Uncropped and unprocessed blots

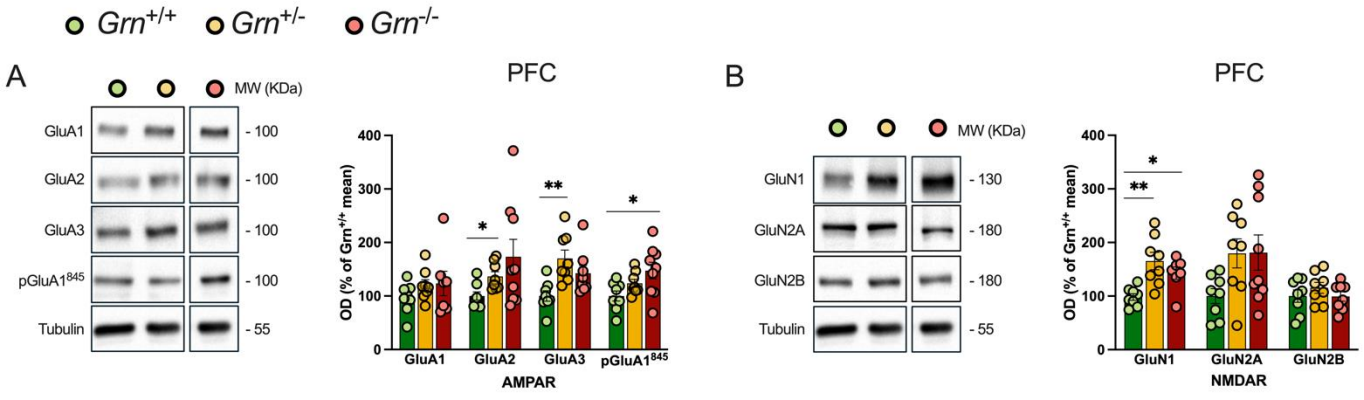
PGRN-related lysosomal dysfunction drives behavioral alterations, region-specific inflammation, and glutamatergic synaptic impairments in a Grn knockout mouse model of frontotemporal dementia

This PowerPoint file contains the uncropped western blot images associated with the manuscript submitted to *Neurobiology of Disease*.

For each brain region (*i.e.*, TIF prefrontal cortex, striatum, hippocampus, olfactory bulb and total homogenate striatum), the first slide entitled "*Uncropped blots, AREA*" shows the representative images included in the corresponding manuscript figure. On these slides, the representative bands used in the figures are highlighted with boxes. Next to each representative image, the corresponding original uncropped blot is shown both alone and merged with the colorimetric molecular weight marker to allow marker visualization.

The following slides contain **all uncropped blots** acquired for that specific brain region, including those used as representative images in the manuscript. For each blot, both the original uncropped image and the merged image with the molecular weight marker are provided. The **detected protein, the corresponding molecular weight marker and the sample loading order** are indicated on each slide.

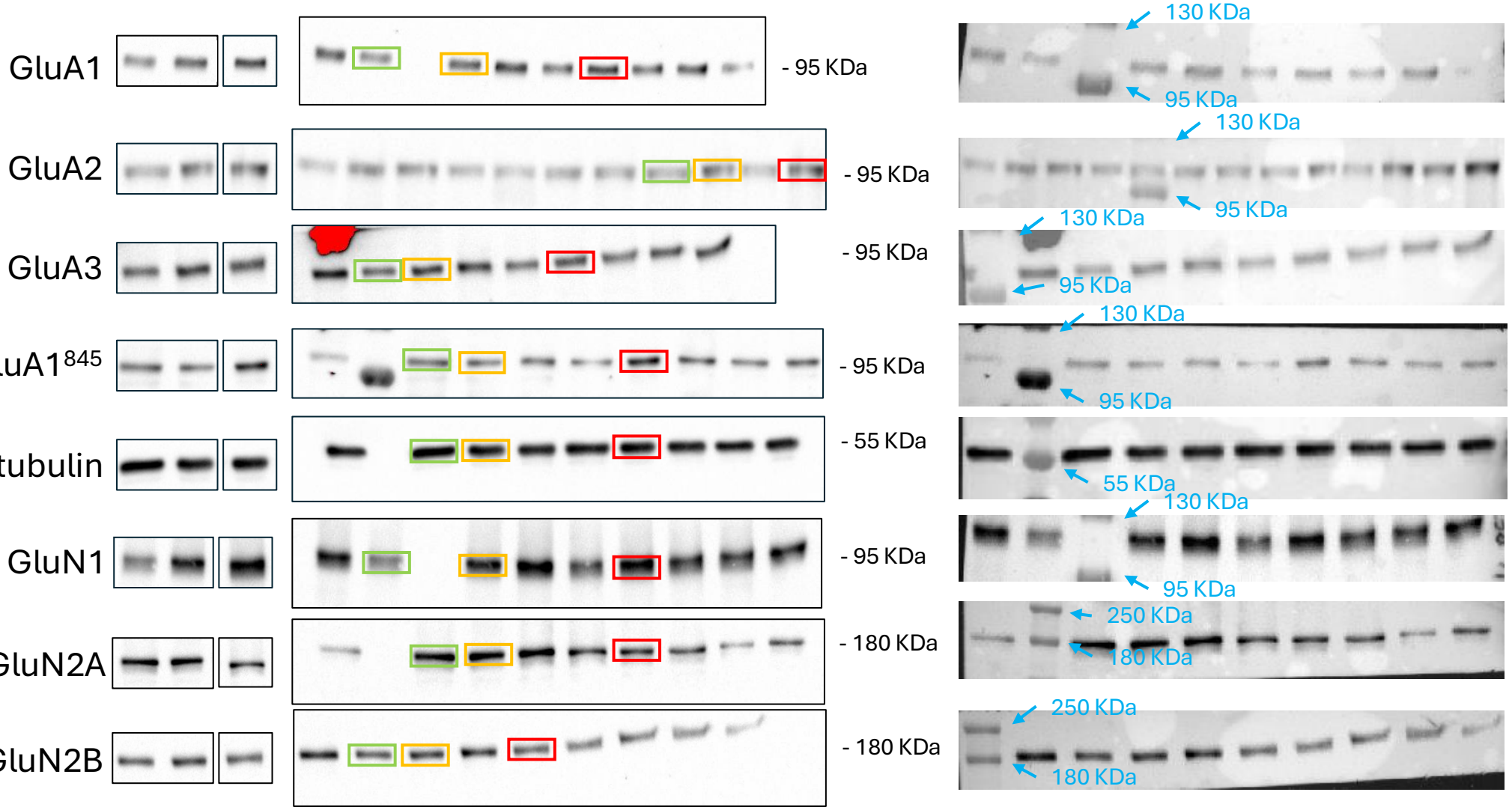
Uncropped blot, **prefrontal cortex**



Boxed regions used in the main figure

Original uncropped blot

Uncropped blot with molecular weight markers



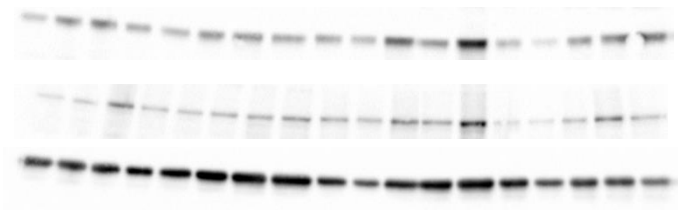
For all bands except GluA2 and GluN2B, lanes 2, 3, and 6 correspond to samples 262+/+, 290+/-, and 265-/-, respectively. For GluA2, lanes 8, 9, and 11 correspond to samples 253+/+, 254+/-, and 249-/-, respectively. For GluN2B, lanes 2, 3, and 5 correspond to samples 262+/+, 290+/-, and 263-/-, respectively. Non-adjacent lanes from the same original blot were juxtaposed for presentation purposes; the corresponding regions are indicated in the annotated uncropped images.

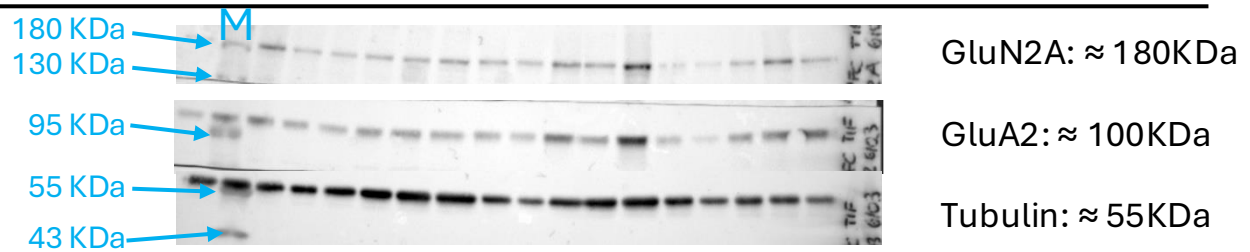
PFC, group 1

For all gels, the loading order was consistent and corresponded to the following samples: **210, 211, 247, 248, 243, 246, 252, 253, 254, 256, 249, 250, 258, 260, 257, 245, 251, and 259**. Samples 210, 211, 252, 253, 258, and 260 were +/+; samples 247, 248, 254, 256, 257, and 245 were +/-; and samples 243, 246, 249, 250, 251, and 259 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide. Due to the low homogeneity of the samples, for group 1, each gel was run in duplicate (set 1 and set 2), and the values obtained were averaged for the final analysis. On the right, the gels are shown without the molecular weight marker; on the left, these images were subsequently merged with the colorimetric image to visualize the marker.

