Supplemental Figure 1. Paucity of αS inclusions in non-motor impaired M83 mice injected in the gastrocnemius muscle with E46K αS fibrils. Representative immunohistochemistry images of spinal cord (SC), brainstem and thalamus (Thal) of M83 mice injected intramuscularly with E46K αS fibrils. Scale bars = 100μm for each respective region shown.

Supplemental Figure 2. Representative images showing co-labeling with 9C10 and anti-p62 antibodies. (A) Double immunofluorescence analysis in the brainstem of an M83 mice following injection in the gastrocnemius muscle with H50Q αS fibrils antibodies 9C10 (green) and anti-p62 (red) or (**B**) in the cortex of an M20 mouse following injection in the hippocampus with H50Q αS fibrils. Merged images with DAPI shown in the right panels. Scale bars = 100 μm.

Supplemental Figure 3. αS inclusion pathology in M83 mice injected in the gastrocnemius muscle with wild-type and mutant αS fibrils. Representative immunohistochemistry images of the pons of M83 mice injected intramuscularly with wild-type, E46K, H50Q, G51D or A53E αS fibrils stained with anti-human αS antibody Syn 211 or N-terminal specific antibody Syn 506. Scale bars = 100 μm.

Supplemental Figure 4. Thioflavin S and argyrophilic αS inclusions following induction of aggregation with αS fibrils. (**A, B**) Representative inclusions stained with Thioflavin S in an M83 mouse following intramuscular injection with H50Q αS fibrils (A) and an M20 mouse following intrahippocampal injection with H50Q αS fibrils (B). (**C, D**) Campbell-Switzer silver stained inclusions in an M83 mouse following intramuscular injection with H50Q αS fibrils (C) and an M20 mouse following intrahippocampal injection with wild-type αS fibrils (D). Arrows depict stained inclusions. Scale bars = 100 μm or 50 μm (inset).

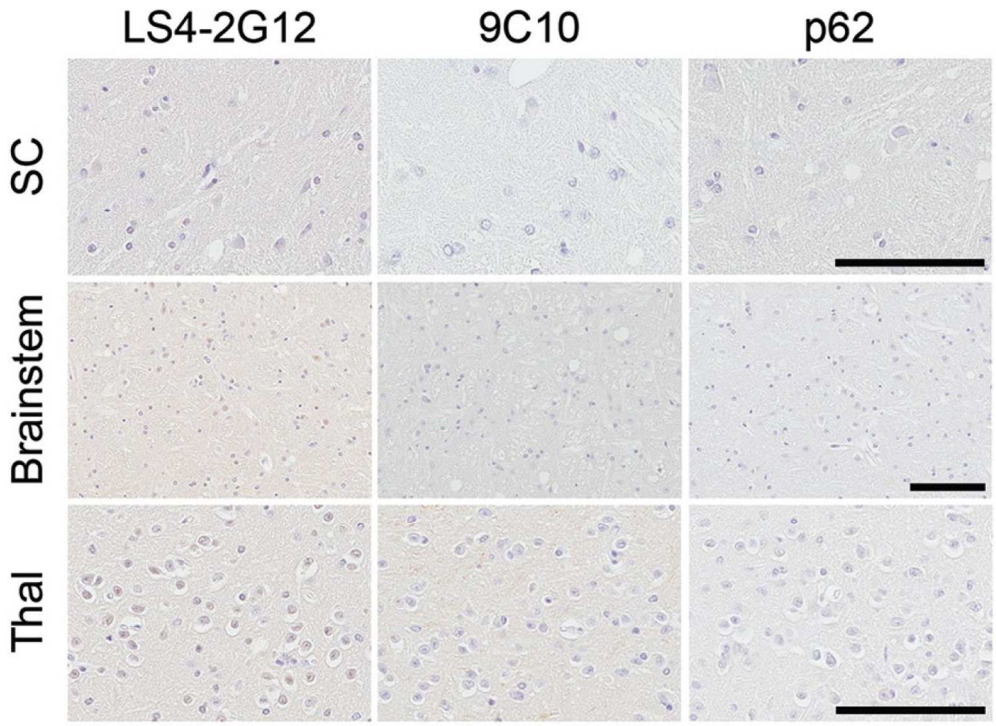
Supplemental Figure 5. αS inclusion pathology in M20 mice following intrahippocampal injection of human wild-type, H50Q, G51D or A53E αS mutant αS fibrils demonstrated by 9C10 immunohistochemistry. Representative 9C10 immunostaining images of the hippocampus (Hipp), entorhinal cortex (Ectx), thalamus (Thal), brainstem and spinal cord (SC) of M20 mice 2 months (A) or 4 months (B) post-intrahippocampal injection of human wild-type, H50Q, G51D or A53E αS fibrils. Scale bars = 50 μm.

