

Investigating The Relationship Between Endo-lysosomal Dysfunction And TDP-43 Pathology In Alzheimer's Disease



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TDP-43 is a major pathology in ADNC and LATE-NC

- TAR DNA-binding protein of 43 kDa (TDP-43) is a key regulator of mRNA and gene expression³. Its pathological mislocalization and aggregation are the central pathology in many neurodegenerative diseases, including ALS, FTLT-DTP, and the majority of Alzheimer's disease (AD) cases^{4,5}.
- In limbic regions of the brain, TDP-43 pathology defines **limbic predominant age-related encephalopathy (LATE)**^{6,7}.
- TDP-43 dysfunction disrupts RNA splicing, leading to cryptic exon inclusion⁴. TDP-43 has been described as having both toxic gain of function and toxic loss of function.
- While TDP-43 has been linked to several endolysosomal proteins (i.e TMEM106B), the relationship between TDP-43 and the endolysosomal network is largely unknown.

SORL1, an endolysosomal protein implicated in AD, may contribute to TDP-43 pathology

- SORL1 is an endolysosomal network (ELN) trafficking protein that binds to the retromer (containing VPS35) and is responsible for the trafficking of many AD-related proteins, such as APP, through the ELN. It has emerged as a major genetic risk factor for AD^{1,2}.
- The R953C (c2857C>T;p.Arg953Cys) SORL1 variant, recently identified in our group, results in loss of function with protein retention in the ER, enlarged early endosomes, and reduced ELN function.
- Notably, **all carriers of this variant exhibit high ADNC alongside LATE, while familial non-carriers with AD lack TDP-43 pathology**
- The R953C variant provides a unique opportunity to investigate ELN contributions to TDP-43 pathology and combined AD+LATE-NC.

To examine how TDP-43 may be related to SORL1 and ELN-driven changes, we (1) assess TDP-43 and ELN dysfunction in R953C variant post-mortem tissue and hiPSC-derived neurons, (2) investigate how toxic gain vs loss of TDP-43 function may contribute to ELN phenotypes

