



Human Total Tau Antibody Bead Kit

INFORMATION SHEET

Catalog #: LHB0041 Description: Human Total Tau Lot:* S053114

*Note: A letter at the end of the lot number signifies an additional packaging of this same lot.

Intended Use

This reagent set comprises the analyte specific components for the measurement of human Tau in CSF and tissue culture media. These reagents are also suitable for the measurement of Tau in cell lysates. The assay may be run alone or in combination with other Neuroscience Antibody Bead Kits from Invitrogen. Buffer reagents needed to complete the reaction are sold separately under Catalog # LNB0001. These reagents are intended for use in the **Luminex® 100™ or 200™ System** only. **This kit is configured for research use only and is not to be used in diagnostic procedures.**

Reagents Provided

1. Antibody Bead Concentrate (10x):

Catalog #: LM134 Description: Ms x Hu Total Tau Beads Lot: S070611 Size: 0.25 mL-100 tests

Bead Region: 69

Form: 0.25 mL 10x bead concentrate solution in storage buffer. Contains 7.5 mM sodium azide as preservative.

Storage: **Light-sensitive material.** Store at 2 - 8°C until the expiration date indicated on the kit.

Note: Beads for Human Total Tau and Human Tau [pT181] or Tau [pS199] cannot be mixed. Each of these analytes must be measured separately.

2. Detector Antibody Concentrate (10x):

Catalog #: DN134 Description: Rbt x Hu Total Tau Lot: S070612 Size: 0.5 mL-100 tests

Form: 0.5 mL of a 10x stock of Detector Antibody Concentrate in Detector Antibody Diluent. Contains 3.3 mM thymol as a preservative. Concentration of antibody is matched to this lot of beads. Do not mix lots of Coated Beads and Detector Antibody.

Storage: Store at 2 - 8°C until the expiration date indicated on the kit.

3. Hu Total Tau Standard (2 vials):

Catalog #: SM046 Description: Rec. Hu Total Tau Lot: S071409 Size: Single use

Form: Lyophilized. This human Tau standard was calibrated against a highly purified recombinant human Tau-441 protein expressed in *E. coli* using the Invitrogen Human Tau ELISA kit (cat. # KHB0041/KHB0042).

Storage: Store at 2 - 8°C. Use within 1 hour after reconstitution. Discard remainder immediately after use.

Concentration of Reconstituted Standard: 5,000 pg/mL

Reconstitution: When working with CSF samples, reconstitute with 0.7 mL Assay Diluent. When working with tissue culture medium samples, reconstitute with a 0.7 mL mixture of 50% Assay Diluent and 50% tissue culture medium. Allow standard to rehydrate for approximately 10 minutes before further dilution.

Recommendations for Combining with Other Standards: When combining with one other standard, reconstitute to 10,000 pg/mL. Combine equal volumes of the two standards. When combining with two other standards, reconstitute to 15,000 pg/mL, and combine equal volumes of the three standards. Up to four standards may be combined.

Recommended Starting Concentrations for Standard Curve: The standard curve should start at 5,000 pg/mL. Make serial 1:3 dilutions in Assay Diluent (CSF and cell lysate samples) or mixture of 50% Assay Diluent and 50% tissue culture medium (tissue culture medium samples). Each well designated for standard receives 50 µL of 1x Detector Antibody followed by 50 µL standard. Each well designated for sample receives 50 µL of 1x Detector Antibody followed by 25 µL Assay Diluent and 25 µL sample. See the Product Insert included in the Neuroscience Buffer Reagent Kit for further information.

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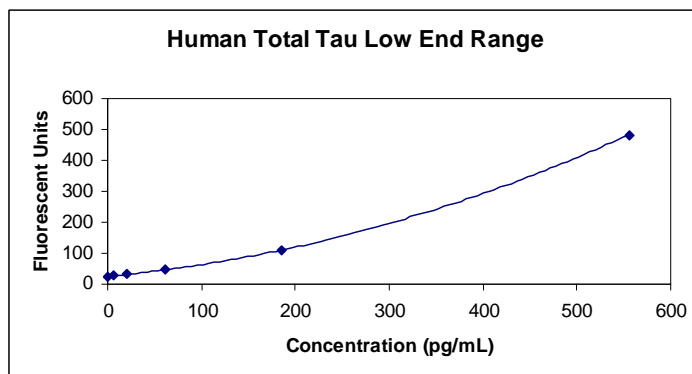
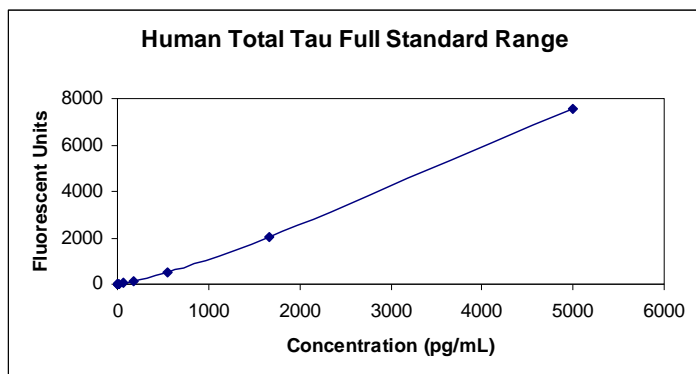
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Performance Characteristics

