

A 3D molecular model of a protein complex, likely an antibody or a chimeric Fcγ receptor. The structure is composed of several subunits, with some colored in shades of blue and green, and others in brown and grey. The background is a light, hazy, and textured surface, possibly representing a cell membrane or a biological environment.

**CNR**

**Istituto di Farmacologia Traslazionale**

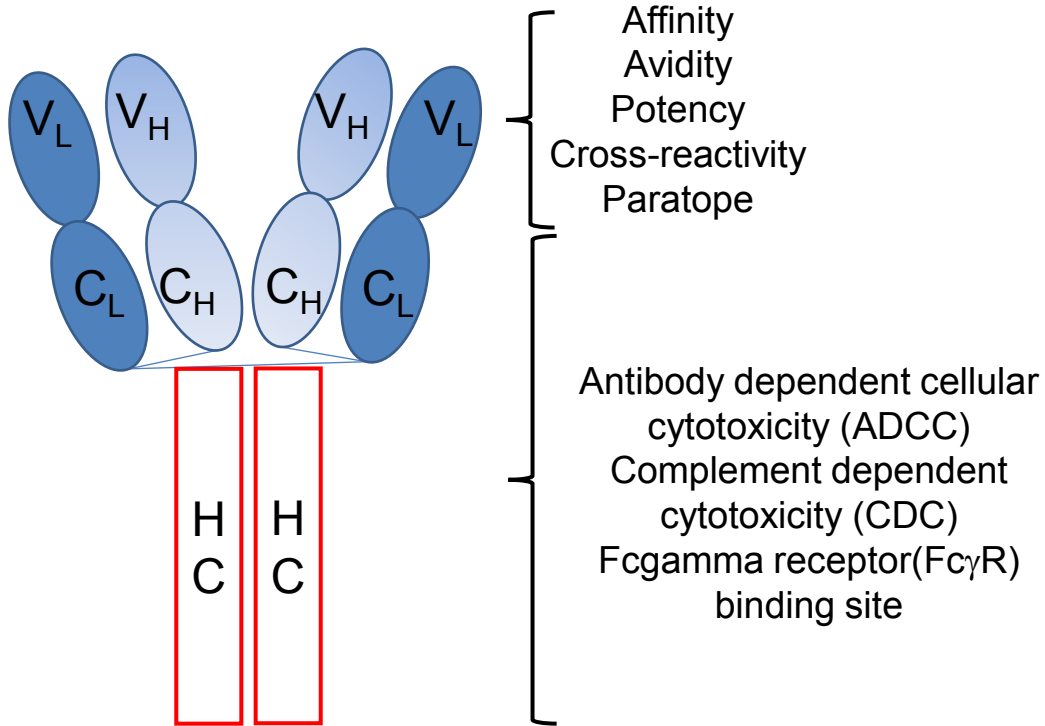
**Laboratorio di Immunologia dei Tumori ed Immunoterapia**

**Combinazioni tra mAbs e molecole  
Fcγ chimeriche: nuove strategie di targeting  
tumorale in oncoematologia**

**Giuseppe Sconocchia**

# Immunoglobulins (IgGs)

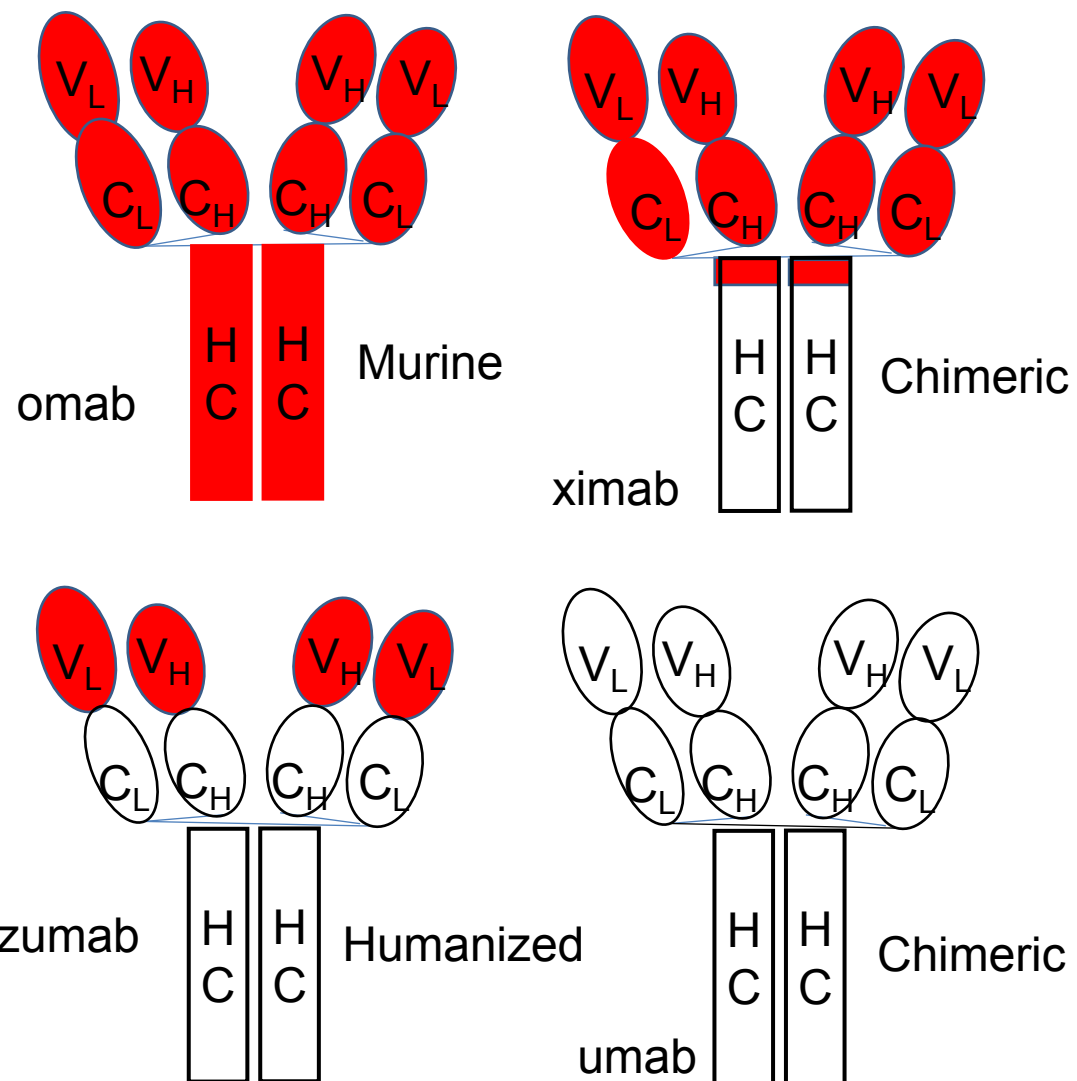
Fragment antigen binding Fab' or [F(ab)<sub>2</sub>]