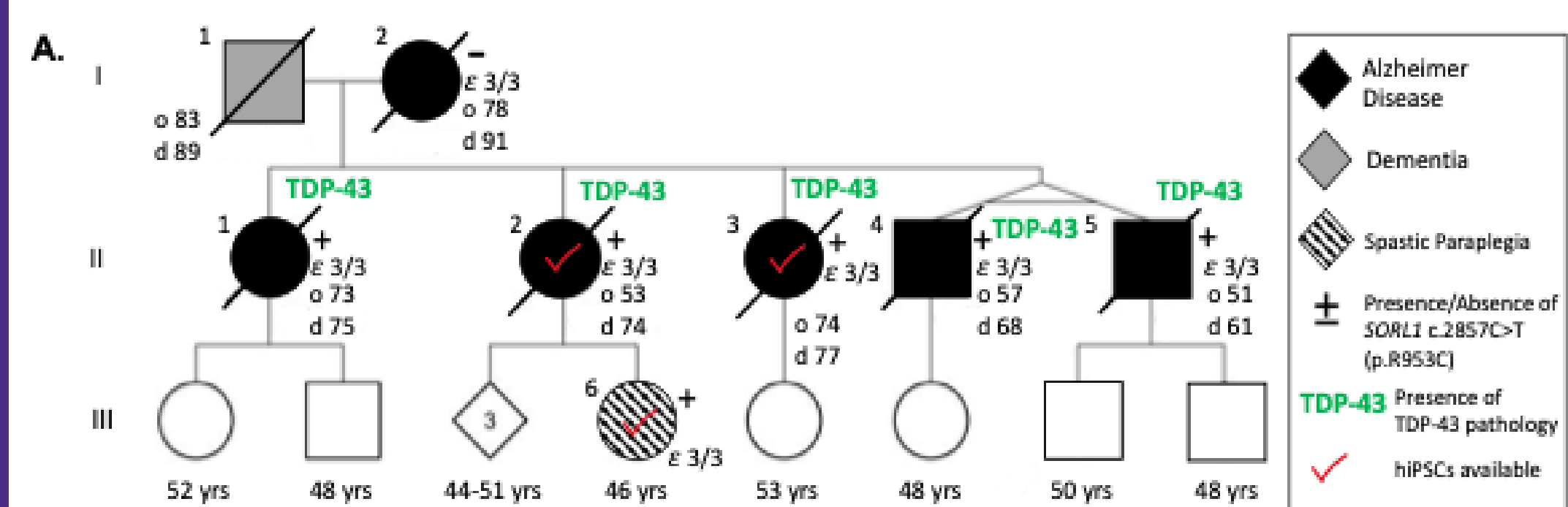


Fig. 1. (A). Pedigree of SORL1 R953C family. TDP-43 indicates presence of LATE-NC on autopsy. Red checkmarks indicate cases where we have reprogrammed hiPSCs available.

