# Approaches to enhance the efficacy of EGFR- directed antibodies


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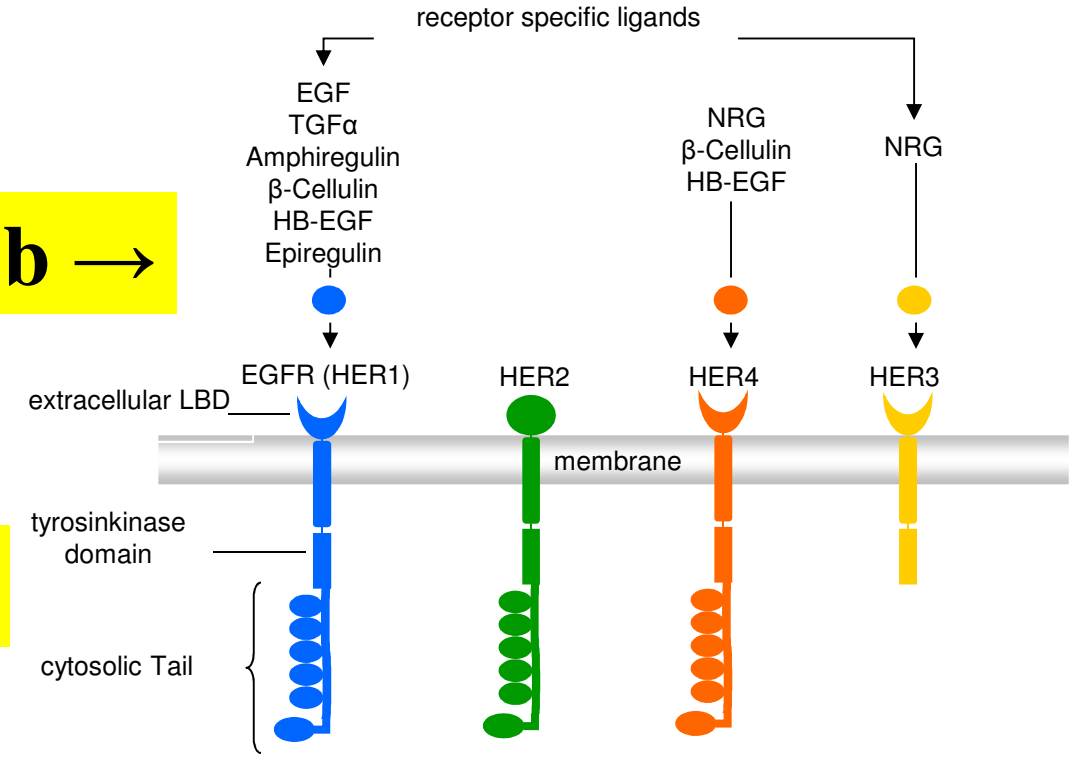
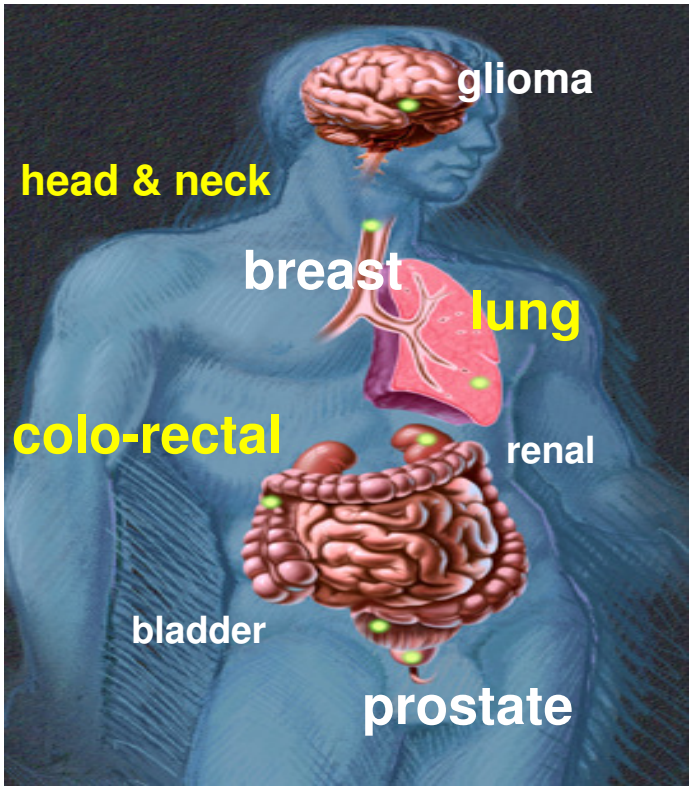
UK SH

# The epidermal growth factor receptor (EGFR) as target antigen for tumor therapy

**mAb →**

**TKI →**

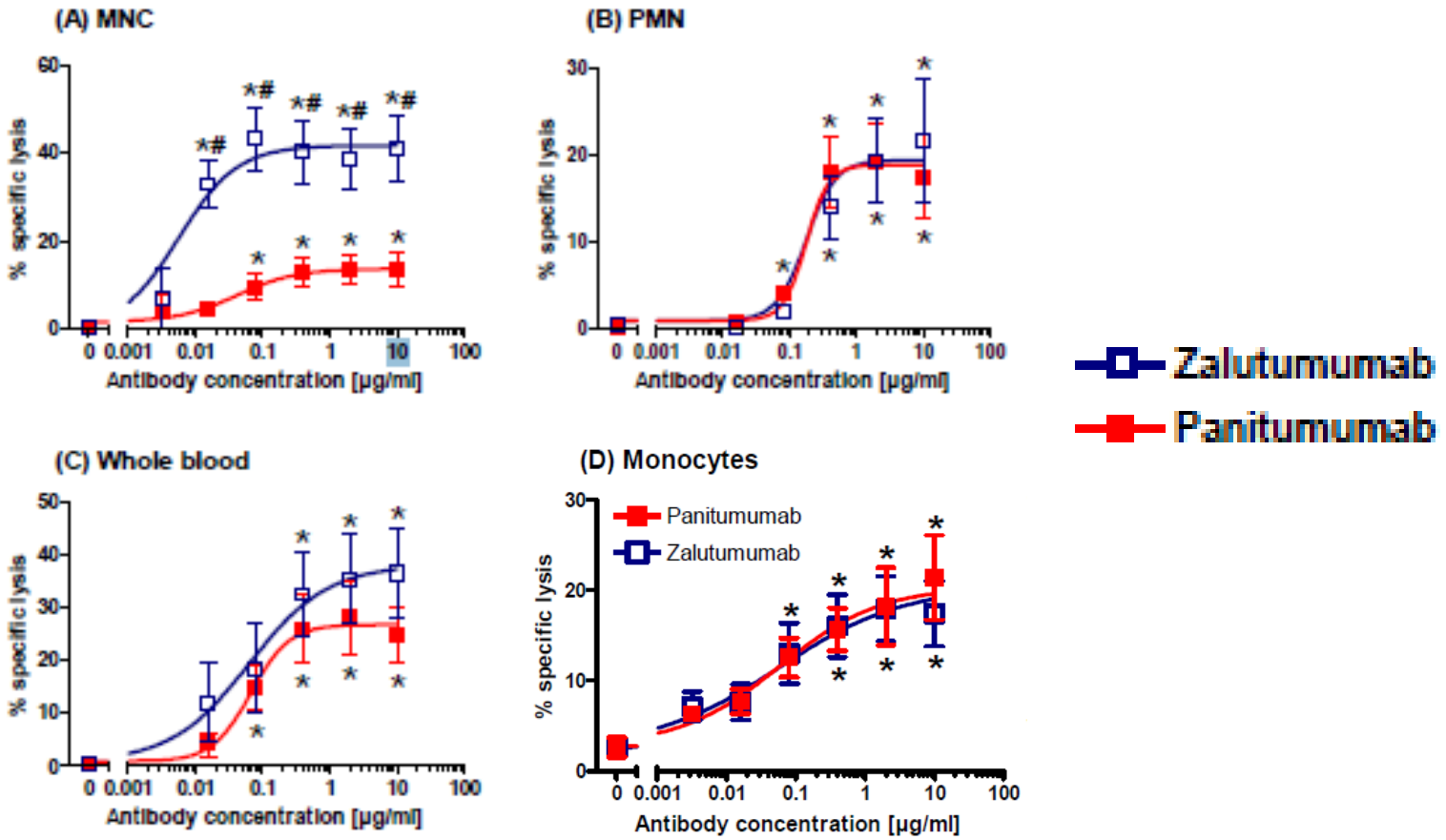



- two approved TKI: erlotinib, gefitinib
- two approved mAb: cetuximab, panitumumab
- different indications and different biomarkers !