Supplemental Figure 6. αS inclusion pathology in M20 mice following intrahippocampal injection of human wild-type, H50Q, G51D or A53E αS mutant αS fibrils demonstrated by p62 immunohistochemistry. Representative p62 immunostaining images of the hippocampus (Hipp), entorhinal cortex (Ectx), thalamus (Thal), brainstem and spinal cord (SC) of M20 mice 2 months (A) or 4

months (B) post-intrahippocampal injection of human wild-type, H50Q, G51D or A53E αS fibrils. Scale bars = 50 μm.

Supplemental Figure 7. αS inclusion pathology in M20 mice following intrahippocampal injection of E46K mutant αS fibrils demonstrated by 9C10 immunohistochemistry. Representative 9C10 immunostaining images of the hippocampus (Hipp), entorhinal cortex (Ectx), thalamus (Thal), brainstem and spinal cord (SC) of M20 mice 4, 6 or 8 months post-intrahippocampal injection of E46K αS fibrils. Scale bars = 50 μm. NP = no pathology present (n = 3), P = pathology present (n = 1).

Supplemental Figure 8. αS inclusion pathology in M20 mice following intrahippocampal injection of E46K mutant αS fibrils demonstrated by p62 immunohistochemistry. Representative p62 immunostaining images of the hippocampus (Hipp), entorhinal cortex (Ectx), thalamus (Thal), brainstem and spinal cord (SC) of M20 mice 4, 6 or 8 months post-intrahippocampal injection of E46K αS fibrils. Scale bars = 50 μm. NP = no pathology present (n = 3), P = pathology present (n = 1).



Supplemental Figure 1. Paucity of αS inclusions in non-motor impaired M83 mice injected in the gastrocnemius muscle with E46K αS fibrils. Representative immunohistochemistry images of spinal cord (SC), brainstem and thalamus (Thal) of M83 mice injected intramuscularly with E46K αS fibrils. Scale bars = 100μm for each respective region shown.

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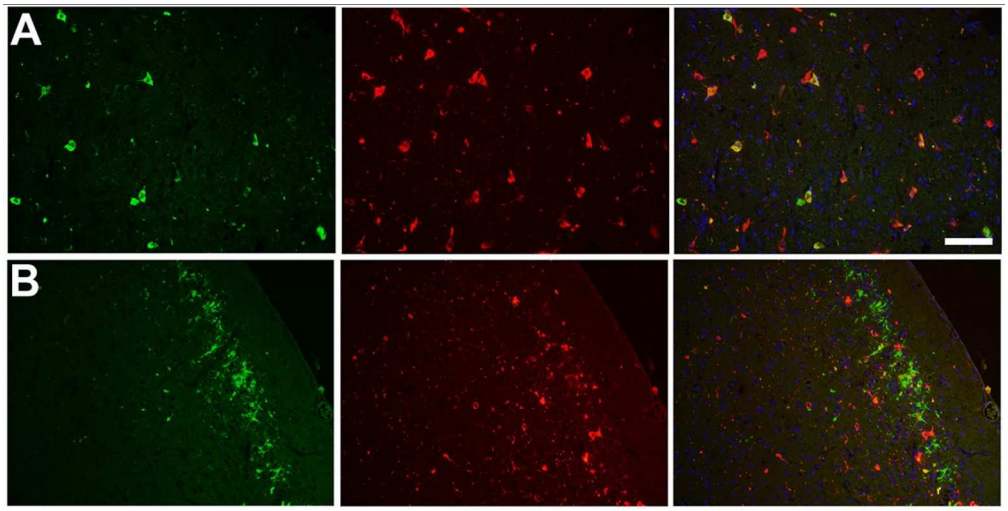

Supplemental Figure 2. Representative images showing co-labeling with 9C10 and anti-p62 antibodies. (A) Double immunofluorescence analysis in the brainstem of an M83 mice following injection in the gastrocnemius muscle with H50Q αS fibrils antibodies 9C10 (green) and anti-p62 (red) or (B) in the cortex of an M20 mouse following injection in the hippocampus with H50Q αS fibrils. Merged images with DAPI shown in the right panels. Scale bars = 100 μm.

