

Monitoring of cytokeratin-positive cells in patients with prostate cancer

Longitudinal monitoring of cytokeratin (CK)-positive cells in patients with prostate cancer after radiation therapy

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BACKGROUND

The **nPAC™ CTC IF Kit** includes antibodies directed against various cytokeratins, CD45, and other markers that are specific to white blood cells (WBC). DAPI is used to stain cell nuclei. The **nCyte Dx® platform** automatically scans the entire sample, acquires images, and displays any event where CK+ and DAPI+ are co-located. Images are presented to the user in a gallery format for final classification of the captured cell. An event is classified as a CTC when its morphological features are consistent with that of a cell and it exhibits the correct phenotypes, i.e., CK+, DAPI+, and CD45-.

In this ongoing study, we are monitoring CK+ cell counts in primary prostate cancer patients before and after radiation therapy with the **nCyte Dx® system**. Nine out of 14 patients received hormone therapy that started before radiation (highlighted in green). Blood was collected at various time points and was analyzed by the **nCyte Dx® system**.

RESULTS

MICROSCOPIC ENUMERATION OF CK+ CELLS

	Number	iPSA	PSA (A)	TIME POINT A				TIME POINT B				TIME POINT C				TIME POINT D					
				CK+ CD45-	CK+ CD45+	CK+ CD45-, apoptotic	CK+ CD45+, apoptotic	CK+ CD45-	CK+ CD45+	CK+ CD45-, apoptotic	CK+ CD45+, apoptotic	CK+ CD45-	CK+ CD45+	CK+ CD45-, apoptotic	CK+ CD45+, apoptotic	CK+ CD45-	CK+ CD45+	CK+ CD45-, apoptotic	CK+ CD45+, apoptotic		
PROSTATE CANCER PATIENTS	#1	9.08	7.62	0	1	0	0							1	2	0	1	0	20	0	1
	#2	18.3	9.83	2	8	0	0	58	160	4	7	0	2	0	0	0	0				
	#3	7.84	8.73	0	3	0	0	0	3	0	1	0	1	0	0	0	0	0	3	0	2
	#4	17	17.6	33	68	0	1	0	3	0	0	2	3	0	0						
	#5	13	0.71	0	0	0	0	0	2	0	0	1	3	0	0						
	#6	25	9.49	1	4	0	0	0	0	0	0	0	0	0	0						
	#7	6.6	6.6	0	1	0	2	0	5	0	0	0	0	0	0						
	#8	4.31	4.61	0	1	0	1	0	0	0	0										
	#9	10.9	4.01	0	0	0	0	0	0	0	2										
	#10	21	3.05	0	2	0	1	0	3	0	0										
	#11	12.2	0.37	0	3	0	0														
	#12	6.85	6.85	0	2	0	0														
	#13	15.4	1.9	0	0	0	0														
	#14	23.5	1.09	0	0	0	1														
CONTROLS	#1			0	2	0	0														
	#2			0	0	0	0														
	#3			0	1	0	1														
	#4			0	5	0	0														
	#5			0	2	0	1														
	#6			0	3	0	0														

iPSA = initial PSA (ng/ml) before radiation and hormone therapy;
PSA(A) = PSA before radiation therapy and after starting of hormone therapy (green highlighted); green highlighted samples are patients that received hormone therapy.

