



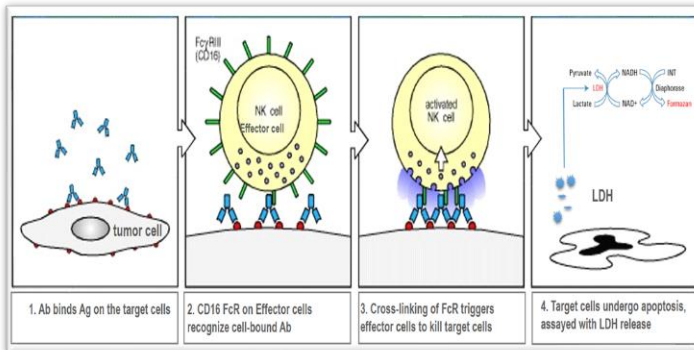
IMMUNE FUNCTIONAL ASSAY: ADCC and CDC

ANTIBODY-DEPENDENT CELL-MEDIATED CYTOTOXICITY AND COMPLEMENT DEPENDENT CYTOTOXICITY

Antibody therapy has been proven to be highly powerful for cancer treatment. Two important mechanisms used by antibody drugs to kill targeted tumor cells are Antibody-Dependent Cell-Mediated Cytotoxicity (ADCC), and Complement Dependent Cytotoxicity (CDC). We currently offer validated human PBMC-based and natural killer (NK) cell-based ADCC assay. The readout is endpoint-driven (target cell lysis). The CDC assay uses normal human or rabbit serum as the source of complement. By implementing strict QC standards, we can provide the assurance of the efficacy and potency profiles of therapeutic antibodies and other immunomodulatory agents.

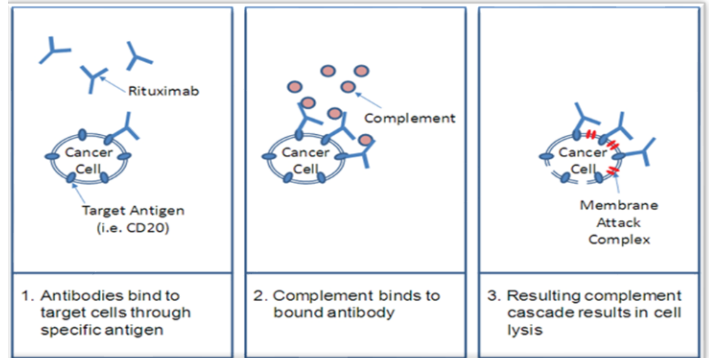
ADCC

In ADCC assay, antibody-bound target cells are cocultured with NK cells or other immune cells to initiate cell killing.



CDC

In CDC assay, antibody-bound target cells are cultured with human or rabbit complement to initiate cell killing.



HOW DOES OUR ASSAY WORK?

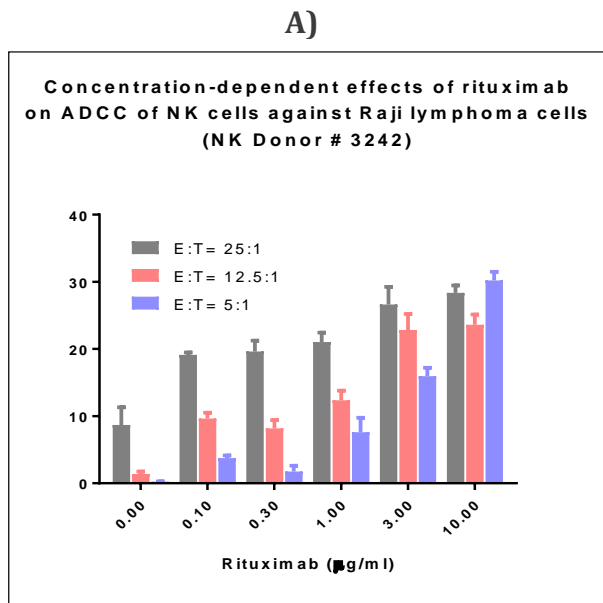
1. PBMC preparation, or of NK cell enrichment (or pretreatment with test molecules if needed)
2. Incubation of target tumor cells with antibody
3. Coculture of Effector and target cells (E:T) for few hours
4. LDH assay to examine the death of tumor cells
5. ADCC calculation:

$$\% \text{ specific lysis} = \frac{(\text{experimental release} - \text{spontaneous release})}{(\text{maximum release} - \text{spontaneous release})} \times 100\%$$

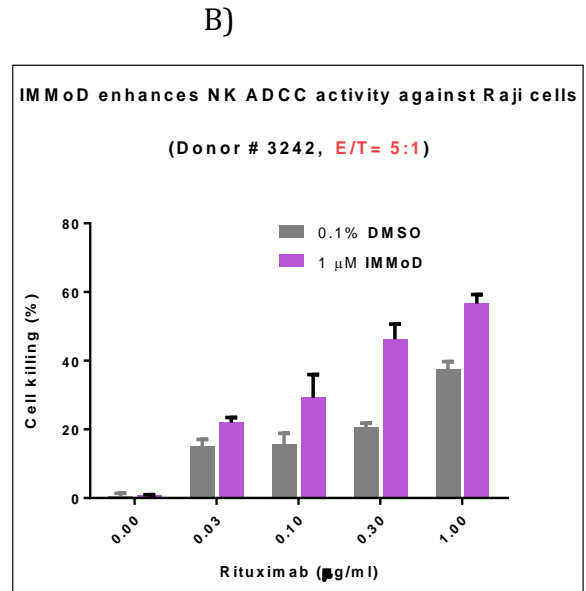


OUR SERVICE FEATURES

- ❖ **High throughput:** 96-well format
- ❖ **Robust and highly reproducible** assay for small to large scale screening
- ❖ **Validated target cells provided:** EGFR, Her2, CD20, SLAMF7, CD38, CTLA 4, ... positive cancer cells available
- ❖ **Both small molecules or large molecules (Abs)** can be assayed
- ❖ **Multiple endpoints:** specific cell lysis, and NK cell activation marker expression (including CD94, NKG2D, NKp30, NKp46, DNAM-1, 2B4, KIR2DL1/S1, intracellular granzyme B, and perforin)
- ❖ **State-of-art platforms:** CytoFLEX S (Flow cytometer with 4 lasers 13 colors) for characterization of effector cells.
- ❖ **Prescreened effector cells** (ADCC low or high) to fit specific projects
- ❖ **Single agents or combinations:** Flexible assay design to fit specific projects
- ❖ Fully validated and quality-controlled
- ❖ Multiple concentrations in triplicate
- ❖ Positive and negative control included
- ❖ Timely data delivery
- ❖ 20+ years of experience: Expert data analysis and interpretation, scientific and technical support
- ❖ **Optional tests:**
 - ✓ Multiple donors of effector cells to address donor-to-donor variability.
 - ✓ Donor PBMC genotype analysis of FcγRIIIa 158V/F polymorphism
 - ✓ FACS binding analysis of antibody to target cells



Example. A) Rituximab induces human NK cell ADCC activity against human Raji lymphoma cells in a concentration-dependent manner (using various E:T ratio).



Example. B) The cell adhesion modulator IMMoD increases human NK cell ADCC activity against Raji lymphoma cells at E:T ratio=5:1.