Experimental approach

Generating hiPSC-derived cortical neurons

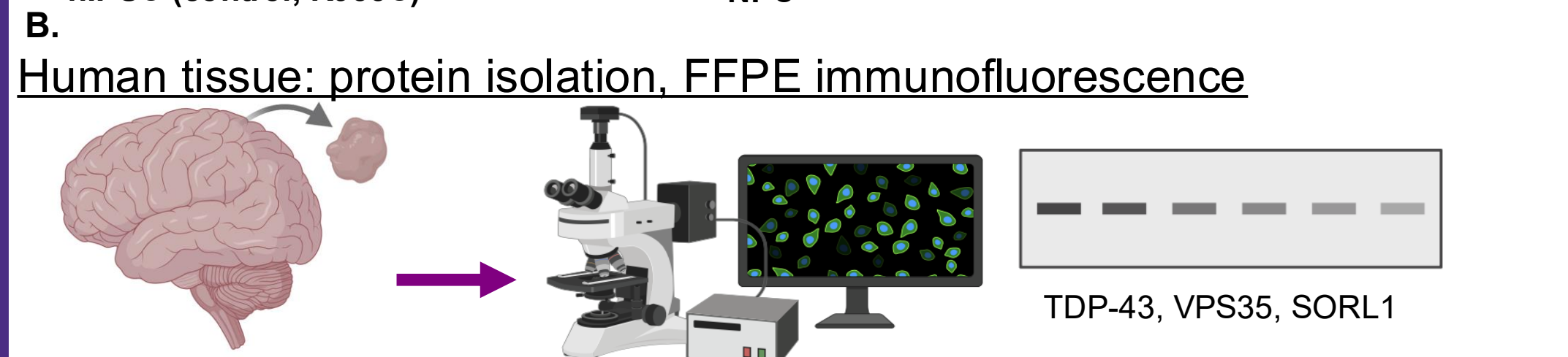
