

# Oligo-Abeta-pE3

Cat.No. 218 511; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

## Data Sheet

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Reconstitution/       100 µg purified IgG, lyophilized. Albumin and azide were added for stabilization.         Storage       For reconstitution add 100 µl µ <sub>2</sub> 0 to get a 1mg/ml solution in PBS. Then aliquot and store at -20°C to -80°C until use.         Antibodies should be stored at +4°C when still lyophilized. Do not freeze!       For detailed information, see back of the data sheet.         Applications       WB: 1: 500 (see remarks)       IP: not tested yet         IIC: not tested yet       IIC: not tested yet         HC: 1: 100       ELISA: external data         Important note       The mouse monoclonal antibody clone 9D5, Cat. No. 218 511, is patented (patent acquires rights to use this product for research purposes only. Any diagnostic and therapeutic in vitro or in vivo use is explicitly excluded.         Clone       9D5         Subtype       IgG2b (k light chain)         Immunogen       Synthetic peptide corresponding to AA 3 to 38 from human Oligo-Abeta-pE3 (UniProt Id: POS067)         Reactivity       Recognizes specific oligomeric structures formed preferentially by Abeta-pE3.         Remarks       WB: We recommend the Invitrogen NativePAGE system in combination with PVDF blotting membranes. Boil membrane after blotting for 3min. This antibody has only been validated with synthetic peptides and not with complex protein samples.         Peptide preparation:       Synthetic Abeta peptides were monomerized in 70 % formic acid, and the solvent was evaporated in a speed-vac immediately. Prior to each experiment, petides were disolved in 0.3 %		
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for usersapplication PCT/EP2011/002739). By purchasing this antibody the customer acquires rights to use this product for research purposes only. Any diagnostic and therapeutic in vitro or in vivo use is explicitly excluded.Clone9D5SubtypeIgG2b (κ light chain)ImmunogenSynthetic peptide corresponding to AA 3 to 38 from human Oligo-Abeta-pE3 (UniProt Id: P05067)ReactivityReacts with: human (P05067), mouse (P12023). Other species not tested yet.SpecificityRecognizes specific oligomeric structures formed preferentially by Abeta-pE3.RemarksWB: We recommend the Invitrogen NativePAGE system in combination with PVDF blotting membranes. Boil membrane after blotting for 3min. This antibody has only been validated with synthetic peptides and not with 	Applications	IP: not tested yet ICC: not tested yet IHC: 1 : 100 IHC-P: 1 : 100
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Amyloid deposits, also called plaques, of Alzheimer's patients consist of several protein components like the amyloid **beta**-peptides (**Abeta**, **Aβ**) 1-40/42 and additional C- and N-terminally truncated and modified fragments. Very abundant are the isoaspartate (isoAsp)-Abeta and **p**yro**glu**tamyl (**pGlu**)-Abeta peptides. The latter are formed by cyclization of the N-terminal glutamate at position 3 or 11 catalyzed by glutaminyl cyclase (QC) resulting in very amyloidogenic and neurotxic variants of Abeta; **Abeta-pE3** and Abeta pE11.

In contrast to extracellular plaques that do not perfectly correlate with Alzheimer´s disease intraneuronal Abeta accumulation and vascular Abeta deposits have gained more and more evidence to be among the crucial factors responsible for progressive neuron loss.

### Selected References for 218 511

Identification of low molecular weight pyroglutamate A{beta} oligomers in Alzheimer disease: a novel tool for therapy and diagnosis.

Wirths O, Erck C, Martens H, Harmeier A, Geumann C, Jawhar S, Kumar S, Multhaup G, Walter J, Ingelsson M, Degerman-Gunnarsson M, et al.

The Journal of biological chemistry (2010) 28553: 41517-24. . WB, IHC, ELISA

Focusing the amyloid cascade hypothesis on N-truncated Abeta peptides as drug targets against Alzheimer's disease. Bayer TA, Wirths O

Acta neuropathologica (2014) 1276: 787-801. . IHC-P; tested species: human

I716F A $\beta$ PP mutation associates with the deposition of oligomeric pyroglutamate amyloid- $\beta$  and  $\alpha$ -synucleinopathy with Lewy bodies.

Sieczkowski E, Milenkovic I, Venkataramani V, Giera R, Ströbel T, Höftberger R, Liberski PP, Auff E, Wirths O, Bayer TA, Kovacs GG, et al.

Journal of Alzheimer's disease : JAD (2015) 441: 103-14. . IHC; tested species: human

Oxidative Stress during the Progression of  $\beta$ -Amyloid Pathology in the Neocortex of the Tg2576 Mouse Model of Alzheimer's Disease.

