# **Product Information**

# Interleukin-6 (human) EIA Standard

Item No. 483364

### **IMPORTANT**

Water used to prepare all EIA reagents and buffers must be deionized and free of trace organic contaminants ('UltraPure'). Use activated carbon filter cartridges or other organic scavengers. Glass distilled water (even if double distilled), HPLC-grade water, and sterile water (for injections) are not adequate for EIA. UltraPure water may also be purchased (Item No. 400000).

### **Laboratory Procedures**

The Interleukin-6 (IL-6) (human) EIA Standard you have purchased contains IL-6 (human) EIA Standard as a lyophilized powder. For long term storage, we suggest that the IL-6 (human) EIA Standard be stored as supplied at -20°C; it will be stable for at least one year. When ready to use, reconstitute the standard with 2 ml EIA Buffer (see buffer preparation instructions below). The concentration of this solution (the bulk standard) will be 5 ng/ml.

To prepare the standard for use in EIA: Obtain eight clean test tubes and number them #1 through #8. Aliquot 950 µl EIA Buffer to tube #1 and 500 μl EIA Buffer to tubes #2-8. Transfer 50 μl of the bulk standard (5 ng/ml) to tube #1 and mix thoroughly. Serially dilute the standard by removing 500 µl from tube #1 and placing in tube #2; mix thoroughly. Next, remove 500 µl from tube #2 and place it into tube #3; mix thoroughly. Repeat this process for tubes #4-7. Do not add any IL-6 to tube #8. This tube is the zero point vial, the lowest point on the standard curve. We recommend that you store these diluted standards for no more than 24 hours.

### **Buffer Preparation**

### 1. Phosphate Buffer

Prepare a 1.0 M phosphate buffer solution by combining 133 g K<sub>2</sub>HPO<sub>4</sub> and 32.15 g KH<sub>2</sub>PO<sub>4</sub> and diluting to a total volume of 1.0 liter with UltraPure water. The pH of this solution will be 7.4.

Combine 100 ml of the phosphate buffer prepared above with 100 mg sodium azide, 23.4 g sodium chloride, 370 mg tetrasodium EDTA, and 1 g bovine serum albumin (Sigma A7030 or equivalent). Stir at room temperature until completely dissolved and dilute to a total volume of 1.0 liter with UltraPure water. Add 0.1 ml Polysorbate 20 and mix thoroughly. This EIA Buffer may also be purchased from Cayman as a 10X concentrate buffer (Item No. 400060).

### 3. Wash Buffer

Combine 10 ml of the phosphate buffer prepared above with 0.5 ml Polysorbate 20. Bring to a final volume of 1.0 liter with UltraPure water. This Wash Buffer may also be purchased from Cayman as a 400X concentrate buffer (Item No. 400062).

### Suggested Assay Protocol

This standard has been tested and formulated to work exclusively with ACE™ reagents. This standard may not perform as described if used with other assay reagents or protocols. NOTE: This is an abbreviated protocol. If you are not familiar with this assay, please contact us for a complete protocol.

- 1. Wash plate once with Wash Buffer.
- 2. Add 100 µl of Standard or sample to the appropriate wells.
- 3. Add 100 µl Conjugate (Item No. 483360) to all wells except Blk.
- 4. Incubate for 90 minutes at 4°C.
- 5. Wash plate five times with Wash Buffer.
- 6. Add 200 µl Ellman's Reagent to each well.
- 7. Develop until S1 wells are visibly yellow.
- 8. Read absorbance between 405-420 nm.
- Dilute samples may require longer development times.

Blk-Blank; S1-S8-Standards; 1-7-Samples

## Related Product

Interleukin-6 (human) EIA Kit - Item No. 583361

WARNING: THIS PRODUCT IS FOR LABORATORY RESEARCH ONLY: NOT FOR ADMINISTRATION TO HUMANS. NOT FOR HUMAN OR VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

### MATERIAL SAFETY DATA

This material should be considered hazardous until information to the contrary becomes available. Do not ingest, swallow, or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. This information contains some, but not all, of the information required for the safe and proper use of this material. Before use, the user must review the complete Material Safety Data Sheet, which has been sent via email to your institution.

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