LONGITUDINAL MONITORING OF CK+ CELLS IN TWO PATIENTS

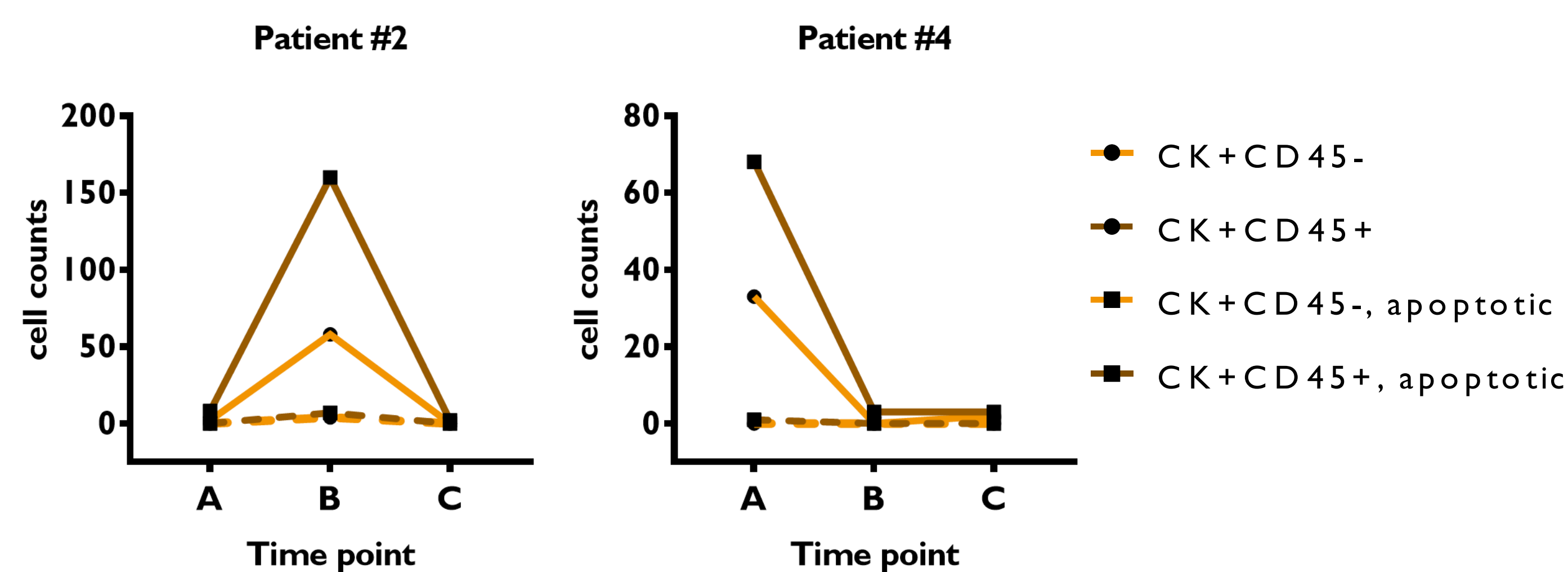


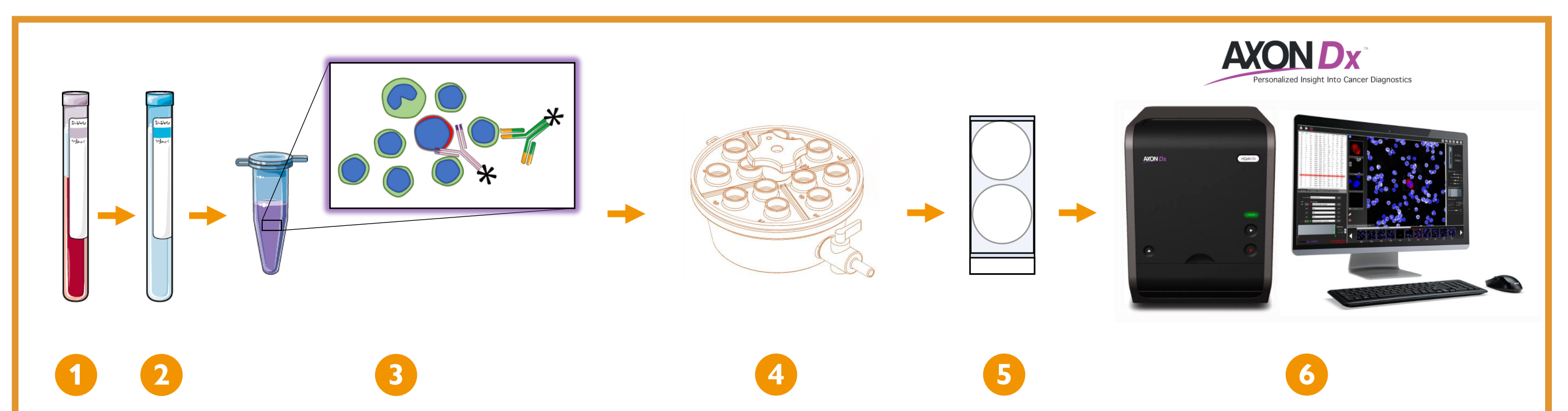
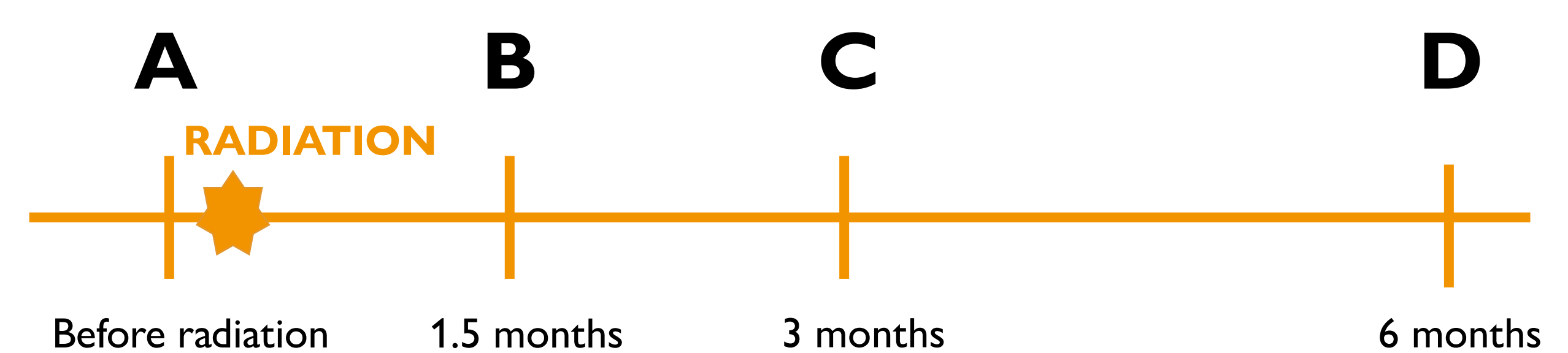
Figure 1: Detection of CK+ cells at three different time points (A=before radiation therapy, B=1.5 months after radiation therapy, C=3 months after radiation therapy).

CONCLUSION

The preliminary results of this study show, that the **nPAC™ CTC IF Kit**, combined with the **nCyte Dx® system**, is a promising superior tool to quantify CK+ cells and monitor their presence in peripheral blood samples from primary prostate cancer patients receiving hormone and/or radiation therapy.

METHODS

TIME POINTS OF BLOOD COLLECTION



ONCOLAB BLOOD COLLECTION KIT (OL60100):

- 1 Collect blood in preservation tube
- 2 Transfer 6 ml of blood into BD Vacutainer® CPT™ tube and isolate PBMCs

ONCOLAB CTC IF STAINING KIT (OL50100):

- 3 Perform sample fixation, permeabilization, and staining

ONCOLAB VACUUM SAMPLING MANIFOLD (OL90300)

- 4 Collect cells by filtration

ONCOLAB CRC IF MATERIALS KIT (OL50900):