modified from: Ciardiello et al., NEJM 2008

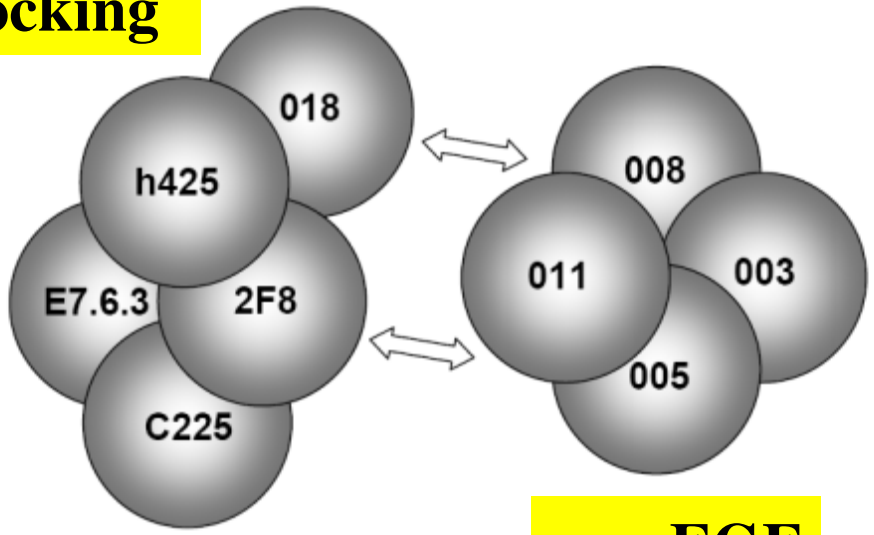
# Panitumumab recruits myeloid effector cells



