TDP43 inclusions are re-routes to autophagy by the activity of the small chaperone HspB8.

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TDP43 is one of the most important protein involved in the pathogenesis of amyotrophic lateral sclerosis (ALS). In ALS patient neurons TDP43 mislocalizes in the cytoplasm forms inclusions that contain also two fragments of TD43 of 35 and 25 kDa. The presence of these fragments is linked to ALS pathogenesis.

In these study we analyzed TDP43 and its disease associated fragments of 35 and 25 kDa in NSC34 cells. By immunofluorescence we observed that TDP25 fragment had a higher clearance than the full length form, but when unsufficiently removed it formed large inclusions in the cytoplasm that colocalized with p62 bodies. In Filter Retardation Assay TDP43 fragments were not detectable due to their higher clearance, while in Western Blot we detect a higher quantity of SDS soluble TDP25. We isolate TDP25 insoluble inclusion using a detergent with a higher solubility power than PBS. By a NP40 extraction we found that most of the TDP25 was detected in the NP40 insoluble fraction.

Inhibition of degradative systems with MG132 and 3MA showed that all the three forms of TDPs were degraded by the proteasome while only TDP25 was degraded by autophagy.

HspB8 is a protein involved in the cargo delivery to autophagy. It forms a complex with Hsp70 and Bag3 that drives the aggregates to p62 that insert them in autophagosomes. As TDP25 colocalized with p62 we hypotized that the aggregates could be reroutes to autophagy by this pathway. We overexpress HspB8 and found that it greatly reduce NP40 insoluble fraction of TDP25.

In conclusion we found that autophagy is involved in removing aggregates of TDP25 fragments and that facilitating autophagy by overexpressing HspB8 greatly reduced the accumulation of ALS associated TDP43 fragments.

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