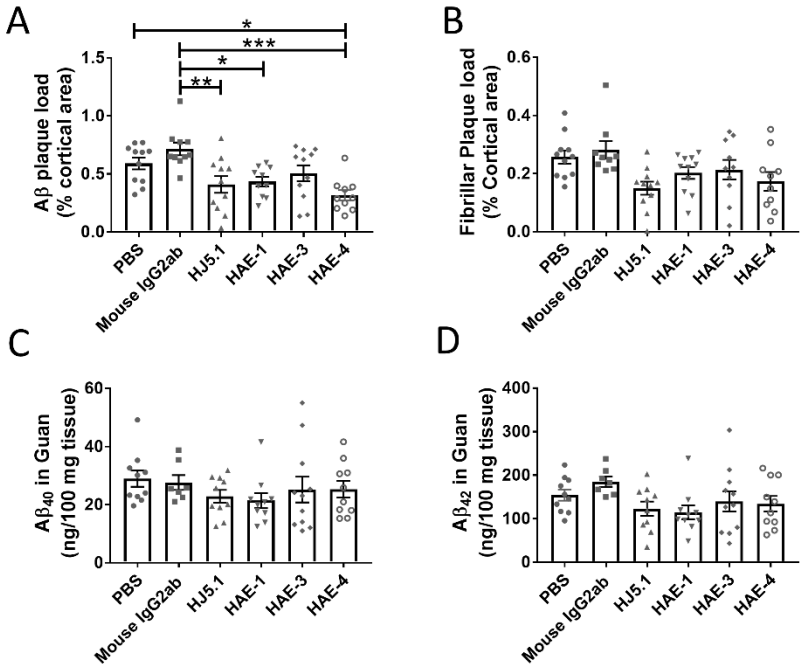
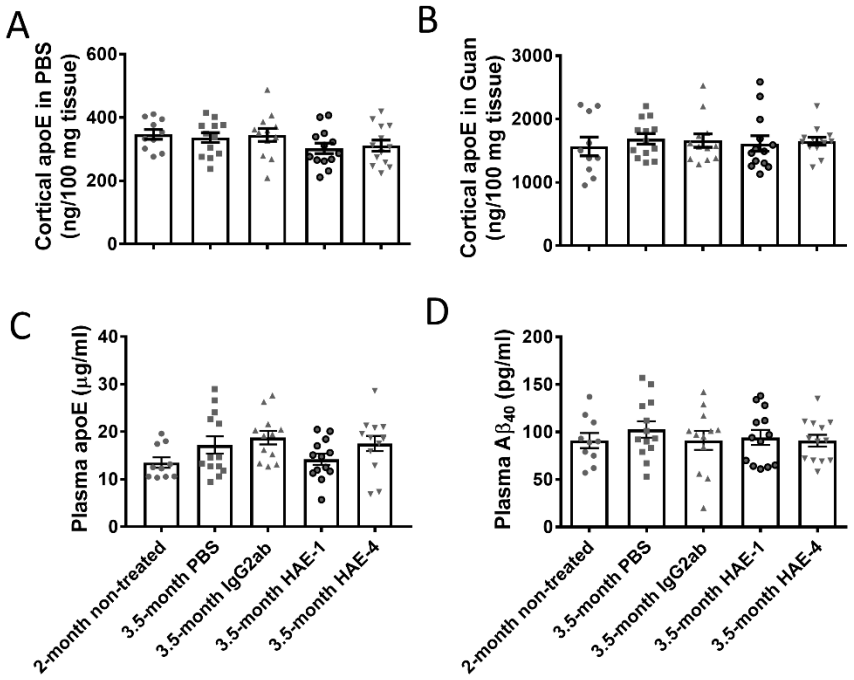
Figure S1



