

Instructions for Generating a Calibration Curve with Pre-Formed Oligomeric A β Peptides

This sheet provides detailed instructions for generating a calibration curve with PE-1750-1000, for use in the Biosensis Oligomeric A β ELISA Kit (BEK-2215). Note that PE-1750-1000 is supplied as 2 x 500 ng vials.

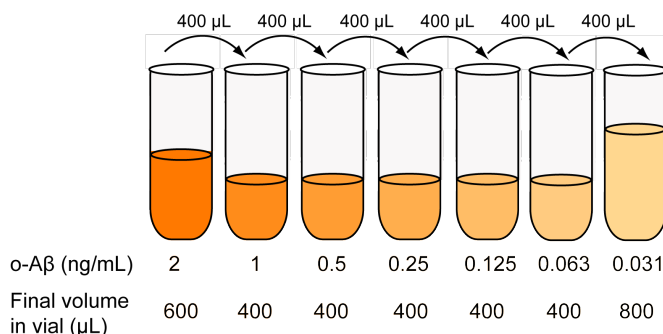
1. Equilibrate one 500 ng vial to room temperature; the lyophilized material may appear wet, which is due to the hygroscopic nature of the stabilizing buffer components.
2. Add 1 mL of Assay Diluent B to the vial.
3. Vortex and bring the lyophilized material into solution.
4. The concentration of oligomeric A β is now 500 ng/mL, label the tube accordingly.

IMPORTANT: While the concentration of monomeric A β peptide used to form the oligomeric complexes is accurately determined, the precise formation, size and number of oligomers cannot be quantified by any known method. We recommend results should be presented as "A β peptide equivalents; ng/mL (Arbitrary Units)".

5. Prepare a standard curve within 2.0 – 0.03 ng/mL, with 1:2 serial dilution. The volumes used for the dilution series depends on the number of repeats per oligomeric A β peptide standard. For triplicate measurement (100 μ L per well) of each oligomeric A β standard (o-A β) concentration, you may want to follow this procedure:

- I. Label 7 tubes with 2 ng/mL, 1 ng/mL, 0.5 ng/mL, 0.25 ng/mL, 0.125 ng/mL, 0.0625 ng/mL and 0.0313 ng/mL, respectively
- II. Aliquot 400 μ L of the assay diluent into each tube except the tube labeled "2 ng/mL"
- III. Dilute the 500 ng/mL o-A β stock solution 250-fold (4 μ L of o-A β stock solution added to 996 μ L assay diluent); the concentration of o-A β is 2 ng/mL
- IV. Take 400 μ L from the "2 ng/mL" tube and transfer to the tube labeled as "1 ng/mL"
- V. Repeat step IV. for each consecutive concentration until the last tube "0.0313 ng/mL" is prepared

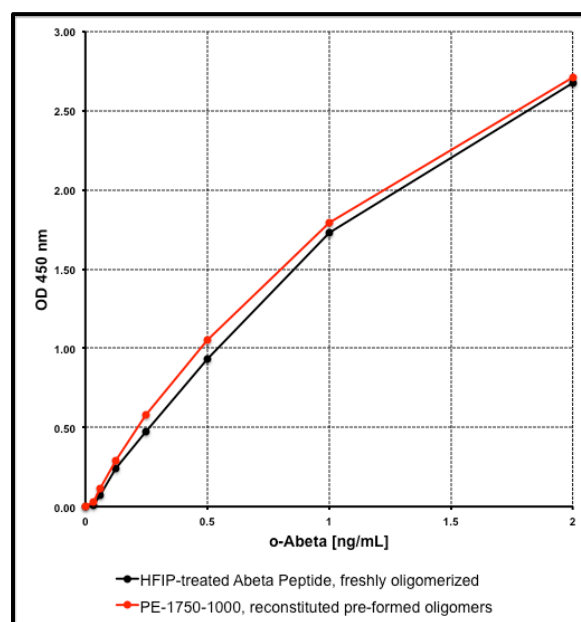
Note: Pipet up- and down and vortex to mix; to avoid foaming, use only a very brief vortex.



6. The calibration curve standards are now ready to be loaded into the microplate wells and should be used within 1 hour; refer to the BEK-2215 kit insert for the assay procedure and instructions on calculating results.
7. The reconstituted 500 ng/mL stock solution may be stored at 2-8° for up to 2 days, and should not be frozen for best results.

Typical Data

Oligomeric A β peptide calibration curves were generated with the HFIP-treated A β peptide supplied in the Oligomeric A β ELISA kit (BEK-2215), also available as PE-1749-50, and compared against pre-formed, lyophilized oligomeric A β peptides (PE-1750-1000).



This graph demonstrates the stability of pre-formed oligomeric A β peptides and the usefulness of PE-1750-1000 as calibration curve standard for BEK-2215 and other oligomeric A β ELISA kits.