Porcellotti S, Fanelli F, Fracassi A, Sepe S, Cecconi F, Bernardi C, Cimini A, Cerù MP, Moreno S Oxidative medicine and cellular longevity (2015) 2015: 967203. . **IHC** 

Oligomeric pyroglutamate amyloid-β is present in microglia and a subfraction of vessels in patients with Alzheimer's disease: implications for immunotherapy. Wirths O, Hillmann A, Pradier L, Härtig W, Bayer TA Journal of Alzheimer's disease : JAD (2013) 354: 741-9. . **IHC** 

Antibody 9D5 recognizes oligomeric pyroglutamate amyloid-β in a fraction of amyloid-β deposits in Alzheimer's disease without cross-reactivity with other protein aggregates. Venkataramani V, Wirths O, Budka H, Härtig W, Kovacs GG, Bayer TA Journal of Alzheimer's disease : JAD (2012) 292: 361-71. . **IHC** 

Intraneuronal A $\beta$  as a trigger for neuron loss: can this be translated into human pathology? Bayer TA, Wirths O Biochemical Society transactions (2011) 394: 857-61. .

### **Selected General References**

Pyroglutamate-Aβ 3 and 11 colocalize in amyloid plaques in Alzheimer's disease cerebral cortex with pyroglutamate-Aβ 11 forming the central core. Sullivan CP et al. Neurosci. Lett. (2011) PubMed:22001577

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/218511</u> or scan the QR-code.



# FAQ - How should I store my antibody?

### **Shipping Conditions**

• All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freezedried) and are stable in this form without loss of quality at ambient temperatures for several weeks.

## Storage of Sealed Vials after Delivery

- Unlabeled and biotin-labeled antibodies and control proteins should be stored at 4°C before reconstitution. They must not be stored in the freezer when still lyophilized! Temperatures below zero may cause loss of performance.
- Fluorescence-labeled antibodies should be reconstituted immediately upon receipt. Long term storage (several months) may lead to aggregation.
- **Control peptides** should be kept at -20°C before reconstitution.

# Long Term Storage after Reconstitution (General Considerations)

- The storage freezer must not be of the frost-free variety ("no-frost freezer"). This cycle between freezing and thawing (to reduce frost-build-up), which is exactly what should be avoided. For the same reason, antibody vials should be placed in an area of the freezer that has minimal temperature fluctuations, for instance towards the back rather than on a door shelf.
- Aliquot the antibody and store frozen (-20°C to -80°C). Avoid very small aliquots (below 20 μl) and use the smallest storage vial or tube possible. The smaller the aliquot, the more the stock concentration is affected by evaporation and adsorption of the antibody to the surface of the storage vial or tube. Adsorption of the antibody to the surface leads to a substantial loss of activity.
- The addition of glycerol to a final concentration of 50% lowers the freezing point of your stock and keeps your antibody at -20°C in liquid state. This efficiently avoids freeze and thaw cycles.

## **Product Specific Hints for Storage**

#### Control proteins / peptides

• Store at -20°C to -80°C.

#### **Monoclonal Antibodies**

- Ascites and hybridoma supernatant should be stored at -20°C up to -80°C. Prolonged storage at 4°C is not recommended! Unlike serum, ascites may contain proteases that will degrade the antibodies.
- **Purified IgG** should be stored at -20°C up to -80°C. Adding a carrier protein like BSA will increase long term stability. Many of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

#### **Polyclonal Antibodies**

- **Crude antisera**: With anti-microbials added, they may be stored at 4°C. However, frozen storage (-20°C up to -80°C) is preferable.
- Affinity purified antibodies: Less robust than antisera. Storage at -20°C up to -80°C is recommended. Adding a carrier protein like BSA will increase long term stability. Most of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

#### **Fluorescence-labeled Antibodies**

• Store as a liquid with 1 : 1 (v/v) glycerol at -20°C. Protect these antibodies from light exposure.

# Avoid repeated freeze-thaw cycles for all antibodies!

# FAQ - How should I reconstitute my antibody?

### Reconstitution

- All our purified antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add the amount of deionized water given in the respective datasheet. If higher volumes are preferred, add water as mentioned above and then the desired amount of PBS and a stabilizing carrier protein (e.g. BSA) to a final concentration of 2%. Some of our antibodies already contain albumin. Take this into account when adding more carrier protein. For complete reconstitution, carefully remove the lid. After adding water, briefly vortex the solution. You can spin down the liquid by placing the vial into a 50 ml centrifugation tube filled with paper.
- If desired, add small amounts of azide or thimerosal to prevent microbial growth. This is especially recommended if you want to keep an aliquot a 4°C.
- After reconstitution of fluorescence-labeled antibodies, add 1 : 1 (v/v) glycerol to a final concentration of 50%. This lowers the freezing point of your stock and keeps your antibody in liquid state at -20°C.
- Glycerol may also be added to unlabeled primary antibodies. It is a suitable way to avoid freezethaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.