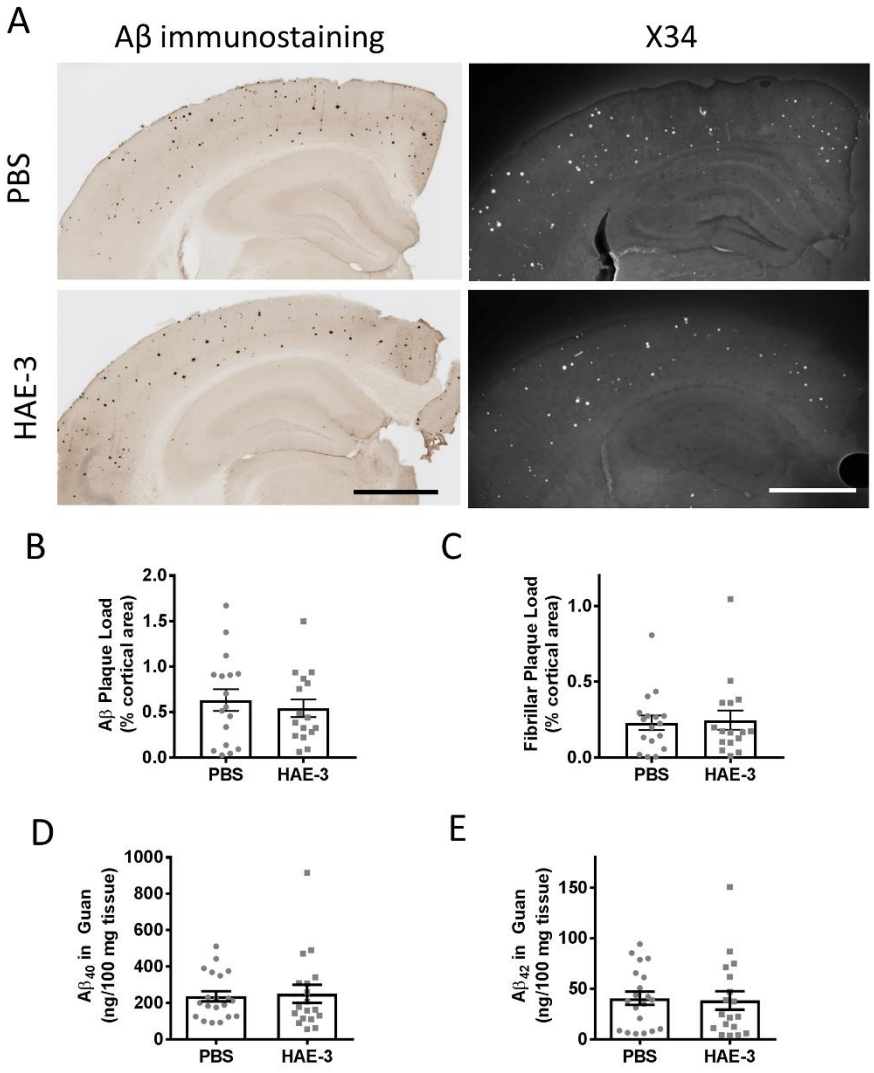
Supplemental Figure 1. Effects of ICV administered anti-apoE antibodies on amyloid pathology in APPPS1-21/APOE4 mice. At the age of 2 months, mice were implanted with a guide cannula connected to a subcutaneous osmotic pump and the antibodies were infused at 0.3 µg/hour for 6 weeks (n=10-11/group, all females) into the lateral ventricle. (A) Aβ in the brain was immunostained with biotinylated anti-Aβ antibody HJ3.4. (B) Fibrillar plaques in the brain were stained using Thioflavin S. (C) Insoluble $A\beta_{40}$ and (D) insoluble $A\beta_{42}$ was assessed by ELISA in neocortical tissue lysates (Guan = Guanidine). One-way ANOVA followed by Tukey post-test was performed to compare different groups. Data were expressed as mean ± SEM. *$p<0.05$, **$p<0.01$, ***$p<0.001$.

Figure S2



Supplemental Figure 2. APPPS1-21/APOE4 mice (2 months old) received weekly IP injection of antibodies at the doses of 50 mg/kg weight for 6 weeks. (A) ApoE in the PBS soluble fraction of cerebral cortex. (B) ApoE in the insoluble (Guan) fraction of cerebral cortex. (C) Plasma apoE levels. (D) Plasma $A\beta_{40}$ levels.