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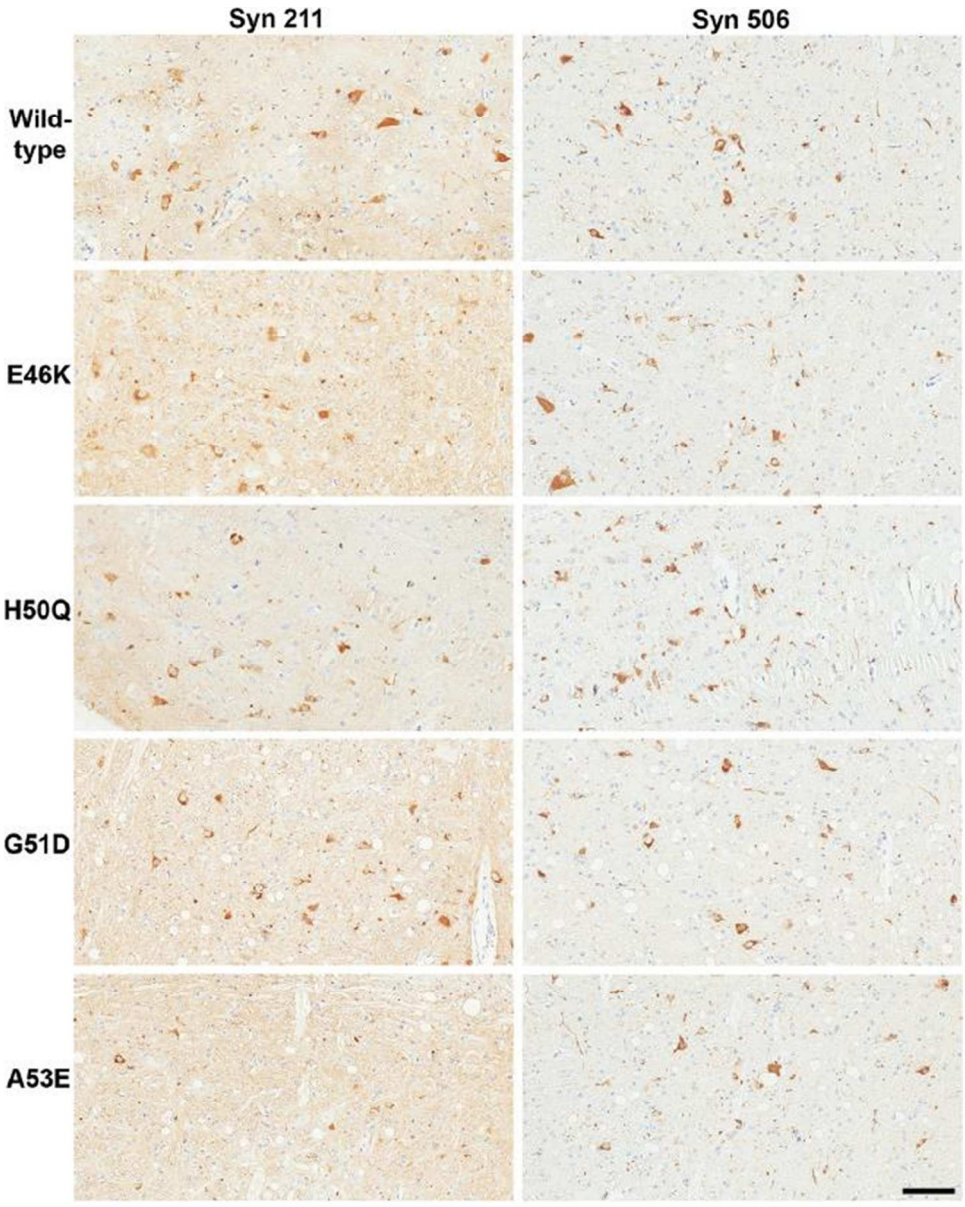