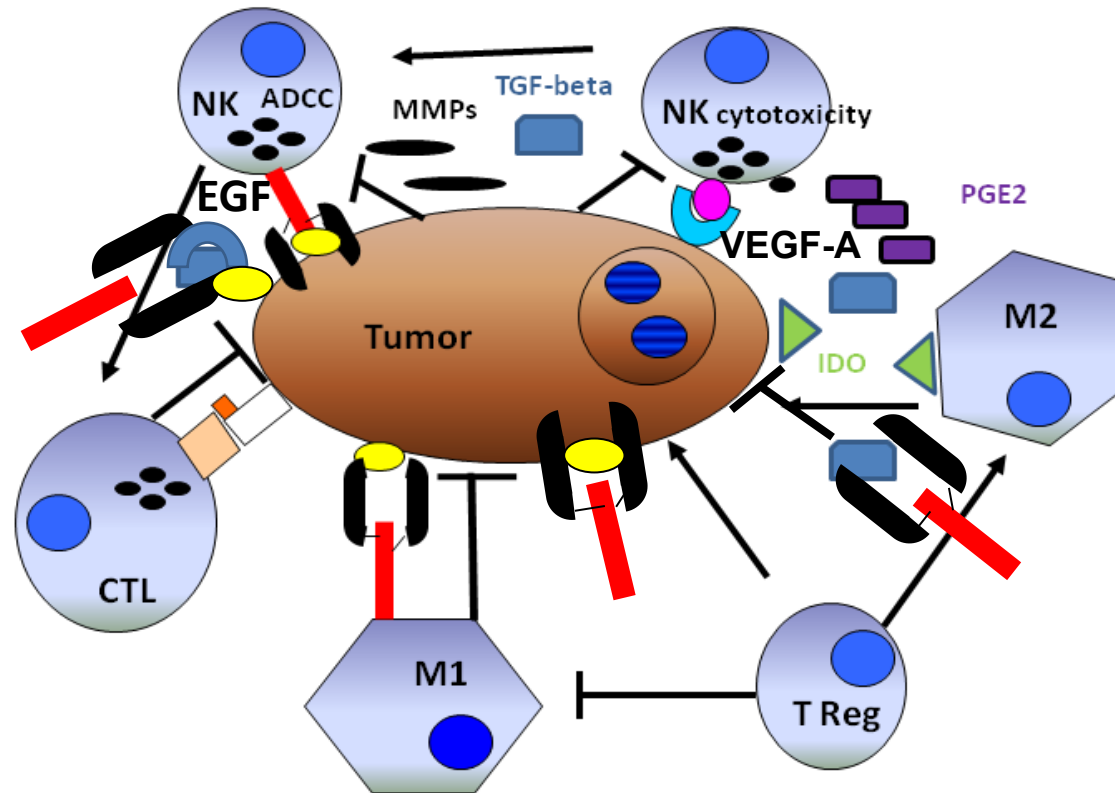
Features	IgG1	IgG2	IgG3	IgG4
Molecular Weight (kDa)	150	150	170	150
Amino acid hinge region	2	4	11	2
Half life (days)	14-21	14-21	7	14-21
Mean serum level (g/l)	6.98	3.8	0.51	0.56
Relative abundance (%)	60	32	4	4

