



Long Distance Neurodegenerative Signaling Mediated by Neurotrophin Receptor p75NTR



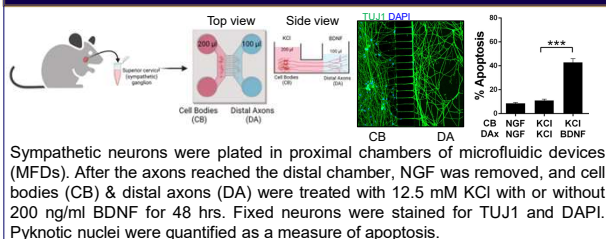
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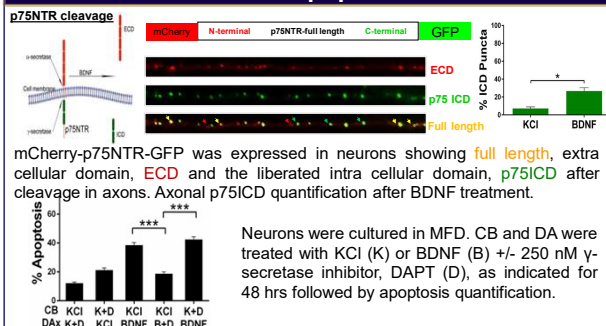
Introduction

- Neurotrophin signaling governs the balance between neuron survival & death.
- Increased expression of pan neurotrophin receptor p75NTR and proneurotrophin, proNGF has been reported in post mortem brain of Alzheimer's disease (AD) patients and mice models.
- Mature neurotrophins bind to their cognate receptor tyrosine kinases to promote long range retrograde survival signaling from distal axons (DA) to the cell body (CB) by forming "signaling endosomes".
- On the contrary, p75NTR activation in distal axons results in retrograde apoptotic signaling in neurons which has been recently realized.
- Despite extensive research on retrograde survival signaling by neurotrophins, the mechanisms for retrograde death signaling mediated by p75NTR remains unclear.
- Here we investigated degenerative signaling endosome biogenesis and their long-distance transport resulting in neuron death. Understanding this mechanism may provide insights into neuron death in Alzheimer's disease.

Retrograde apoptotic signaling in sympathetic neurons



p75NTR cleavage in distal axons is necessary for neuron apoptosis



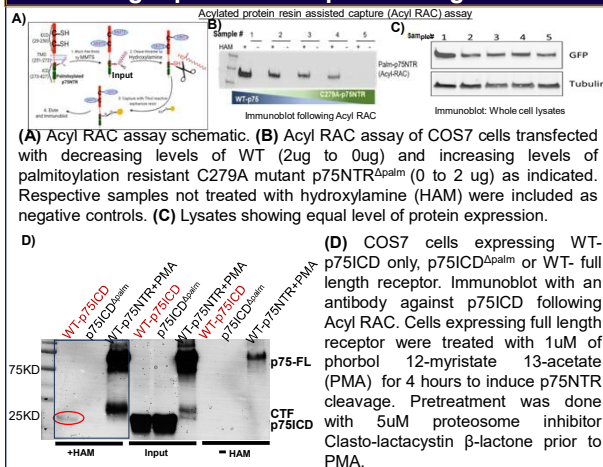
Palmitoylation of p75NTR at well conserved cysteine residue

Regulated intramembrane proteolysis of p75NTR	Conserved Cysteine residue
Human	275 WNSCKQNKQGA 285
Rat	276 WNSCKQNKQGA 286
Mouse	278 WNSCKQNKQGA 288
Chick	262 WNSCKQNKQGA 272
Xenopus	243 YTCCKQKKQLA 251
Zebra fish	265 WNSCKQNKQAA 275

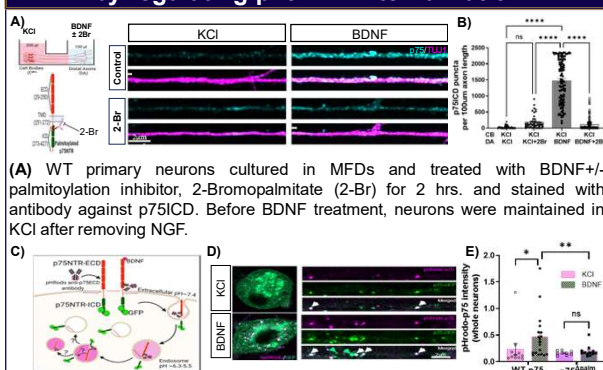
P75NTR is palmitoylated at an evolutionarily conserved cysteine (Barker et al, 1994).

