready.fcs	K14-K19-VIM+	408
SpotMergeFCsready.fcs	Ungated	10,280

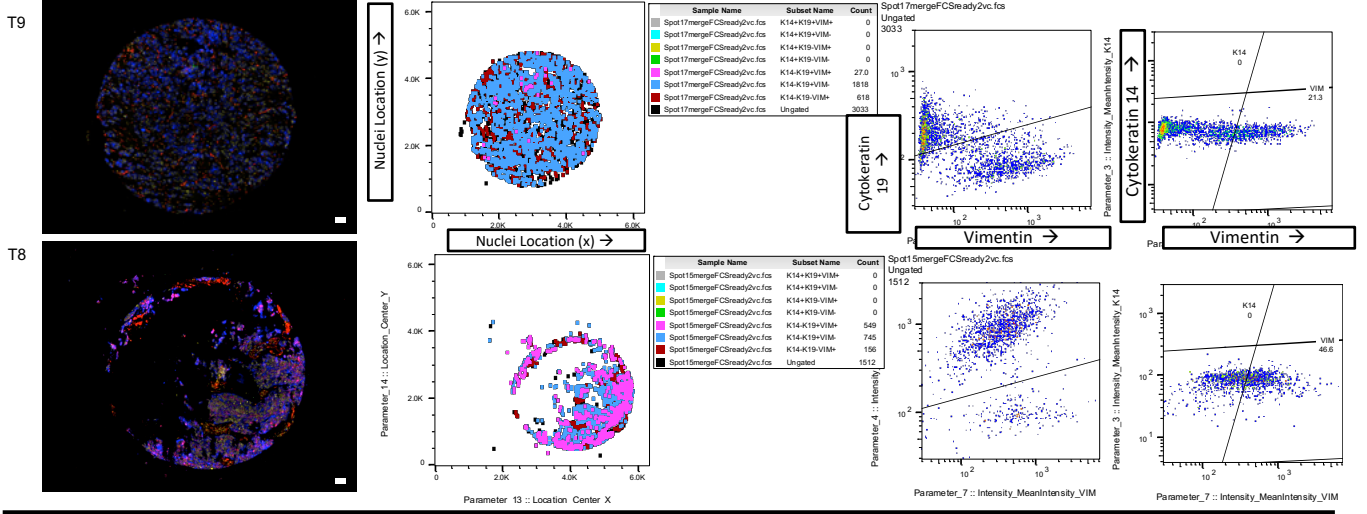


### Triple Negative Tumors

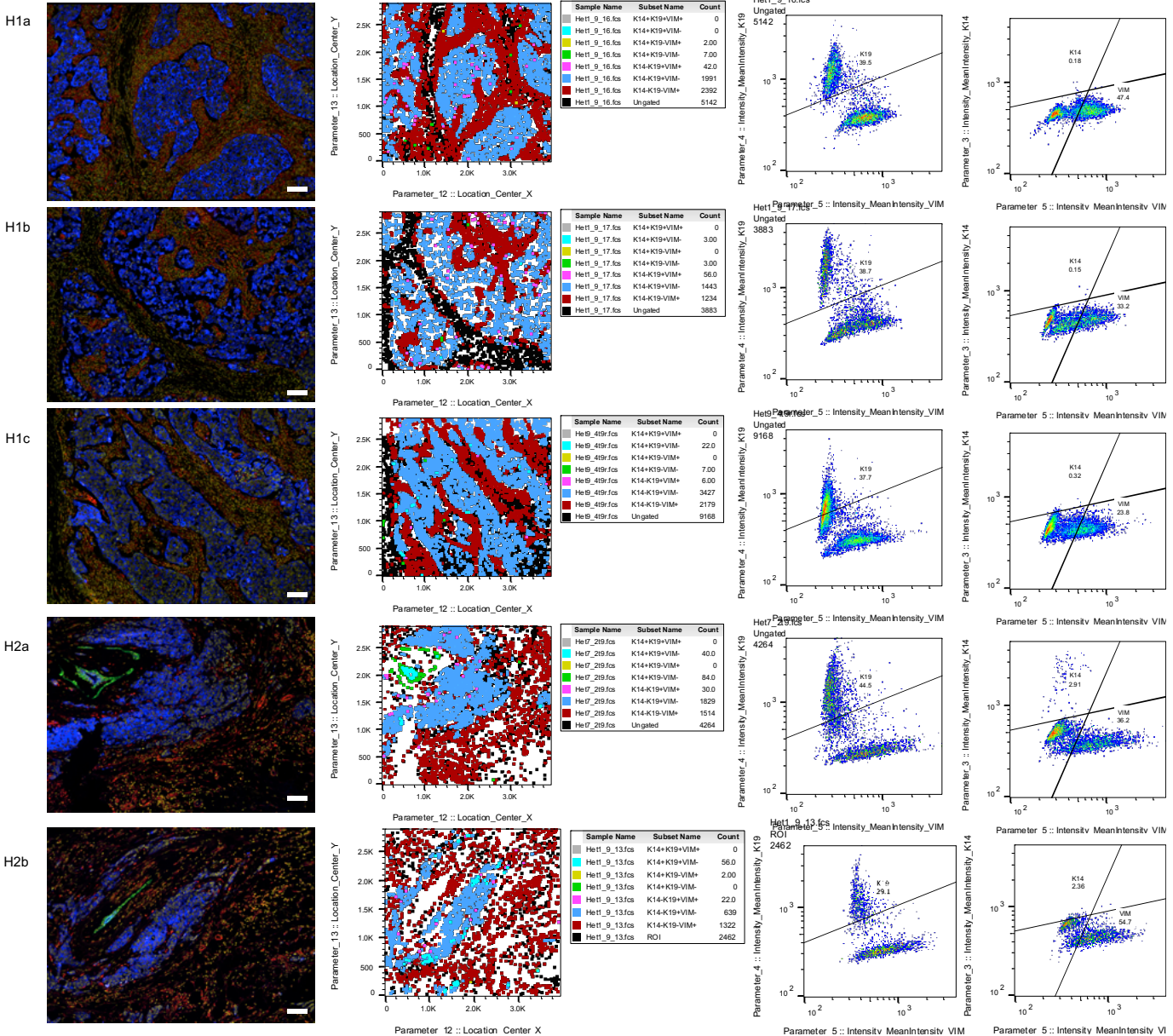
IF: DAPI K19 K14 VIM

Cell Phenotype