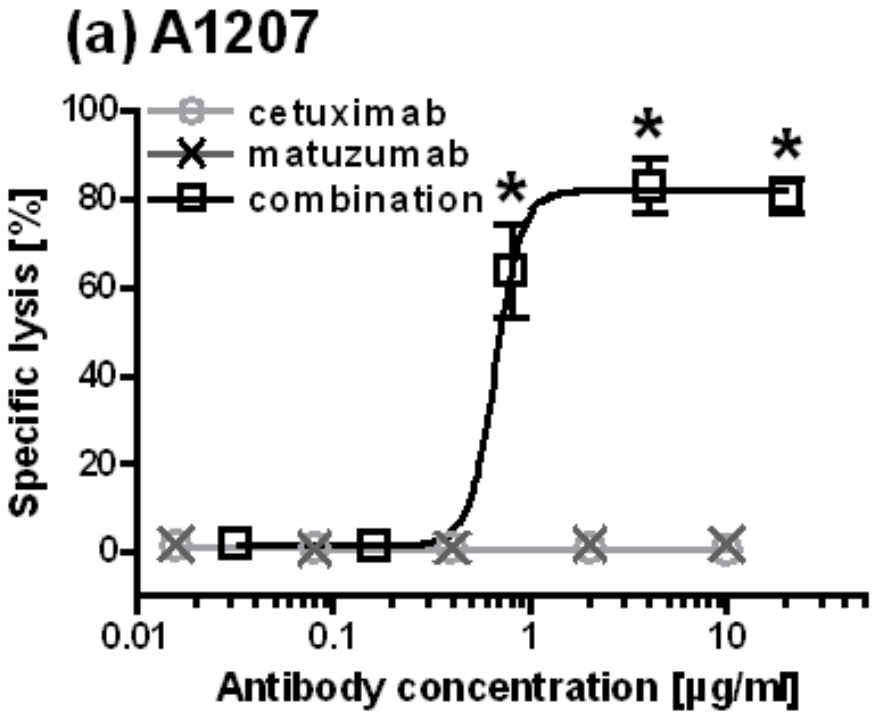

Schneider-Merck T et al. J Immunol. 184:512, 2010

# EGFR antibody combinations trigger CDC

**EGF blocking**



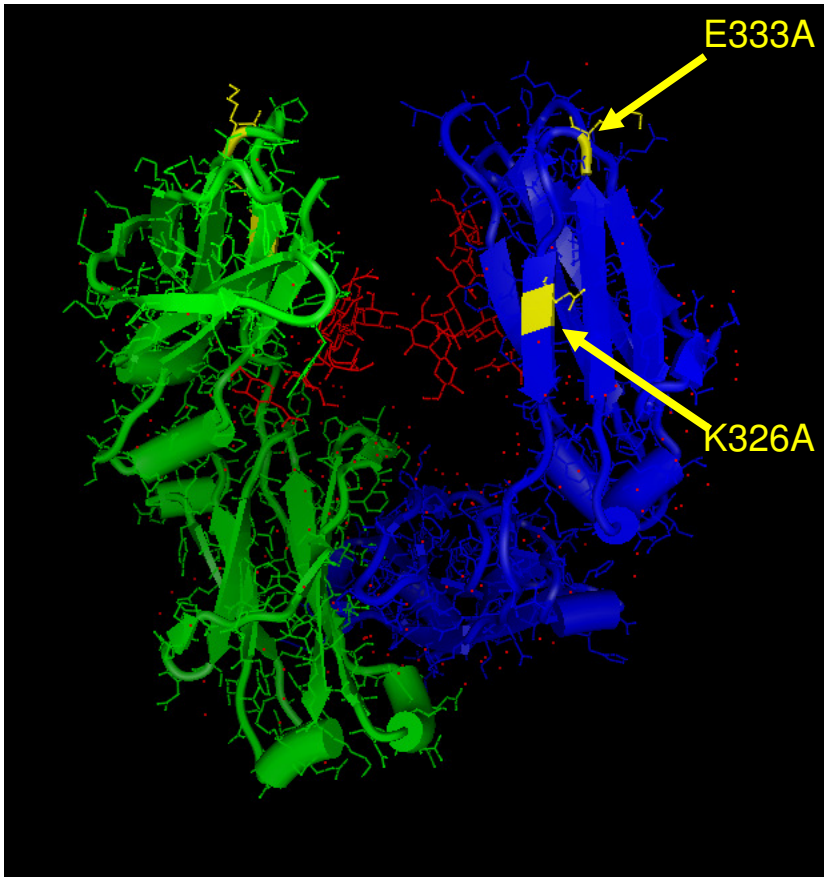

**non EGF blocking**

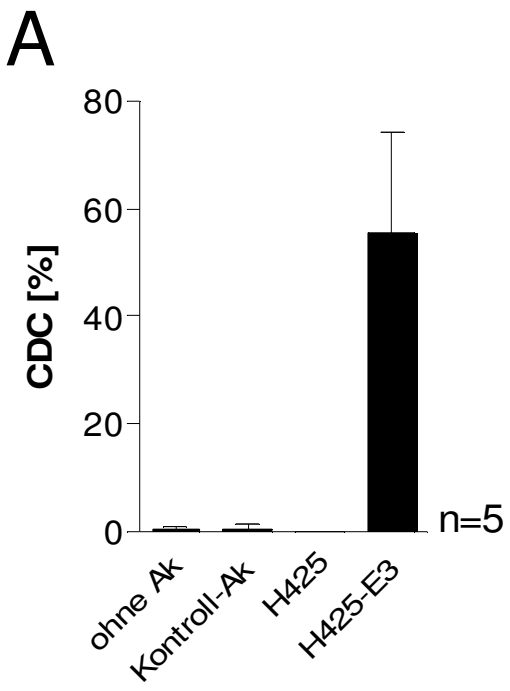


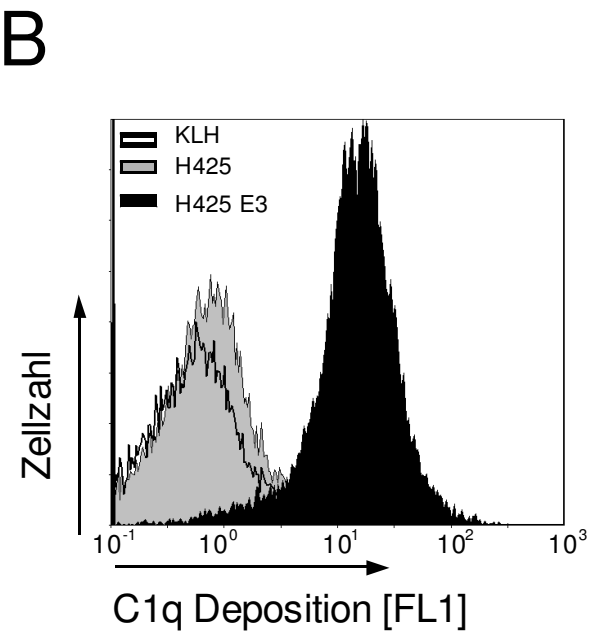

**complement fixation and CDC by select Ab combinations**

Dechant M et al Cancer Res. 2008

# Fc engineering enhances CDC



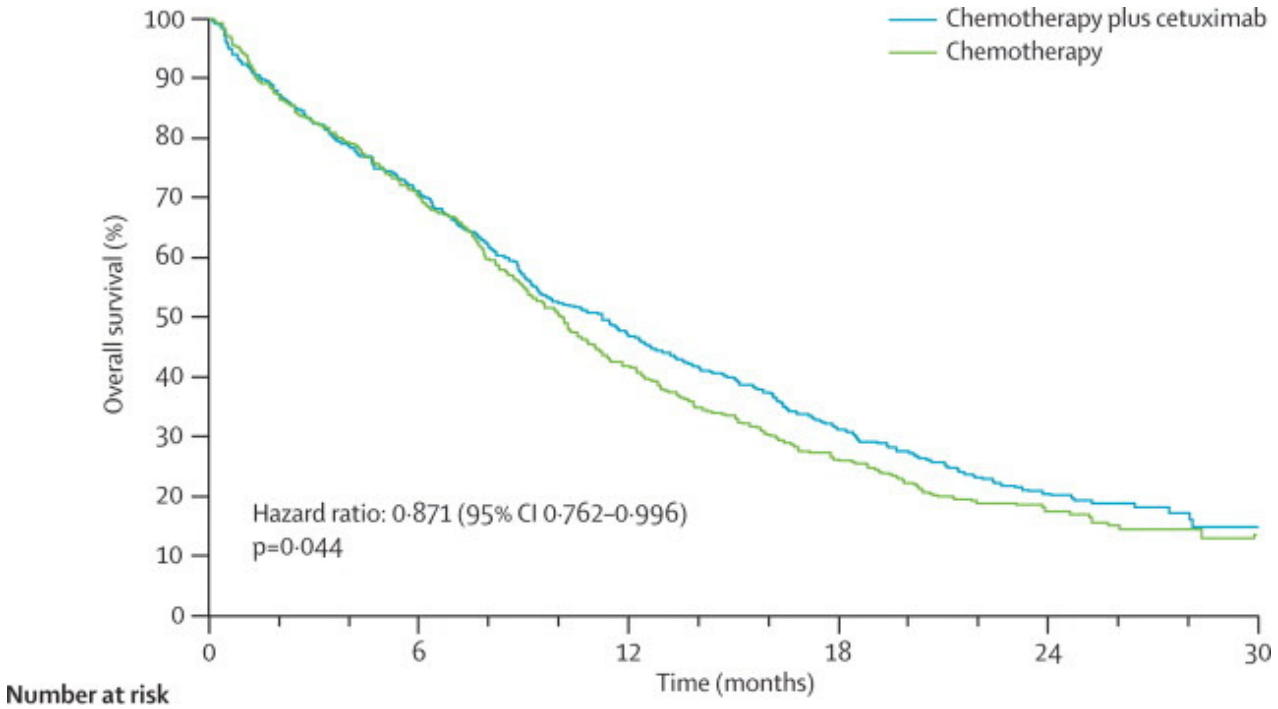




**Idusogie et al., J Immunol, 2001**

# Cetuximab + plus chemotherapy in NSCLC: the FLEX study

- **1125 patients with advanced NCSLC randomized**

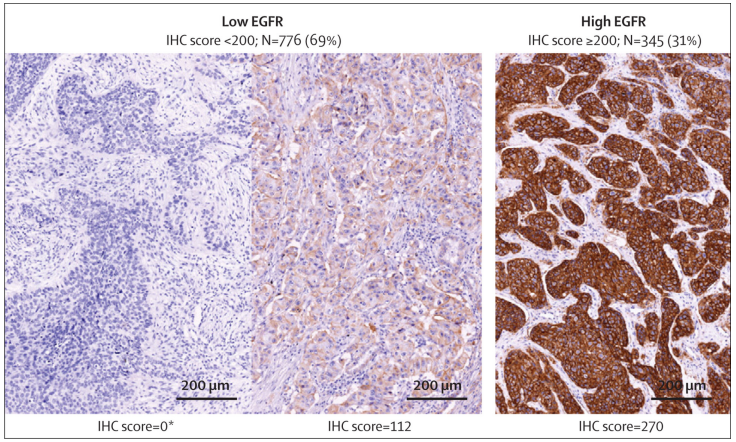



**Pirker R et al. Lancet 373, 1525, 2009**

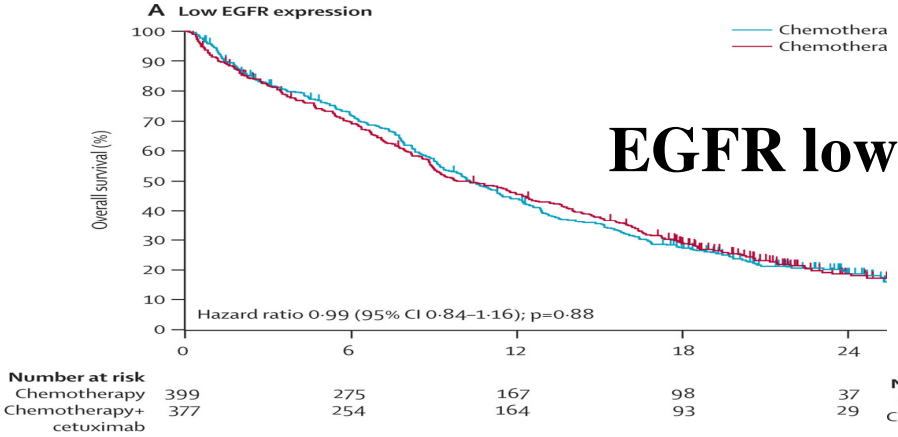
- **EGFR mutations not predictive**

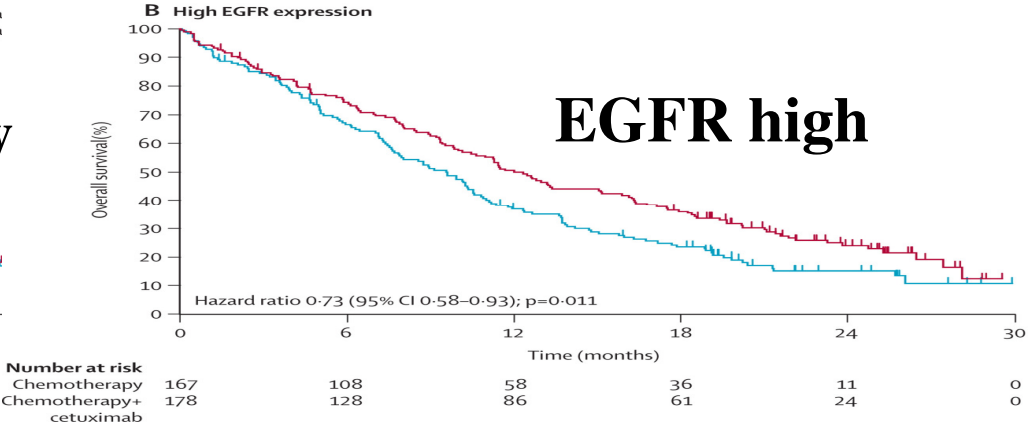
**O'Byrne et al. Lancet Oncol. 12:795, 2011**

# Quantitative EGFR histochemistry in NSCLC




- **retrospective analyses of FLEX data**
- **EGFR-ICH score 0 – 300**
- **high > 200 (30 %)**