# Evolution of Therapeutic monoclonal antibodies (mAbs)

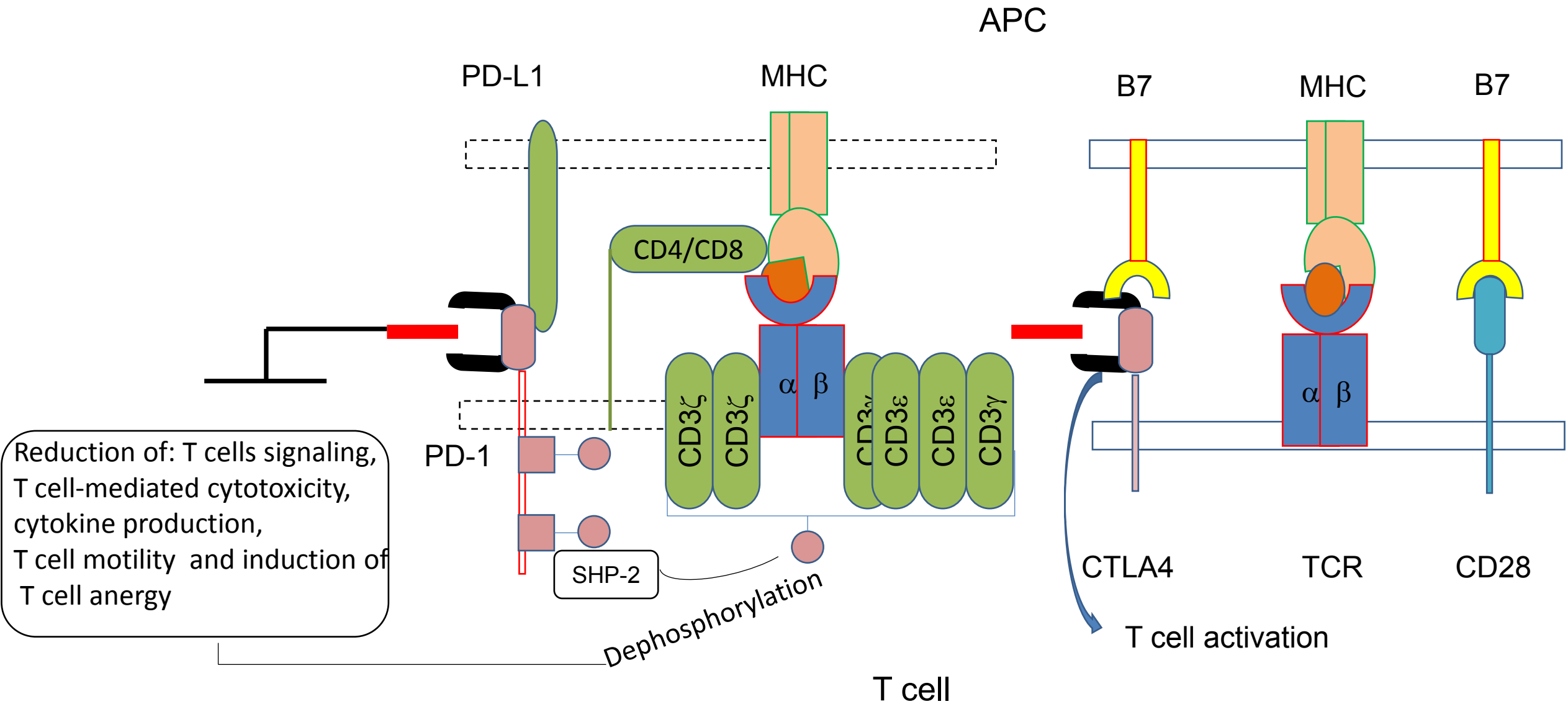
Anti-PD-L1 (Avelumab, Merck)	2017	Anti-PD-L1 (Atezolizumab, Roche)
Anti-PD-1 (Pembrolizumab, Keytruda, Merck)	2015	
	2014	Anti-PD-1 (Nivolumab, Opdivo, BMS)
Anti-CTLA4 (Ipilimumab, YERVOY, BMS)	2011	
	2007	Chimeric-anti-EGFR (Cetuximab, Erbitux, BMS)
Human-anti-EGFR (Panitumumab, Vectibix, Amgen)	2006	
	2004	Humanized-anti-VEGF-A (Bevacizumab, Avastin, Genetech)
<sup>131</sup> I Mouse anti-CD20 (Tositumomab, Bexxar, GSK)	2003	
	2001	Anti-CD52 (Alemtuzumab, Campath, Genzyme)
Humanized anti-CD33 (Gentuzumab-ozog, Mylotarg, Pfizer)	2000	
	1998	Humanized-Trastuzumab (Herceptin, Genetech)
Chimeric-Anti-CD20 (Rituximab, MabThera, Roche; Rituxan, Biogen Inc.)	1997	
	1990	mAb engineering methodology



# Types of anti-tumor activity of naked mAbs



# The immune check point of T cell activation



# Therapeutic mAbs improve the clinical course of solid and hematological malignancies

Therapeutic mAbs	Target	Therapeutic activity
Rituximab	CD20	Improves the clinical outcome of aggressive, indolent, low-grade B cell lymphomas and CLL
Cetuximab , panitumumab	EGFR	Benefits mCRC patients without KRAS and NRAS mutations. Cetuximab is also active in Head & Neck cancers
Trastuzumab	EGFR2 (HER2)	Early stage, metastatic and refractory BC and stomach cancer
Bevacizumab	VEGF-A	Useful in mCRC and recurrent GBM
Ipilimumab, and pembrolizumab and nivolumab	CTLA, and PD-1 respectively	Doubled the OS of mMelanoma patients and induced remission in refractory HL patients
Atezolizumab	PDL-1	Improves the OS in NSCLC patients. New treatment for advanced bladder cancer with 27% of response in a recent study
Pembrolizumab	PD-1	Improves OS in NSCLC patients. Since october 2016, iia the 1st line treatment of PDL-1+ NSCLC
Nivolumab	CTL4	Doubled the OS of patients with CHT resistant head & neck cancer

# Limitations of therapeutic mAb-based immunotherapy

- Impairment of a direct anti-tumor activity of mAbs by KRAS and NRAS mutations involved in the signaling pathway of EGFRs.
- Lack of effective biomarkers indicating which subpopulation of cancer patients take advantage from check point inhibitor-based immunotherapy
- Inadequate success of check point inhibitor-based immunotherapy in patients without hypermutated cancers.