(K19 / K14 / VIM): + / - / - - / + / - - / - / + + / - / + - / + / - - / + / + - / - / + / + + / + / + + - / - / -



### HER2+ Tumors

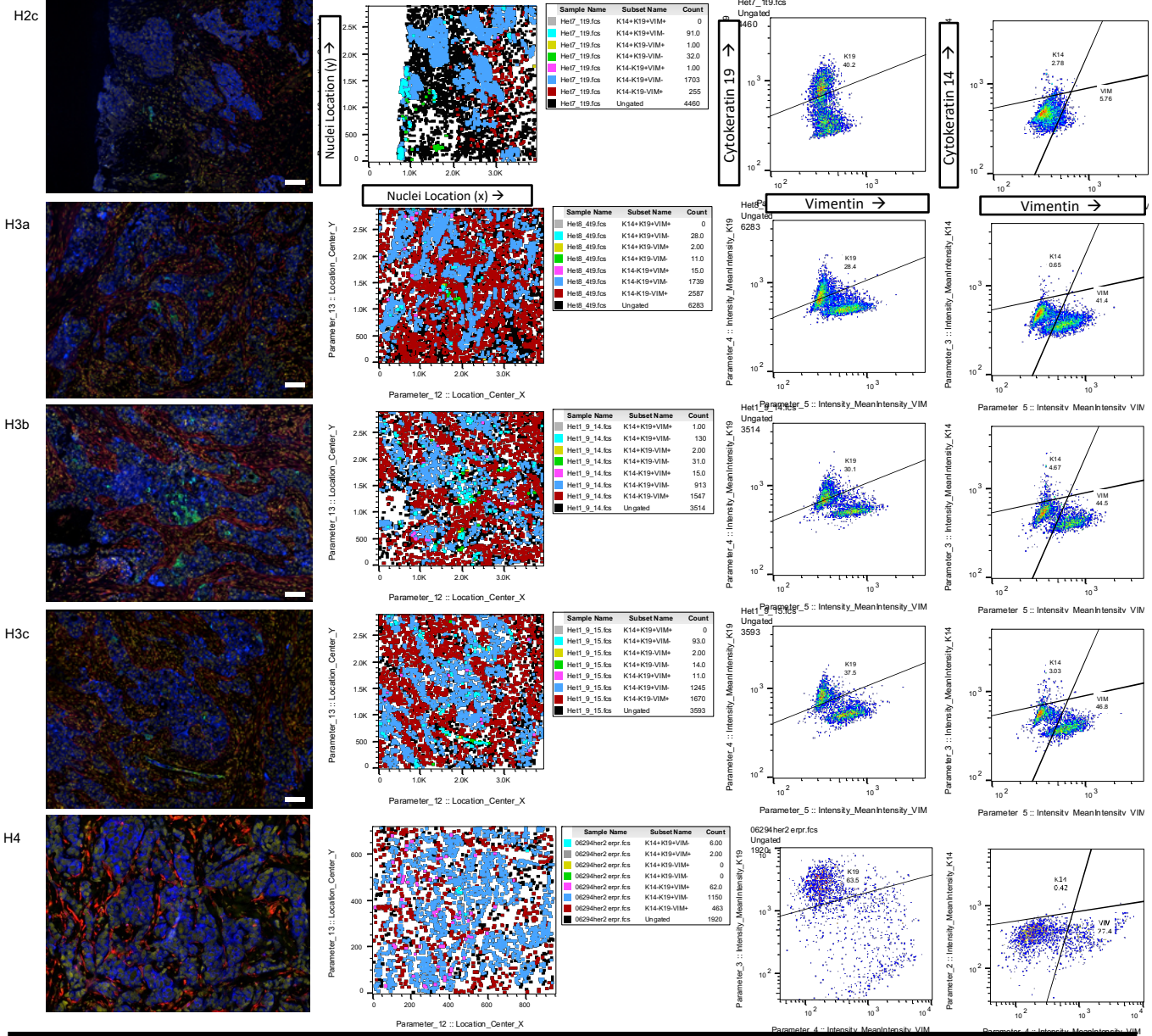


## HER2+ Tumors

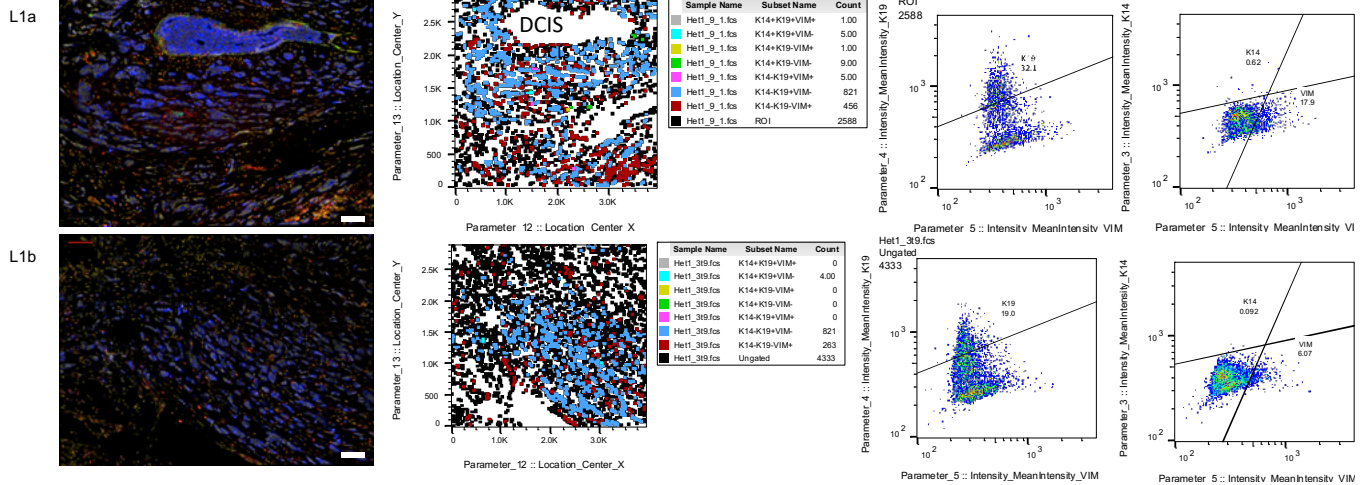