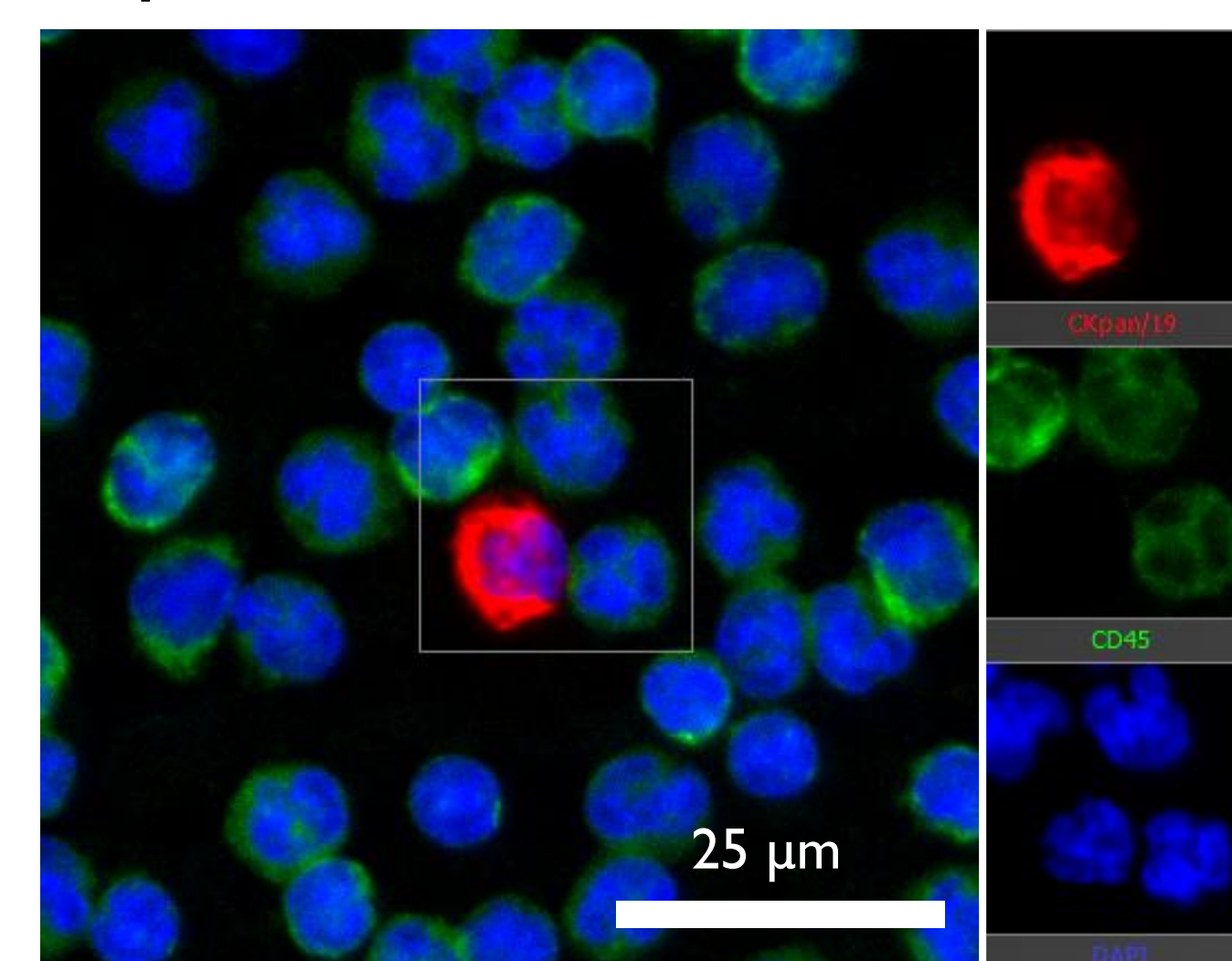
- 5 Mount filters on slides

nCyte nAble® SOFTWARE & nCyte® PLATFORM (OL90100)