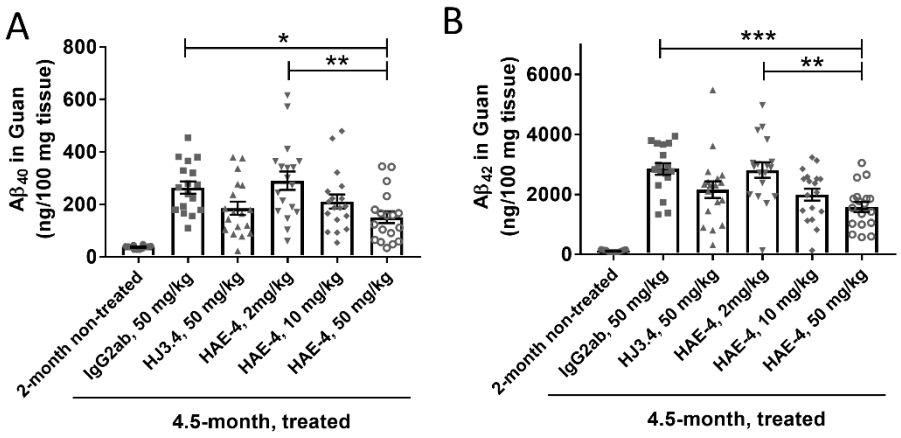
Figure S3



Supplemental Figure 3. Effects of IP administered anti-apoE HAE-3 on Aβ pathology in APPPS1-21/APOE4 mice. At the age of 2 months, the mice were weekly IP injected with 50 mg/kg of HAE-3 or PBS (n=18-20 group, equal numbers of males and females). The mice were sacrificed at the age of 3.5 months and the Aβ pathology in the brain was assessed using histology and biochemical approaches. (A) Representative Aβ staining with anti-Aβ antibody HJ3.4 (left panel) and representative X34 staining for fibrillar plaques (right panel). Scale bar =1

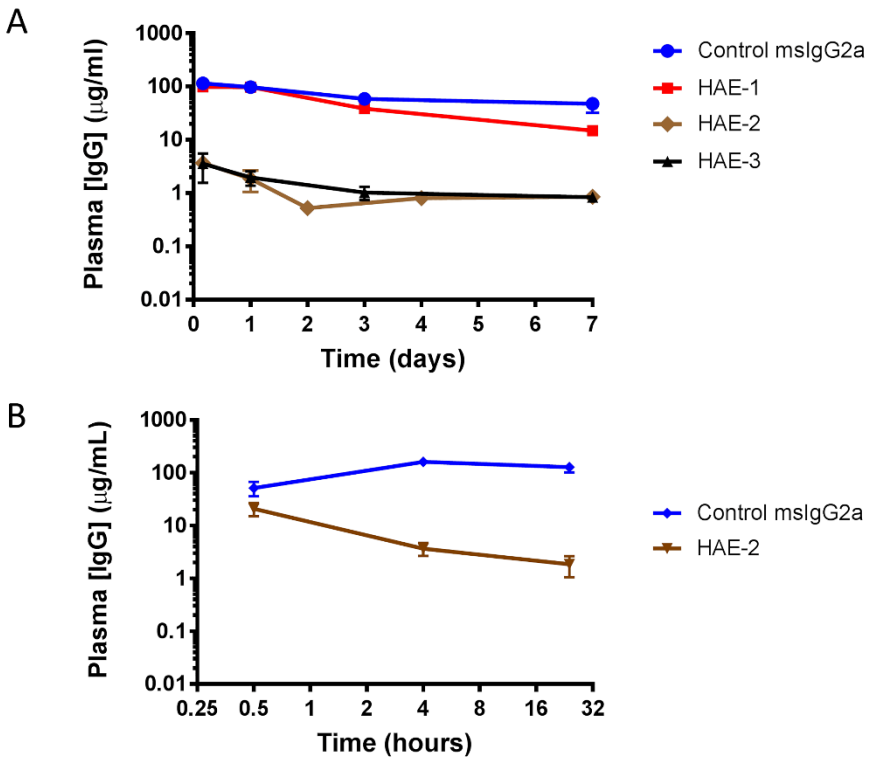
mm. (B) Quantification of HJ3.4 staining. (C) Quantification of X34 staining. (D) $A\beta_{40}$ and (E) $A\beta_{42}$ was measured by ELISA in insoluble fractions of tissue lysates (Guan = Guanidine) by ELISA. Data were expressed as mean ± SEM.