IF: DAPI K19 K14 VIM

Cell Phenotype

(K19/K14/VIM): +/+/+ -/+/- -/+/- -/+/- +/+/+ +/+/+ -/+/-



## ER+ Tumors

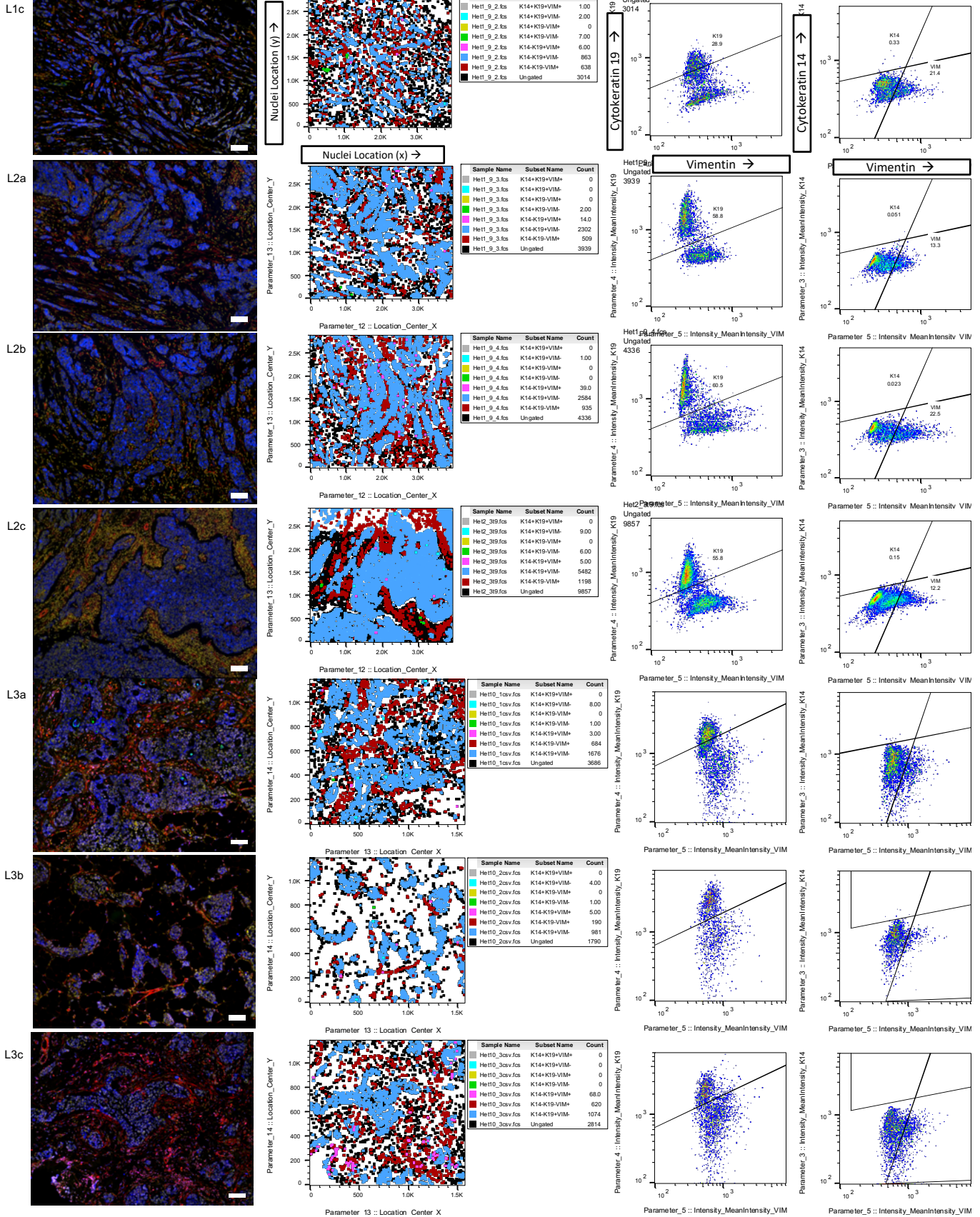


# Luminal Tumors

IF: DAPI K19 K14 VIM

Cell Phenotype (K19 / K14 / VIM):