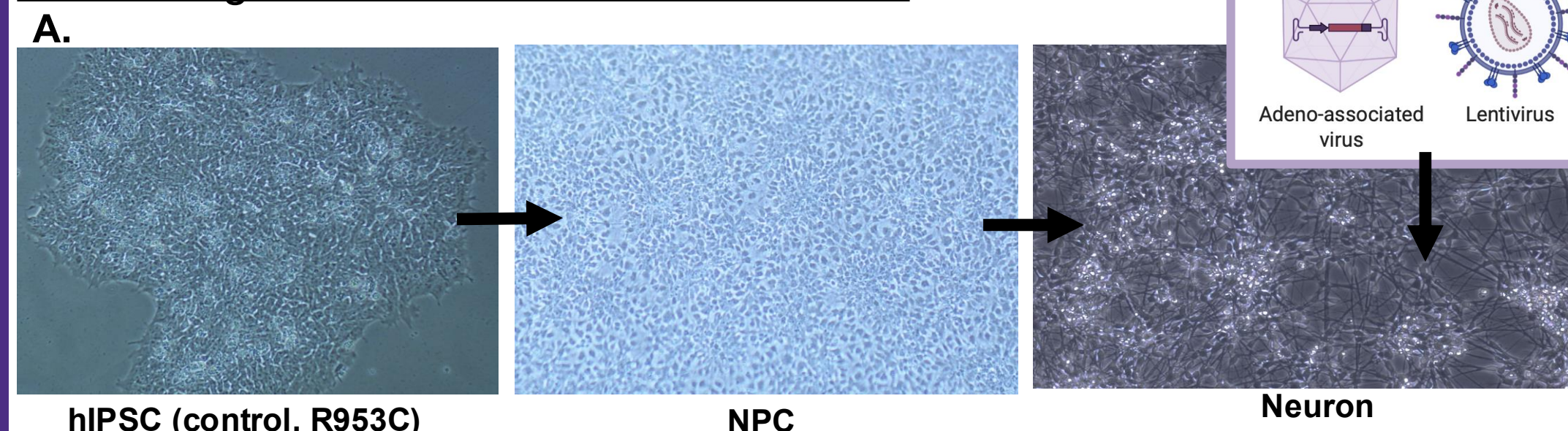
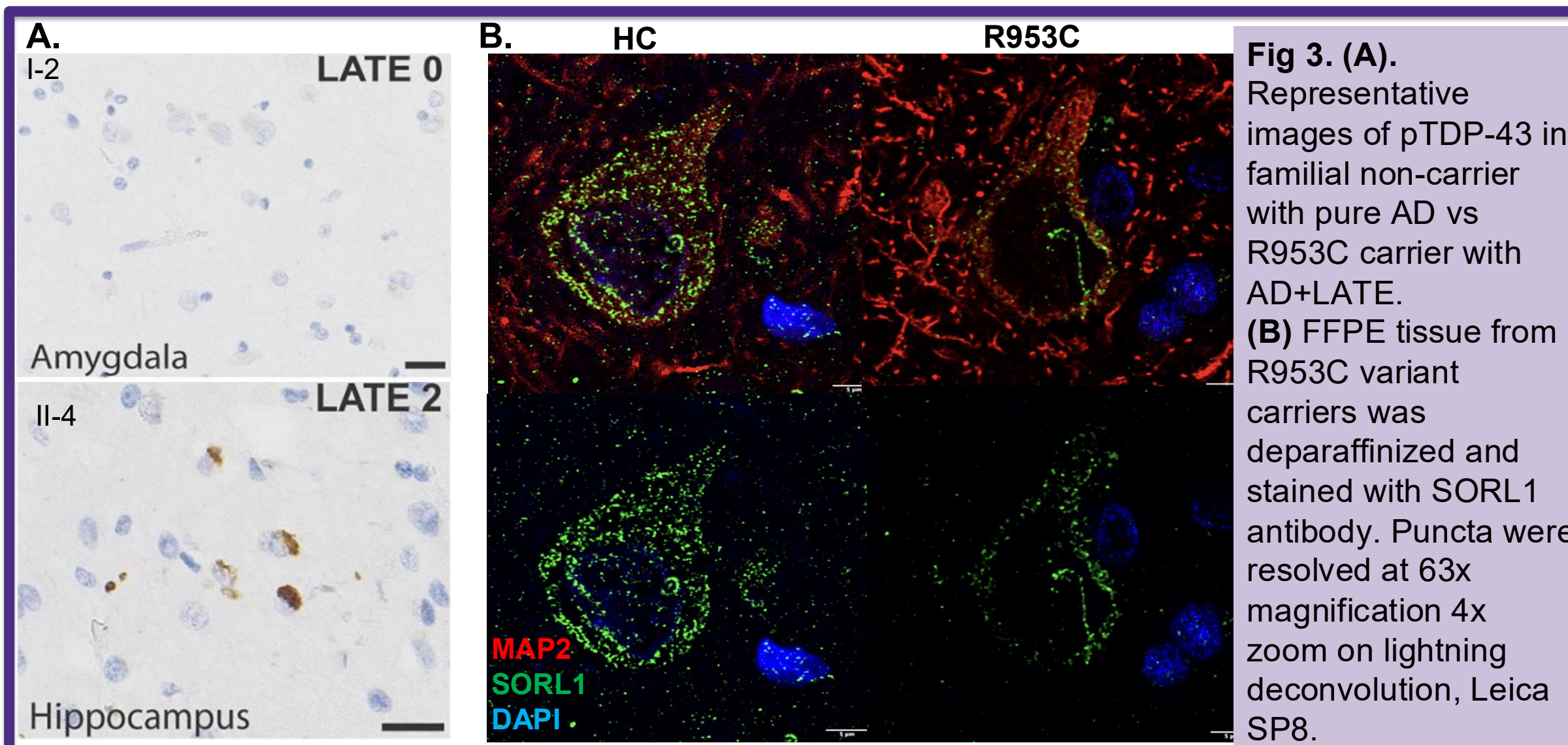
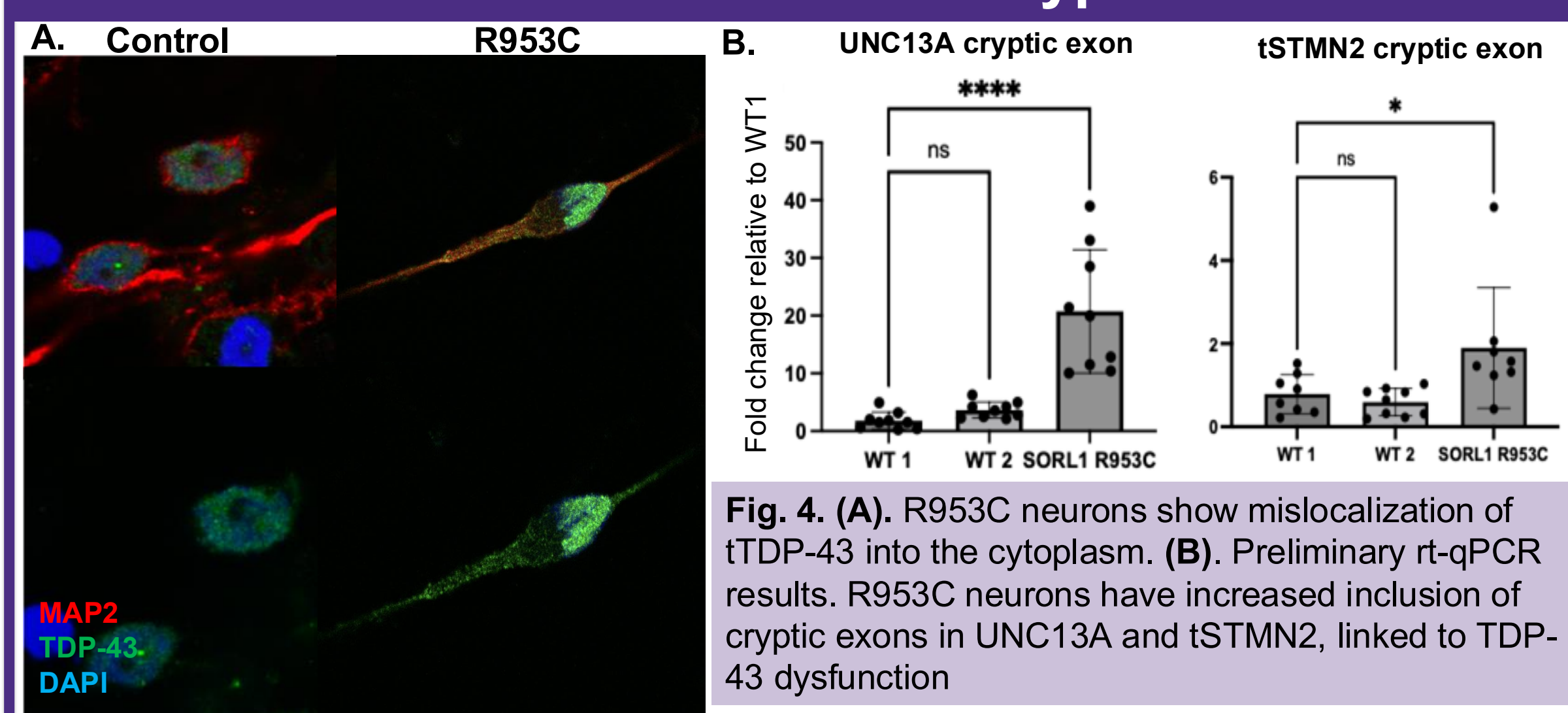