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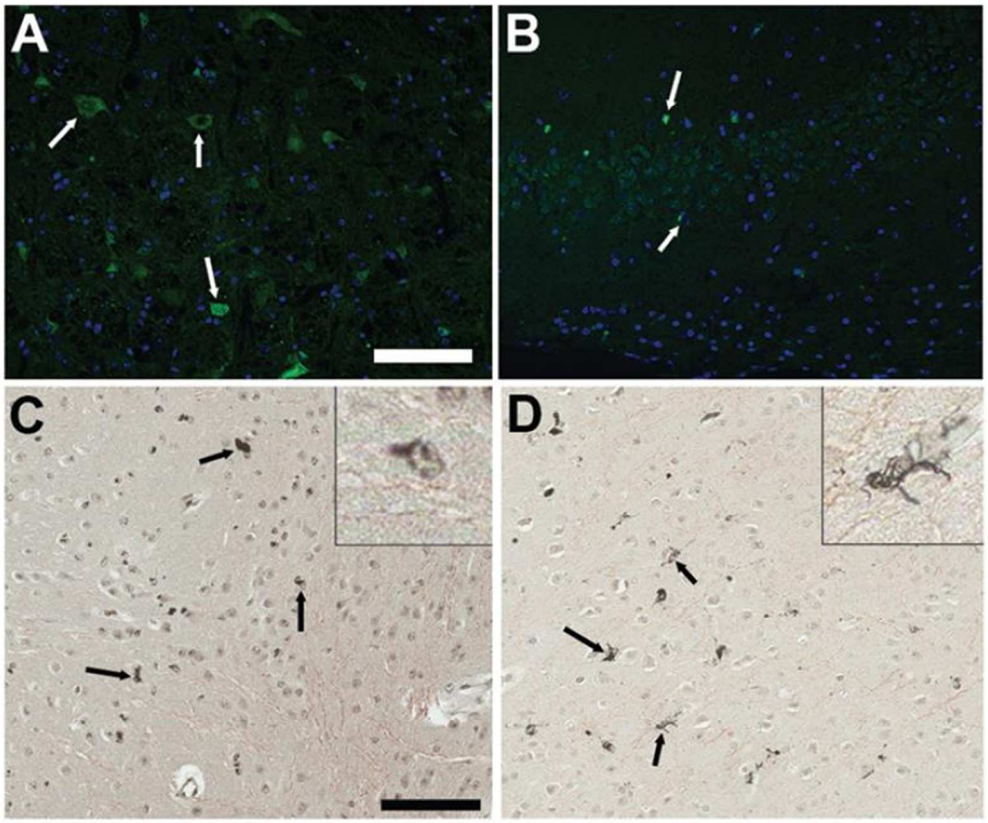

Supplemental Figure 4. Thioflavin S and argyrophilic αS inclusions following induction of aggregation with αS fibrils. (A, B) Representative inclusions stained with Thioflavin S in an M83 mouse following intramuscular injection with H50Q αS fibrils (A) and an M20 mouse following intrahippocampal injection with H50Q αS fibrils (B). (C, D) Campbell-Switzer silver stained inclusions in an M83 mouse following intramuscular injection with H50Q αS fibrils (C) and an M20 mouse following intrahippocampal injection with wild-type αS fibrils (D). Arrows depict stained inclusions. Scale bars = 100 μm or 50 μm (inset).