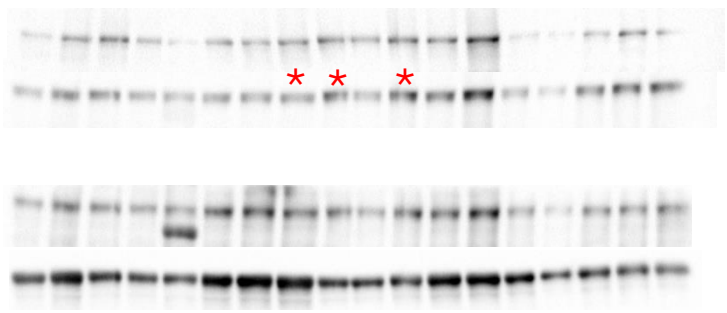
Gel 1

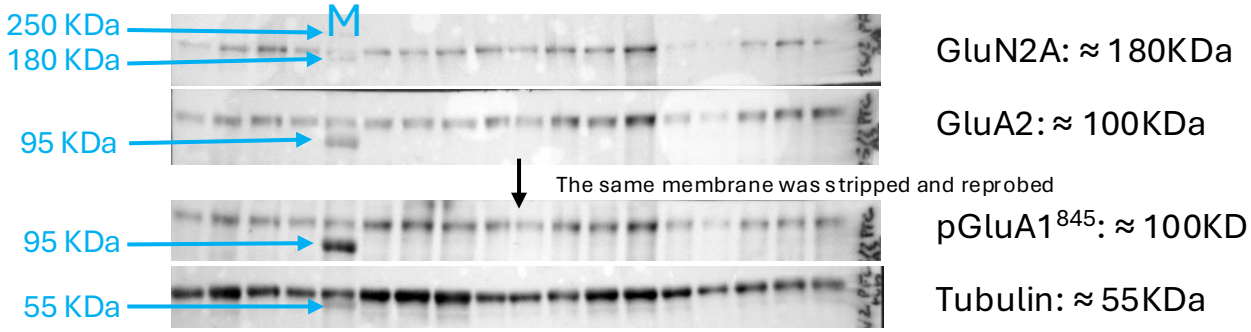
Set 1





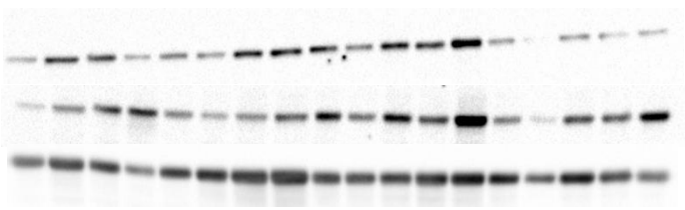
Set 2

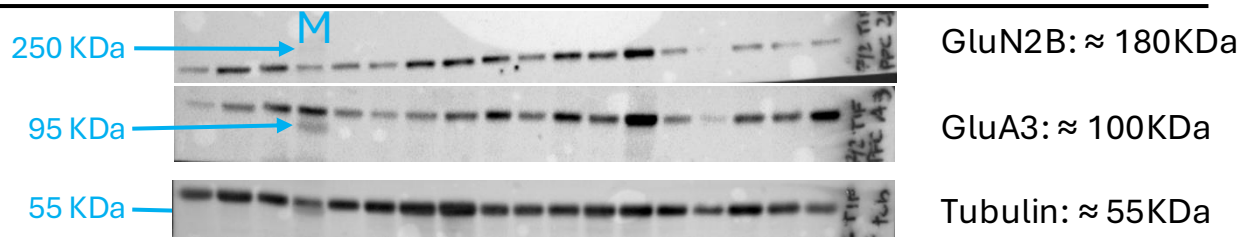




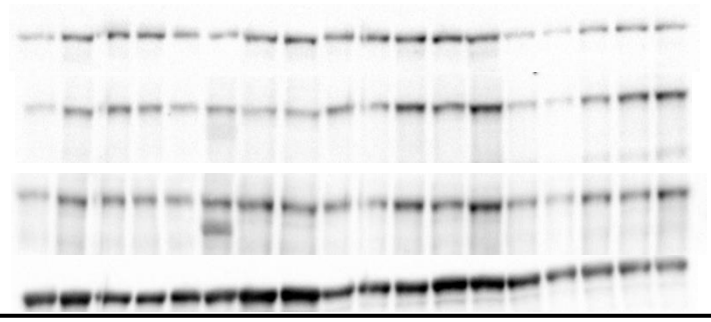
Gel 2

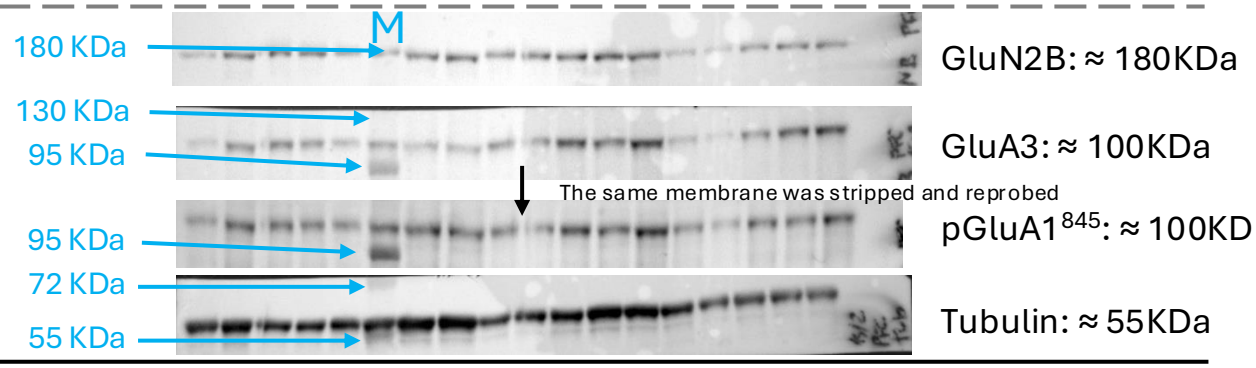
Set 1





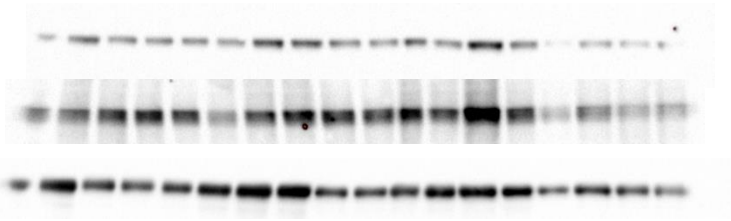
Set 2

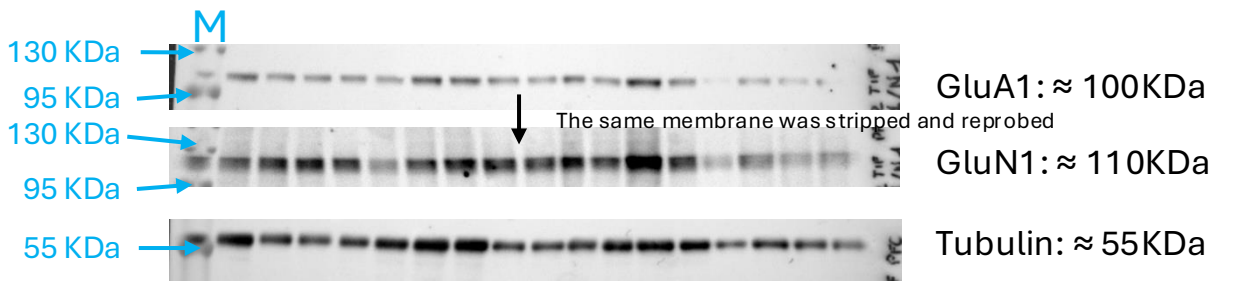




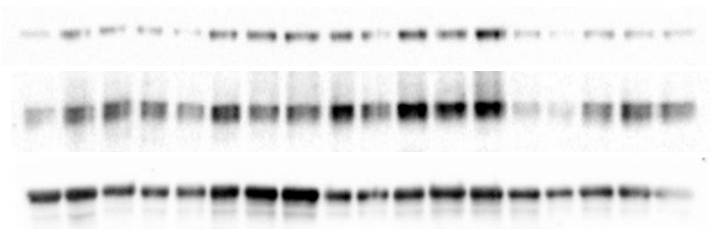
Gel 3

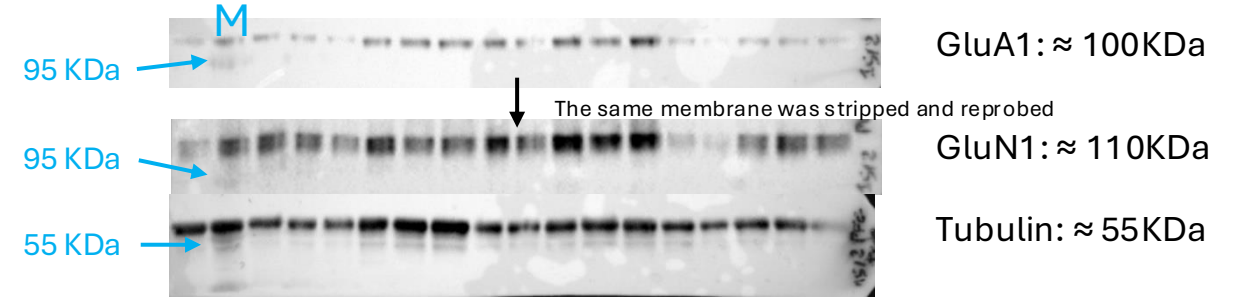
Set 1





Set 2

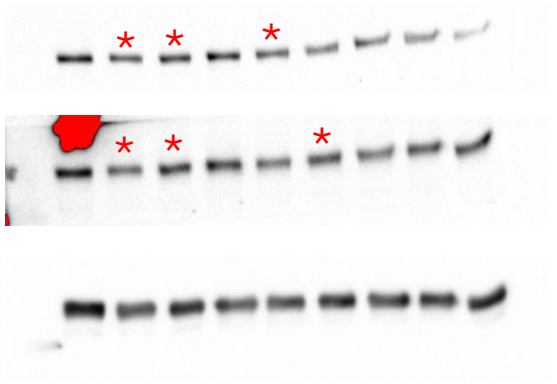


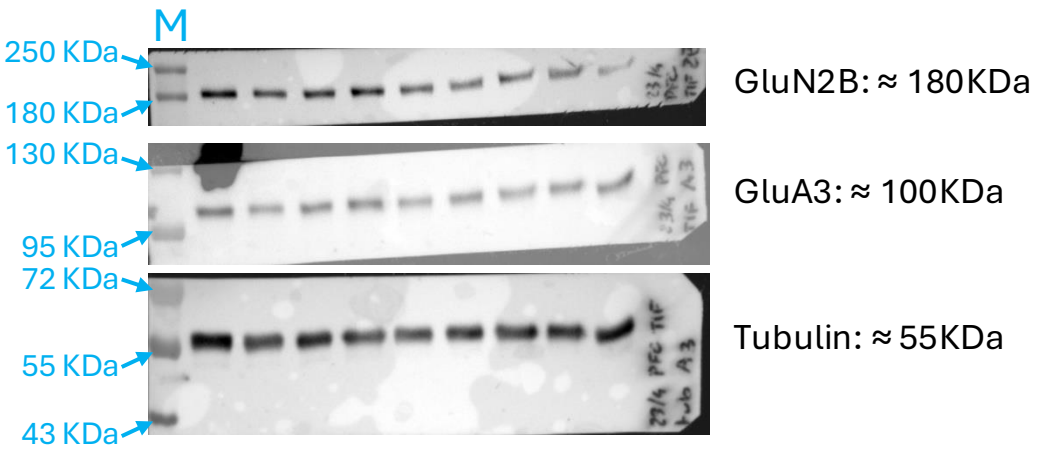


PFC, group 2

For all gels, the loading order was consistent and corresponded to the following samples: **261, 262, 290, 291, 263, 265, 264, 293, and 266**. Samples 261, 262, and 264 were +/+; samples 290, 291, and 293 were +/-; and samples 263, 265, and 266 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide. On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

Gel 1



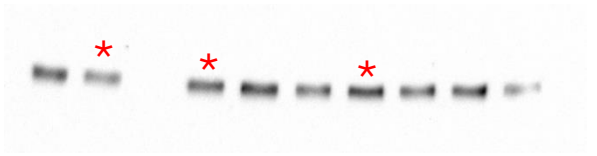


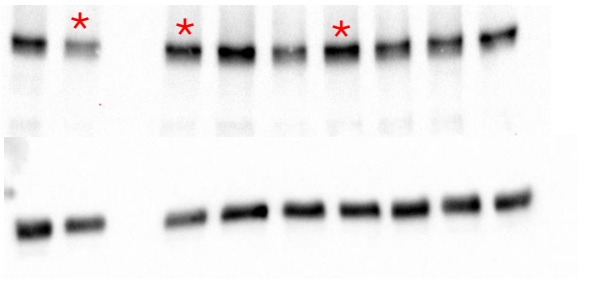
GluN2B: ≈ 180KDa

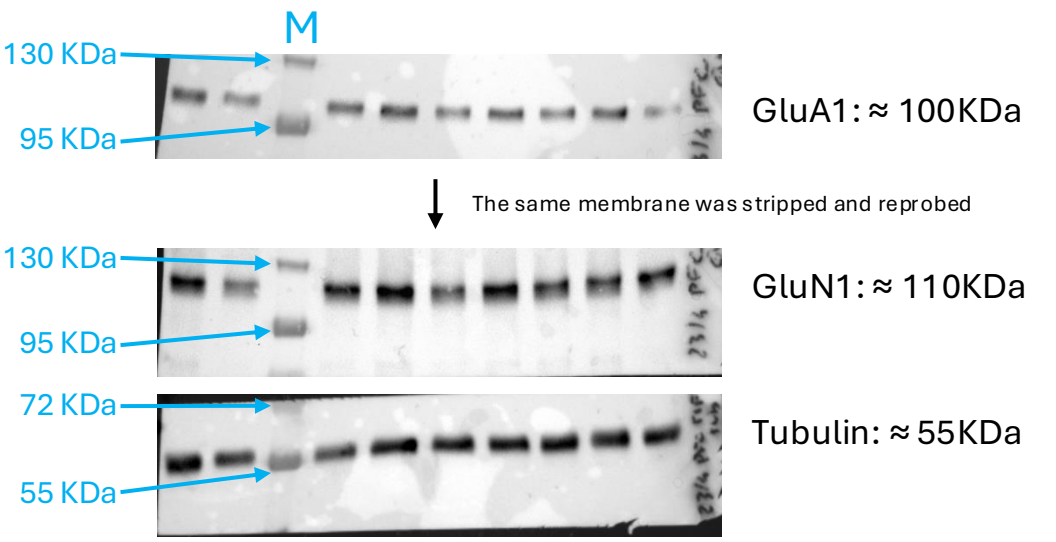
GluA3: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 2