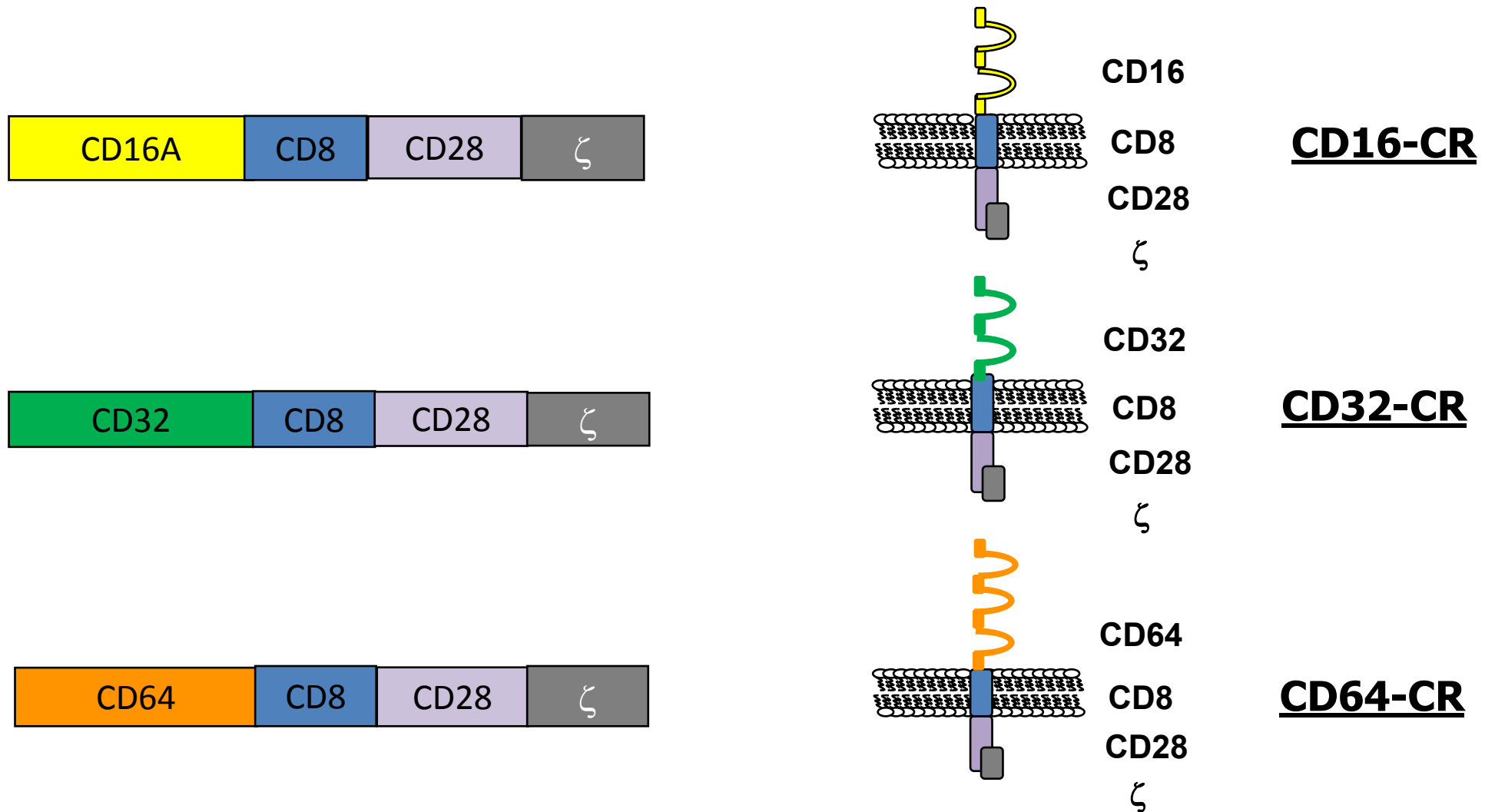
# Overcoming mAb limitations transferring ADCC function from NK cells to T cells

- NK cells armed with anti-EGFR mAbs kill KRAS mutated cancer cells
- The presence of specific Fc $\gamma$ RIII polymorphisms on NK cells predict favorable in patients with EGFR + malignancies.
  
- Leukemia and solid tumor cells induce NK cell damage
- NK cells barely infiltrate the tumor microenvironment
  
- CD8+ T cells are significantly present in colorectal lesions, and their infiltration is associated with a favorable prognosis

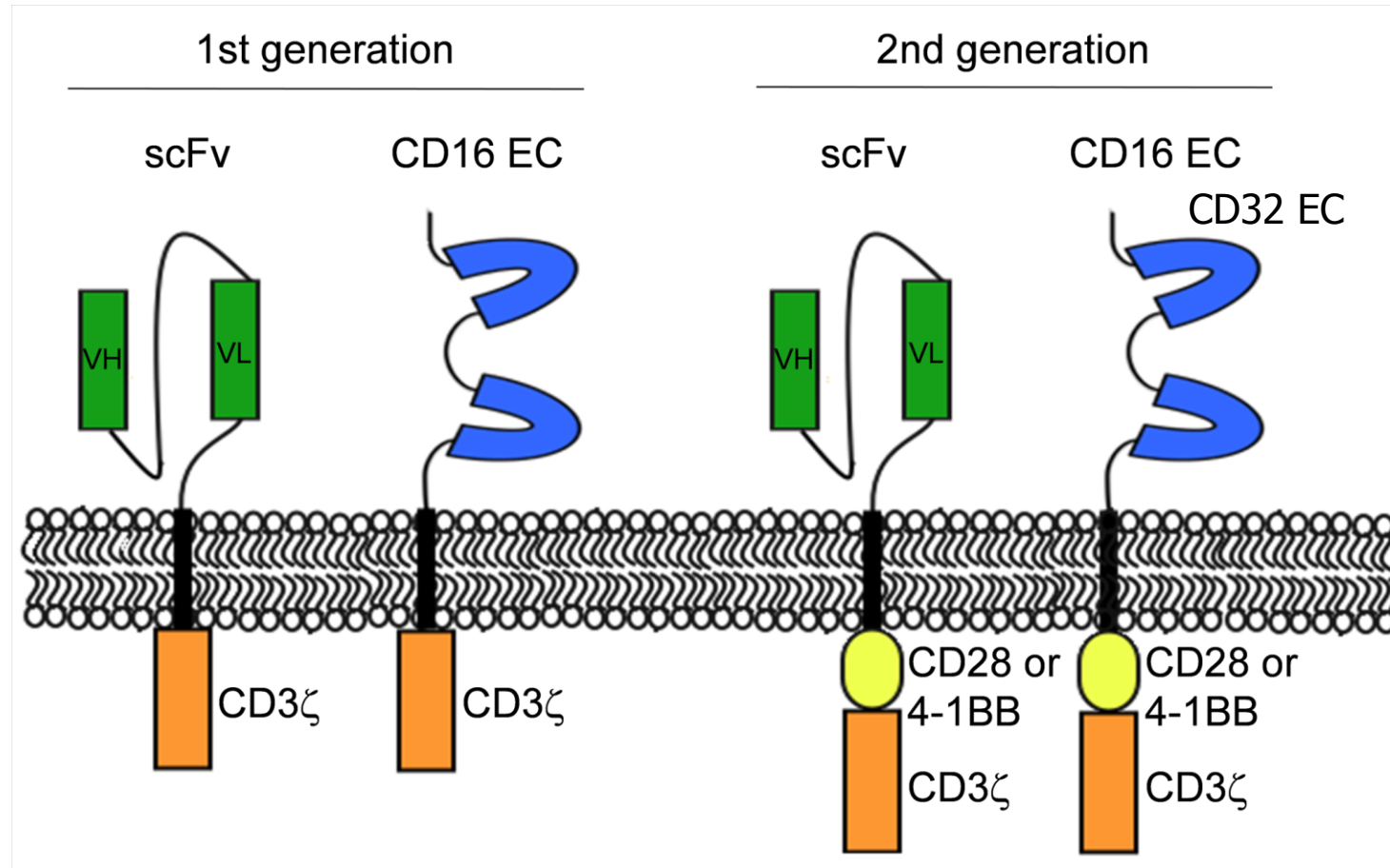
**We and other investigators propose to enhance the anti-tumor potential of mAbs by engineering cytotoxic T cells with Fc $\gamma$ Rs**