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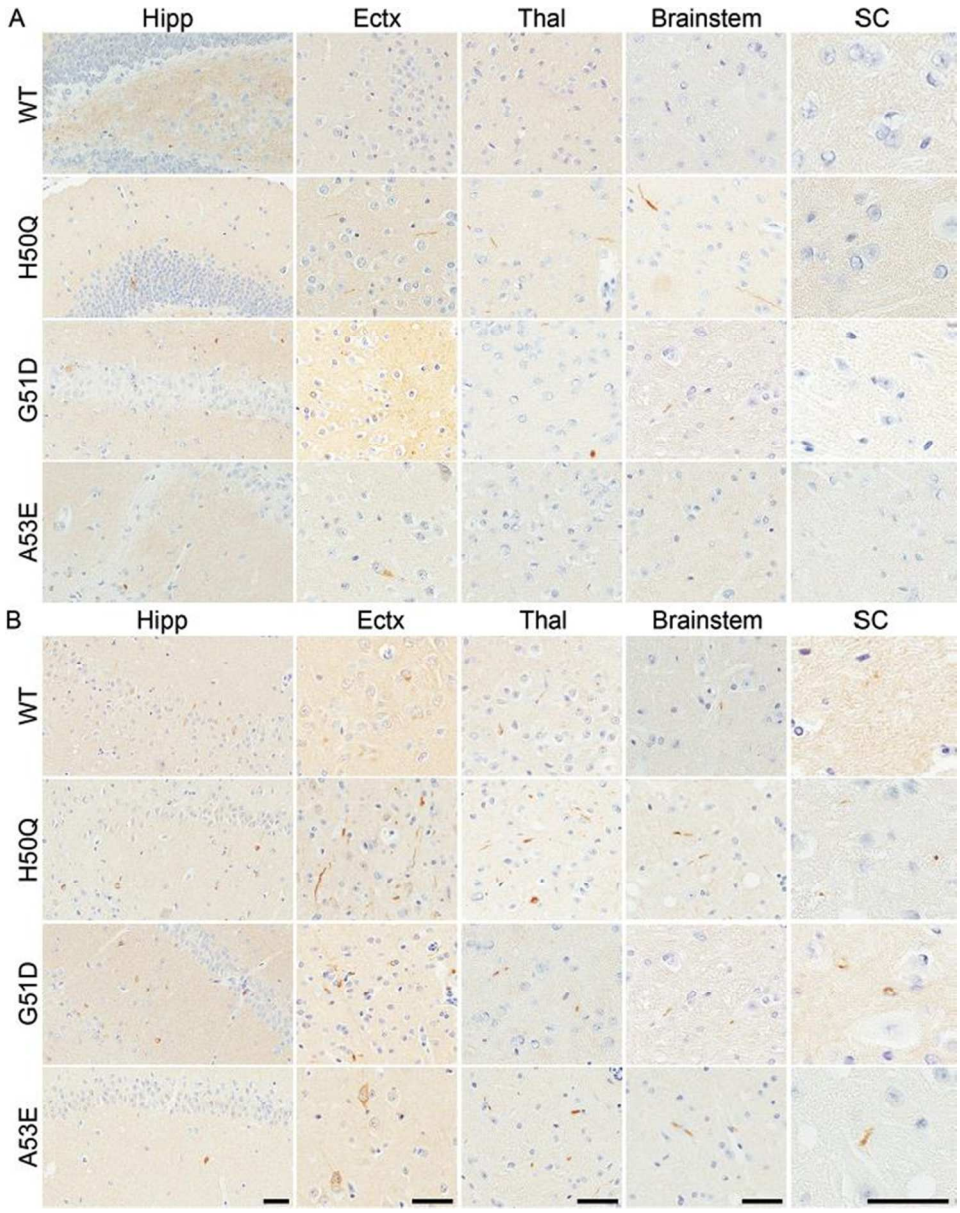

