Pathological mechanism in ALS

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LARGE AND SMALL EXTRACELLULAR VESICLES MAY CONTRIBUTE TO THE PROPAGATION OF ALS AND FTD CARRYING TOXIC TDP SPECIES AND POTENTIALLY HARMFUL MIRNAS

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BACKGROUND-AIM

Extracellular vesicles (EVs) are spherical particles, classified in large (LVs) and small vesicles (SVs), composed by a bilayer proteolipid membrane and carryig proteins, RNA and DNA. In our previous studies we demonstrated that both LVs and SVs play a role in the disposal of the insoluble neurotoxic TAR DNA-binding protein 43 (TDP-43) and its C-terminal fragments of 35 (TDP-35) and 25 KDa (TDP-25), the main components of the Amyotrophic Lateral Sclerosis (ALS) and Frontotemporal Dementia (FTD) associated pathological aggregates. Moreover, we observed an increased in their secretion after the impairment of the protein quality control (PQC) system [i.e. chaperone proteins, the ubiquitin proteasome system (UPS) and the autophagic pathway], a common condition observed both in ALS and FTD. Since TDP-43 is an RNA-binding protein, also involved in miRNA biogenesis, and knowing that EVs also contain miRNAs, we wondered whether PQC impairment could also affect the miRNA content of EVs.

METHODS

We studied the miRNA content of LVs and SVs isolated from the culture medium of immortalized neuronal NSC34 cells treated or not with MG132 and NH4Cl (respectively UPS and autophagy inhibitors). miRNA libraries were generated using Small RNA-Seq Library Prep Kit (Lexogen) and sequenced on a NextSeq 500/550 (Illumina). Interaction prediction was carried out on TarBase v.8 database.

RESULTS

We found a total of 91 Differentially Expressed (DE) (log Fold Change (FC) >1 and <-1) microRNAs in treated-EVs compared to untreated EVs. No DE miRNA were found in NH4Cl-LVs, only 7 miRNA were DE in MG132-LVs and of the 82 miRNAs in MG132-SVs and 66 in NH4Cl-SVs, 43 were in common. Interestingly, one of the most enriched pathway targeted by commonly DE SVs-miRNAs is the prion disease.

CONCLUSIONS

In conclusion, from our observations, we can assume that, in disease condition, EVs, enriched in both toxic TDP-43 species and potentially harmful miRNA, may contribute to the propagation of the disease from affected to healthy cells.