Figure S4



Supplemental Figure 4. Two-month-old APPPS1-21/APOE4 mice were IP injected with a weekly dose of IgG2ab (50 mg/kg), anti-Aβ HJ3.4 (50 mg/kg) or HAE-4 (2, 10 and 50 mg/kg) for 12 doses (n=17-18/group, equal numbers of males and females). A group of 2 month old animals (n=12) were harvested to determine the baseline level of Aβ. At the age of 4.5 months (3 days after the last dose), the mice were perfused with ice-cold PBS containing 0.3% heparin. The cerebral cortices were sequentially homogenized with cold PBS and 5 M guanidine buffer in the presence of 1× protease inhibitor mixture. The (A) $A\beta_{40}$ and (B) $A\beta_{42}$ in guanidine fraction were determined by ELISA. One-way ANOVA followed by Tukey post-test was performed to compare different groups. Data were expressed as mean ± SEM. *$p<0.05$, **$p<0.01$, ***$p<0.001$.

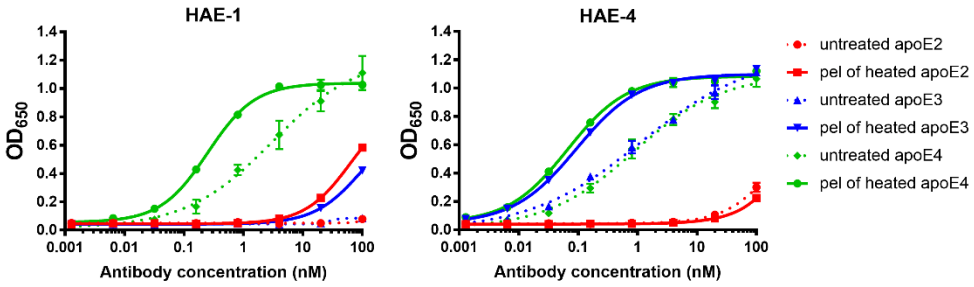
Figure S5



Supplemental Figure 5. Plasma antibody concentrations of HAE-1, HAE-2, HAE-3, or control anti-Her2 msIgG2a following IP injection at 10 mg/kg into APOE4 KI mice. (A) Plasma samples were collected by submandibular puncture at various time points. Assessment of plasma antibody concentrations were performed using coated recombinant apoE4 (5 µg/ml) to capture dosed HAE-1, -2, -3, and recombinant Her2 protein (R&D) (1 µg/ml) to capture the control antibody. Plates were blocked with 3% BSA in TBS/0.1%Tween for 1 hour and washed 3x before incubation with plasma samples at 1:2500 dilution. Bound antibodies were detected with HRP-anti-Mouse IgG. The standard curve range was 0.49 – 1000 ng/mL and fitted with a four-parameter logistic function. (B) Because HAE-2 and HAE-3 levels decreased quickly after

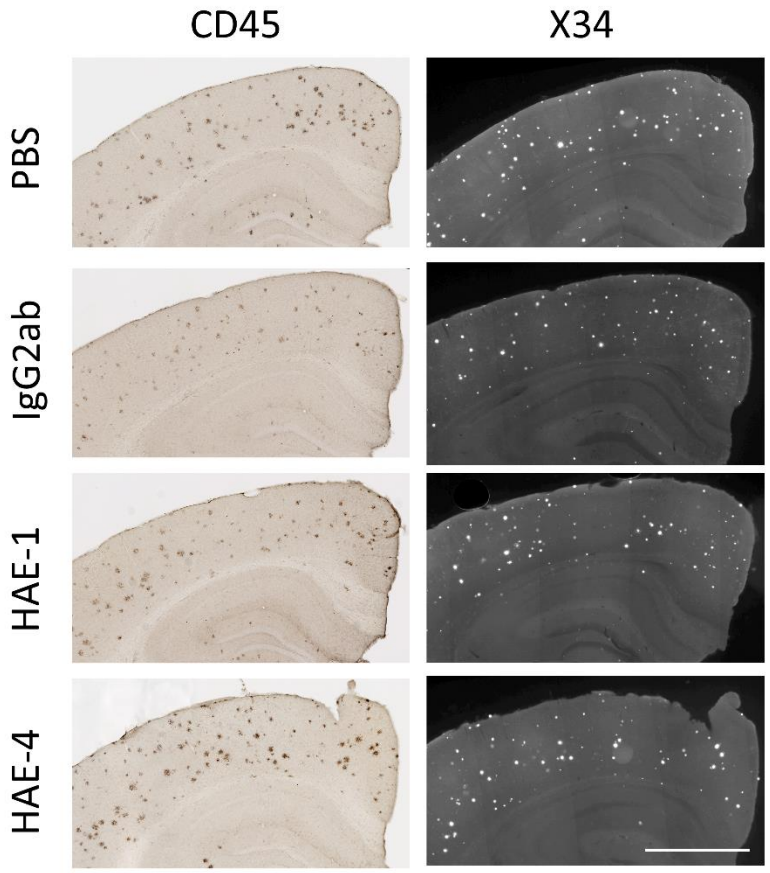
dosing, a seprate PK study at shorter time points was performed with HAE-2 to demonstrate higher levels of the antibody were present after 30 minutes.