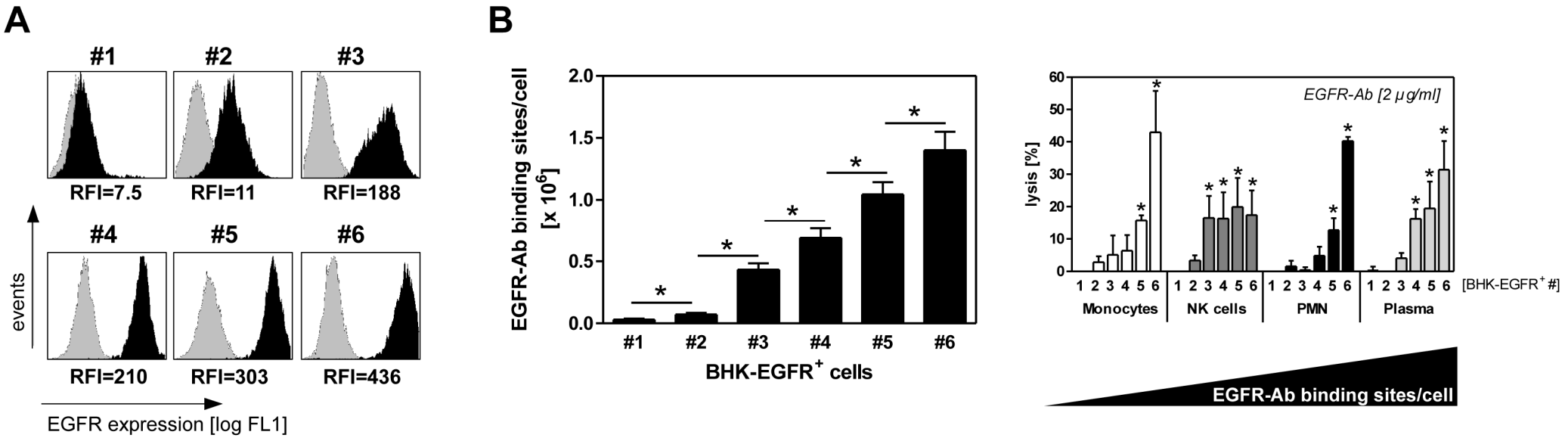



**impact of EGFR expression on different MoA ?**

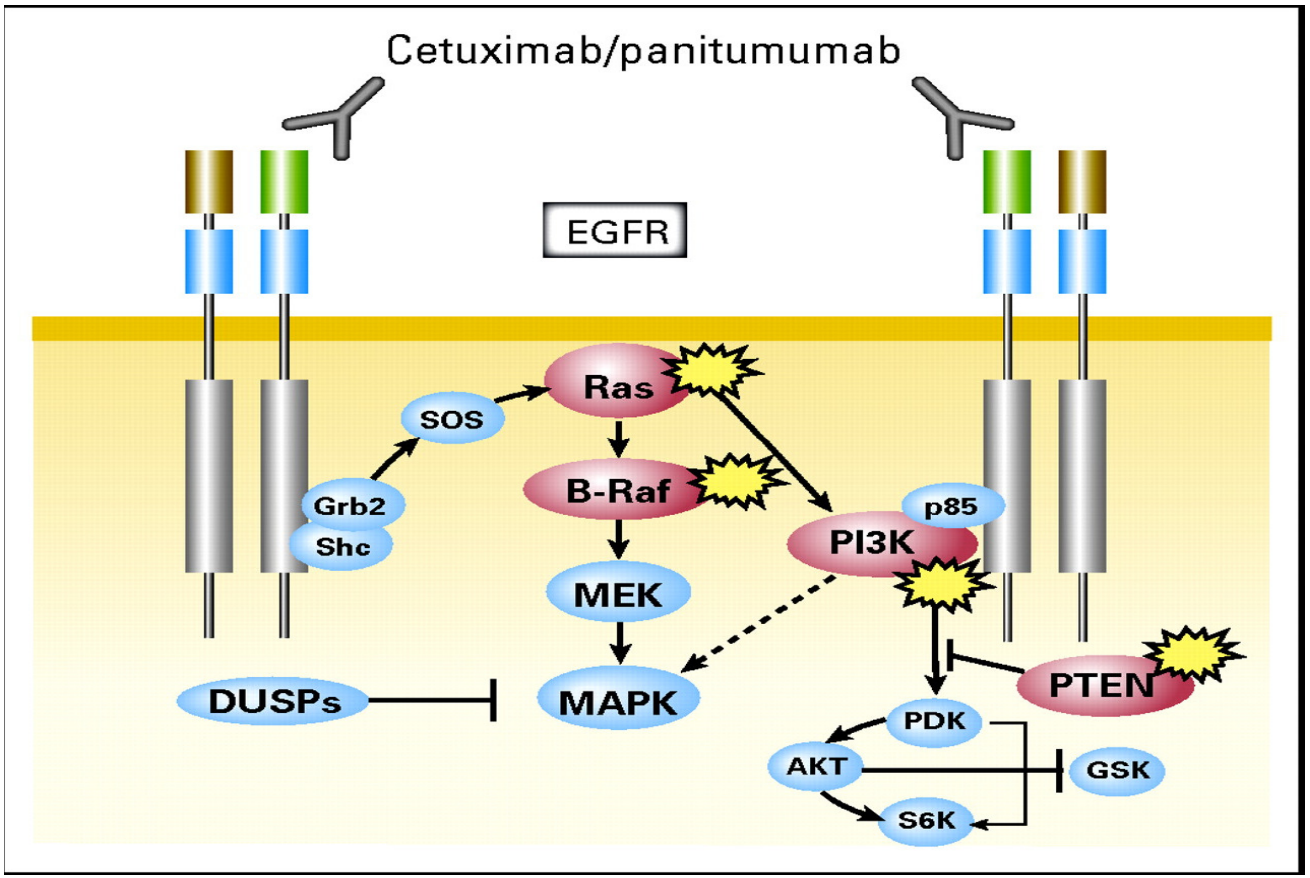
Pirker R et al. Lancet Oncol. 13:33 2012

# EGFR expression and mAb effector mechanisms




- **EGFR expression levels correlate with Fc- mediated killing**

# KRAS activation by-passes EGFR blockade

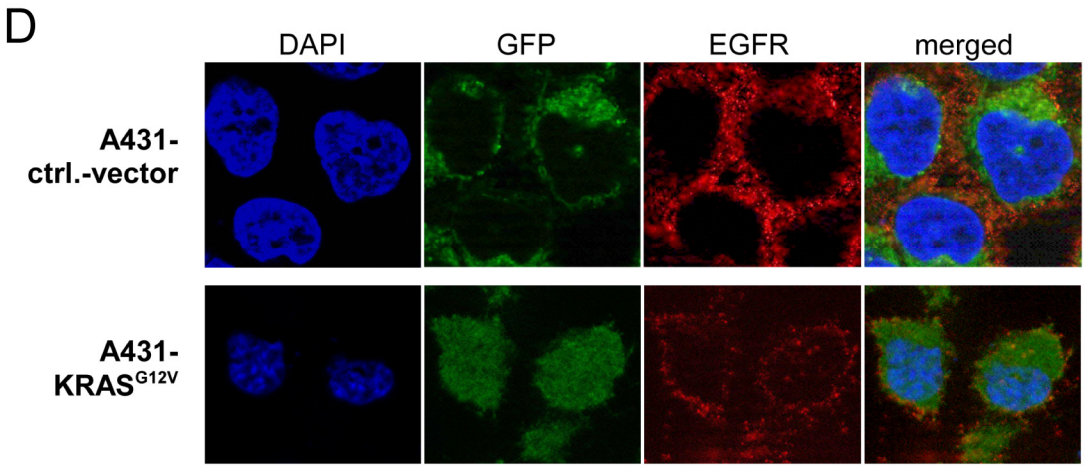


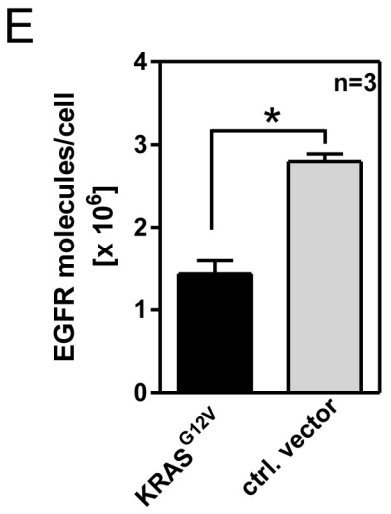

Bardelli A and Siena S. J Clin Oncol 28:1254, 2010

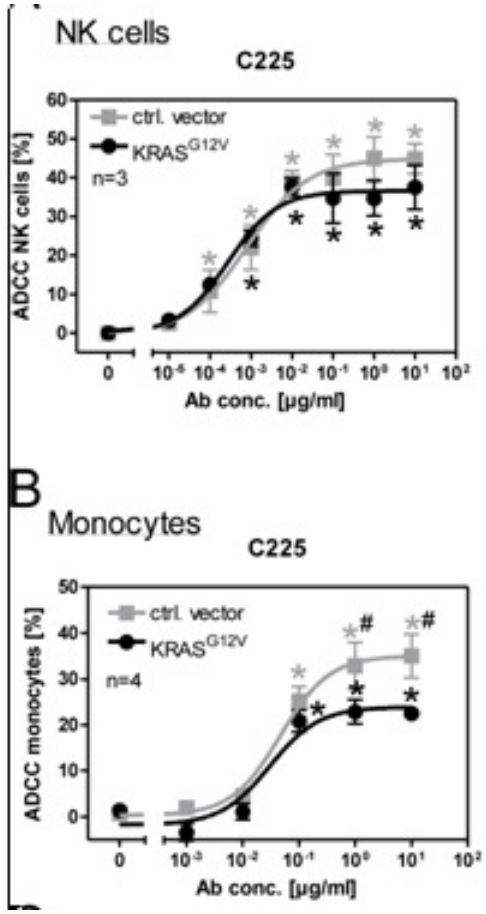
**Does KRAS activation impact ADCC and CDC ?**

