**ESTROGEN RECEPTOR ALPHA validation file**

**Target:** Estrogen receptor alpha  
**Clone name:** CRET94D  
**Description:** mouse monoclonal  
**Antigen used:** MBP-CRE-ERT recombinant protein  
**Isotype:** IgG1  
**Antigen Species:** human and mouse  
**Localization:** nuclear  
**Positive control:** tonsil  
**Storage Buffer:** Preservative: Sodium Azide. Constituents: Tissue culture supernatant  
**Storage:** Aliquot and store at 4C. Do not freeze

### Characterization

To confirm that CRET94D mAb recognized human Estrogen receptor protein, immunohistochemistry on frozen cytospins preparations of CRE-ERT expressed in Hek293T was performed. Cytospin preparation of E4F1 transfected cells was used as negative control.
Applications

**WB- Western Blotting:** CRET94D mAb is able to detect estrogen receptor protein by WB.

**Dilution:** 1:5

Predicted molecular weight: **66kDa**
Observed molecular weight: **66kDa**

**Lanes**
- Lane 1 Hek-CRE-ERT (20ug) (+)
- Lane 2 HBL100 cell line (100ug) (-)
- Lane 3 MCF7 cell line (100ug) (+)
- Lane 4 Human tonsil (100ug) (-)
- Lane 5 ZR75-1 cell line (human breast carcinoma) (100ug)(+)

Predicted molecular weight: **66kDa**
Observed molecular weight: **66kDa**

**Lanes**
- Lane 1 Mouse uterus wt (100ug)
- Lane 2 Mouse uterus KO Estrogen Receptor Alpha (100ug)
- Lane 3 Mouse uterus KO Estrogen Receptor Beta (100ug)

**IP- Immunoprecipitation:** not done

**IHC-F-Immunohistochemistry (frozen):** not done
**IHC-P-** **MOUSE**-Immunohistochemistry (paraffin):** CRET94D antibody can be used to detect mouse Estrogen receptor protein in paraffin embedded tissues.

**Tissue sample:** mouse ovary (wt, KO beta subunit and KO alpha subunit)

**Dilution:** neat supernatant

**Detection kit used:** OminMap Standard RiboCC No Heat (Ventana)

---

**IHC-P-** **Immuno**histochemistry (paraffin):** CRET94D antibody can be used to detect human estrog**en** receptor protein in paraffin embedded tissue samples.

**Tissue sample:** Human breast cancer

**Dilution:** 1:40

**Antigen retrieval:** 20 min ER2 (Tris-EDTA)

**Antibody incubation:** 30 minutes

**Detection kit used:** Novolink kit (BondMax Leica)
IF-Immunofluorescence: not done

FC-Flow cytometry: not done