

Materials List for:

Multiplexed Immunofluorescence Analysis and Quantification of Intratumoral PD-1⁺ Tim-3⁺ CD8⁺ T Cells

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URL: <https://www.jove.com/video/56606>

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Materials

Name	Company	Catalog Number	Comments
Vectra 3 Automated Quantitative Pathology Imaging	Perkin Elmer	CLS142338	
inForm cell analysis 2.1.	Perkin Elmer	CLS135781	
R software	https://www.r-project.org		
Dakopen delimiting pen	Dako	S2002	
Tris Buffer Salin TBS Tablets	Takara, Bio Inc.	TAKT9141Z	pH7.6 100 tablet
Tris Buffer Salin Tween 20 TBS(+Tween20)	Takara, Bio Inc.	TAKT9142Z	pH 7.6 100 tablets
Biotin blocking system	Dako	X0590	Avidin 0.1% and Biotin 0.01%
normal donkey serum	Jackson ImmunoResearch	017-000-001	5% vol./vol. concentration
Fluoroshield with DAPI	Sigma-aldrich	F6057	1.5 µg/mL concentration
Knittel glass coverslip	Knittel Gläser,	100039	24x60 mm 100 cover slips
Rabbit anti-CD8 Clone P17-V	novus	NBP1-79055	use at 4µg/mL
Mouse anti-PD-1 Clone NAT	abcam	ab52587	use at 2 µg/mL
Goat anti-Tim-3	R&D	AF2365	use at 3 µg/mL
Rabbit anti-PD-L1 Clone SP142	Roche	7309457001	use at 1 µg/mL
Mouse AF647 labeled pan- Keratin Clone C11	Cell Signalling	4528	use at 0.5 µg/mL
Goat anti-human gal9	R&D	AF2045	use at 0.3 µg/mL
Cyan 5 conjugated donkey anti-rabbit	Jackson ImmunoResearch	711-175-152	use at 5 µg/mL
Biotinylated F(ab'2) donkey anti-mouse IgG	Jackson ImmunoResearch	715-066-150	use at 3 µg/mL
Alexa Fluor488 conjugated donkey anti-goat IgG	abcam	ab150133	use at 5 µg/mL
Cy3 labeled streptavidin	Amersham	PA43001	use at 3 µg/mL
negative control mouse IgG1	Dako	X0931	use at 2 µg/mL

IgG from goat serum	Sigma-aldrich	I5256	use at 3 µg/mL
IgG from rabbit serum	Sigma-aldrich	I5006	use at 4µg/mL