# Impact of mutated KRAS on ADCC

- wildtype A431 versus A431 transfected $KRAS^{G12V}$



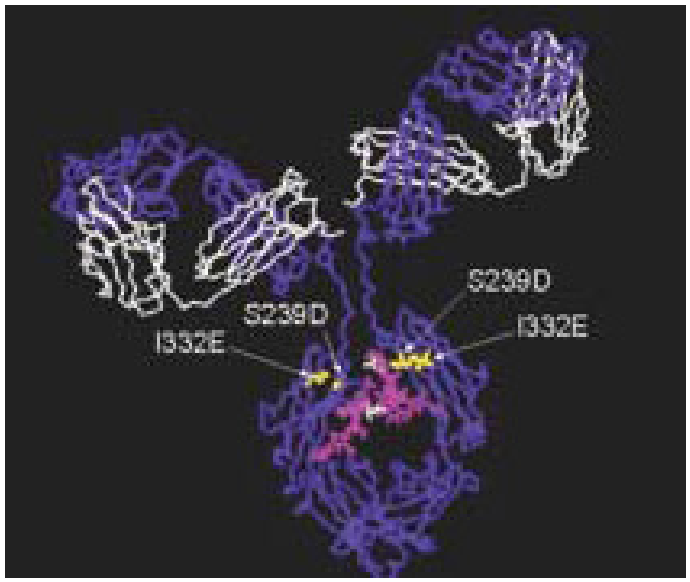




- mutated KRAS reduces EGFR expression
- impairs ADCC and CDC

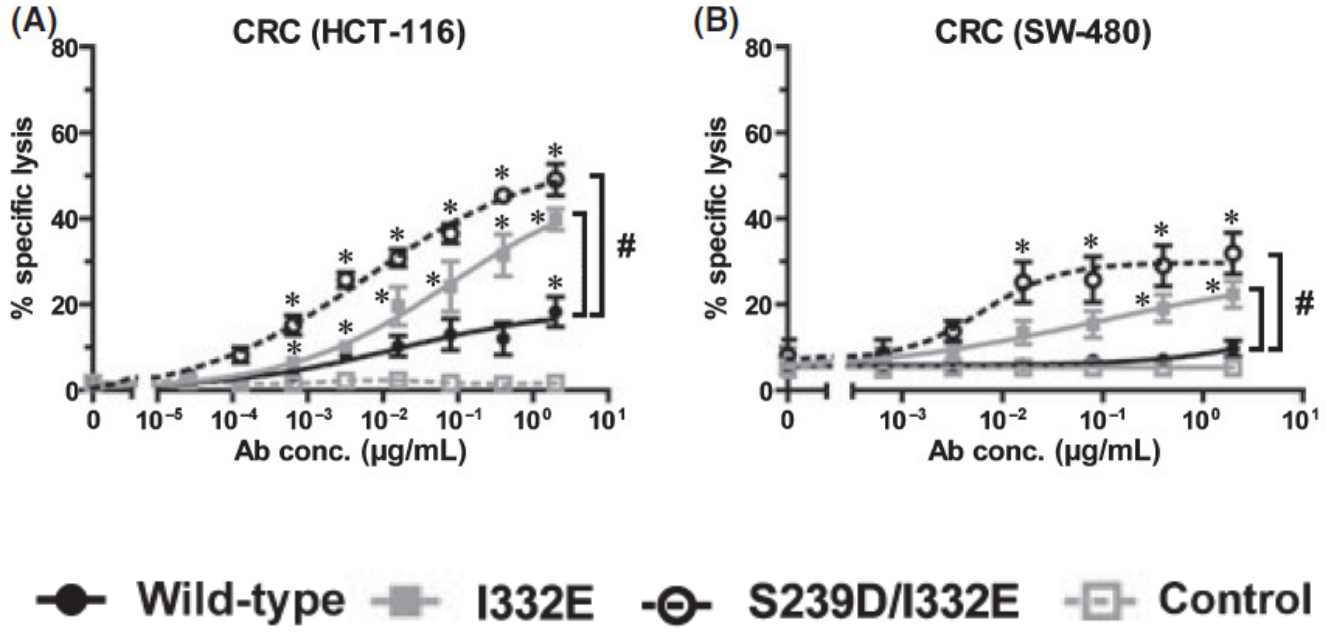
Derer S et al Neoplasia 2012

# Fc protein- engineering improves killing of KRAS mutated tumor cells




| | FcγRIII* |
|---|---|
| wt | 1 |
| single | 5 |
| double | 40 |

* fold enhancement

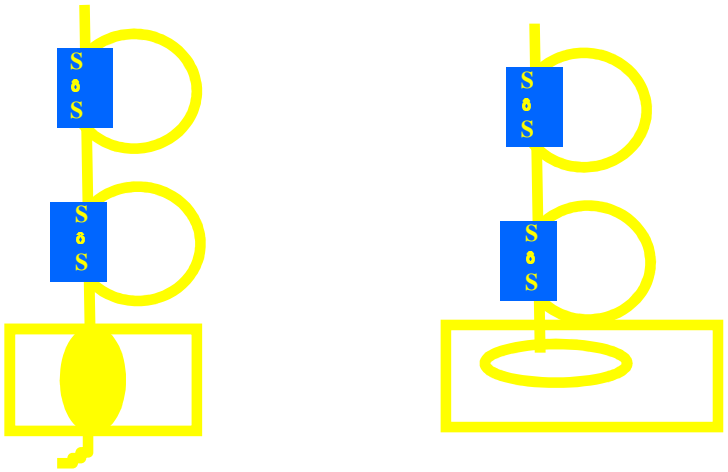



Schlaeth et al Cancer Sci 101:1080, 2010

# Enhancing the affinity for FcγRIIIa: caveat !

**Human FcγRIII (CD16)**

**IIIa** **IIIb**



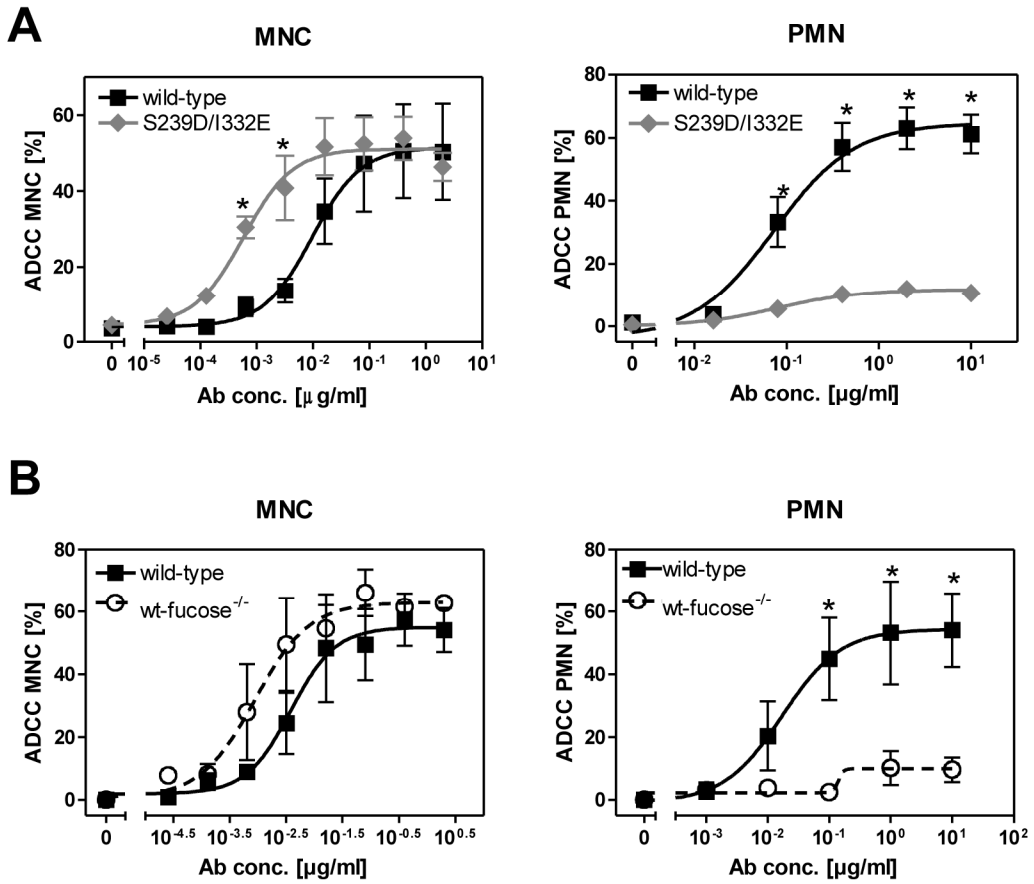
**transmembrane** **GPI-linked**

**NK** **PMN**

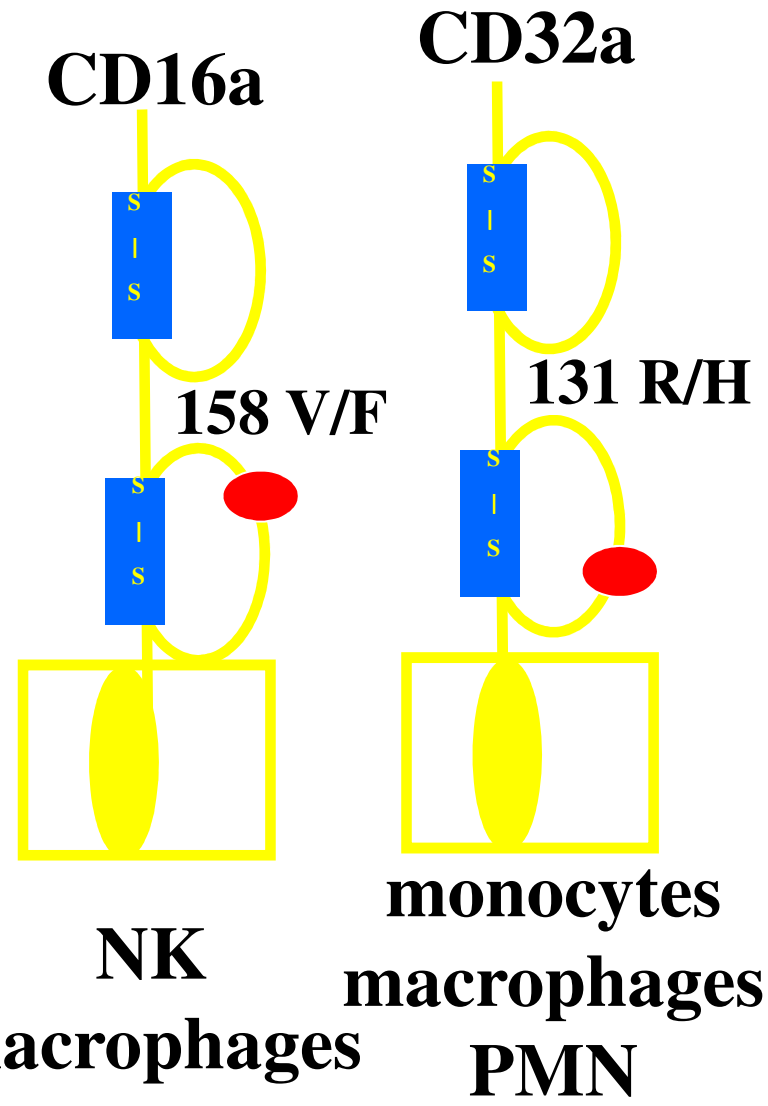
- **highly homologous**
- **similar affinities for IgG**
- **both polymorphic**