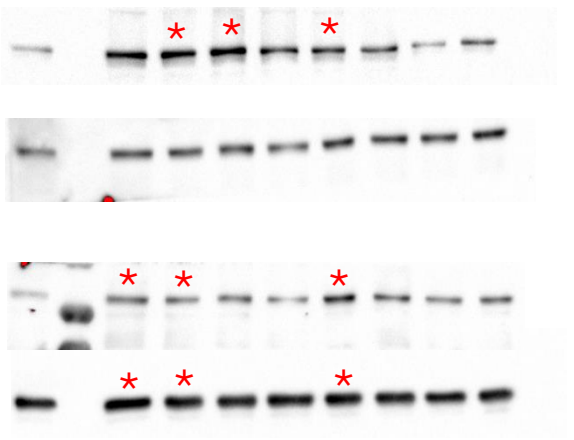


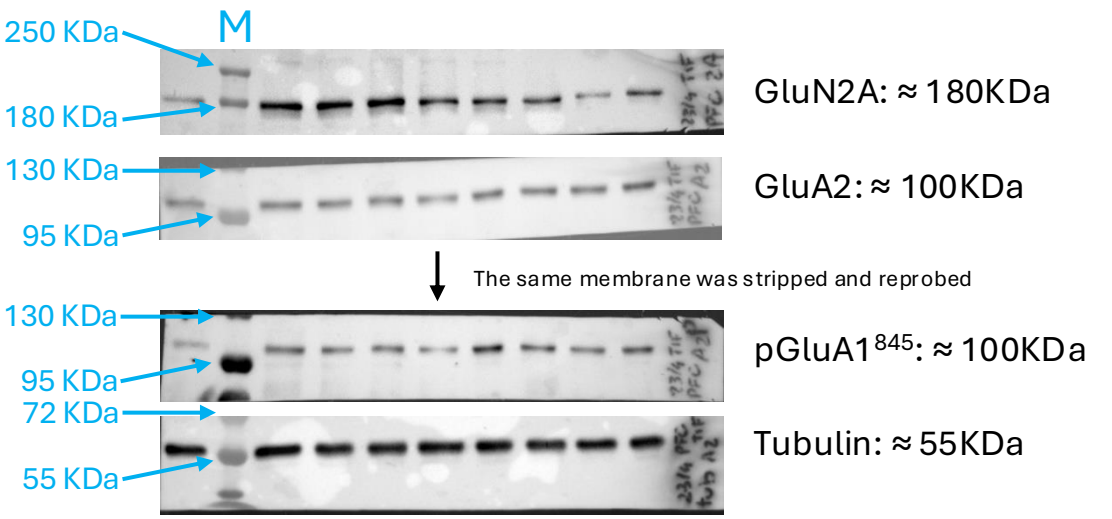
GluA1: ≈ 100KDa

GluN1: ≈ 110KDa

Tubulin: ≈ 55KDa

Gel 3





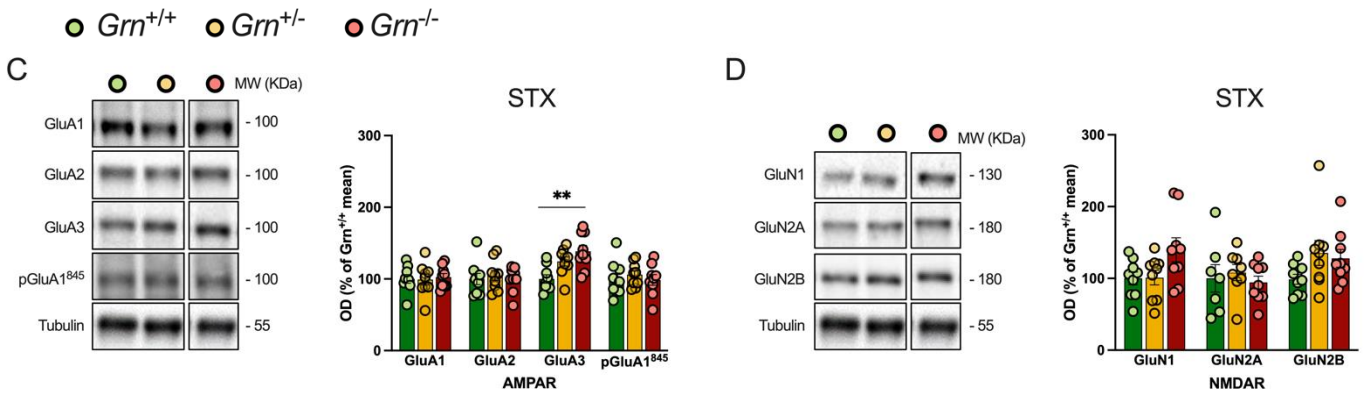
GluN2A: ≈ 180KDa

GluA2: ≈ 100KDa

pGluA1^{845}: ≈ 100KDa

Tubulin: ≈ 55KDa

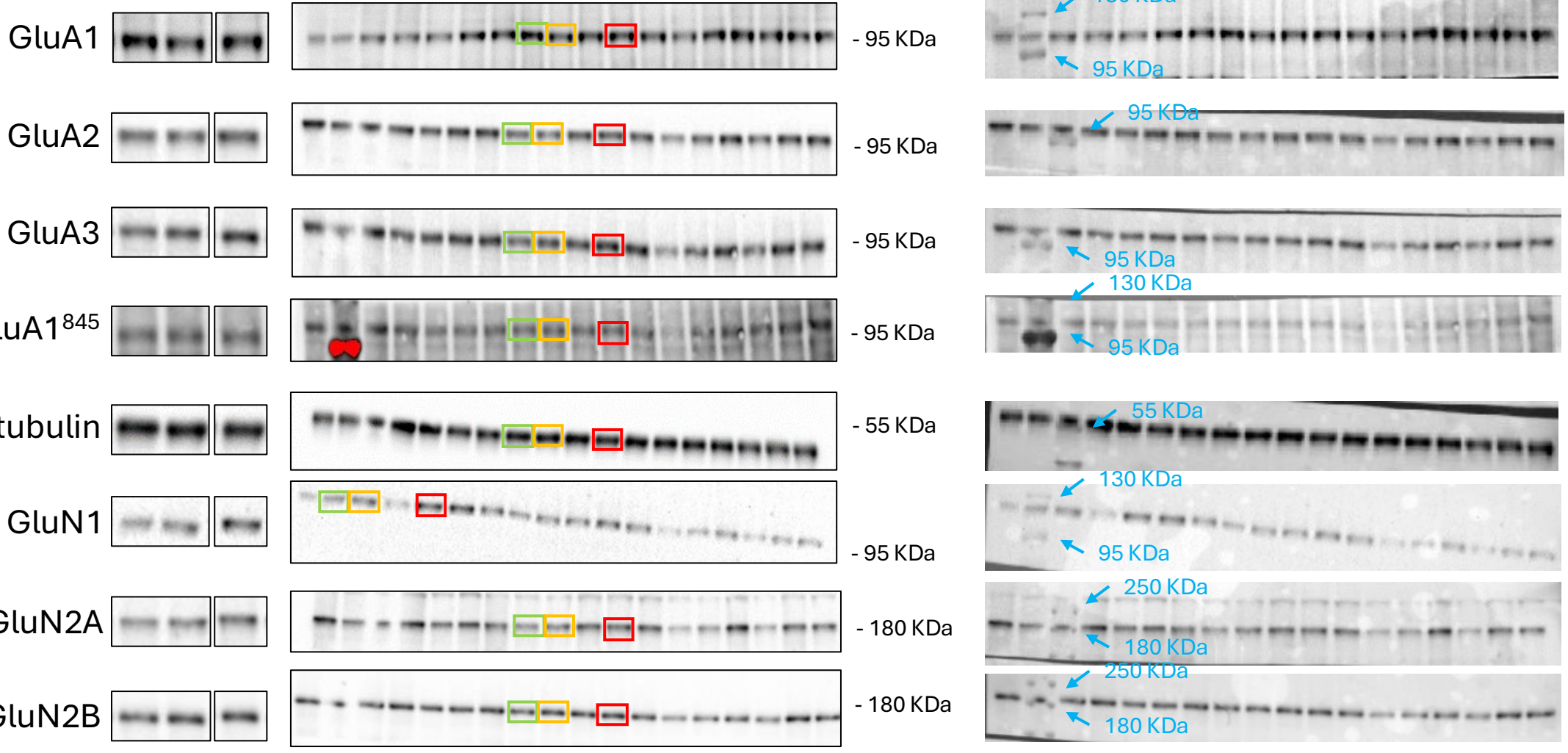
Uncropped blot, **striatum**



Boxed regions used in the main figure

Original uncropped blot

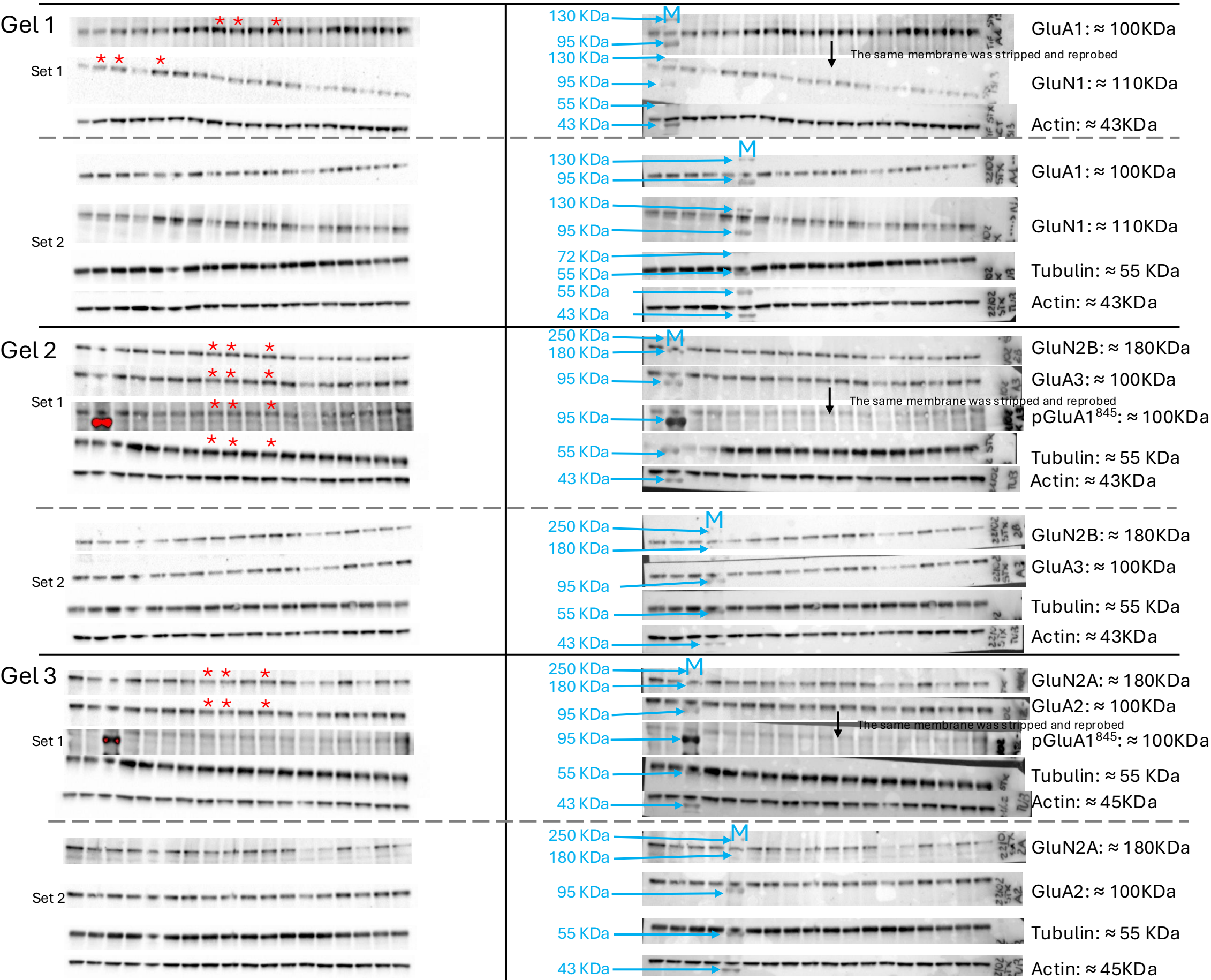
Uncropped blot with molecular weight markers



For all bands except GluN1, lanes 8, 9, and 11 correspond to samples 253+/+, 254+/-, and 249-/-, respectively. For GluN1, lanes 2, 3, and 5 correspond to samples 211+/+, 247+/-, and 243-/-, respectively. Non-adjacent lanes from the same original blot were juxtaposed for presentation purposes; the corresponding regions are indicated in the annotated uncropped images.

Stx, group 1

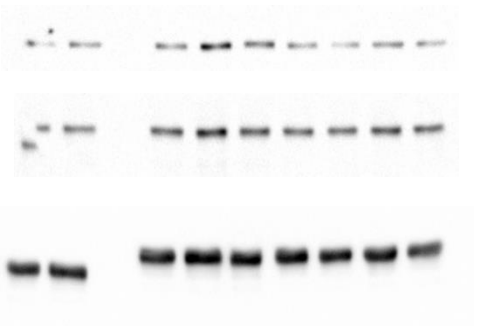
For all gels, the loading order was consistent and corresponded to the following samples: **210, 211, 247, 248, 243, 246, 252, 253, 254, 256, 249, 250, 258, 260, 257, 245, 251, and 259**. Samples 210, 211, 252, 253, 258, and 260 were +/+; samples 247, 248, 254, 256, 257, and 245 were +/-; and samples 243, 246, 249, 250, 251, and 259 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide. Due to the low homogeneity of the samples, for group 1, each gel was run in duplicate (set 1 and set 2), and the values obtained were averaged for the final analysis. On the right, the gels are shown without the molecular weight marker; on the left, these images were subsequently merged with the colorimetric image to visualize the marker.