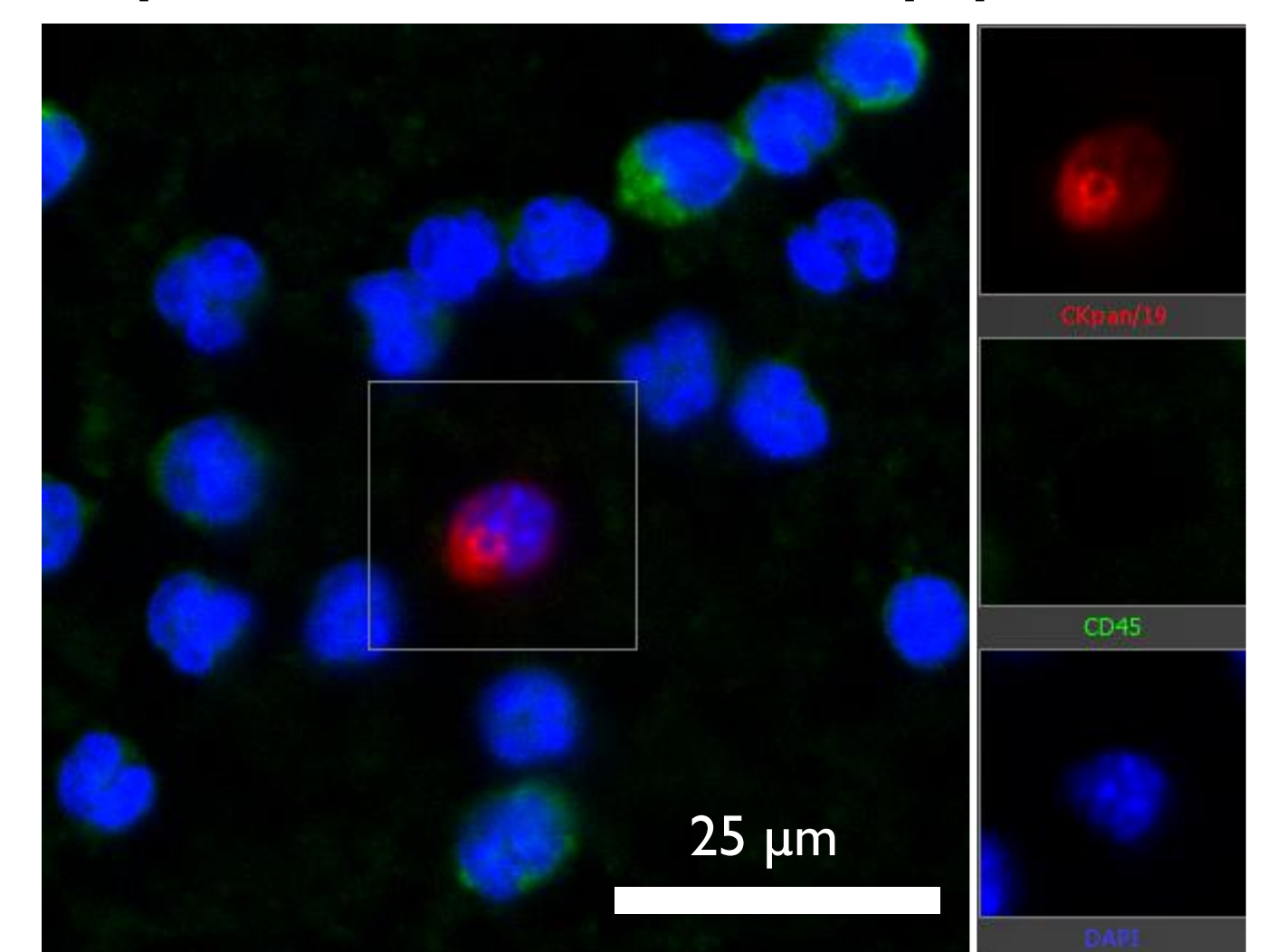
- 6 Analyze sample

CHARACTERIZATION OF EVENTS // EXAMPLES

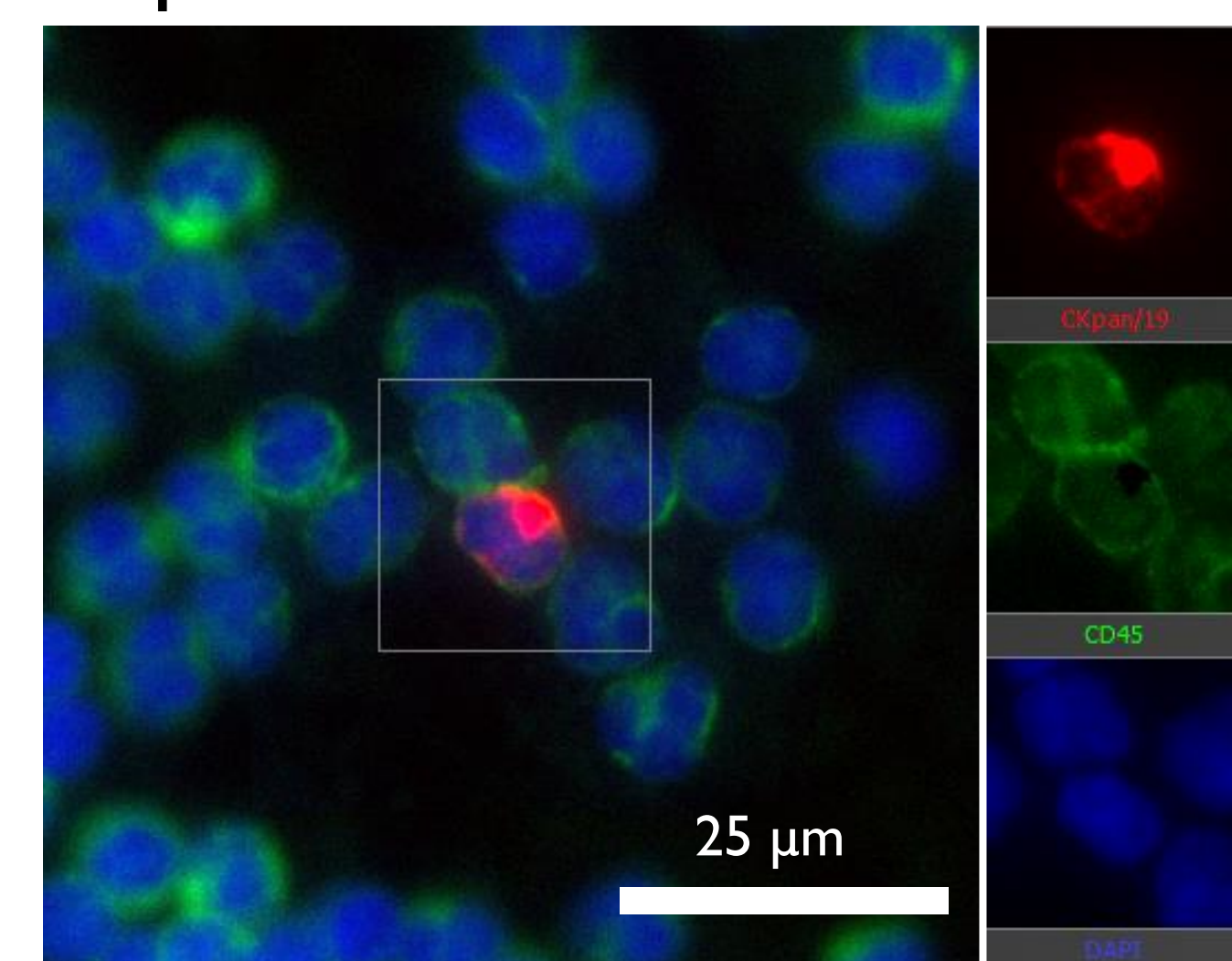