- **IIIb more abundant**
- **IIIb <u>not</u> cytotoxic**

**Peipp et al Blood 112:2390, 2008**

# Enhancing FcγRIII affinity abrogates PMN ADCC



# Effector cells for antibody therapy



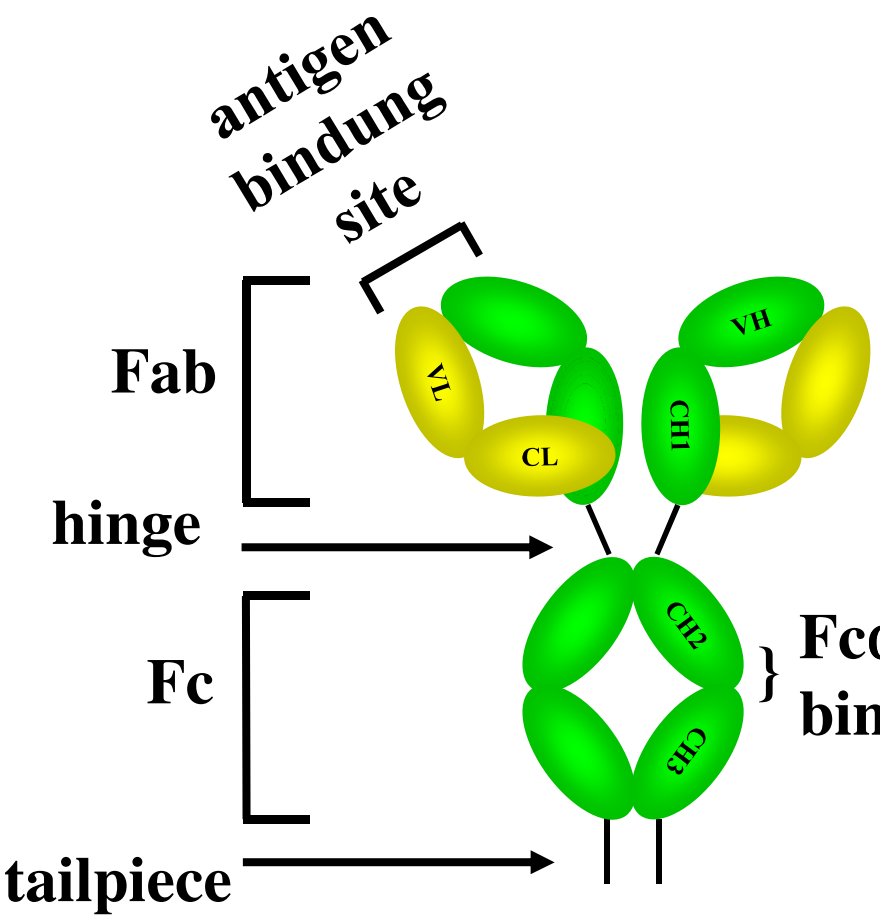

## NK cells:

- kill via FcγRIIIa (CD16)
- low numbers
- inhibited by HLA class I expression
- expansion associated with side effects

## Myeloid cells:

- kill via FcγRIIa (CD32)
- predominant leukocyte tumor infiltrate
- HLA class I- independent killing
- expanded by G-CSF or GM-CSF

# Human IgA antibodies

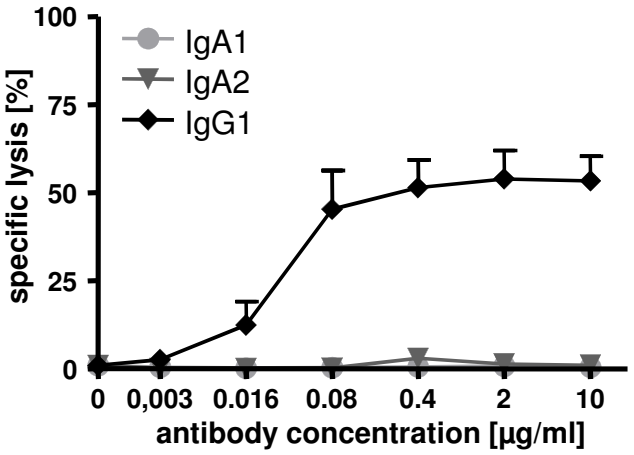



- most abundantly produced Ig
- protects serosal surfaces
- serum half life approx. 7 days
- two isotypes: $IgA_1$ and $IgA_2$
- therapeutic potential not explored

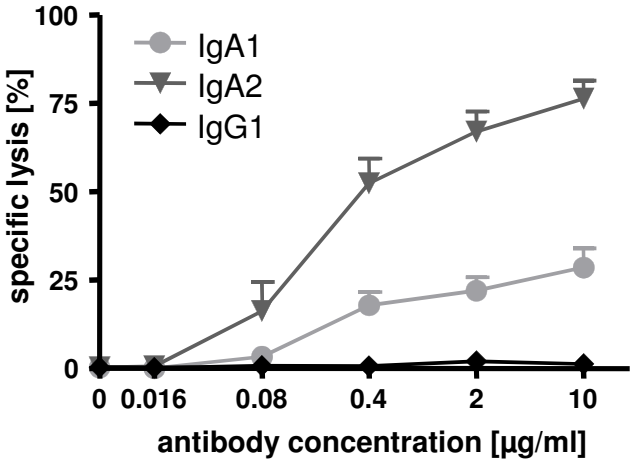
⇒ C225-$IgG_1$, -$IgA_1$, -$IgA_2$
in serum-free CHO-K1