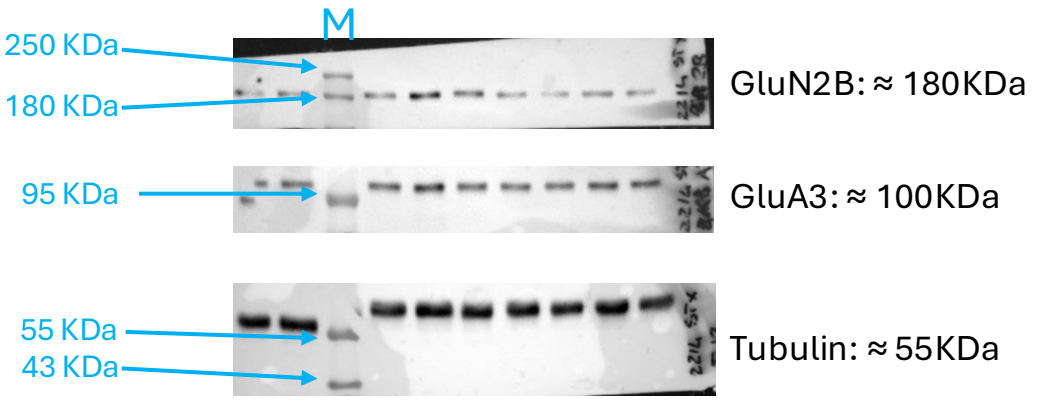


Stx, group 2

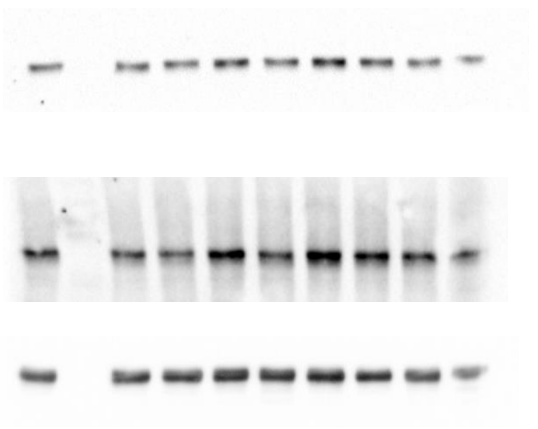
For all gels, the loading order was consistent and corresponded to the following samples: **261, 262, 290, 291, 263, 265, 264, 293, and 266**. Samples 261, 262, and 264 were +/+; samples 290, 291, and 293 were +/-; and samples 263, 265, and 266 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