CKpan+ CD45- DAPI+ event



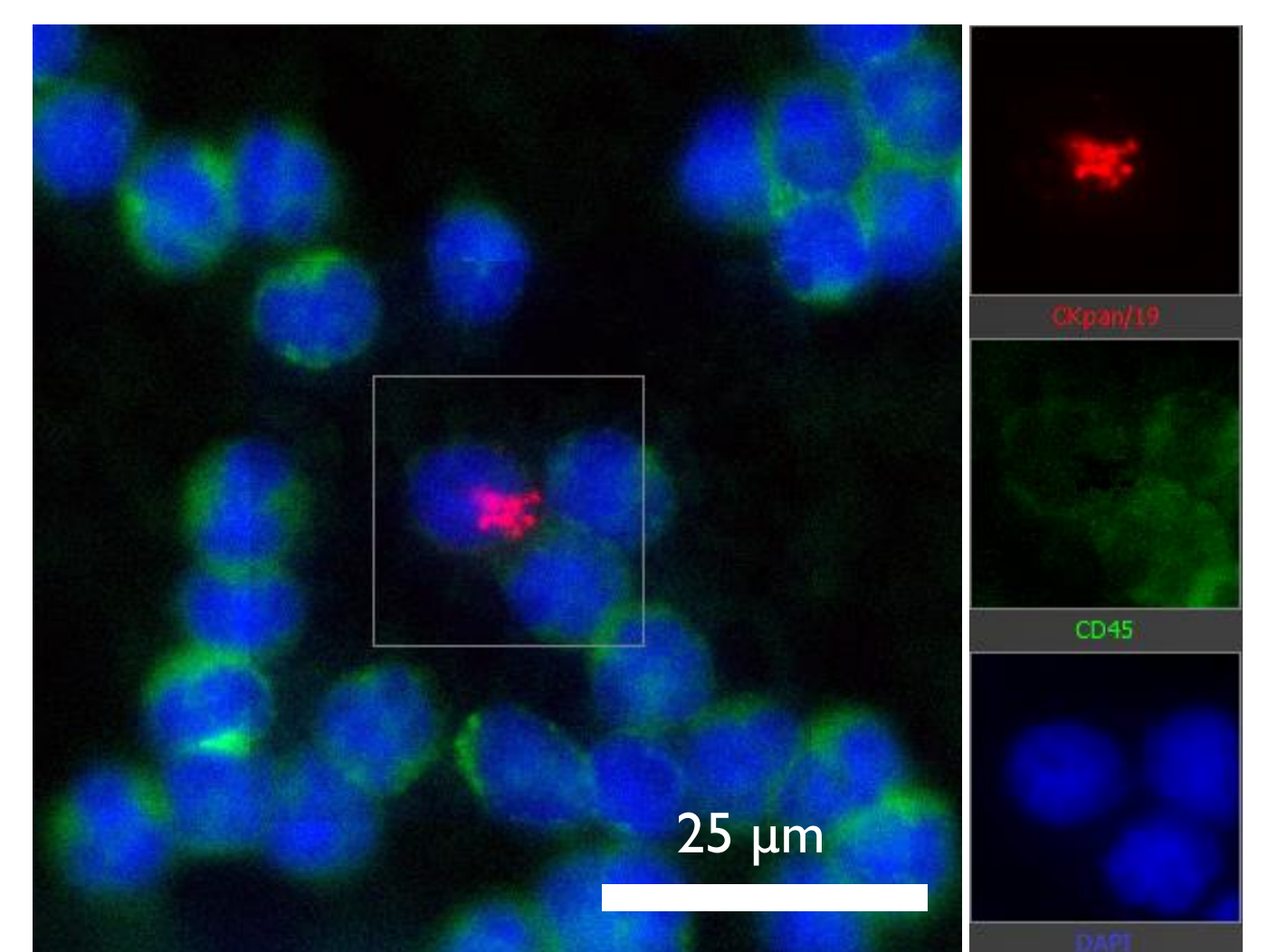
CKpan+ CD45+ DAPI+ event, apoptotic



CKpan+ CD45+ DAPI+ event



CKpan+ CD45- DAPI+ event, apoptotic



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