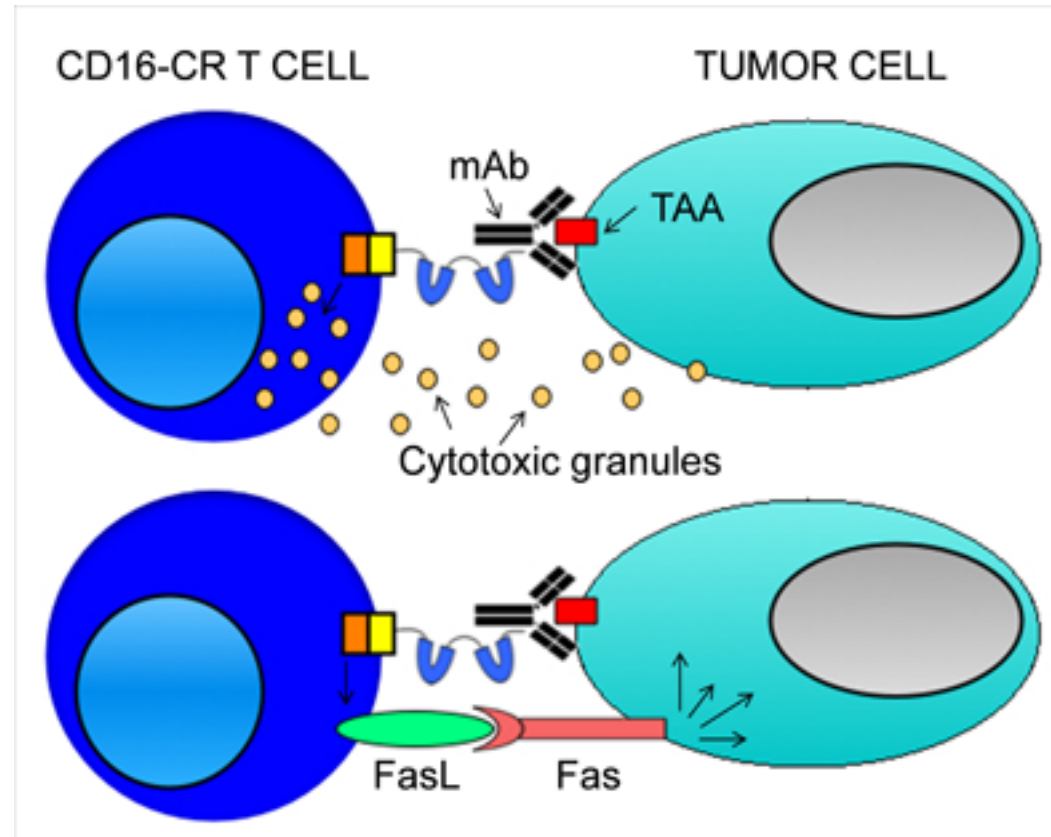
# Fc $\gamma$ Chimeric Receptors (Fc $\gamma$ -CR)



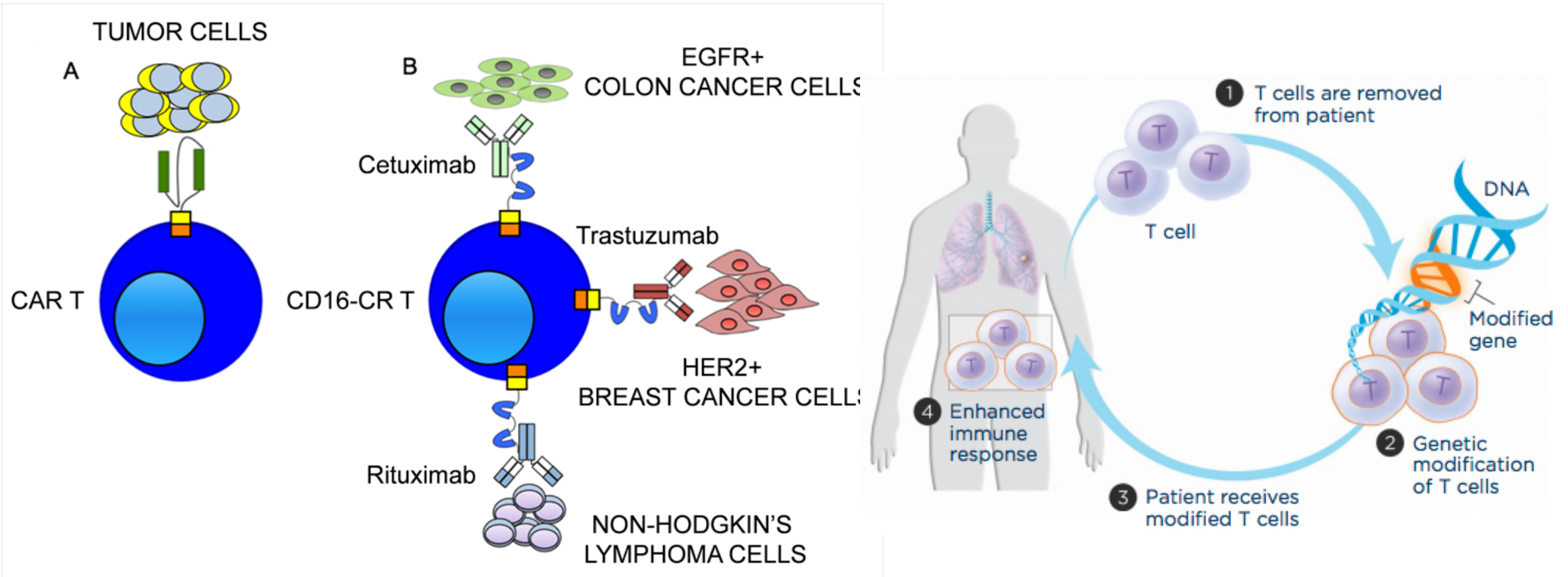
# Therapeutic mAbs confear multiple tumor specificity to Fc $\gamma$ -CR-engineered T cells