■ +/+/+ 
 ■ +/+/- 
 ■ -/+/- 
 ■ -/+/+ 
 ■ +/+/- 
 ■ -/+/+ 
 ■ +/+/+ 
 ■ -/-/-

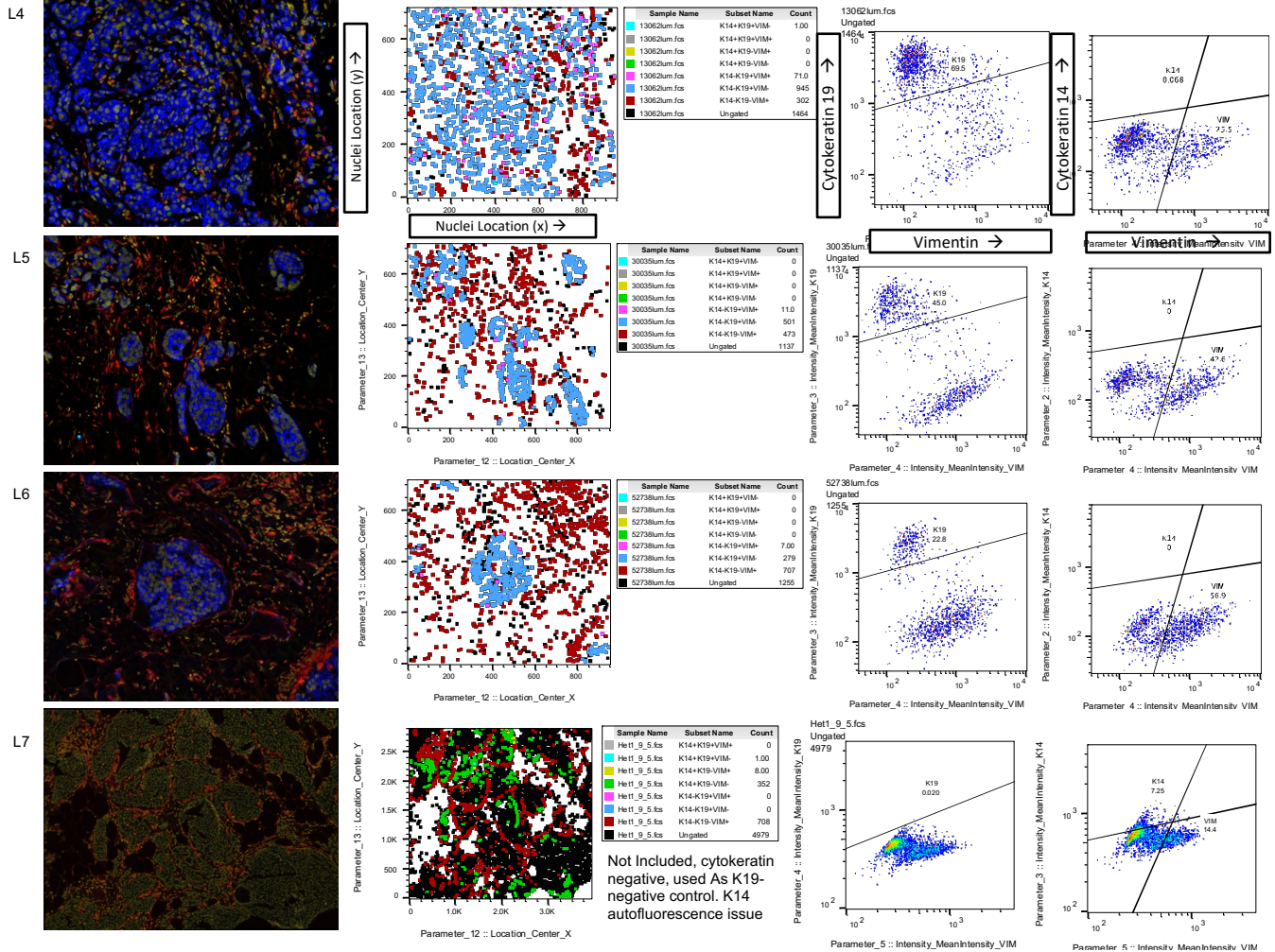


# Luminal Tumors

IF: DAPI K19 K14 VIM

Cell Phenotype

(K19 / K14 / VIM): + / - / - - / + / - - / - / + - / - / + + / + / + - / - / -