Analytical Sensitivity: The minimum detectable dose of Human Total Tau is <22 pg/mL. This was determined by adding two standard deviations to the mean MFI obtained when the zero standard was assayed 32 times.



Typical Standard Curve

Species Reactivity: This Human Total Tau Antibody Bead Kit is specific for human Tau. It does not cross-react with rat or mouse. Other species were not tested.

Specificity: Buffered solutions of a panel of substances with concentrations ranging between 1 and 30 ng/mL were assayed with the Invitrogen Human Total Tau Antibody Bead Kit. The following substances were tested and found to have no cross-reactivity: human A β 40, A β 42, IL-1 α , IL-1 β , IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IFN- α , IFN- γ , GM-CSF, TNF- α , IL-12, IL-13, IL-15, IL-17, MCP-1, MIP-1 α , MIP-1 β , Eotaxin, RANTES, IP-10, MIG.

Precision:

	Intra-assay (n=16)	Inter-assay (n=32)
Mean (pg/mL)	465.43	464.66
SD	16.62	20.57
%CV	3.57%	4.43%

Linearity: CSF (diluted 1:2 in Assay Diluent) and tissue culture medium (DMEM containing 10% FBS) were spiked with human Total Tau and serially diluted in Assay Diluent or 50/50 Assay Diluent/tissue culture medium over the range of the assay. Linear regression analysis of samples versus the expected concentration yielded a correlation coefficient of 1.0 for CSF and a correlation coefficient of 1.0 for tissue culture medium.

Recovery: CSF diluted 1:2 in Assay Diluent averaged 94.3%. Tissue culture medium containing 10% FBS averaged 97.3%.

Correlation to ELISA: A correlation coefficient of 0.85 was calculated when values for natural Tau from human CSF, obtained with the Hu Total Tau Antibody Bead Kit, were compared with the Invitrogen ELISA for Hu Total Tau (cat. # KHB0041/KHB0042). With natural sample, Hu Total Tau Antibody Bead Kit (pg/mL) \times 0.32 = Hu Total Tau ELISA (pg/mL). Correlation of results obtained with the Hu Tau Antibody Bead Kit to one's own system should be determined to arrive at an appropriate multiplication factor.

Cell Lysate Sample Preparation:

In addition to CSF and tissue culture medium samples, this kit has been used to detect Hu Tau from lysates made from SH-SY5Y cells, prepared in Cell Lysis Buffer (Invitrogen Cat. # FNN0011: 10 mM Tris, pH 7.4, 100 mM NaCl, 1 mM EDTA, 1 mM EGTA, 1 mM NaF, 20 mM Na₄P₂O₇, 2 mM Na₃VO₄, 1% Triton X-100, 10% glycerol, 0.1% SDS, 0.5% deoxycholate, 1 mM PMSF [added fresh from 0.1 M stock in DMSO], 1% Triton X-100, 10% glycerol, 0.1% SDS, 0.5% deoxycholate, 1 mM PMSF [added fresh from 0.1 M stock in DMSO], and diluted at least ten-fold in Assay Diluent. To produce a lysate, incubate cells with cell lysis buffer (1-2 \times 10⁸ cells/mL is recommended) on ice for 30 minutes, vortexing at 10 minute intervals, then clarify the lysate by centrifugation at 13,000 RPM for 10 minutes. Cell lysates may be stored at -80°C for up to three months with one freeze/thaw cycle. Optimization of cell stimulation and cell lysis procedures may be required for each specific application.

By purchasing this Kit, which contains fluorescently labeled microsphere beads authorized by Luminex® Corporation ("Luminex®"), you, the customer, acquire the right under Luminex's patent rights to use this Kit or any portion of this Kit, including without limitation the microsphere beads contained herein, only with Luminex's laser based fluorescent analytical test instrumentation marketed under the name Luminex® 100™ or 200™. This product is covered by one or more of the following U.S. patents: 6,046,807.

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