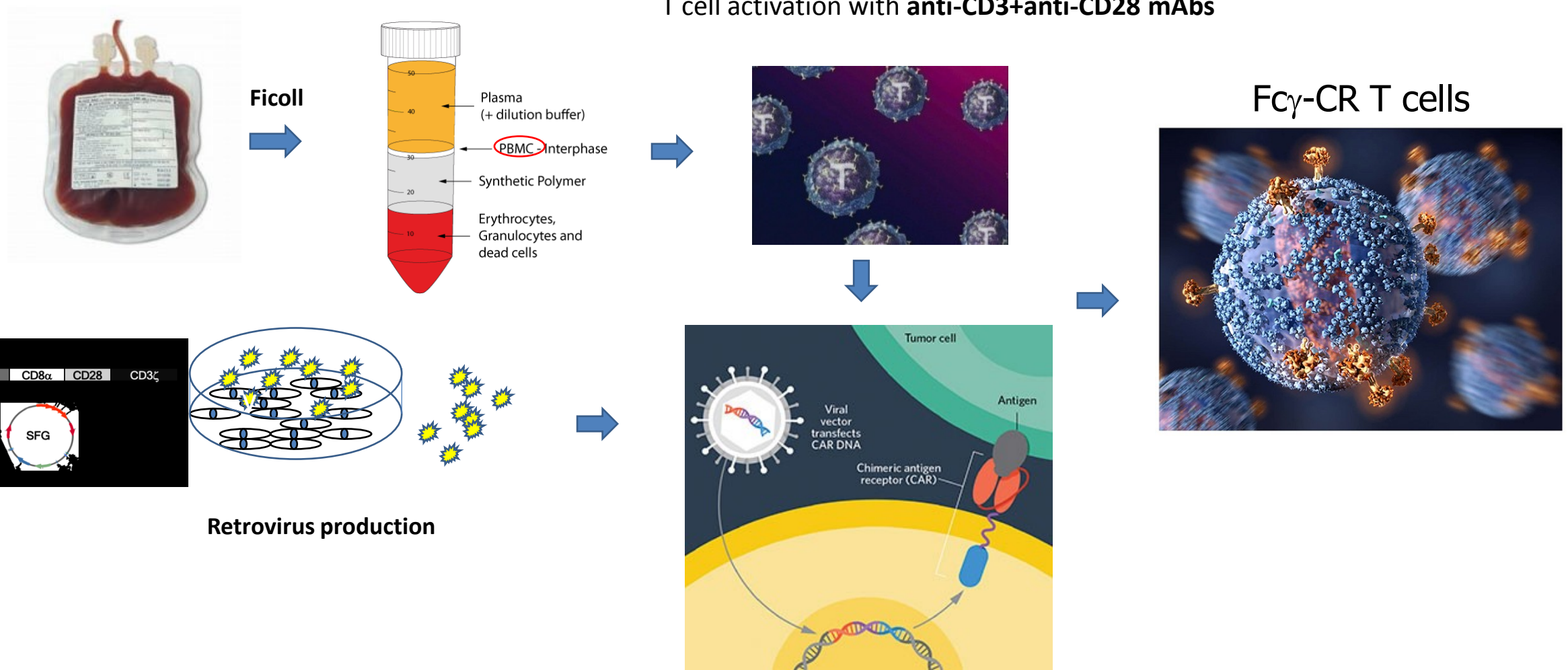
# Mechanisms of Fc $\gamma$ -CR T cell anti-tumor activity