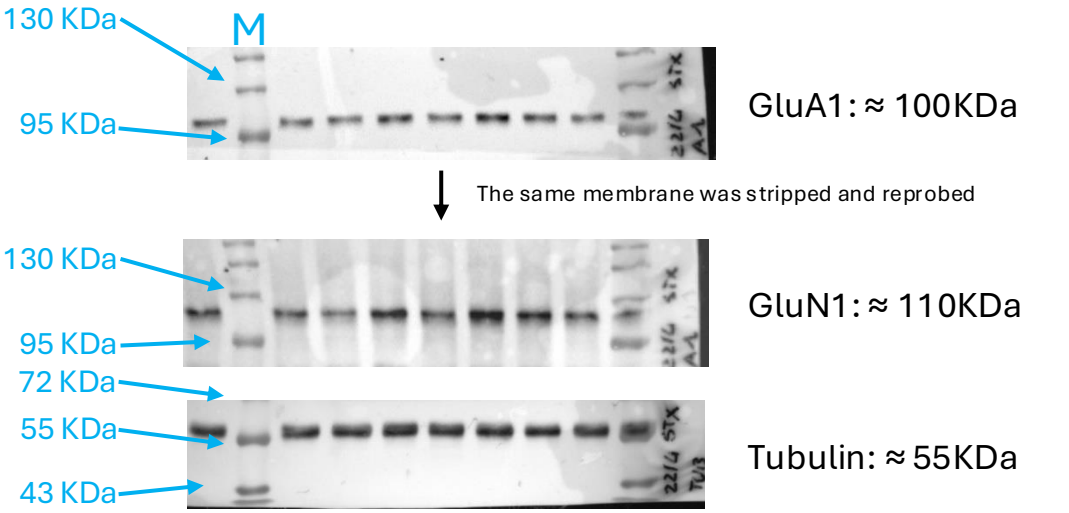
Gel 1



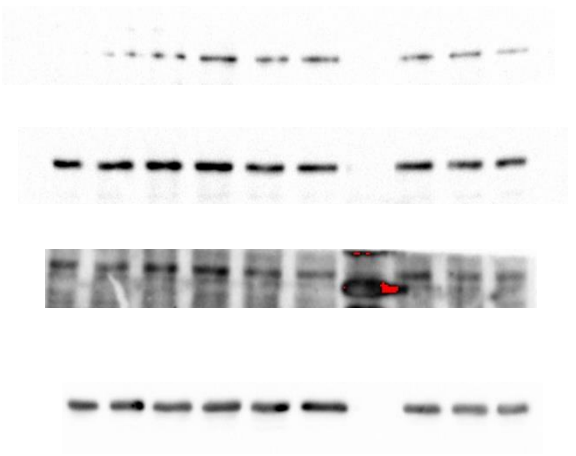


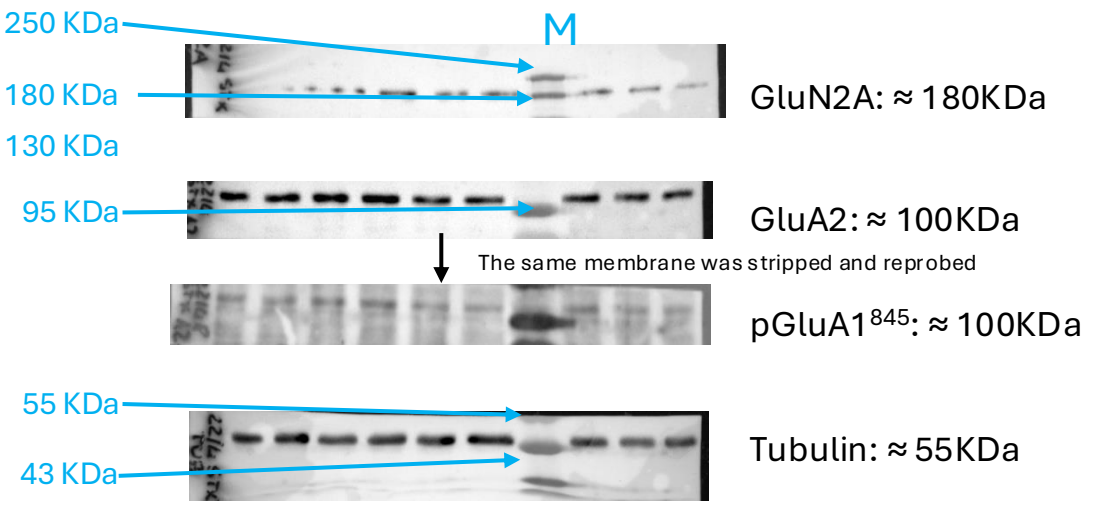
Gel 2



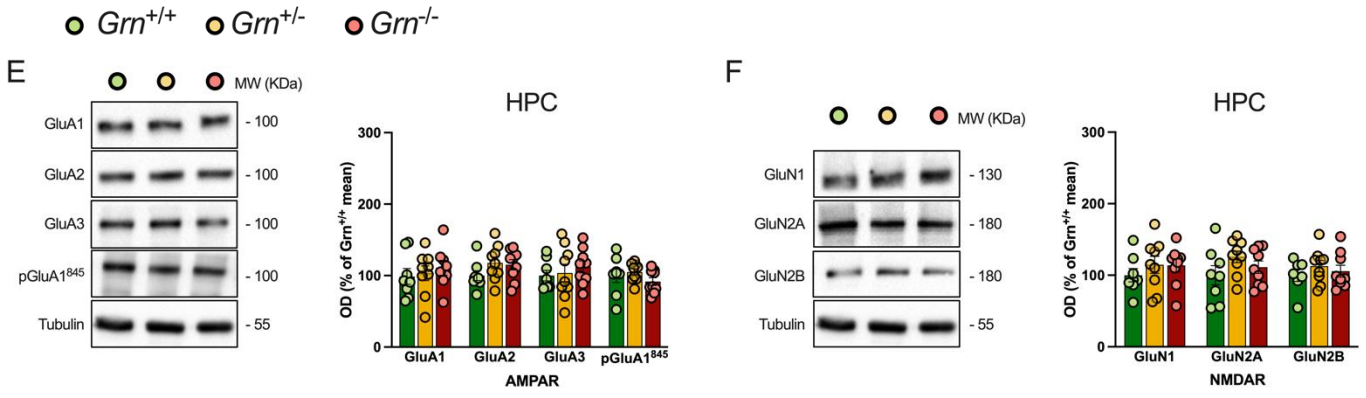


Gel 3





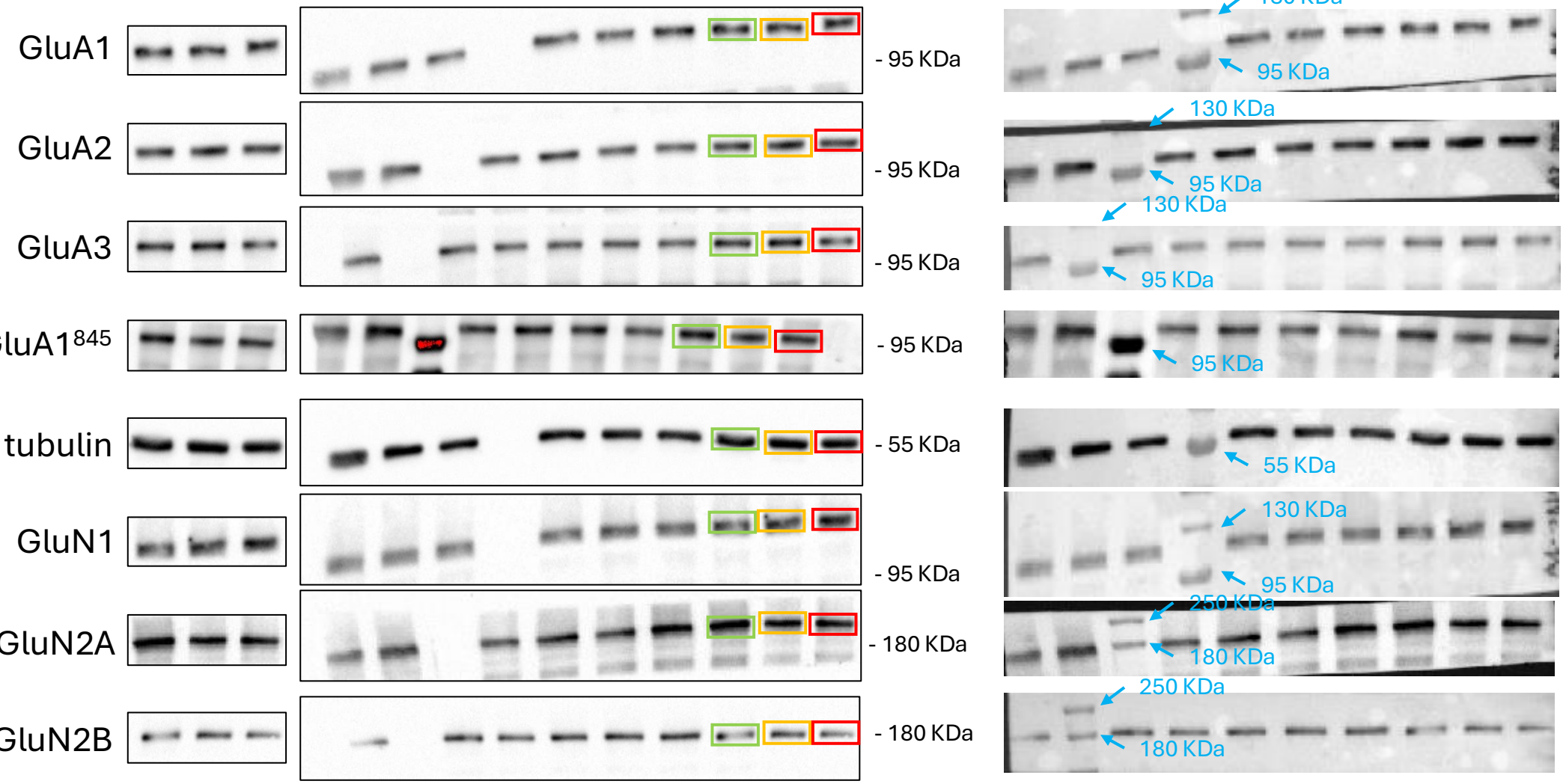
Uncropped blot, **hippocampus**



Boxed regions used in the main figure

Original uncropped blot

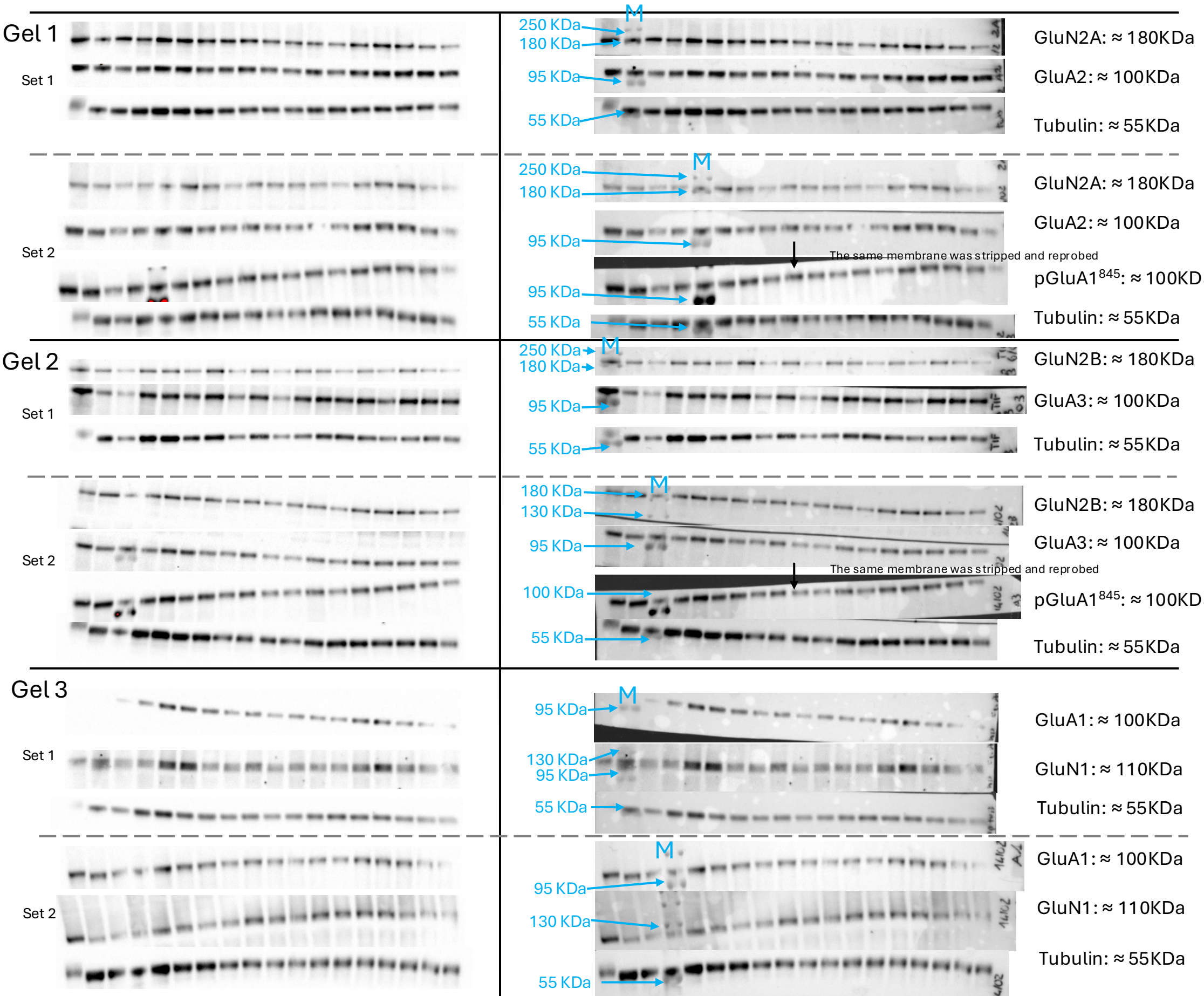
Uncropped blot with molecular weight markers



For all bands lanes 7, 8, and 9 correspond to samples 264+/+, 293+/-, 266-/-

Hp, group 1

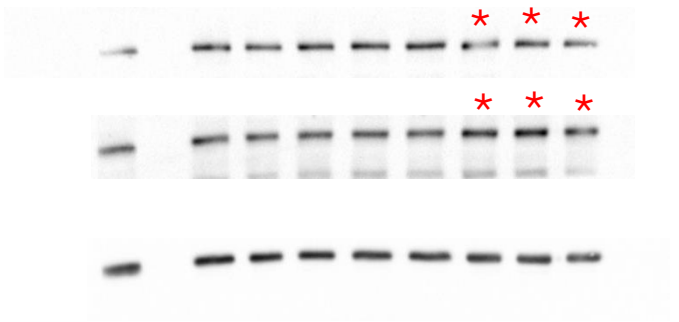
For all gels, the loading order was consistent and corresponded to the following samples: **210, 211, 247, 248, 243, 246, 252, 253, 254, 256, 249, 250, 258, 260, 257, 245, 251, and 259**. Samples 210, 211, 252, 253, 258, and 260 were +/+; samples 247, 248, 254, 256, 257, and 245 were +/-; and samples 243, 246, 249, 250, 251, and 259 were -/-.. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Due to the low homogeneity of the samples, for group 1, each gel was run in duplicate (set 1 and set 2), and the values obtained were averaged for the final analysis. On the right, the gels are shown without the molecular weight marker; on the left, these images were subsequently merged with the colorimetric image to visualize the marker.

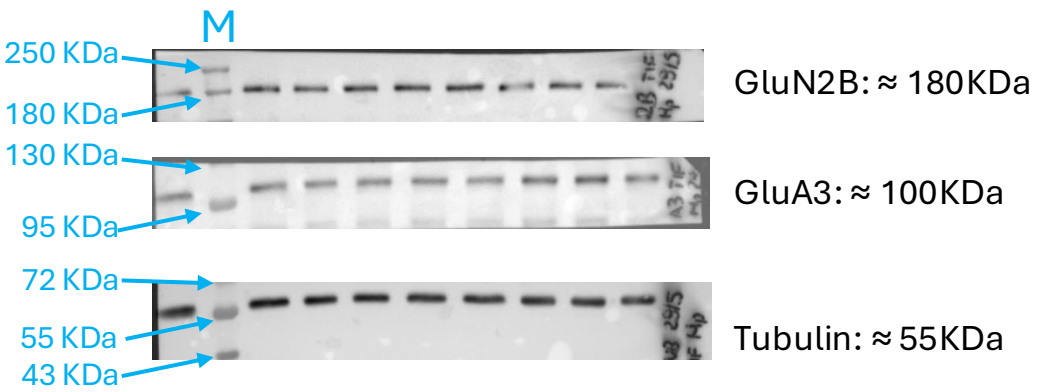


Hp, group 2

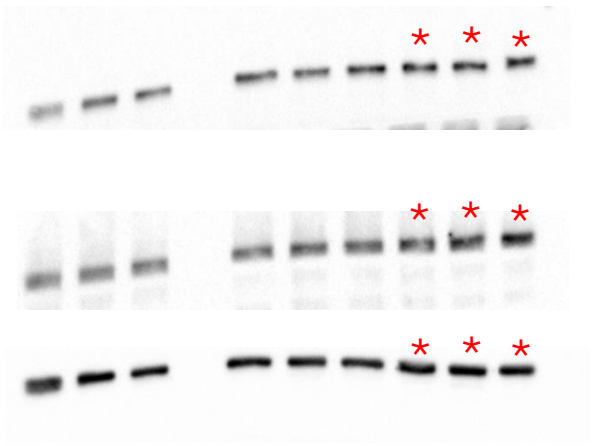
For all gels, the loading order was consistent and corresponded to the following samples: **261, 262, 290, 291, 263, 265, 264, 293, and 266**. Samples 261, 262, and 264 were +/+; samples 290, 291, and 293 were +/-; and samples 263, 265, and 266 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide. On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