Fig. 2. (A) Standard protocols in the Young lab for the generation of hiPSC-derived cortical neurons. After differentiation, downstream assays are used to assess protein expression and localization for TDP-43 and ELN markers. Neurons are treated with Syn hTDP-43 AAV at 1x10⁴ vg/cell for 3 days. For lentivirus shRNA TDP-43 knock-down, neurons are treated with 1x vg/cell for 14 days (B). Pipeline to assess protein expression in human tissue after autopsy.

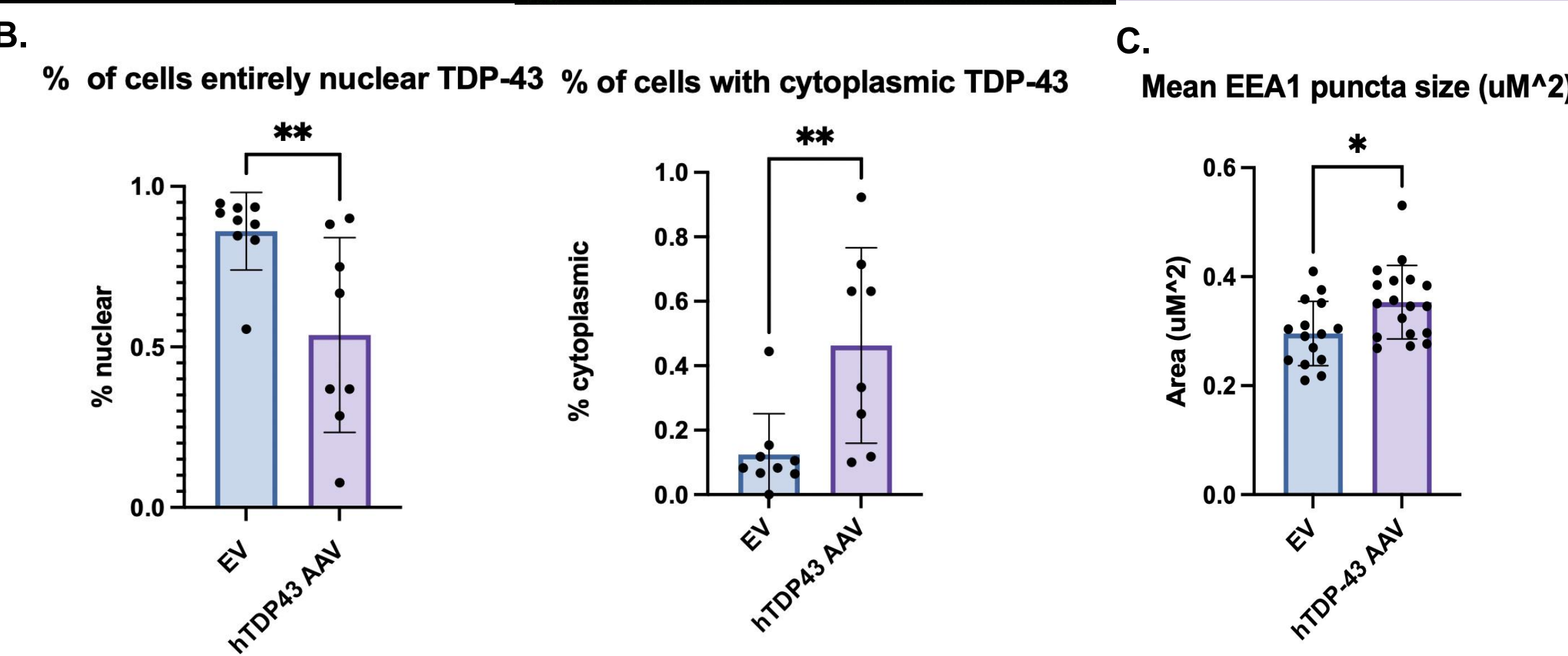
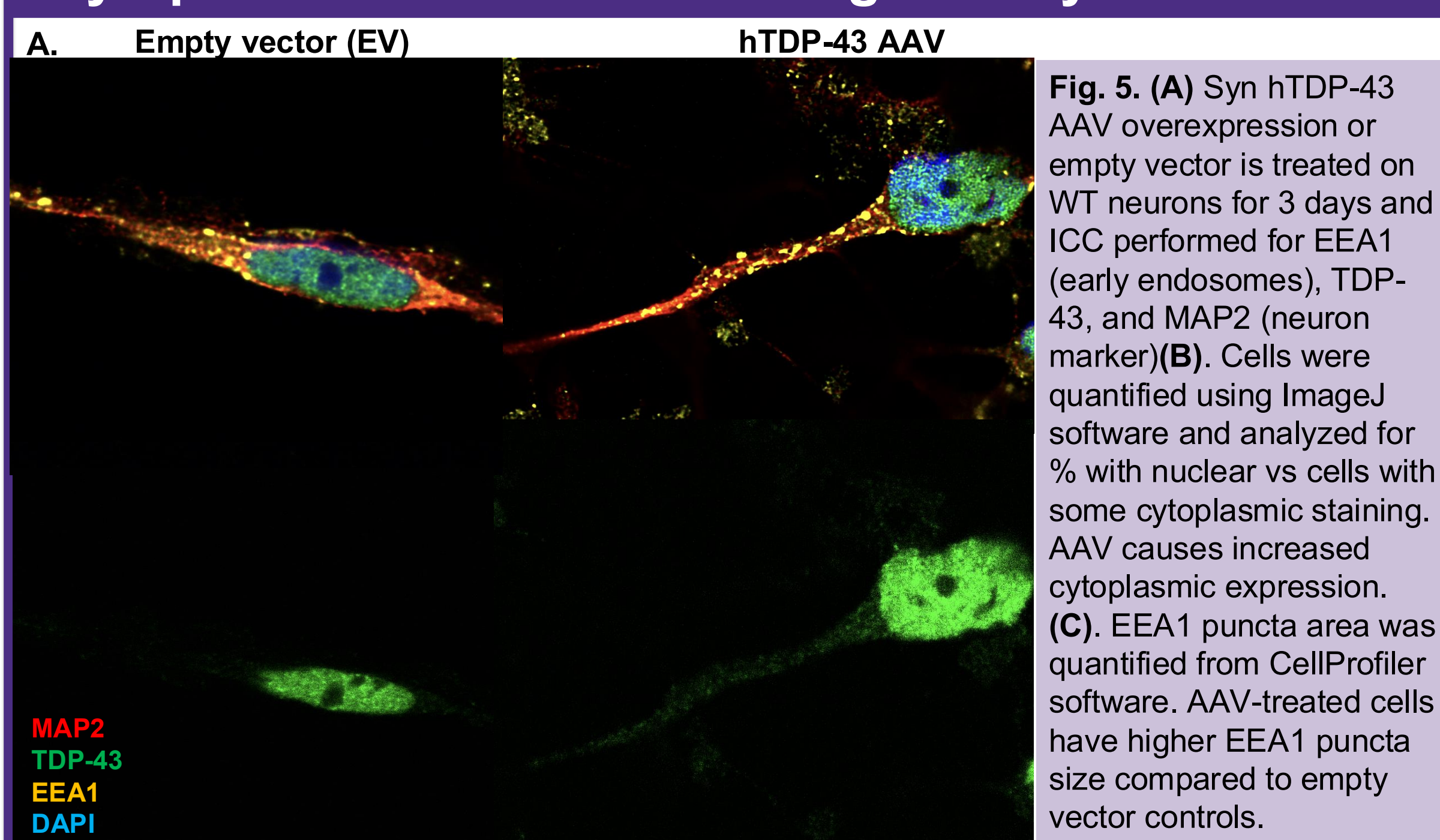
SORL1 R953C carriers show TDP-43 pathology and reduced SORL1 immunofluorescence