Figure S6



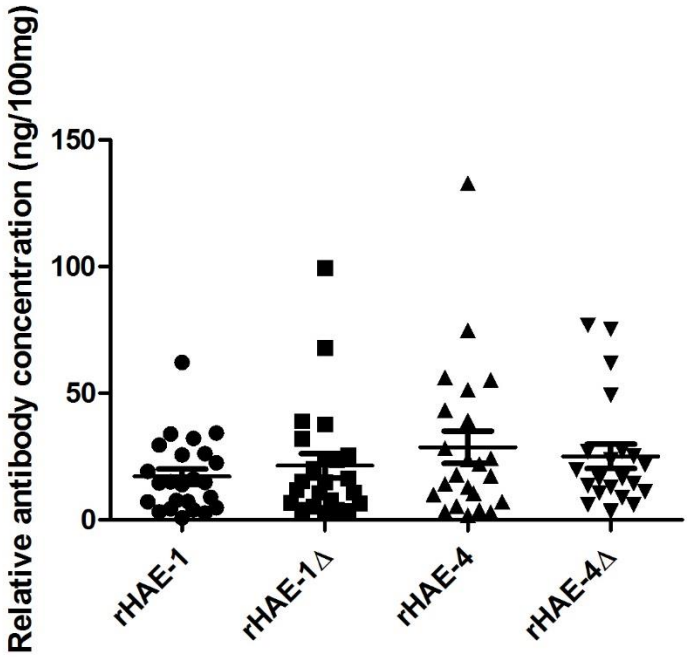
Supplemental Figure 6. Binding of HAE-1 and HAE-4 to untreated recombinant and heat-induced aggregates of apoE2, apoE3 and apoE4. The aggregated apoE was induced by incubating apoE at 1 mg/ml concentration at 40°C for 24 hours. The aggregates were then recovered in the pellet fraction following ultracentrifugation at 186,000 g for 1 hour. For ELISA, the untreated apoE or heat-induced aggregates of apoE was coated directly to ELISA plates at 0.5 µg/ml overnight at 4°C. After three washes with PBS, the wells were blocked with 1% BSA-PBS for 1 hour at room temperature with shaking at 500 rpm. The blocked wells were washed once with PBS and subsequently loaded with HAE antibodies at serial concentrations (starting at 100 nM with 5-fold dilutions thereafter). Bound HAE antibodies were detected with HRP-labeled goat anti-mouse IgG (Jackson Immunoresearch) and visualized with TMB substrate at OD_{650} (reaction stopped with BioFX stop solution). The results show that HAE-1 only detected apoE4 and had some preference for heat-treated apoE4, and HAE-4 detected both apoE3 and apoE4 and preferred heat treated apoE.

Figure S7



Supplemental Figure 7. Staining of microglia and X34 after acute immunization of HAE-4. At the age of 4 months, the APPPS1-21/APOE4 mice received 4 IP injections of 50 mg/kg weight of antibodies every three days. The mice were sacrificed 24 hours after the final injection and the plaques were stained with X34 and the activated microglia was stained with CD45 (Bar =1mm).

Figure S8



Supplemental Figure 8. Relative antibody concentration in the cortex of APPPS1-21/APOE4 mice expressing recombinant (r) HAE-1, rHAE-1 with D265A mutation (Δ), rHAE-4, and rHAE-4Δ. APPPS1-21/APOE4 mice were injected at P0 with AAV2/8 that express full length rHAE-1 and rHAE-4 with or without the D265A mutation. At the age of 3.5 months, antibody concentration in the PBS soluble fraction of cortex was measured by ELISA. The relative level of each antibody was calculated by using its hybridoma-derived, purified antibody as a standard.

Uncut gels for Figure 4