# Monomeric IgA antibodies

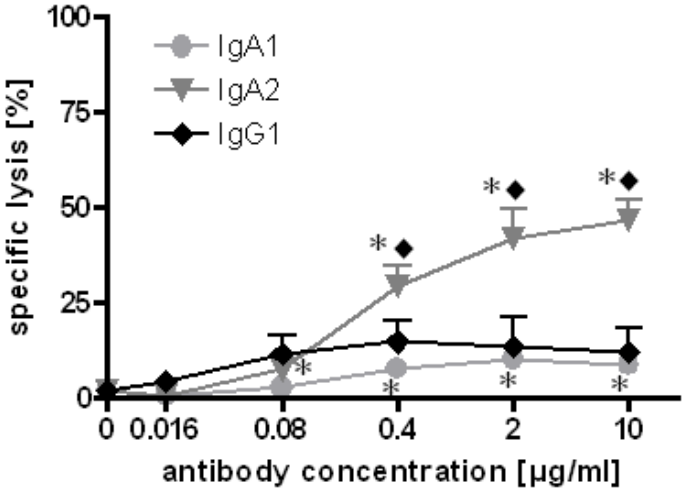
## NK cells




## PMN




## whole blood

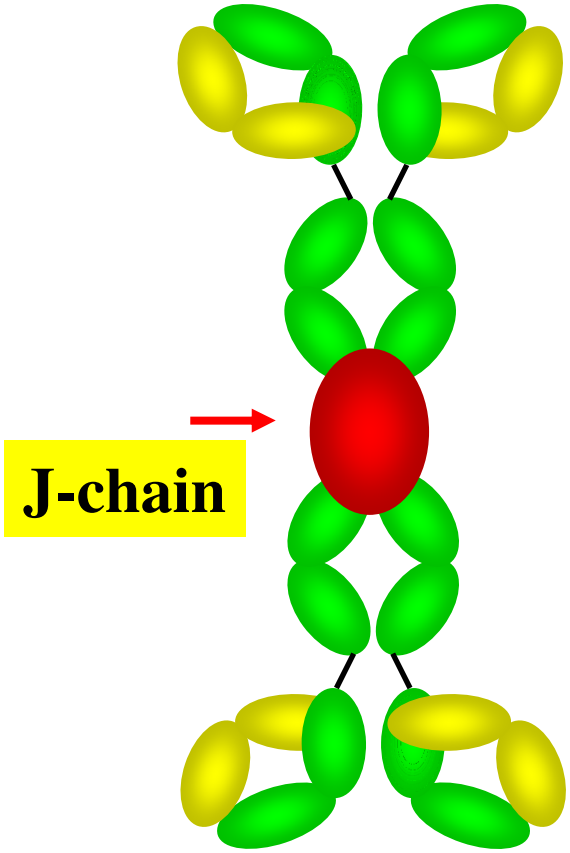



**ADCC by $IgA_2$ is enhanced compared to $IgG_1$ and $IgA_1$**

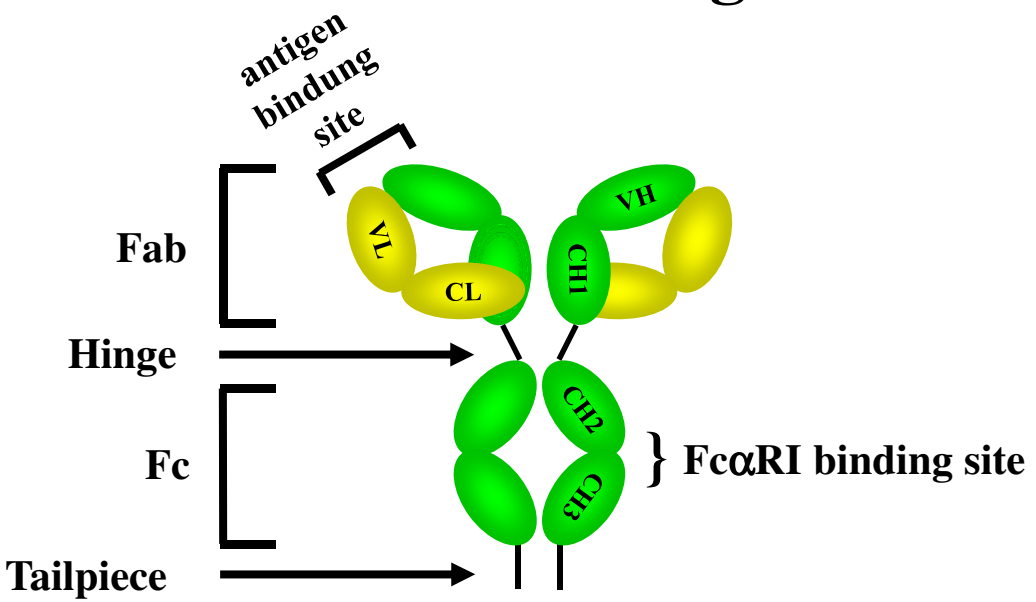
Dechant M et al. J. Immunol. 179:2936, 2007