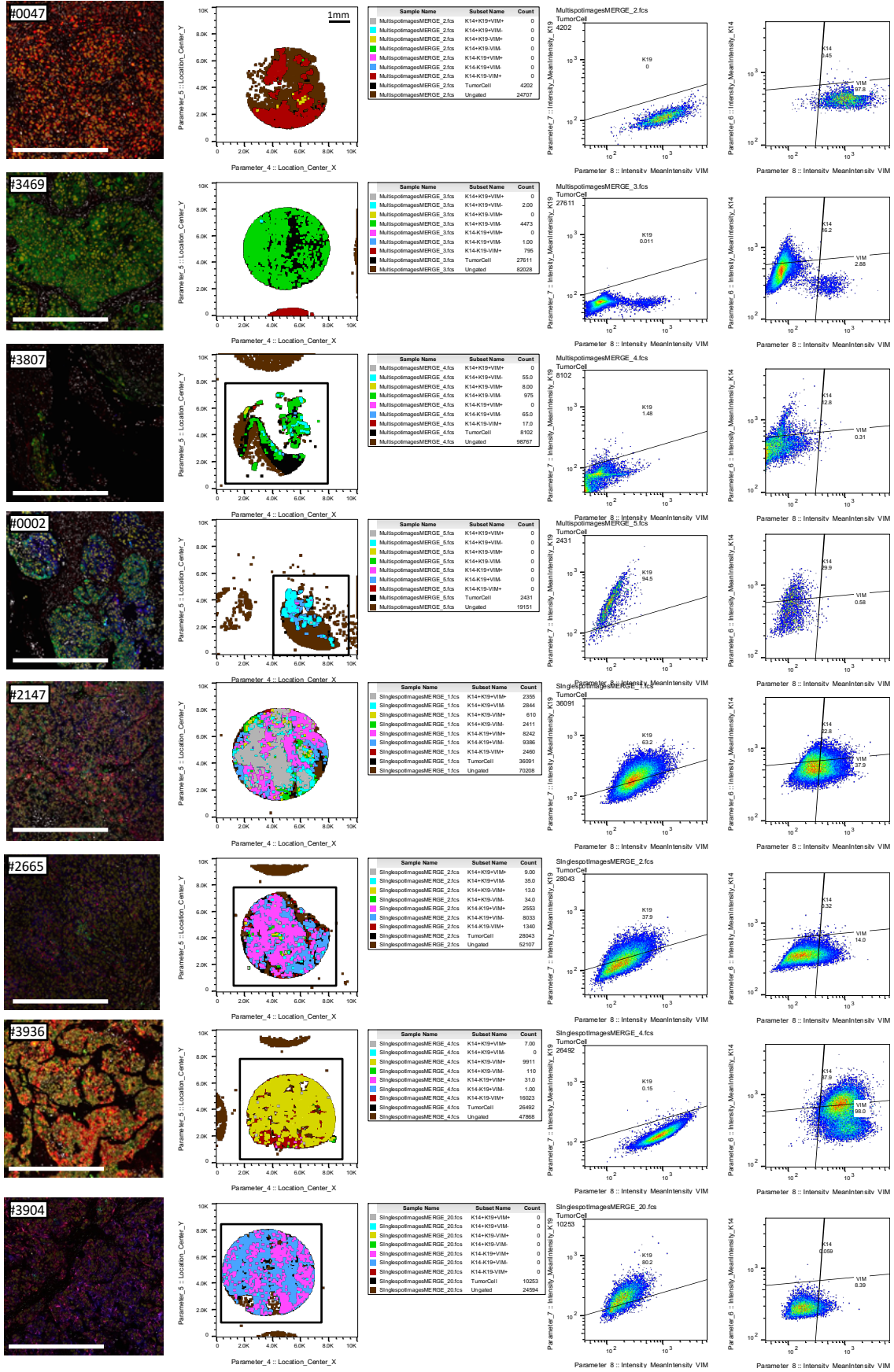


# Patient-Derived Xenograft Tumors

Ku80+ Tumor Cell Phenotype (K19 / K14 / VIM):

IF:  DAPI  K19  K14  VIM  Ku80

■ + / - / -   
 ■ - / + / -   
 ■ - / - / +   
 ■ + / - / +   
 ■ + / + / -   
 ■ - / + / +   
 ■ + / + / +   
 ■ Ku80-negative cell