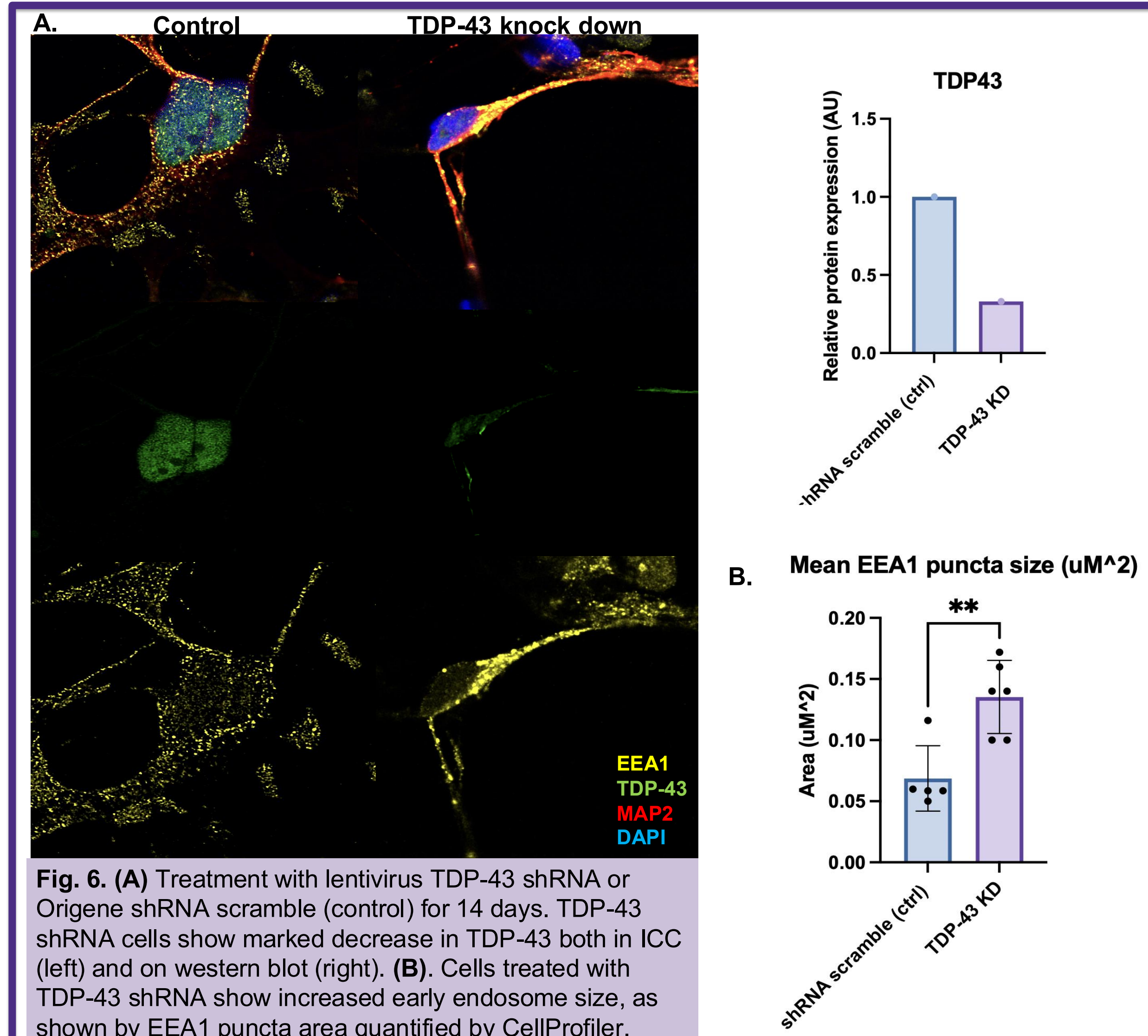
hiPSC-derived neurons from R953C carriers show mislocalized tTDP-43 and cryptic exons



