Hexanucleotide repeat expansions in C9orf72 impair microglial activation and result in a defective glial response in ALS

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Although the involvement of microglia in amyotrophic lateral sclerosis (ALS) is recognized, the precise underlying molecular mechanisms remain elusive. We generated single nuclei transcriptomic profiles from the spinal cord and motor cortex of sporadic (sALS) and C9orf72 (C9-ALS) ALS patients. We confirmed that C9orf72 is highly expressed in microglia and observed that the hexanucleotide repeat expansion (HRE) results in haploinsufficiency specifically in these cells. Whereas sALS microglia transitioned towards disease-associated profiles, C9orf72 HRE microglia exhibited a diminished response, with deficits in phagocytic and lysosomal pathways. We confirmed these lysosomal alterations in cortical and spinal cord tissue, as well as in C9orf72-deficient induced pluripotent stem cell (iPSC)-derived microglia, with a disrupted lysosomal distribution and an increased number of intracellular structures consistent with storage lysosomes. Furthermore, we observed a diminished response of astrocytes in C9orf72 HRE carriers and found that spinal astrocytes are distinct from cortical

astrocytes and show a particular enrichment for ALS upregulated genes. We provide in silico predictions pointing to altered cell-cell interactions between microglia and astrocytes. This complex interplay highlights possible variations in cellular responses between sporadic and inherited ALS variants, providing valuable insights for patient stratification and for selecting appropriate treatments.