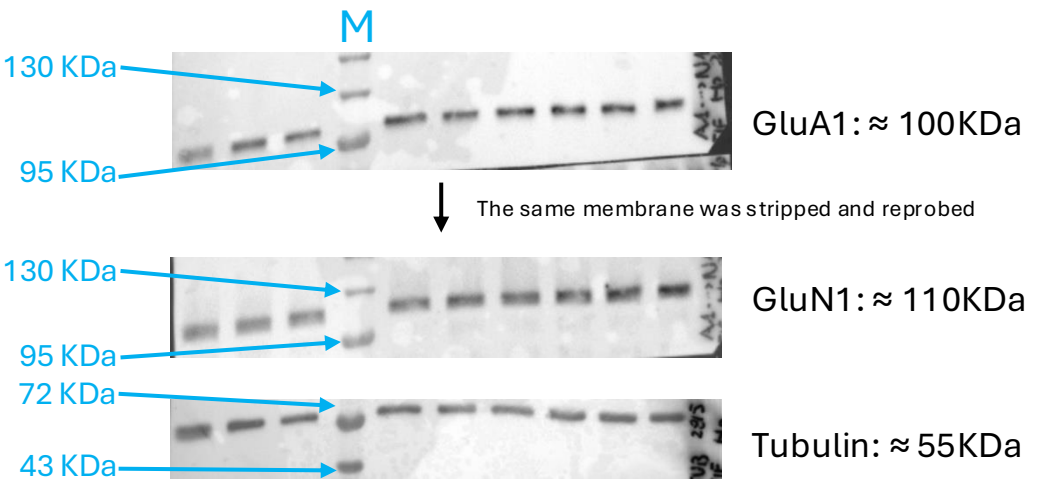
Gel 1



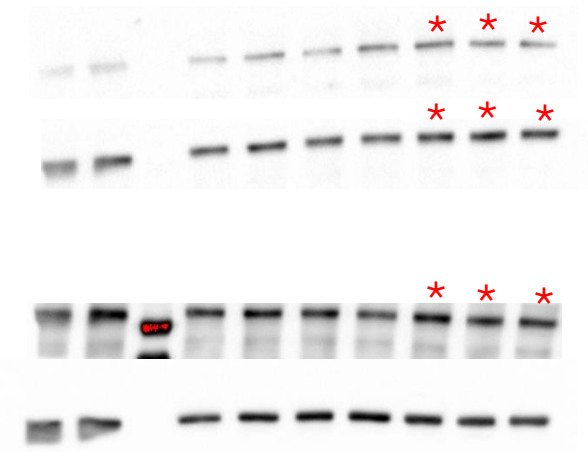


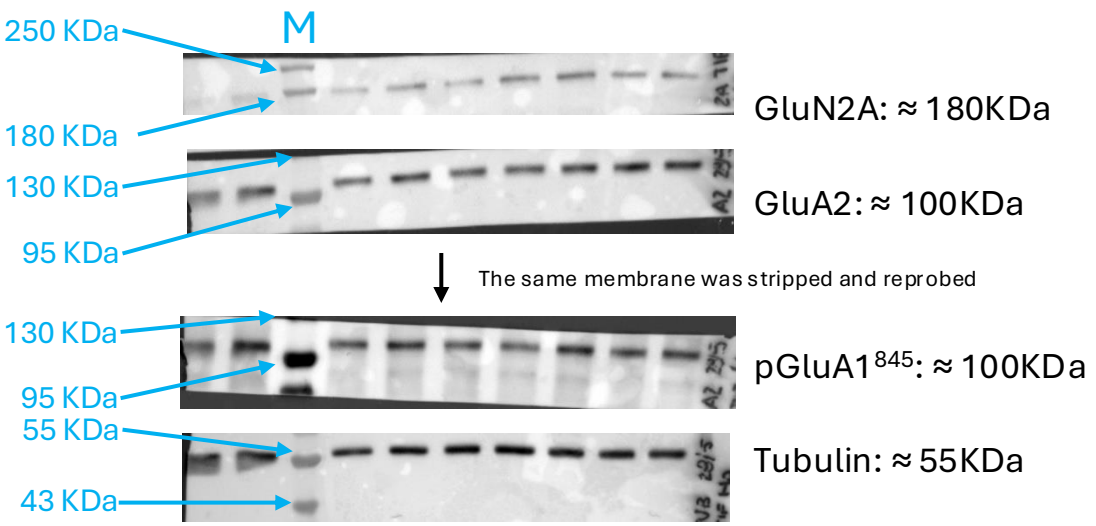
Gel 2



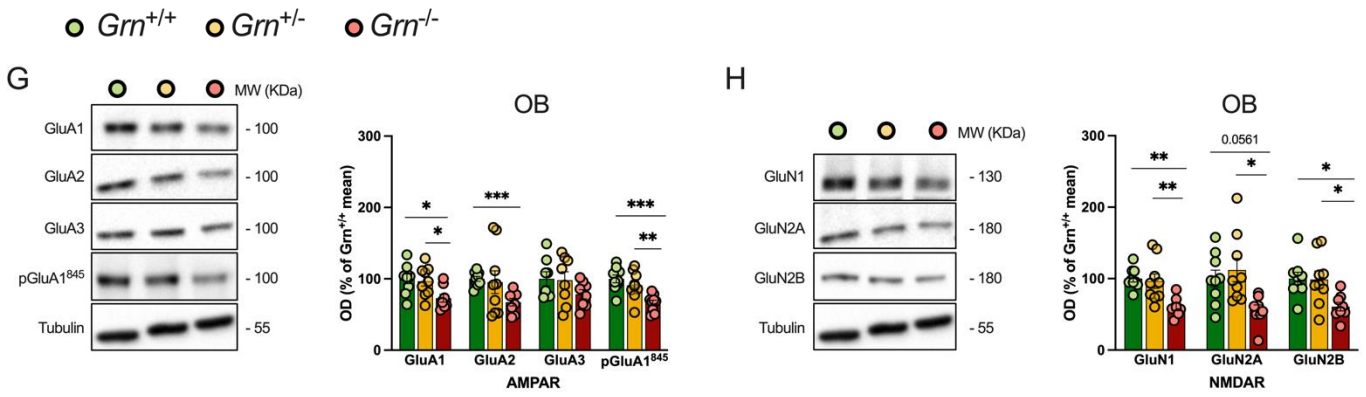


Gel 3





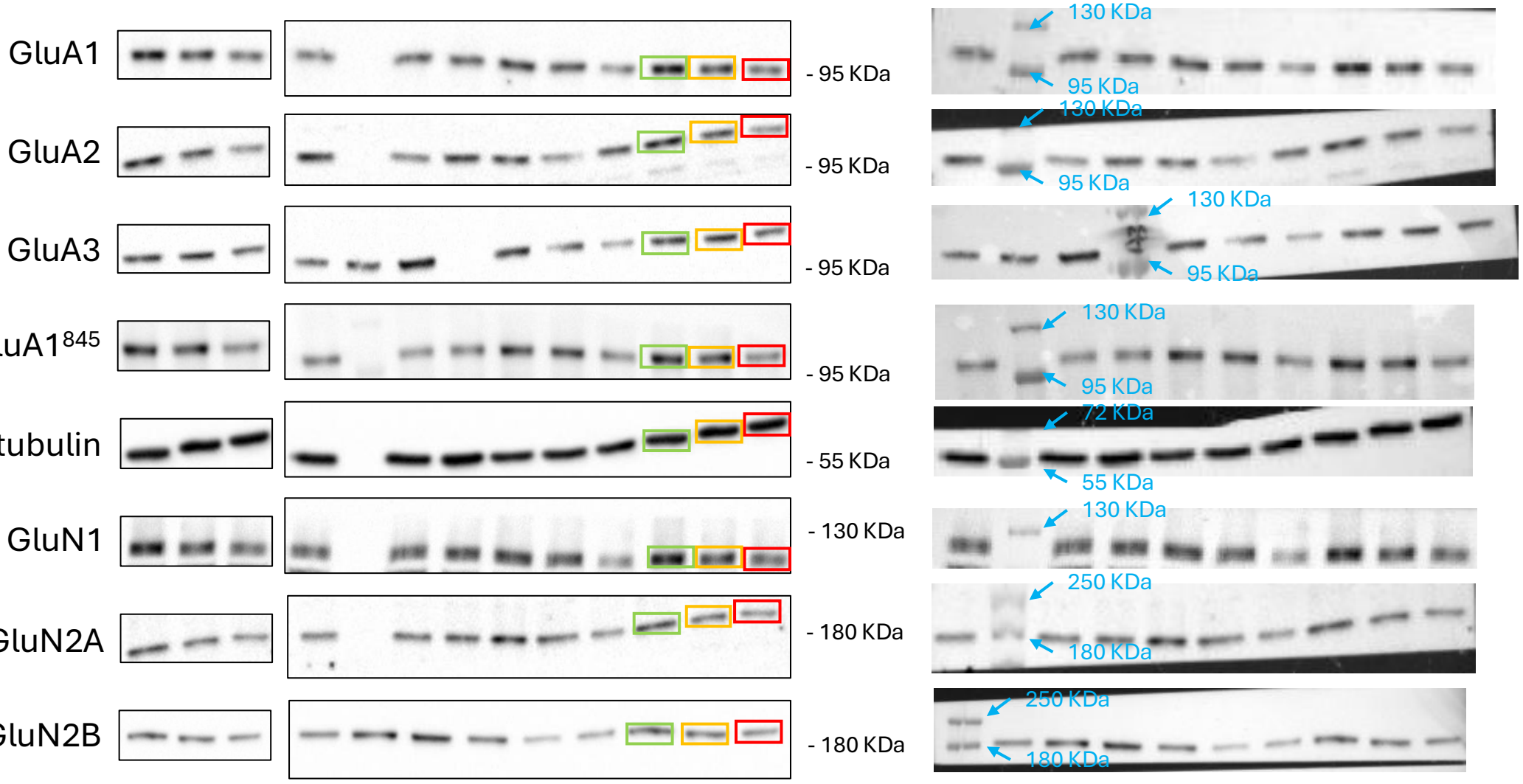
Uncropped blot, **olfactory bulb**



Boxed regions used in the main figure

Original uncropped blot

Uncropped blot with molecular weight markers

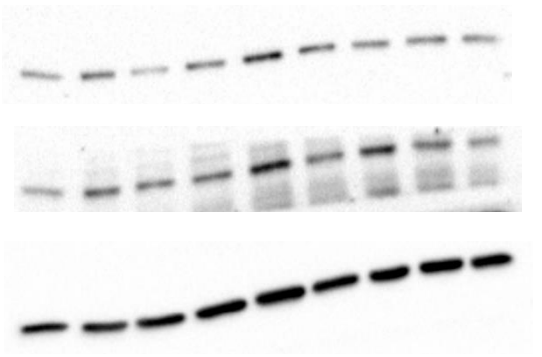


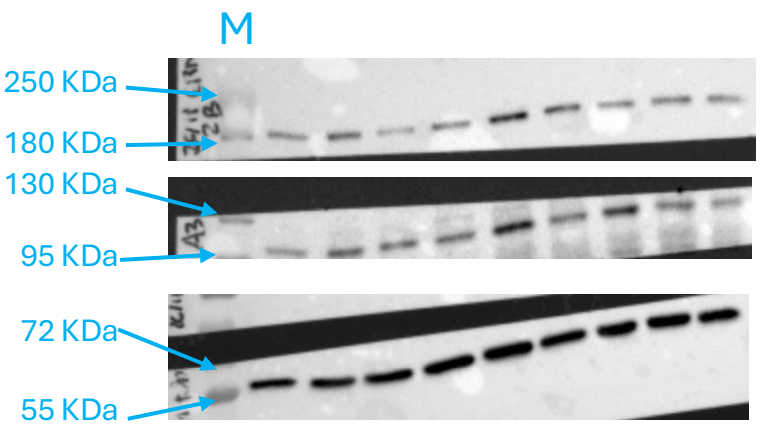
For all bands except GluA3 and GluN2B, lanes 7, 8, and 9 correspond to samples 538+/+, 534+/−, and 507−/−, respectively. For GluA3 and GluN2B, lanes 7, 8, and 9 correspond to samples 630+/+, 634+/−, and 620−/−, respectively.

Ob, group 1

For all gels, the loading order was consistent and corresponded to the following samples: **527, 514, 461, 522, 516, 466, 528, 521, 502**. Samples 527, 522, and 528 were +/+; samples 514, 516, and 521 were +/-; and samples 461, 466, and 502 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide
On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

Gel 1



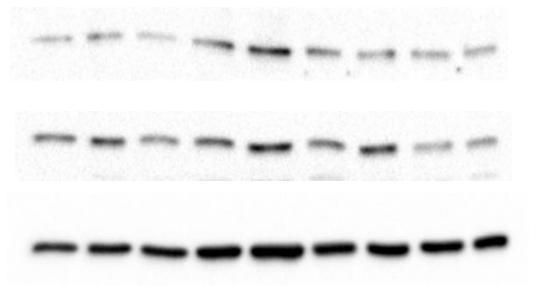


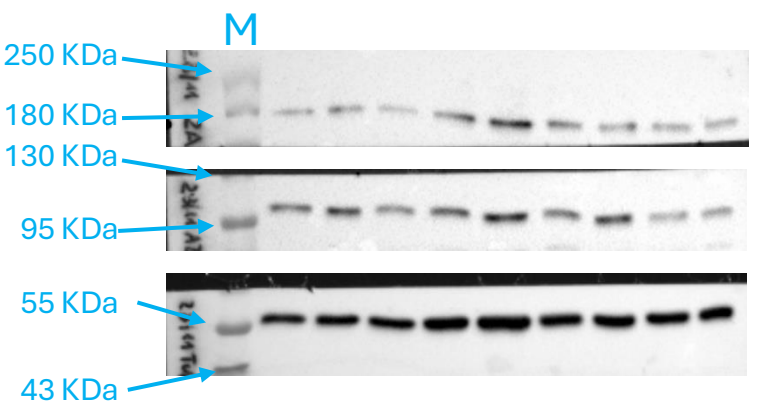
GluN2B: ≈ 180KDa

GluA3: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 2



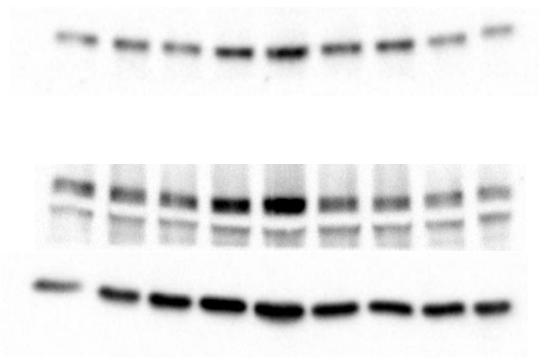


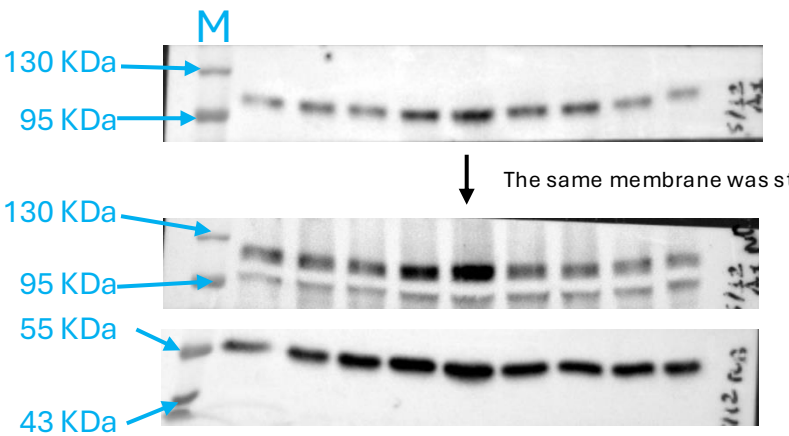
GluN2A: ≈ 180KDa

GluA2: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 3



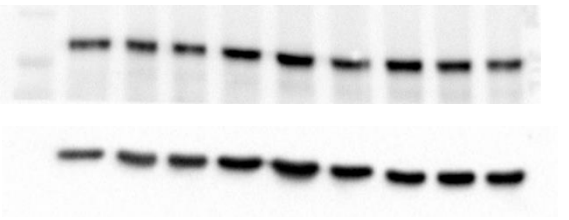


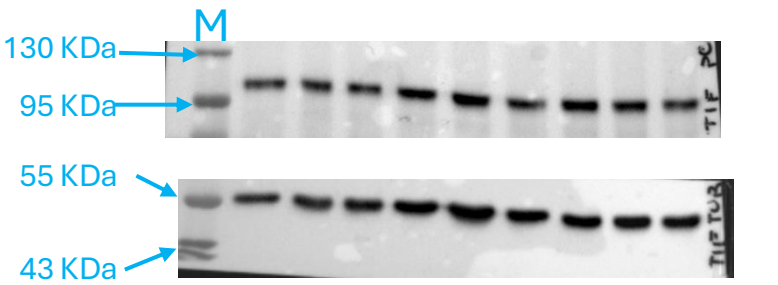
GluA1: ≈ 100KDa

GluN1: ≈ 110KDa

Tubulin: ≈ 55KDa

Gel 4





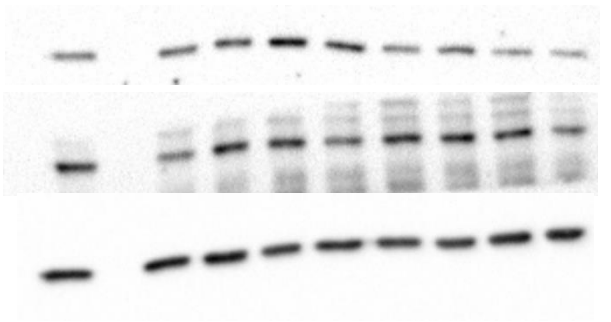
pGluA1^{845}: ≈ 100KDa

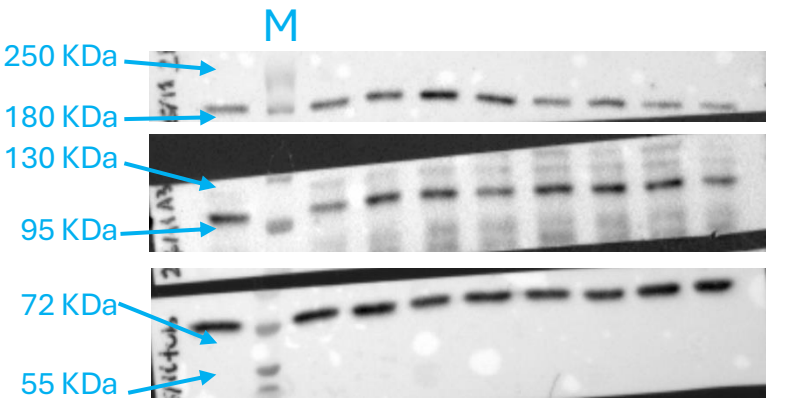
Tubulin: ≈ 55KDa

Ob, group 2

For all gels, the loading order was consistent and corresponded to the following samples: **536, 523, 503, 537, 532, 540, 538, 534, 507**. Samples 536, 537, and 538 were +/+; samples 523, 532, and 534 were +/-; and samples 503, 540, and 507 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide
On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

