

## Product Specification Sheet

### Human Her-2/neu(erbB-2) protein Antibodies (Herceptin biosimilar)

|   |  |                     |
|---|--|---------------------|
| <input type="checkbox"/> <b>Cat #</b> HER35-M | Humanized anti-human Her2/ErbB2/Neu protein IgG (Herceptin Biosimilar) | <b>SIZE:</b> 100 ul |
| <input type="checkbox"/> <b>Cat #</b> HER21-C | Recombinant human Her-2/neu(erbB-2)-Fc protein control for WB          | <b>SIZE:</b> 100 ul |

HER2/neu (also known as ErbB-2, ERBB2) is a protein (protein accession # P04626; 1255 aa, ~185 kda, chromosome 17q21.1) highly expressed in breast cancers. It is a cell membrane surface-bound receptor tyrosine kinase and is normally involved in the signal transduction pathways leading to cell growth and differentiation. **Herceptin**, a fully humanized monoclonal antibody (IgG1k) that neutralized Her2 and used to treat some breast cancer patients. Herceptin is produced in CHO cell culture. It binds to the domain IV of the extracellular segment of the HER2/neu receptor. Humanized antibodies are 'Animal Antibodies' that have been engineered by recombinant DNA-technology to reduce the overall content of the animal-portion of IgG so as to increase acceptance by humans or minimize 'rejection'. The portion of the mouse IgG that remains in the 'humanized IgG' or even the human portion may be recognized as foreign by humans and may result into the generation of "Human Anti-Drug Antibodies (HADA or Human Anti-Mouse Antibodies or HAMA). Some patients receiving Herceptin developed some form of anti-Herceptin (HADA) response. The presence of anti-Drug antibody (e.g., Human Anti-Herceptin IgG) may limit the long-term usage the humanized antibody (Herceptin). The prevalence of anti-drug antibodies are highly dependent upon the nature of sample, duration of therapy, and sensitivity of the assay. Therefore it is necessary to monitor the presence of anti-Herceptin antibody levels in patients receiving long-term Herceptin immunotherapy

**Protein name** Receptor tyrosine-protein kinase erbB-2 [Precursor]  
Synonyms EC 2.7.10.1, p185erbB2, C-erbB-2, NEU proto-oncogene, Tyrosine kinase-type cell surface receptor HER2, MLN 19, CD340 antigen HER2, NEU, NGL  
**Gene name** Name: ERBB2

#### Sources of antigen and antibodies

|                        |  |
|------------------------|--|
| <b>Antigen</b>         | Recombinant purified human ErbB2 extracellular domain 23-652 aa  |
| <b>Ab Host/type</b>    | Humanized monoclonal IgG1k expressed in CHO cells, Aff pure IgG ( <b>cat # HER35-M</b> ) supplied in PBS, pH 7.4, 0.1% BSA and 0.05% azide ~50-100 ug/100 ul |
| <b>2-ab</b>            | <b>Goat Anti-Human IgG-HRP cat # 10120 (AP, biotin, FITC conjugates also available) or 2007-G1-1 (human IgG1, myeloma)</b>                                   |
| <b>-ve control IgG</b> | Cat # 20007-1, Human (non-immune) Serum IgG, purified, suitable for ELISA, Western, IHC as -ve control   |

Human ErbB2.neu protein (23-652aa) was expressed in NSO cells as human IgG-Fc (100-330aa) his-tag fusion protein and purified (>95%). Recombinant ErbB2-Fc chimeric protein (#HER21-C) is ~130 kda under reducing conditions. For Western blot +ve control (**Cat # HER21-C**) is supplied in SDS-PAGE sample buffer (reduced). Load 10 ul/lane of **HER21-C** for good visibility with antibody Cat # **HER35-M**. Store at -20oC in suitable size aliquots. SDS may crystallize in cold conditions. It should redissolve by warming before taking it from the stock. It should be heated once prior to loading on gels. If the product has been stored for several

weeks, then it may be preferable to add 5 ul of fresh 2x sample buffer per 10 ul of the **HER21-C** solution prior to heating and loading on gels. This preparation is not biologically active. It is not suitable for ELISA or other applications where native protein is required. Do not freeze, thaw, or heat repeatedly

#### Form & Storage of Peptide and Antibodies

##### Affinity pure IgG

solution  lyophilized powder in PBS pH 7.4 +0.1% BSA, 0.05% azide  
**Reconstitute** powder in PBS in water

##### Storage

**Short-term:** Liquid, unopened, undiluted vials for less than a week at 4oC and powder up to several months at 4oC.

**Long-term:** at -20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20oC or below.

**Shipping:** 4oC for solutions and room temp for lyophilized items.

#### Recommended Usage

**Western blot:** Optimal dilution must be determined by user. We suggest initial testing of antibody at 1:200-1:1000 using ECL

**ELISA** (1:10-50K; 10-100 ng of control protein/well).

**Immunohistochemistry:** We suggest testing of aff pure IgG at 1:100-1:500 ug/ml using paraffin embedded sections.

**Flow cytometry** – use 5-10 ul per sample (~1-3 x10<sup>5</sup> cells) in ~200 ul.

#### Specificity and crossreactivity

Antibody # HER31-M is specific for human ErbB2 with no reaction with human EGFR, ErbB3 or ErbB4. Other species not tested. Human recombinant ErbB2 proteins (#HER21-R-10 or HER23-R10) can be used as positive control.

**General References:** (1) Yamamoto T (1986) Nature 319, 230-234; Semba K (1985) PNAS 82, 6497-6501; Akiyama T (1986) Science 232, 1644-1646; Bargmann CI (1986) Nature 319, 226-230; Coussens L (1985) Science 230, 1132-1139; Doherty JK (1999) PNAS 96, 10689-10874

*This product is for In vitro research use only.*

#### Related items

|                    |  |
|--------------------|--|
| 200-510-HLG        | Herceptin/Trastuzumab ELISA Kit for human, 96 tests    |
| 200-520-HAG        | Human Anti-Herceptin/Trastuzumab Antibody (ADA)        |
| ELISA Kit 96 tests |  |
| 200-525-HAG        | Mouse Anti-Herceptin/Trastuzumab Antibody (ADA)        |
| ELISA Kit 96 tests |  |
| 200-530-HER        | Human Her2/neu/ErbB2/CD340 protein ELISA kit, 96 tests |
| HER35-M            | 141121A  |

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