# Patient-Derived Xenograft Tumors

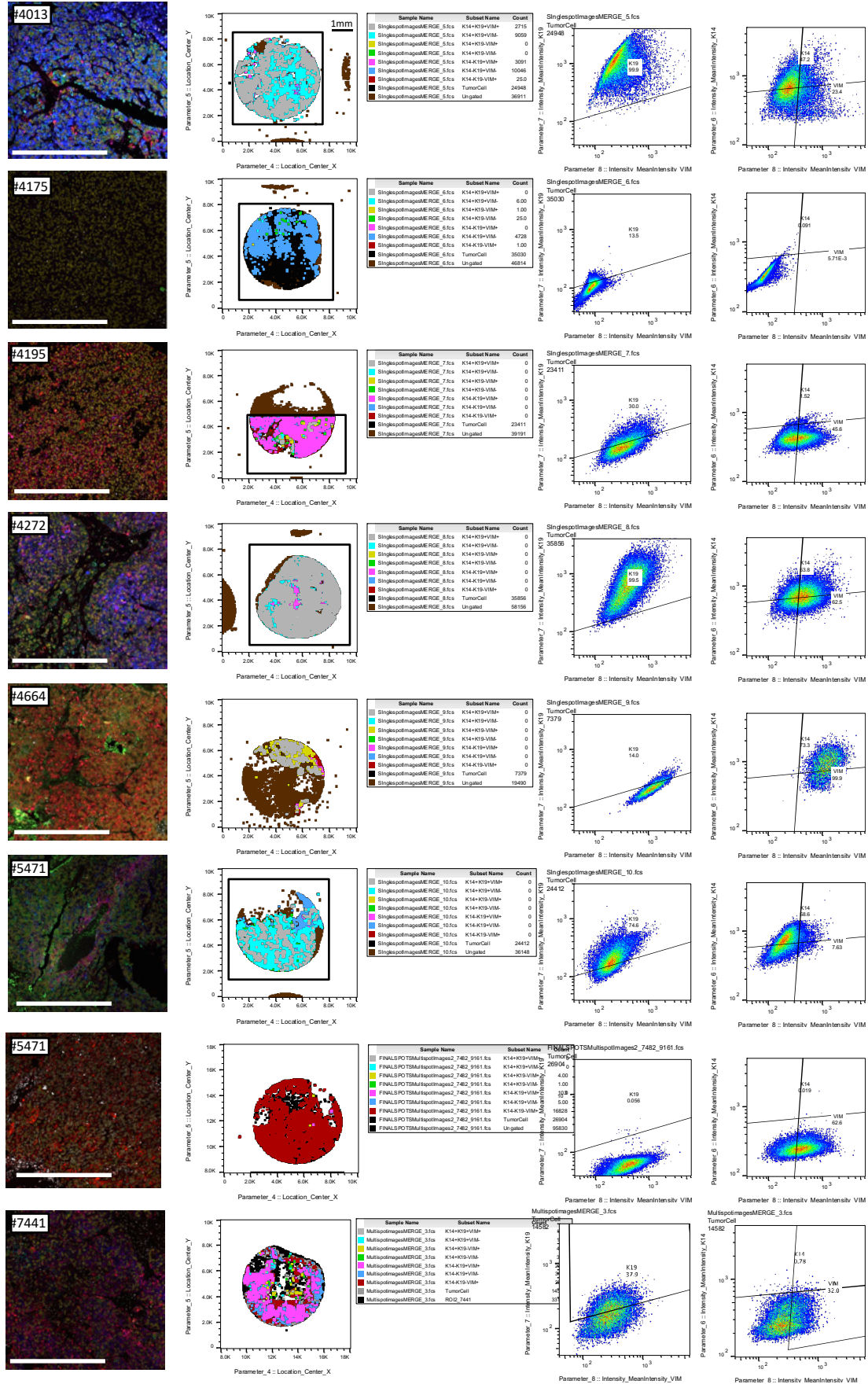
Ku80+ Tumor Cell Phenotype

(K19 / K14 / VIM):

■ + / - / -   
 ■ - / + / -   
 ■ - / - / +   
 ■ + / - / +   
 ■ + / + / -   
 ■ - / + / +   
 ■ + / + / +