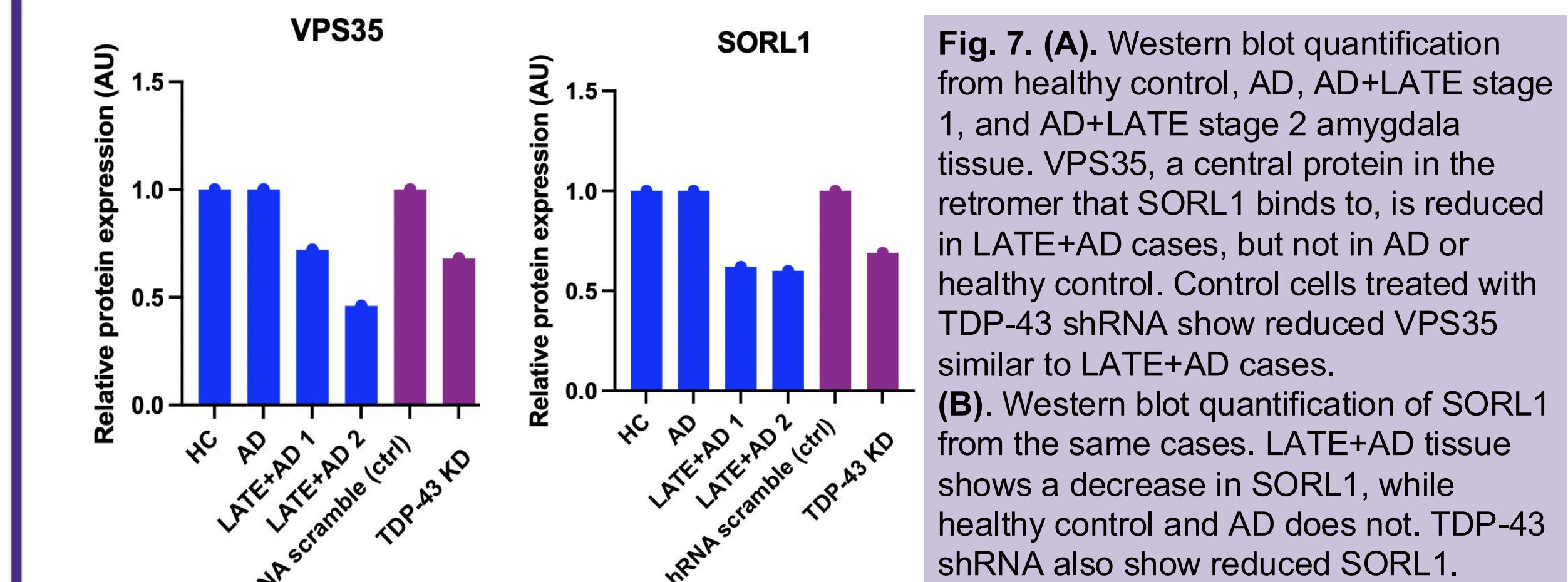
TDP-43 overexpression in control neurons results in cytoplasmic TDP-43 and enlarged early endosomes



TDP-43 shRNA treatment in control cells results in decreased tTDP-43 and enlarged early endosomes



Retromer component, VPS35, as well as SORL1 may be reduced in AD+LATE human cases and TDP-43 LOF models in vitro



Conclusions and future directions

The presence of TDP-43 in the SORL1 R953C family provides a unique opportunity to investigate TDP-43 dynamics. Our findings point towards potential mechanisms of TDP-43 development in AD. Here, we show that

- SORL1 R953C tissue and hiPSC-derived cortical neurons have reduced SORL1 and increased cytoplasmic TDP-43.
- While both AAV and lentivirus experiments in control cells show enlarged early endosomes, they're likely through different mechanisms.
- Treatment with AAV overexpression demonstrates potential toxic gain of function (cytoplasmic mislocalization, enlarged early endosomes), while lentivirus shRNA knock-down shows potential toxic loss of function (reduced VPS35, reduced SORL1, enlarged early endosomes likely due to VPS35 and SORL1 reduction).

References and Acknowledgements

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