Gel 1



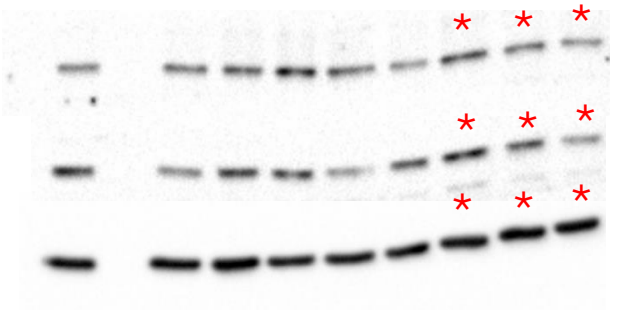


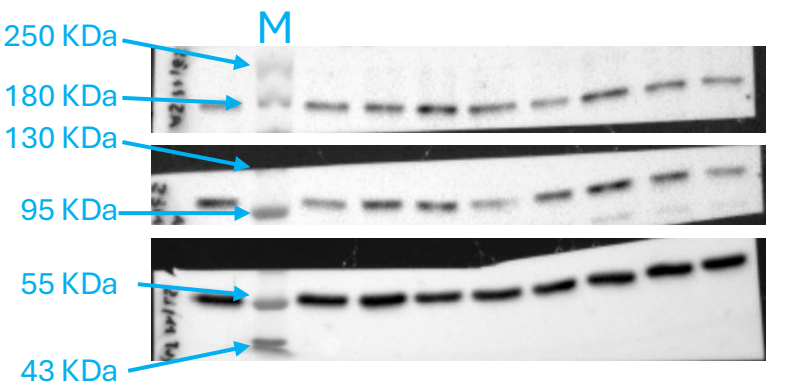
GluN2B: ≈ 180KDa

GluA3: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 2



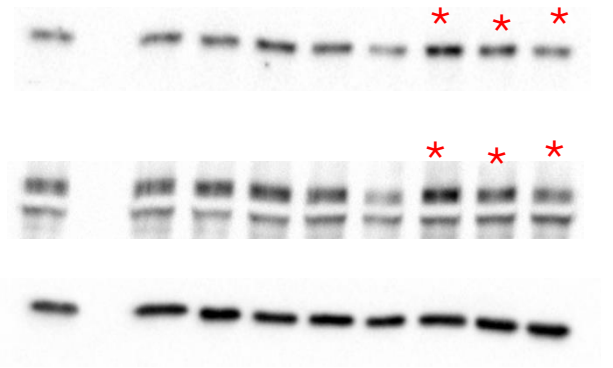


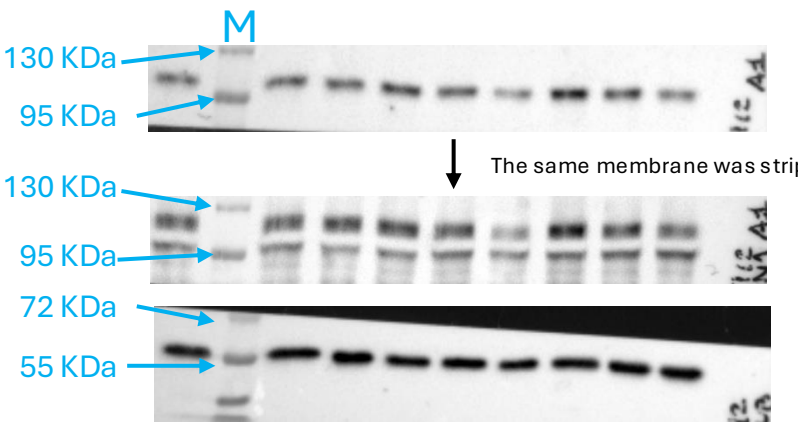
GluN2A: ≈ 180KDa

GluA2: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 3



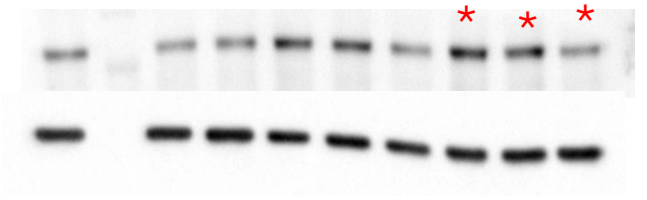


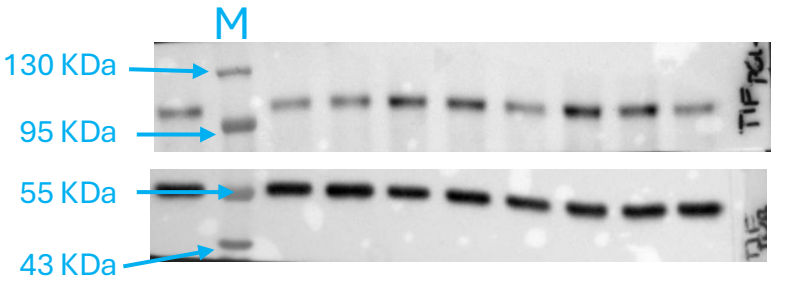
GluA1: ≈ 100KDa

GluN1: ≈ 110KDa

Tubulin: ≈ 55KDa

Gel 4





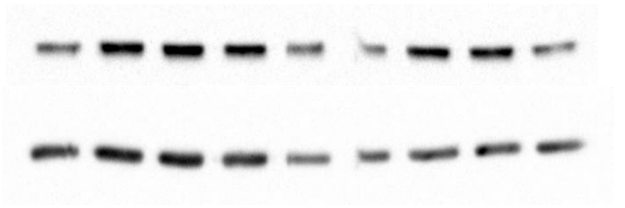
pGluA1^{845}: ≈ 100KDa

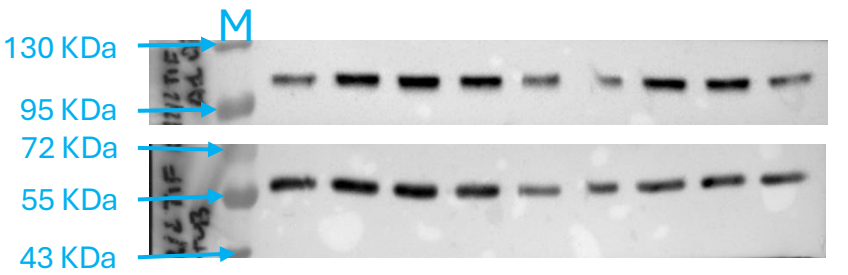
Tubulin: ≈ 55KDa

Ob, group 3

For all gels, the loading order was consistent and corresponded to the following samples: **626, 627, 604, 621, 613, 619, 630, 634, 620**. Samples 626, 627, and 630 were +/+; samples 604, 621, and 634 were +/-; and samples 613, 619, and 620 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide
On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