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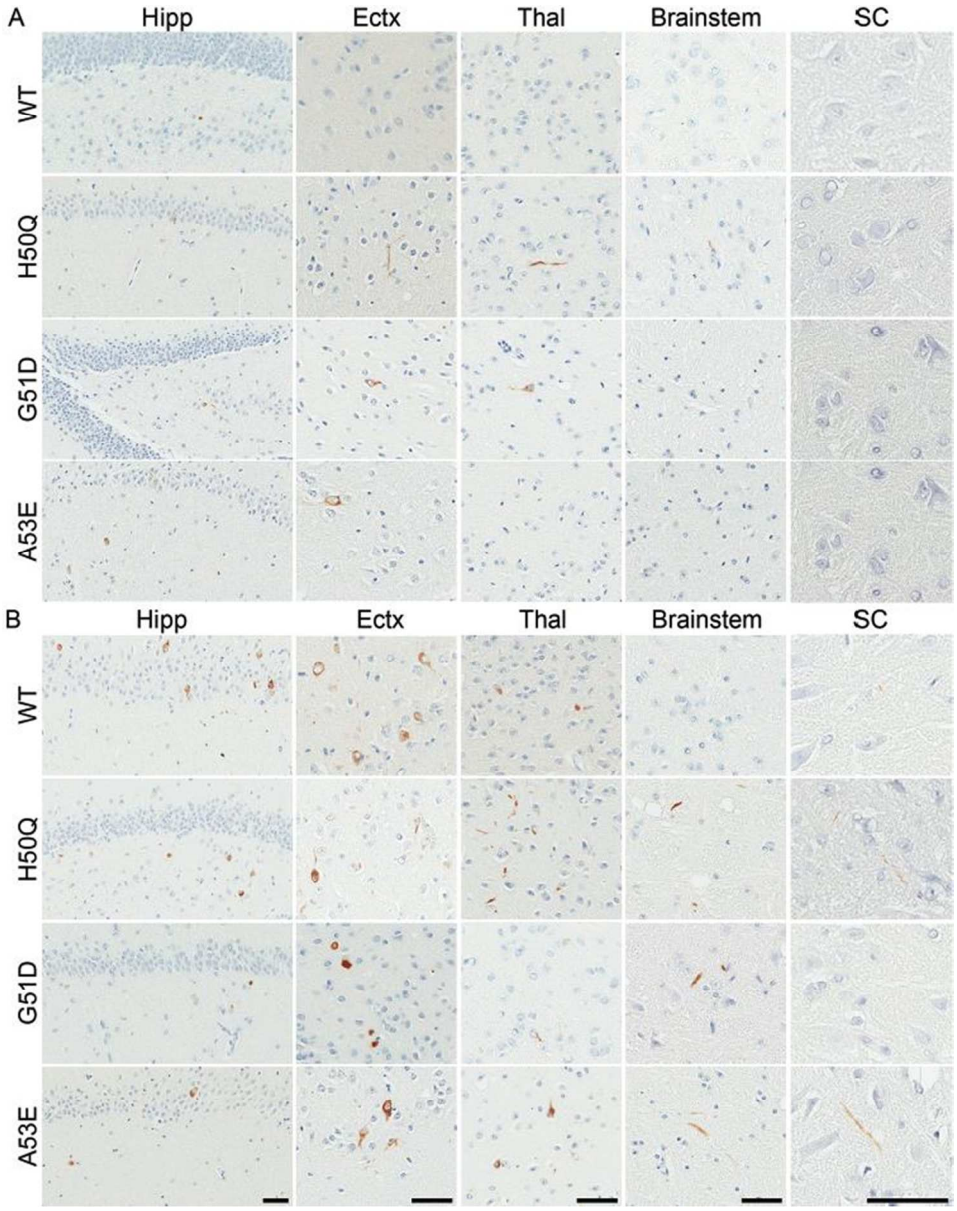

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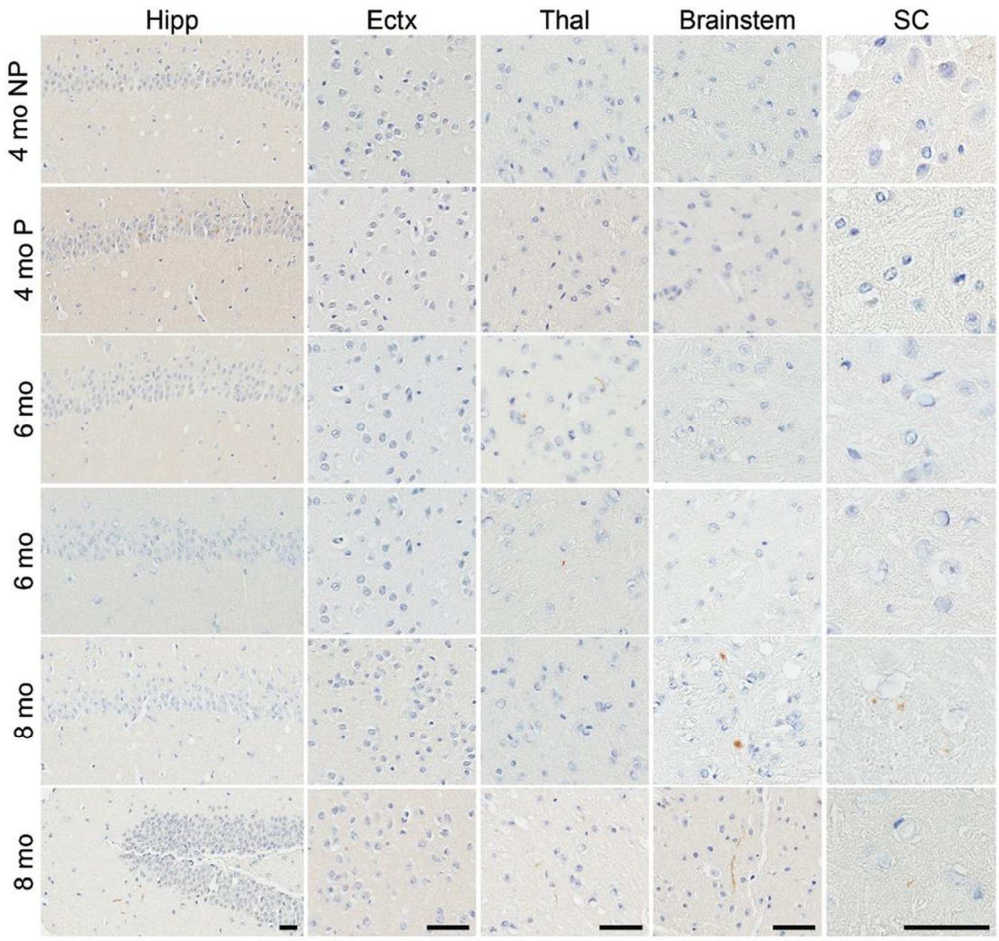