IF: ■ DAPI ■ K19 ■ K14 ■ VIM ■ Ku80

■ Ku80-negative cell

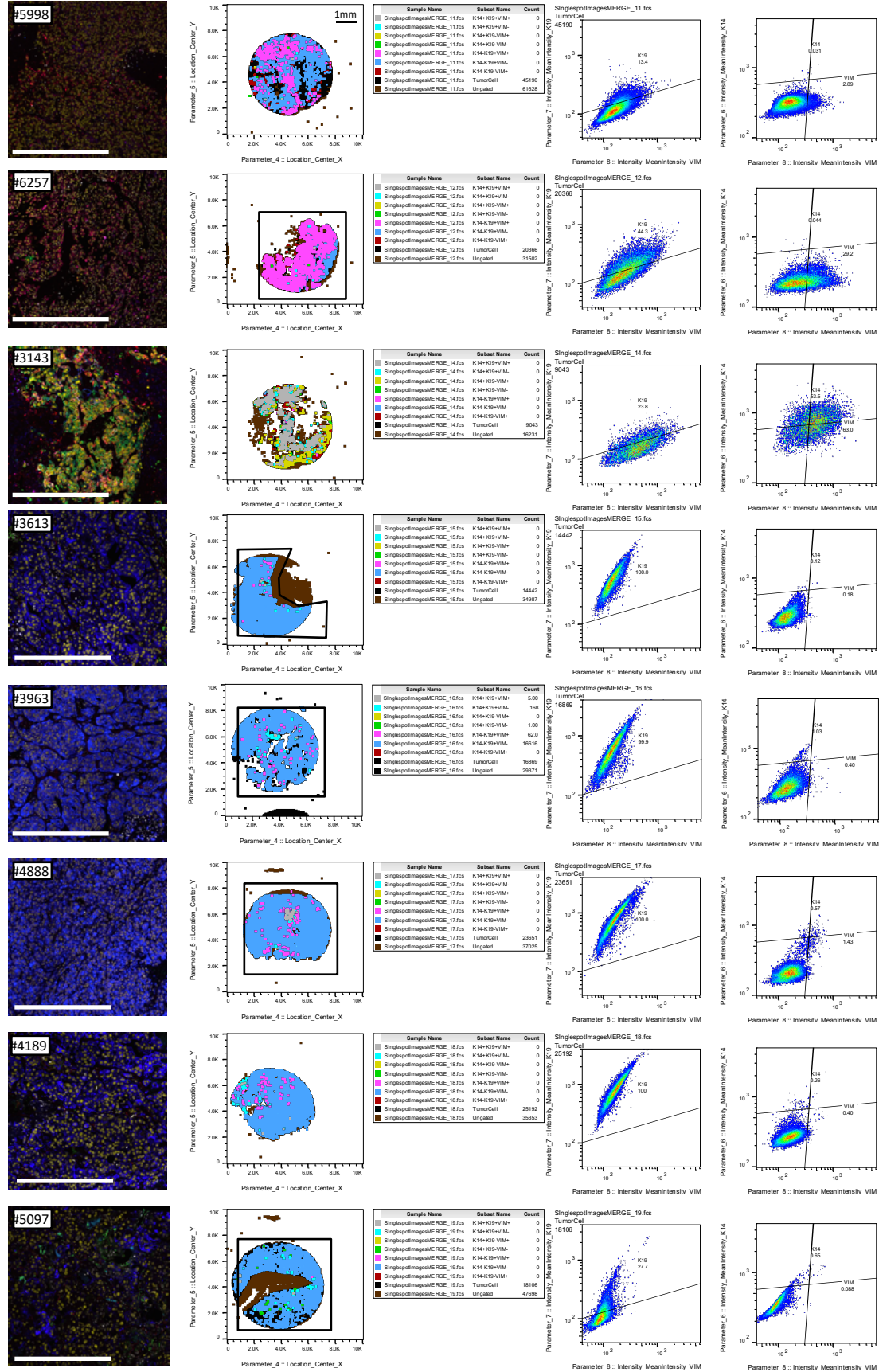


# Patient-Derived Xenograft Tumors

## Ku80+ Tumor Cell Phenotype (K19 / K14 / VIM):

IF:  DAPI  K19  K14  VIM  Ku80

■ + / - / - ■ - / + / - ■ - / - / + ■ + / - / + ■ + / + / - ■ - / + / + ■ + / + / +  
■ Ku80-negative cell



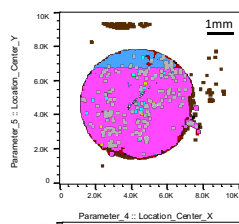
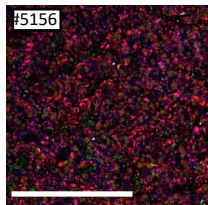
# Patient-Derived Xenograft Tumors

Ku80+ Tumor Cell Phenotype

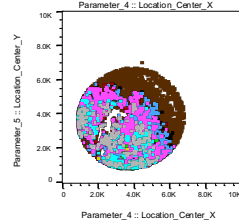
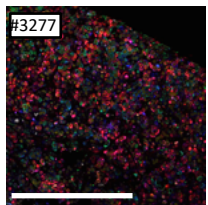
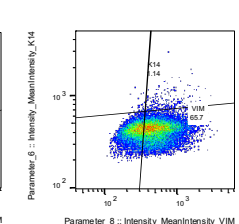
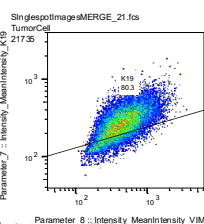
(K19 / K14 / VIM): ■ + / - / - ■ - / + / - ■ - / - / + ■ + / - / + ■ + / + / - ■ - / + / + ■ + / + / +

IF: ■ DAPI ■ K19 ■ K14 ■ VIM ■ Ku80

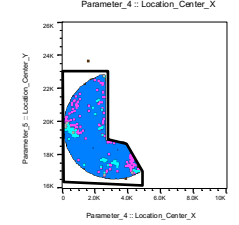
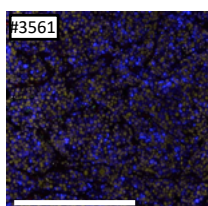
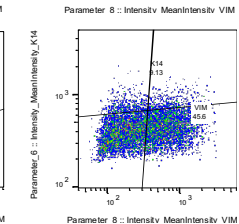
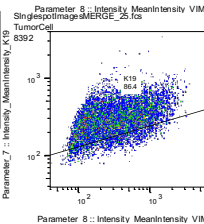
■ Ku80-negative cell



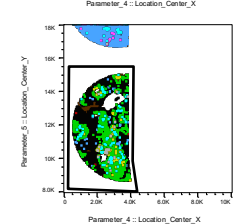
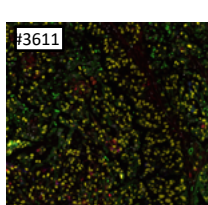
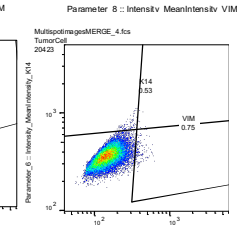
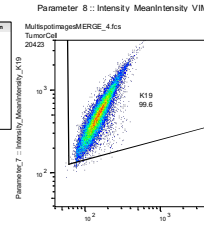
Sample Name	Subst Name	Count
SinglespotImageMERGE_21.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_21.fcs	K14+K19-VIM-	0
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SinglespotImageMERGE_21.fcs	Un gated	39487