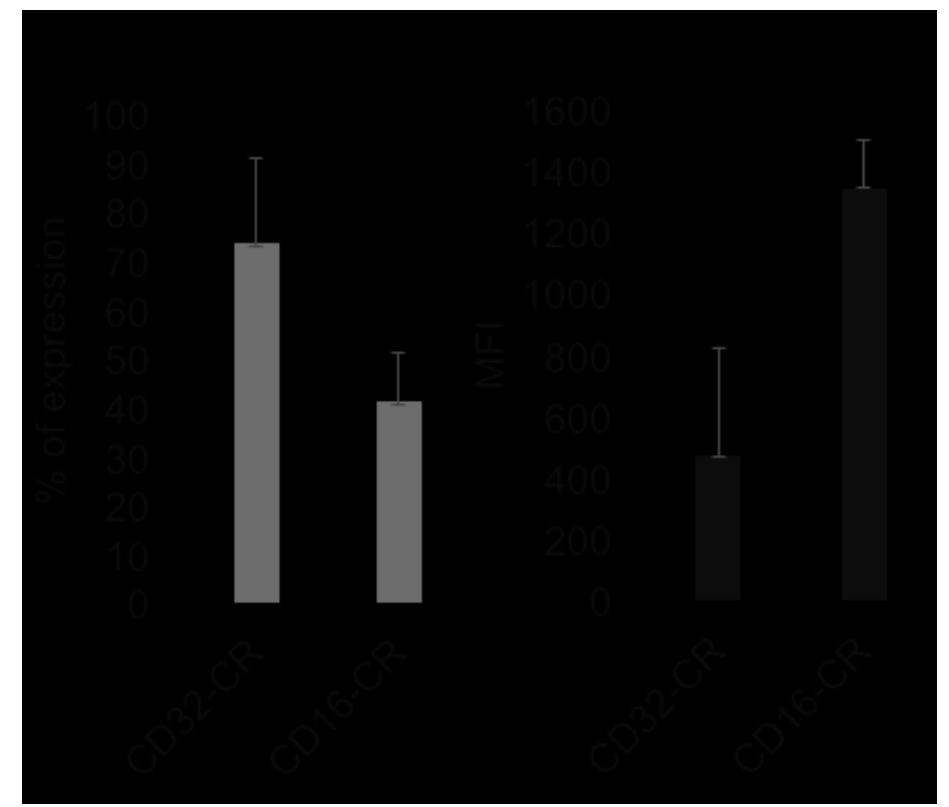
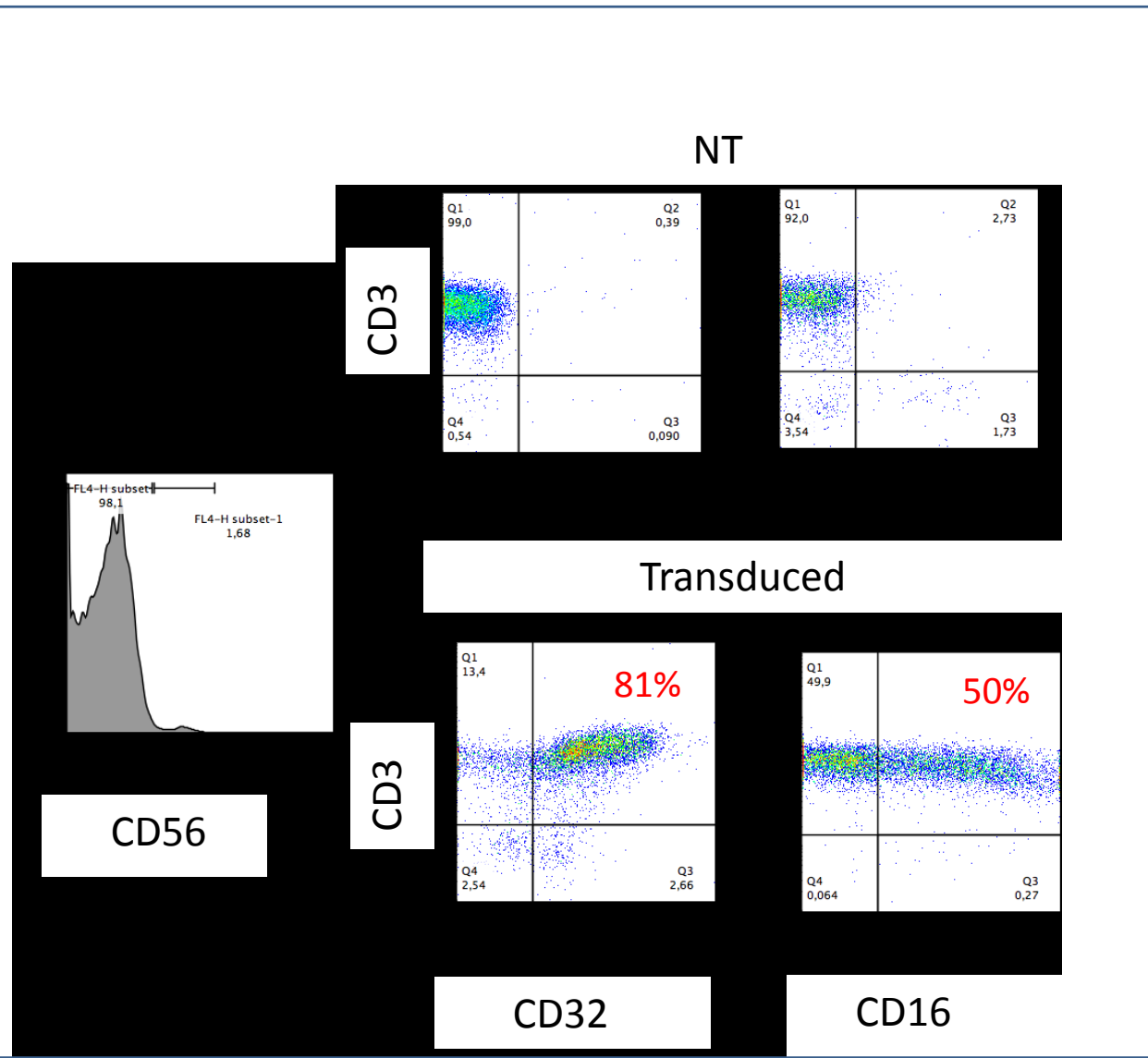
# Therapeutic mAbs confer multiple tumor specificity to Fc $\gamma$ -CR-engineered T cells