# Dimeric IgA antibodies for cancer therapy

## dimeric IgA



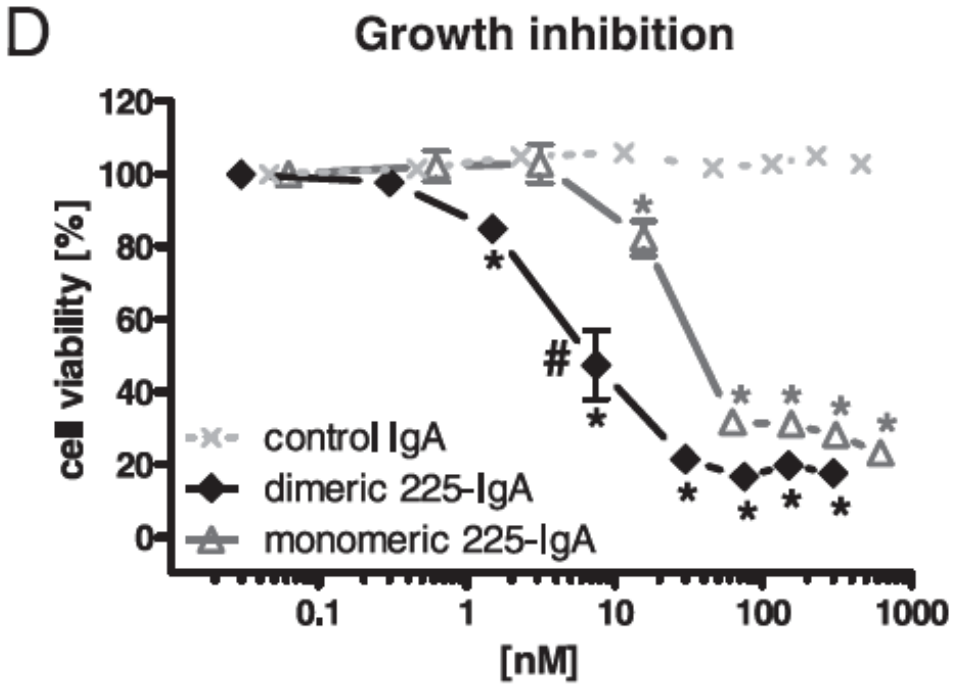

## monomeric IgA



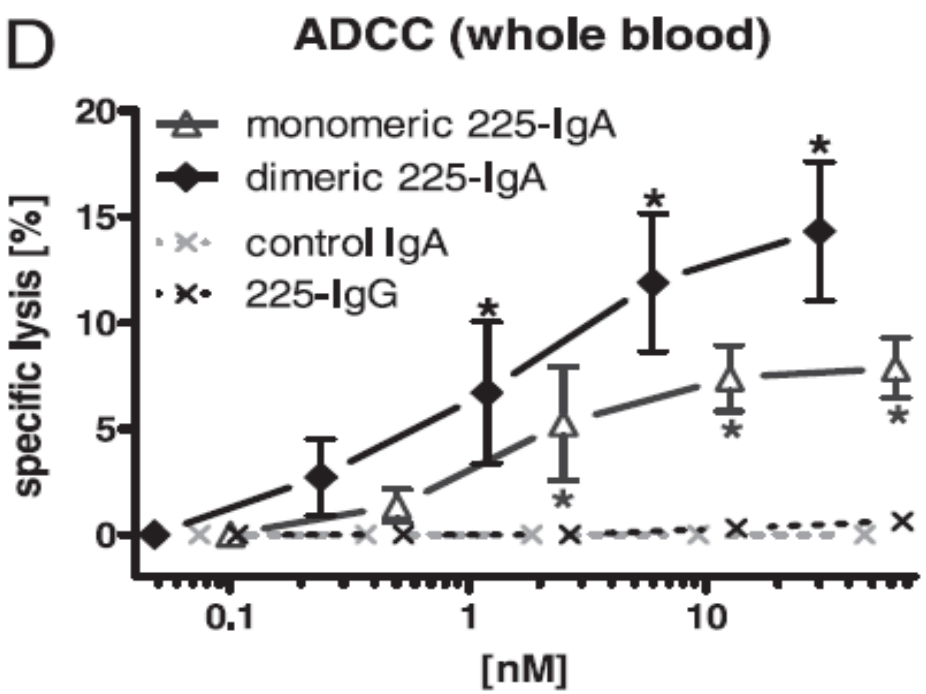

- improved signaling in tumor cells ?
- triggering of effector cells ?
- favourable biodistribution ?

# Fab- and Fc- mediated effector functions are enhanced by dimeric IgA

**Fab- mediated**




**Fc- mediated**

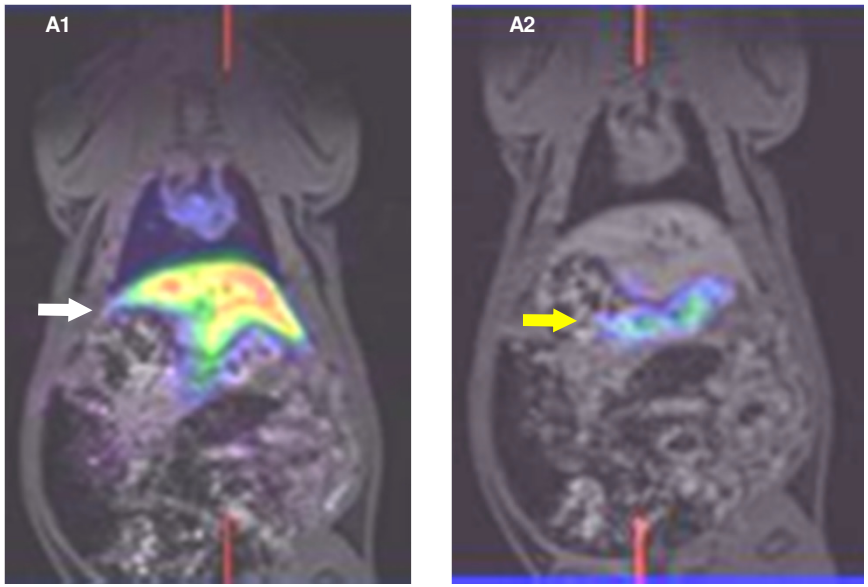



Lohse S et al. J. Immunol. 186:3770, 2011

# Dimeric IgA targets the mucosal system

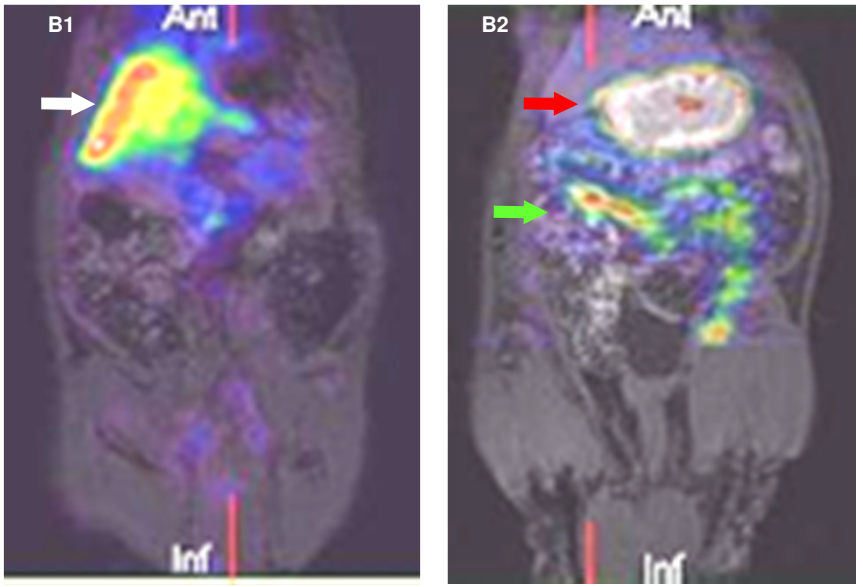
## monomeric IgA

10 min. 5 h




## dimeric IgA

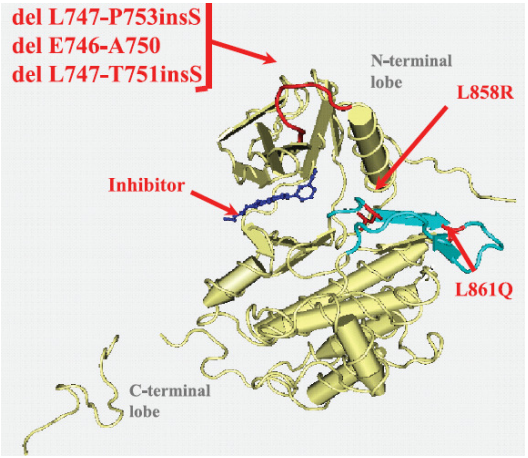
10 min 5 h



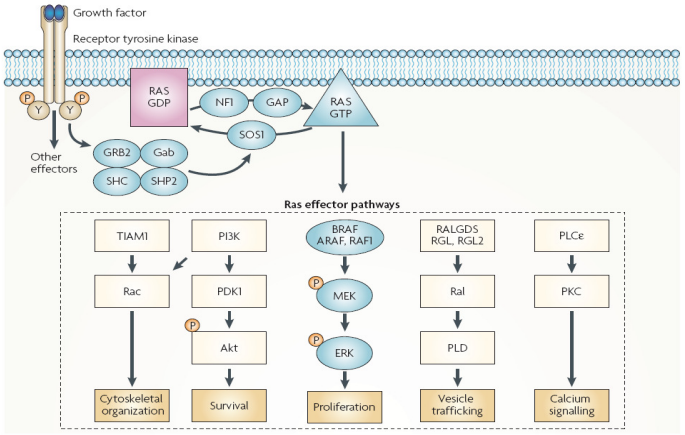
# Conclusions :

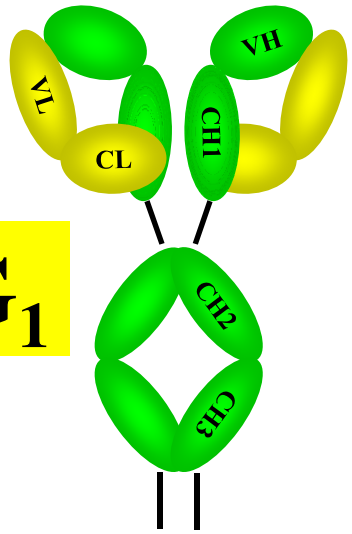
1. EGFR mutations
   - affect response to TKI
   - irrelevant for Ab
2. KRAS mutations
   - impact Ab therapy
   - can be overcome
3. EGFR expression levels
   - relevant for Ab in NSCLC (?)
   - critical for Fc- mediated MoA
4. myeloid cell recruitment
   - suboptimal with $IgG_1$
   - occurs with $IgG_2$
   - improved with IgA

**EGFR mutations**



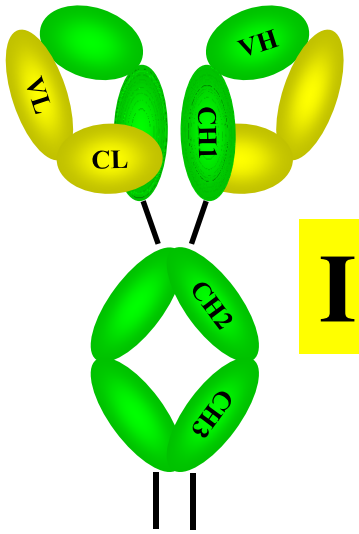

**KRAS mutations**




**$IgG_1$**

**NK cells**




**$IgA_2$**

**myeloid cells**

# Co-workers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