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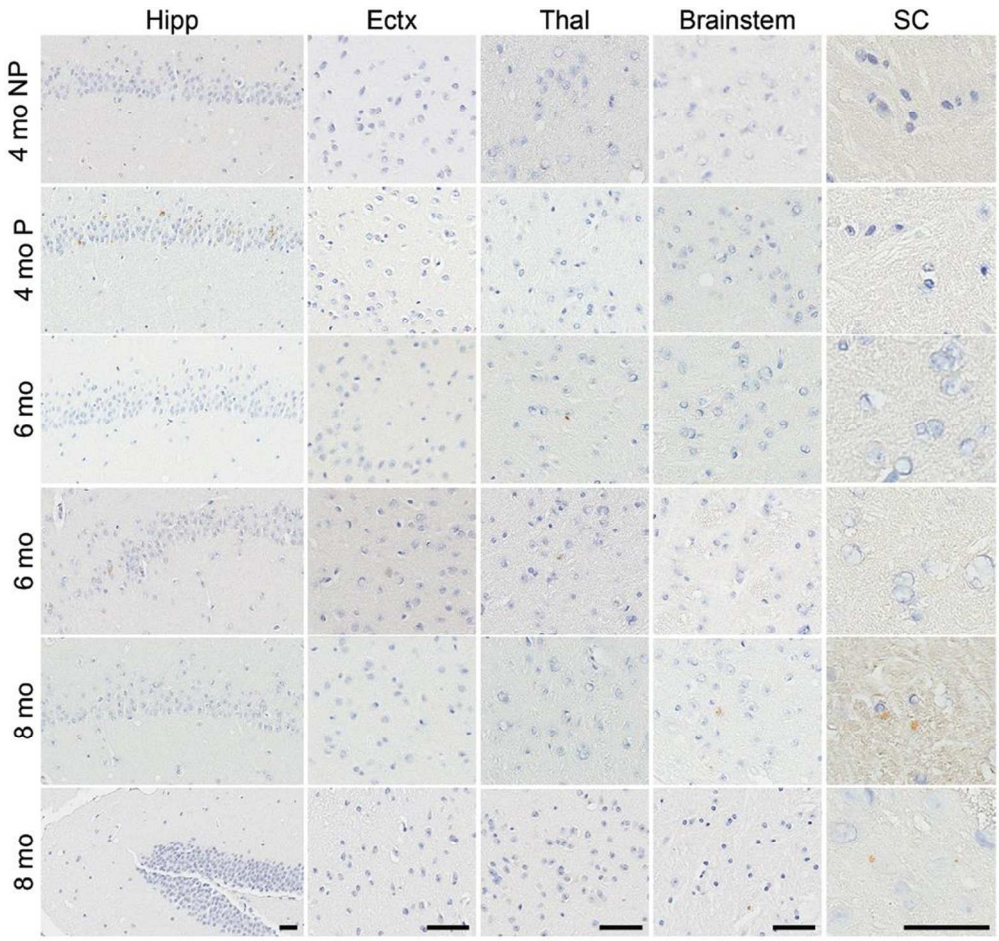

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