Gel 1

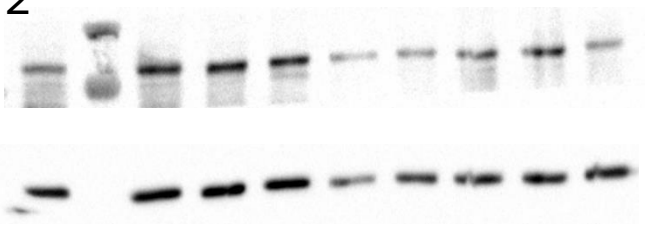


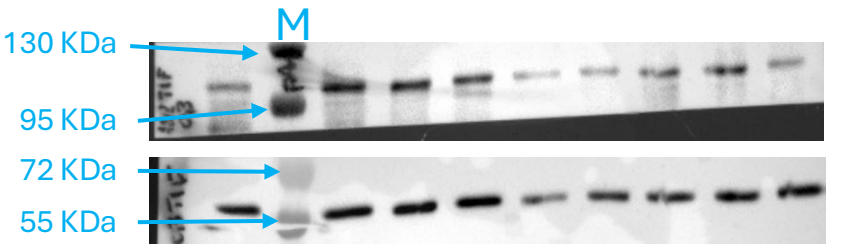


GluA1: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 2

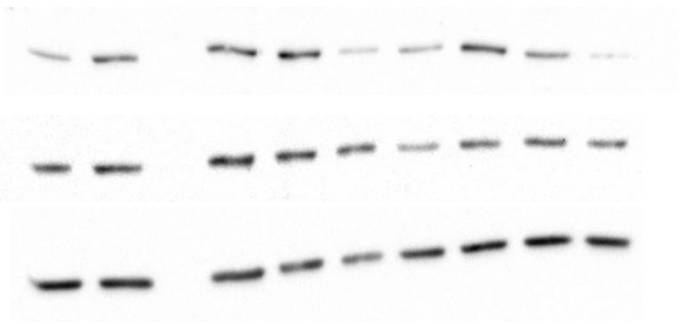


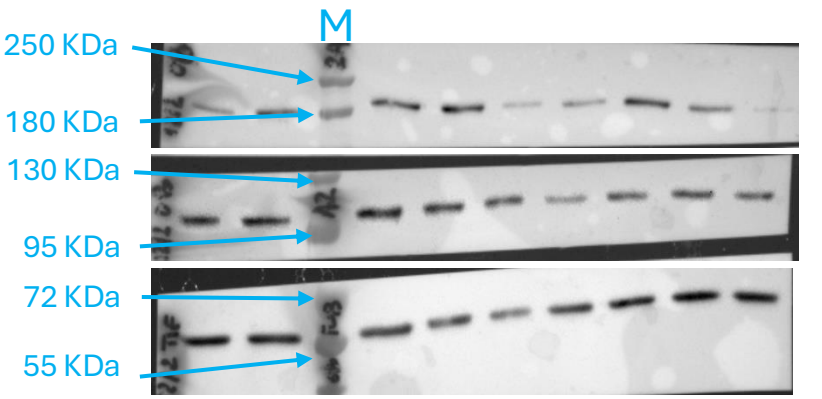


pGluA1^{845}: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 3



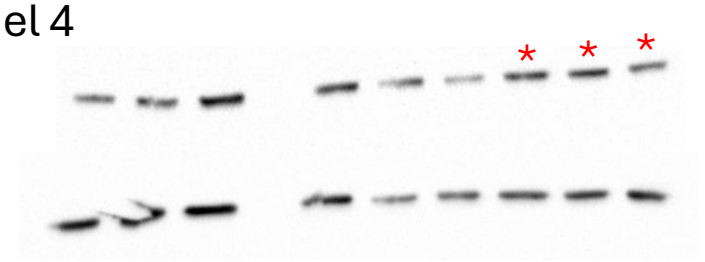


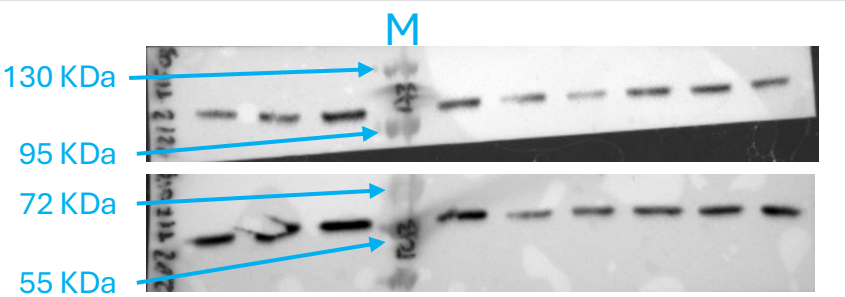
GluN2A: ≈ 180KDa

GluA2: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 4

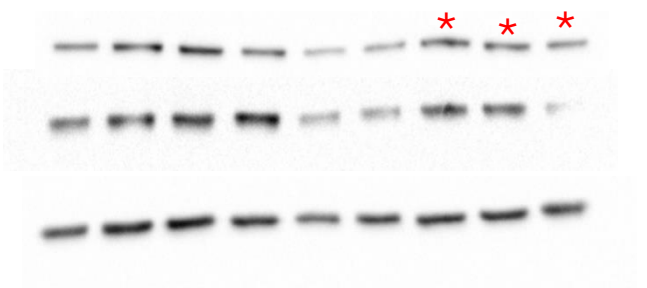


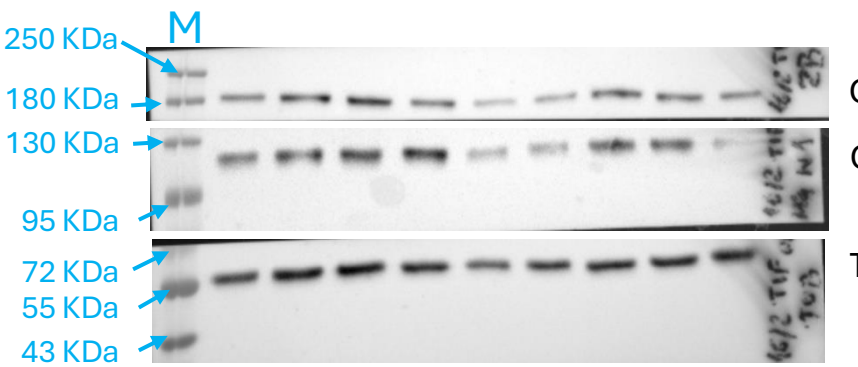


GluA3: ≈ 100KDa

Tubulin: ≈ 55KDa

Gel 5



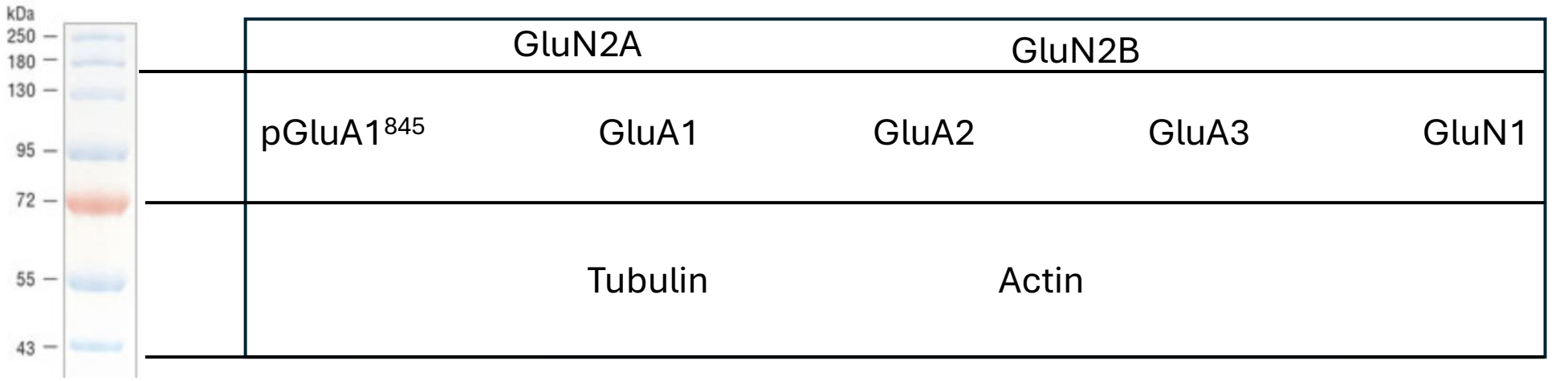


GluN2B: ≈ 180KDa

GluN1: ≈ 110KDa

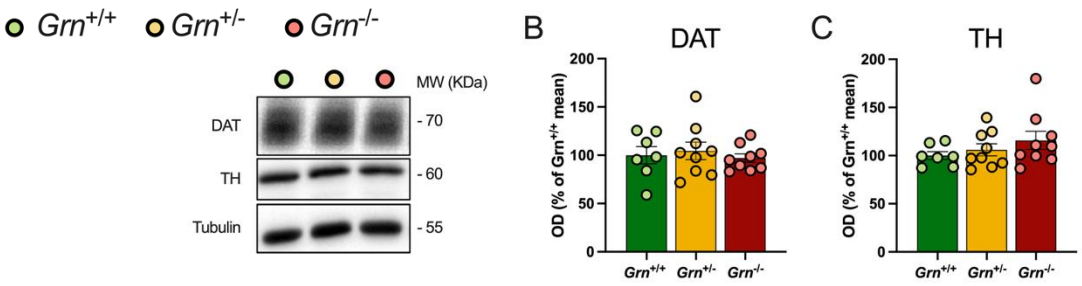
Tubulin: ≈ 55KDa

Molecular weight estimation was performed using the Color Prestained Protein Standard, Broad Range (10–250 kDa) ladder (NEB #P7719), used for all gels/blots.



The same cropping strategy was applied throughout all TIF experiments, as indicated here, according to the molecular weight ranges corresponding to the target proteins analyzed.

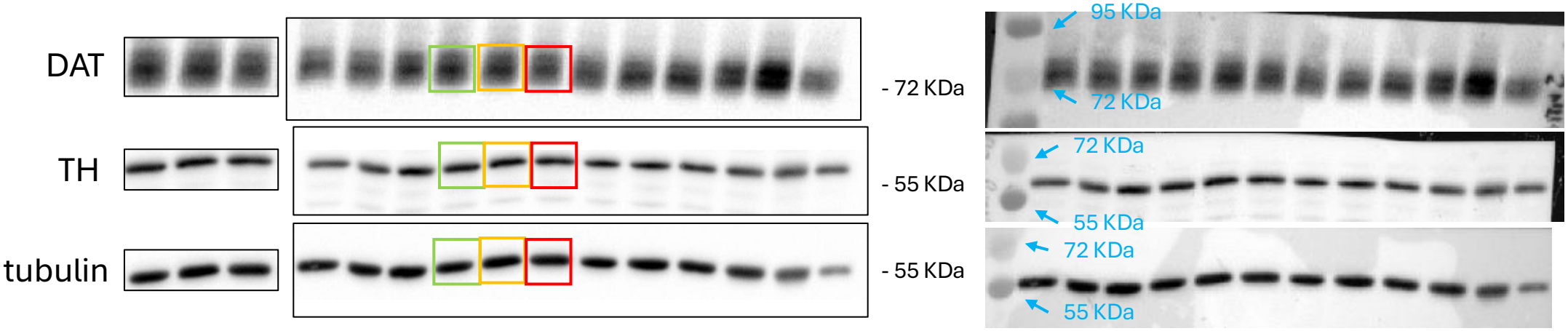
Uncropped blot, **striatum**



Boxed regions used in the main figure

Original uncropped blot

Uncropped blot with molecular weight markers

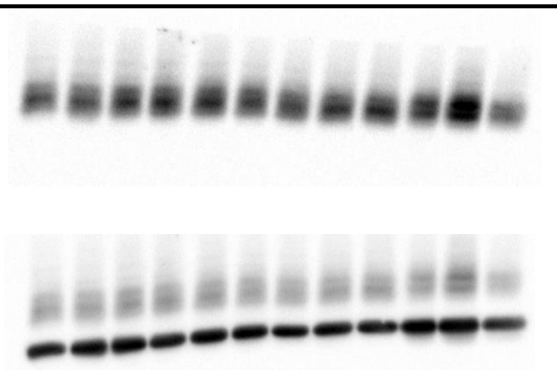


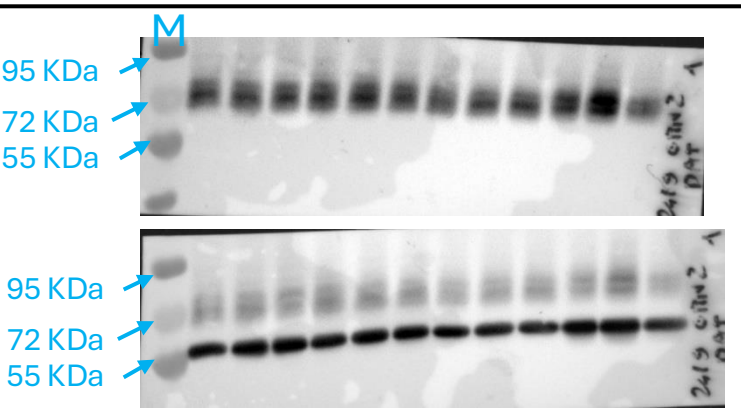
For all bands lanes 4, 5, and 6 correspond to samples 262+/+, 291+/-, 265-/-

Stx, total homogenate, all samples

For all gels, the loading order was consistent and corresponded to the following samples: **261, 290, 263, 262, 291, 265, 264, 293, 266, 211, 245, 243 (PART 1), 210, 254, 246, 252, 257, 249, 253, 247, 250, 256, 251, 248, 259 (PART 2).** Samples 261, 262, 264, 211, 210, 252, 253 were +/+; samples 290, 291, 293, 245, 254, 257, 247, 256, 248 were +/-; and samples 263, 265, 266, 243, 246, 249, 250, 251, and 259 were -/-. Sample identities are not indicated separately on each gel, as the sample loading order was identical across all experiments. Bands marked with an asterisk (*) were selected as representative bands and are also highlighted as representative in the previous slide. On the right, the gels are shown without the molecular weight marker; these images were subsequently merged with the colorimetric image to visualize the marker.

Part 1

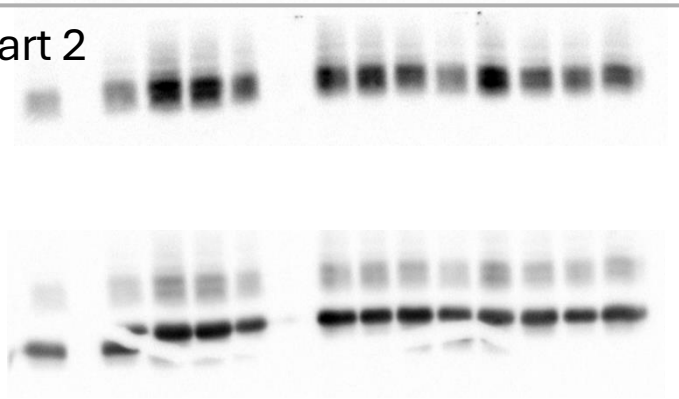


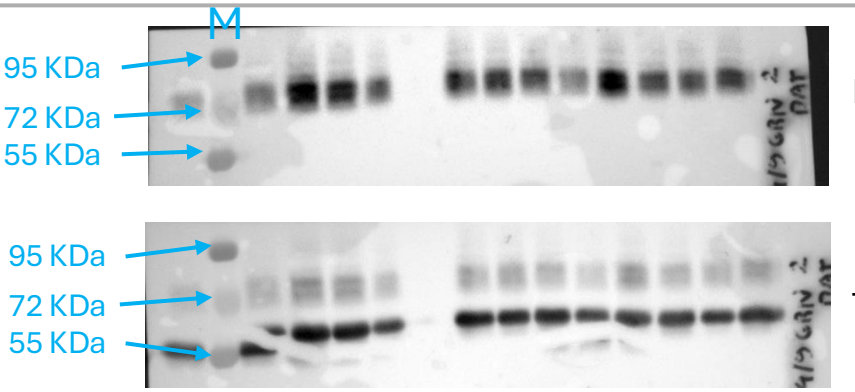


DAT: ≈ 70KDa

Tubulin: ≈ 55KDa

Part 2

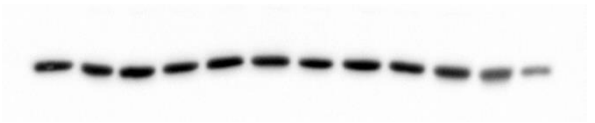


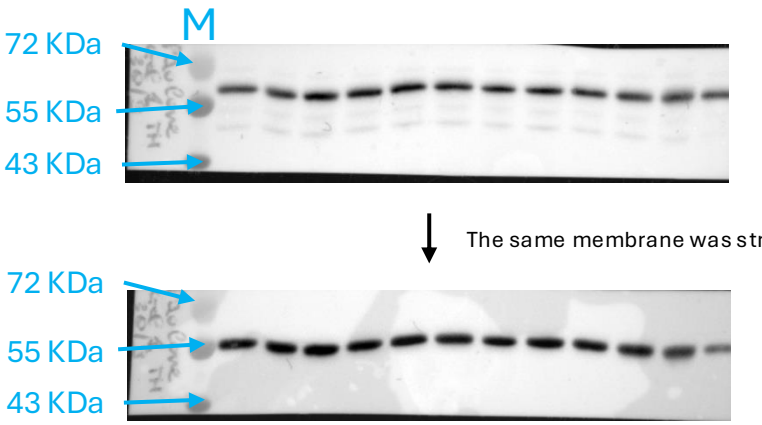


DAT: ≈ 70KDa

Tubulin: ≈ 55KDa

Part 1

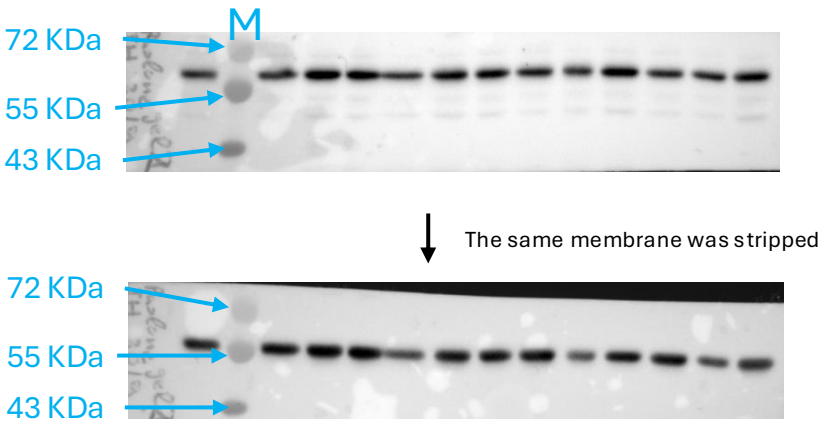




TH: ≈ 60KDa

Tubulin: ≈ 55KDa

Part 2



TH: ≈ 60KDa

Tubulin: ≈ 55KDa