Bioassay
Bioanalytical Contract Laboratory
A Reaction Biology Company

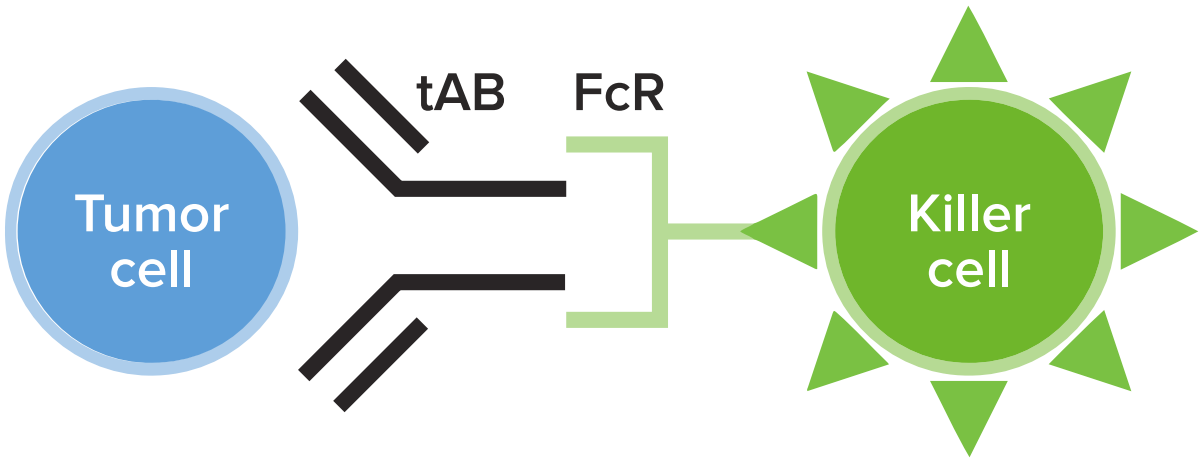
ADCC
The Antibody Dependent Cell mediated Cytotoxicity

Therapeutic antibodies (tAb) and their medical indications have grown fast recently

The Antibody Dependent Cell mediated Cytotoxicity (ADCC) is a mechanism of immune defense attributed to the mode of action of several tAb, in particular tumor cell targeting. In ADCC, tumor cells (recognized by tAb) are targeted by natural killer cells by their FcγRIII (CD16) receptor binding to the Fc portion of the tAb followed by lysis of the target (tumor) cell. Flow cytometry based analytical method

Therapeutic antibodies (tAb) and their medical indications have grown fast recently

- Different target cell lines
- Purified natural killer (NK) cells as effector cells
- NK cells genotyped (158 V/F, 131 H/R) and quality controled by flow cytometry (CD16, CD56 marker expression)
- Established protocols for several tAb
- FACS-based ADCC Assays
- Different stainings of target cells (e.g. PKH26, Calcein AM)



Assay Setup and Evaluation

- Microtiter plate-based (MTP) ADCC Assays
- Different cytotoxicity detection methods (e.g. Calcein AM, LDH, Alamar-Blue®, Delfia®)
- Comparative ADCC potency testing
- Double controlled assay by measurement of fluorescence in supernatant (NK cell lysed) and direct lysis of remaining target cells (detergent lysed)
- Full logistic curve fitting

Figure:
Example for MTP ADCC Assay:

- MabThera® (RTX) was used with 100% and 70% potency level as comparative potency testing
- Calcein AM fluorescence (vertical) and RTX dose (horizontal) are shown on graph as response
- A 4-PL fit was used for potency determination
- Estimated vs stated potency: 69.3% versus 70%
- Relative confidence interval of 12.5%