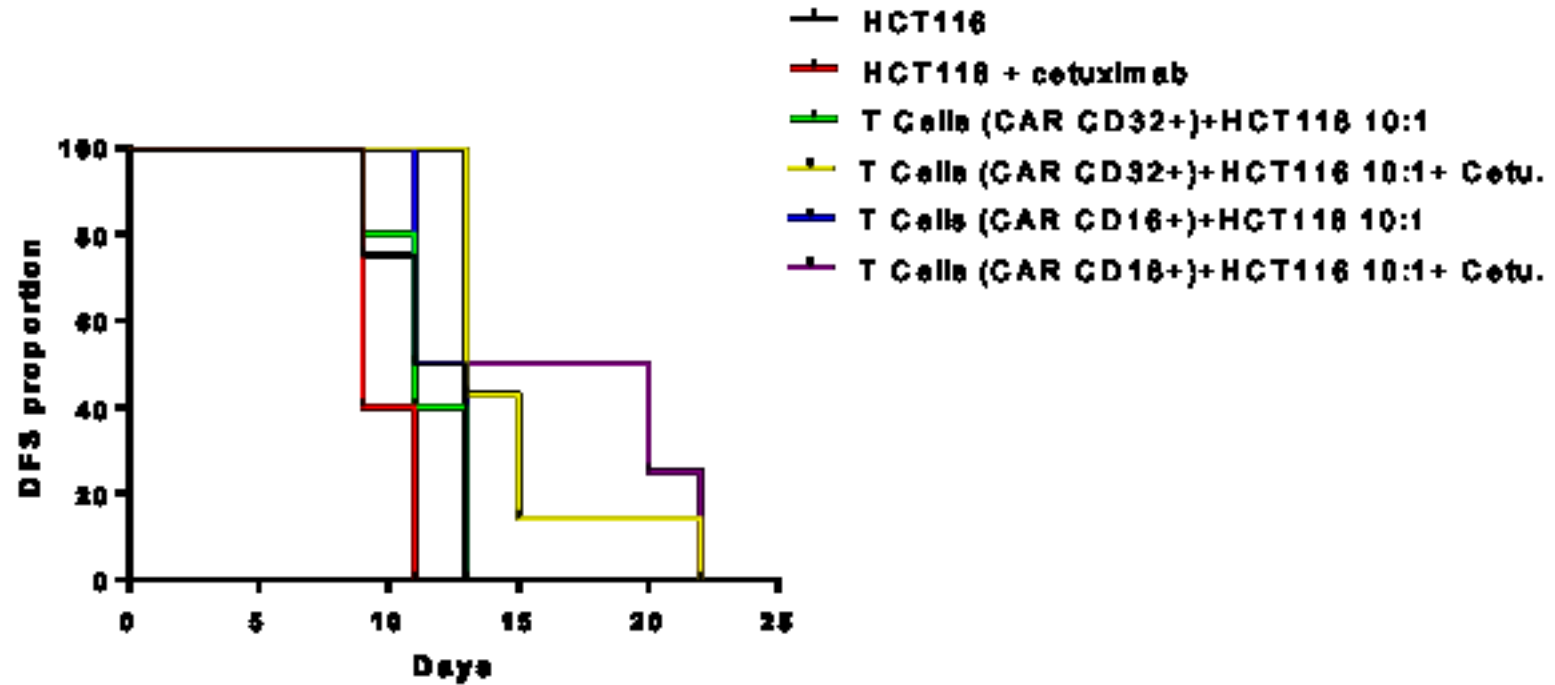
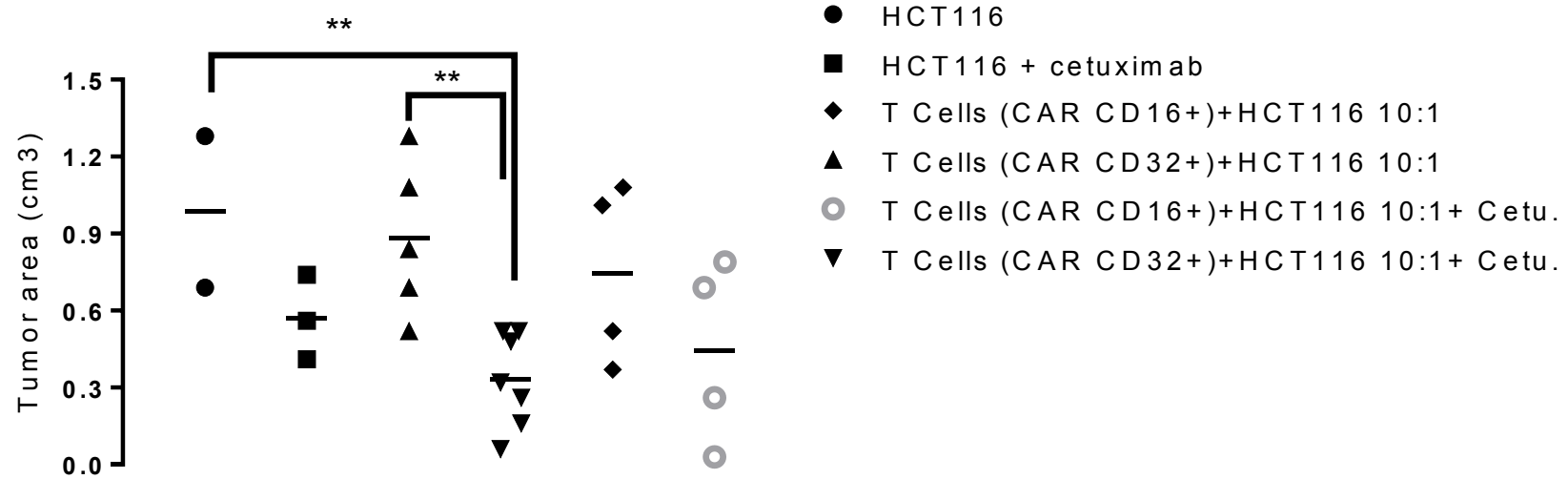
# Gene transfer technology used to engineer T cells to express Fc $\gamma$ -CR



# CD32-CR and CD16-CR are differently expressed on T cells



# Fc $\gamma$ -CR-engineered T cells damaged KRAS-Mutated cancer cells

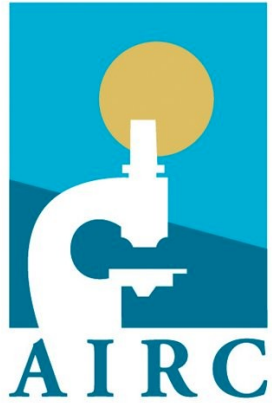


# Future perspectives

- ✓ Optimizing of CD64-CR
- ✓ Improving CD16- and CD32-CRs affinity for IgG isotypes (point mutation CD16<sup>V158</sup>/CD32<sup>H131</sup>)
- ✓ Testing the Fcγ-CR T cells anti-cancer activity against different EGFR+++/*Kras*<sup>M</sup> tumors in vitro and in vivo (es. NSCLC)
- ✓ Testing Fcγ-CR toxicity in non-immunodeficient mice bearing a spontaneous or engrafted CRC.



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