Hypothesis: Upon losing its transmembrane domain following receptor cleavage, palmitoylation facilitates vesicular localization of p75 ICD and retrograde transport of death signals in neurons.

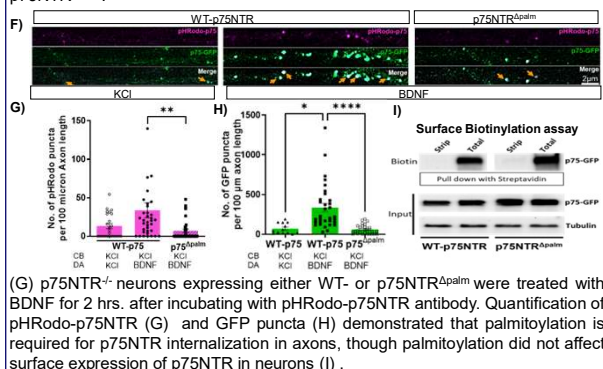
Acyl RAC assay to detect palmitoylation of full length p75NTR and p75ICD fragment



Palmitoylation targets p75ICD to axonal vesicles by regulating p75NTR internalization

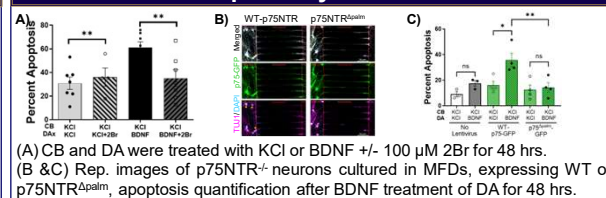


(A) Schematic for pHrodo-p75NTR internalization assay. Antibody against p75NTR-ECD was tagged with a pH sensitive dye pHrodo which fluoresce only at acidic pH. Rep. images showing neurons expressing WT-p75NTR. Quantification of BDNF treated neurons showed reduced internalization of p75NTR^{Δpalm}.

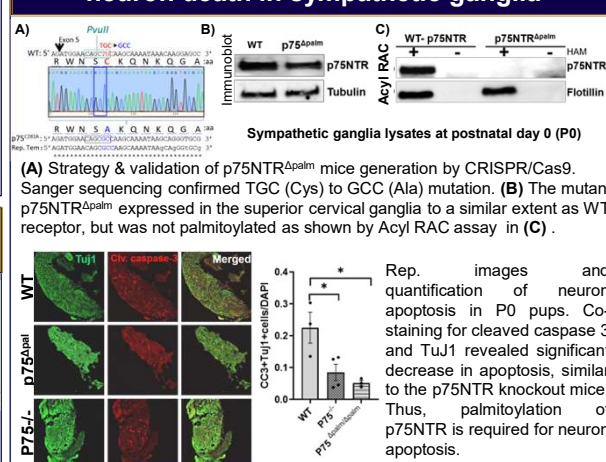


(G) p75NTR^{Δpalm} neurons expressing either WT- or p75NTR^{Δpalm} were treated with BDNF for 2 hrs. after incubating with pHrodo-p75NTR antibody. Quantification of pHrodo-p75NTR (G) and GFP puncta (H) demonstrated that palmitoylation is required for p75NTR internalization in axons, though palmitoylation did not affect surface expression of p75NTR in neurons (I).

Retrograde neuron death require p75NTR palmitoylation



p75NTR palmitoylation is necessary for neuron death in sympathetic ganglia



Summary and Conclusion

- Localized cleavage of p75NTR within axons generated p75ICD as a retrograde death signal.
- p75NTR palmitoylation represents a mechanism for attaching p75ICD to the retrograde transport vesicles resulting in biogenesis of degenerating signaling endosomes and long distance death signaling in neurons.
- Current efforts are focused on determining the role of p75NTR palmitoylation in cholinergic neuron death to understand the mechanism of Alzheimer's disease onset and progression.
- In light of evidence for disrupted neurotrophin signaling in AD and our demonstration of p75ICD as a degenerative signal, axonal transport of both pro-survival and pro-apoptotic signals must be considered in designing therapeutic strategies.

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