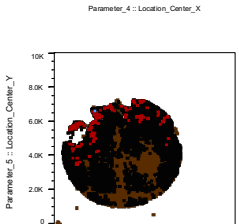
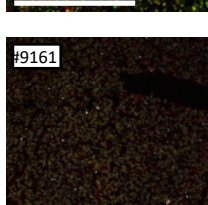
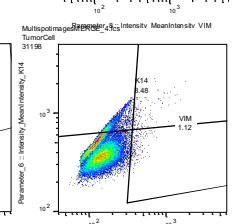
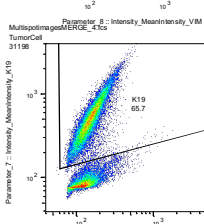
Sample Name	Subst Name	Count
SinglespotImageMERGE_25.fcs	K14+K19-VIM+	0
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SinglespotImageMERGE_25.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_25.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_25.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_25.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_25.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_25.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_25.fcs	TumorCell	6392
SinglespotImageMERGE_25.fcs	Un gated	34244



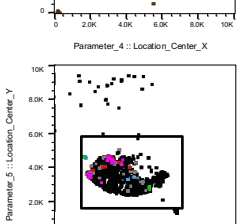
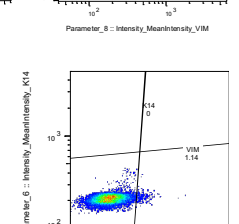
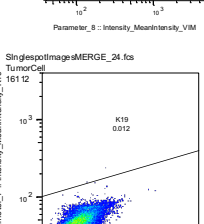
Sample Name	Subst Name	Count	Workspace Annotation
MultipotImageMERGE_4.fcs	K14+K19-VIM+	720	
MultipotImageMERGE_4.fcs	K14+K19-VIM-	101	
MultipotImageMERGE_4.fcs	K14+K19-VIM+	0	
MultipotImageMERGE_4.fcs	K14+K19-VIM-	147	
MultipotImageMERGE_4.fcs	K14+K19-VIM+	2006	
MultipotImageMERGE_4.fcs	K14+K19-VIM-	733	
MultipotImageMERGE_4.fcs	TumorCell	20423	
MultipotImageMERGE_4.fcs	Un gated	27664	



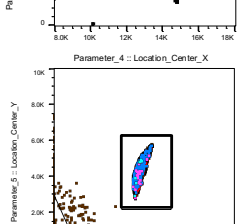
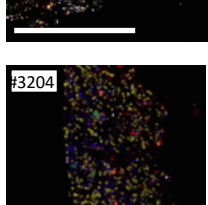
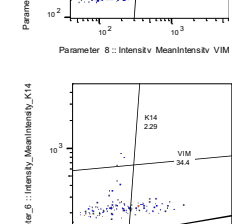
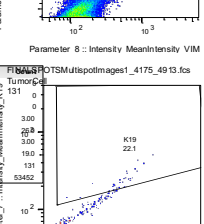
Sample Name	Subst Name	Count
MultipotImageMERGE_4.fcs	K14+K19-VIM+	663
MultipotImageMERGE_4.fcs	K14+K19-VIM-	346
MultipotImageMERGE_4.fcs	K14+K19-VIM+	103
MultipotImageMERGE_4.fcs	K14+K19-VIM-	2132
MultipotImageMERGE_4.fcs	K14+K19-VIM+	152
MultipotImageMERGE_4.fcs	K14+K19-VIM-	1922
MultipotImageMERGE_4.fcs	K14+K19-VIM+	232
MultipotImageMERGE_4.fcs	K14+K19-VIM-	848
MultipotImageMERGE_4.fcs	Un gated	48643



Sample Name	Subst Name	Count
SinglespotImageMERGE_24.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM+	0
SinglespotImageMERGE_24.fcs	K14+K19-VIM-	0
SinglespotImageMERGE_24.fcs	TumorCell	16112
SinglespotImageMERGE_24.fcs	Un gated	54520



Sample Name	Subst Name	Count
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM+	131
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM-	0
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM+	300
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM-	221
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM+	190
FNALS POTSMultipotImages1_4175_4913.fcs	K14+K19-VIM-	131
FNALS POTSMultipotImages1_4175_4913.fcs	TumorCell	5342
FNALS POTSMultipotImages1_4175_4913.fcs	Un gated	5342



Sample Name	Subst Name	Count
MultipotImageMERGE_5.fcs	K14+K19-VIM+	0
MultipotImageMERGE_5.fcs	K14+K19-VIM-	800
MultipotImageMERGE_5.fcs	K14+K19-VIM+	100
MultipotImageMERGE_5.fcs	K14+K19-VIM-	34
MultipotImageMERGE_5.fcs	K14+K19-VIM+	253
MultipotImageMERGE_5.fcs	K14+K19-VIM-	34
MultipotImageMERGE_5.fcs	K14+K19-VIM+	73
MultipotImageMERGE_5.fcs	K14+K19-VIM-	94
MultipotImageMERGE_5.fcs	TumorCell	94
MultipotImageMERGE_5.fcs	Un gated	1919

