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Adlyfe at ICAD 2008

Adlyfe is please to be presenting two posters at ICAD 2008 and the satellite meetings. Please join us for these presentations.

Intranasal Administration Of Pronucleon Peptides For In Vivo Imaging Of Beta Amyloid In Alzheimer'S Disease Nyborg A, Wegrzyn R, Moll J, Nelsen C, Rudolph A, Shivaprasad S, Schieber G, and Duan R
Saturday, July 26, 2008, 2:00 pm - 2:30 pm and Sunday Jul 27, 2008 12:30 pm - 3:00 pm

Background: One of the pathological features of Alzheimer's disease (AD) is the conversion of a normal soluble protein into insoluble beta amyloid aggregates. Aggregation and deposition of amyloid beta in vivo may precede clinical symptoms by many years. In vivo imaging of amyloid plaques would be beneficial for the following reasons: 1) A potential preclinical diagnostic of AD, 2) Determining the efficacy of anti-amyloid therapies, 3) Monitoring effects of AD therapeutics in clinical trials.

We previously reported a novel Pronucleon™ peptide technology that specifically measures beta amyloid. The Pronucleon™ peptide undergoes a sequence-specific conformational change in the presence of the amyloid beta aggregates which can be used to measure beta amyloid in CSF and blood.

Objective: The primary objective is to develop an easily administered imaging agent that will provide an early measure of plaque or amyloid aggregate burden associated with AD.

Methods: The Pronucleon™ peptide was first examined as an ex vivo stain for amyloid plaques. We have since administered (intranasally) Pronucleon peptides in hAPP transgenic mice that develop extensive plaque pathology. Sections from these mice were subjected to ex vivo analyses including fluorescence, Thioflavin S staining, anti-amyloid staining and anti-GFAP staining. The Pronucleon™ peptides contain fluorescent tags that are sensitive to beta amyloid conformational states and results in the ability to image beta amyloid aggregates with a fluorescent microscope.

Results and Conclusions: Ex vivo administration of the Pronucleon™ peptide to tissue section from transgenic mice demonstrates that the peptide labels plaque like material. These, ex vivo, plaque-like images co merge with Thioflavin S staining. Intranasal, in vivo, administration of the fluorescent Pronucleon™ peptide labels plaques in the hippocampus and cortex of transgenic mice. Fluorescent structures had plaque like morphology and co merged with anti-amyloid antibody or Thioflavin S staining. A significant positive correlation was observed between Pronucleon™ peptide staining and Thioflavin S staining. These data suggest that the Pronucleon™ peptide can efficiently cross the blood brain barrier, label plaques, and may be an effective tool for in vivo imaging.



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Specific Conformational Detection Of Beta-sheet Rich Amyloid Beta Proteins Using Conformationally Dynamic Pronucleon™ Peptides

Schieber G, Duan R, Moll J, Nelsen C, Nyborg A, Rudolph A, Shivaprasad S, Soukharev S, and Wegrzyn R
Tuesday Jul 29, 2008 12:30 PM - 3:00 PM

Background: The pathogenesis of many protein disorders is characterized by the conversion of a constitutive soluble protein into a less soluble beta-sheet rich conformation. In the case of Alzheimer's disease (AD), assembly of amyloid beta (Abeta) protein into neurotoxic oligomers is reported to be a likely cause of disease. Diagnostic tools that can distinguish between different conformations of the Abeta protein biomarker would be a unique and valuable contribution to the field.

We have previously reported the use of small conformationally dynamic peptides (Pronucleon™ peptides), that enable the detection of beta sheet amyloid proteins both in prion disease and in AD. These fluorescently-labeled peptides undergo a sequence-specific conformational rearrangement in the presence of the beta-sheet structure of Abeta aggregates. The rearrangement results in a change in the fluorescence profile that can be monitored using standard laboratory instrumentation.

Objective: Toward the overall goal of creating an in vitro diagnostic test that directly measures peptide molecular structures implicated in AD, we tested a system of specifically defined amyloid aggregates and spiked these into clinical matrices including cerebrospinal fluid (CSF). We addressed issues of assay design, dose-response, and target availability in this biological matrix.

Methods: We have created controlled populations of soluble Abeta aggregates prepared from purchased recombinant Abeta. Assay studies were performed using Adlyfe's MPD ("misfolded" protein detection) assay which combines sample and Pronucleon peptide in a 96-well plate and follows the fluorescence transition associated with conformational changes of the Pronucleon peptide in the presence of Abeta aggregates.

Results: In previous studies, we demonstrated reactivity with insoluble Abeta "fiber" aggregate. In this work, we extend our assay to soluble Abeta aggregates. We have evaluated variations in the sequence of the Pronucleon peptides, considering structural regions of Abeta folding and aggregation. We demonstrate detection of synthetic aggregate forms and present data on the translation of assay conditions to clinical samples representing cohorts from subjects with AD or mild cognitive impairment versus age-matched controls.

Conclusion: The Pronucleon peptide technology demonstrates specific recognition of beta-sheet rich Abeta aggregates and the potential to create a diagnostic test for the peptide biomarkers of AD.