Figure 8. Microglial and astrocytic cell staining patterns in 3xTg-AD mice are not overtly affected by amplicon-mediated vaccination. Helper virus-free $HSV_{IE}A\beta_{CMV}2$ **(a-d** and **q-t)**, $HSV_{IE}A\beta_{CMV}$IL-4 **(e-h** and **u-x)**, and $HSV_{IE}1_{CMV}2$ **(i-l** and **y-ab)** were delivered subcutaneously thrice to 3xTg-AD mice beginning at 2 months of age ($1x10^6$ transduction units per vaccination). Coronal mouse brain sections (30 μm) were prepared from vaccinated mice sacrificed at 11 months of age and were processed for F4/80 immunohistochemistry **(a-p)** to assess alterations in microglial activation and GFAP immunohistochemistry **(q-af)** to examine changes in astrocyte staining patterns as a result of amplicon-mediated vaccination. Age-matched, non-vaccinated control (NVC) 3xTg-AD mice (**m-p** and **ac-af** for F4/80 and GFAP staining, respectively) were sacrificed at 11 months of age and brains processed identically for comparison purposes. Images were obtained for four areas of the brain. Area I represents the CA_1 hippocampal region at –1.28 mm from Bregma **(a, e, i, m, q, u, v, ac)**, area II represents the CA_1 region at –2.12 mm from Bregma **(b, f, j, n, r, v, z, ad)**, area III represents the subiculum at –2.75 mm from Bregma **(c, g, k, o, s, w, aa, ae)**, and area IV represents the entorhinal cortex at -2.50 to -3.80 mm from Bregma **(d, h, l, p, t, x, ab, af)**. The scale bar depicted in **m** represents 250 μm.

Supplementary Figure 1. Gross assessment of cellular infiltration into brains of vaccinated 3xTg-AD mice using H&E histochemistry. Helper virus-free $HSV_{IE}A\beta_{CMV}2$ **(a, b)**, $HSV_{IE}A\beta_{CMV}$IL-4 **(c, d)**, $HSV_{IE}1_{CMV}2$ **(e, f)**, and non-vaccinated control (NVC) (**g, h**) were delivered subcutaneously thrice to 3xTg-AD mice beginning at 2 months of age ($1x10^6$ transduction units per vaccination). Coronal mouse brain sections (30 μm) were prepared from vaccinated mice sacrificed at 11 months of age and were processed for H&E histochemistry after

the sections had been stained by AT180 phospho-Tau immunohistochemistry (Figure 7) to assess the extent of basophilic and eosinophilic structures that may be differentially represented among the vaccinated mice. Images were obtained at 10X and 20X magnification, and represent the CA_1 hippocampal region at –1.28 mm from Bregma **(a-h)**. The scale bar depicted in **g** (for panels **a, c, e, g**) and **h** (for panels **b, d, f, h**) represent 250 μm.