Senescent fibro-adipogenic progenitors are potential drivers of pathology in inclusion body myositis

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Background and aims
Inclusion body myositis (IBM) is unique across the spectrum of idiopathic inflammatory myopathies due to its refractoriness to current treatment approaches. One explanation for this resistance may be the engagement of cell-autonomous mechanisms that sustain or promote disease progression of IBM independent of inflammatory activity. In this study, we focused on senescence of tissue-resident cells as potential driver of disease.

Methods
We compared IBM patients to non-diseased controls (NDC) and immune-mediated necrotizing myopathy (IMNM) patients using single-nuclei RNA sequencing, immunohistochemistry and immunofluorescence analysis.

Results

- **A:** UMAP embedding demonstrating distinct clusters of cell types and subtypes. **B:** Clustered dot plot visualization of top-regulated marker genes. The mean expression for each cluster is indicated by colour code. The dot size indicates the percent of expressing cells. Clusters were annotated based on marker genes. **C:** Frequency of CDKN1A+ cells for each cell cluster as indicated for IBM patients and NDC. Differences between groups were analysed by the Kruskal–Wallis test followed by post hoc testing. **D:** Representative immunofluorescence staining of IBM muscle specimen. Muscle slices were incubated with SPIDER-βGAL at a pH of 6. SPIDER-βGAL indicates the activity of the senescence-associated β-galactosidase. SPIDER-βGAL stains green. Senescent FAPs were identified in the perimysium by p21 staining in red.

Results and Conclusion
Histopathological analysis suggested that cellular senescence is a prominent feature of IBM, primarily affecting non-myogenic cells. In-depth analysis by single nuclei RNA sequencing allowed for the deconvolution and study of muscle-resident cell populations. Among these, we identified a specific cluster of fibro-adipogenic progenitors (FAPs) that demonstrated key hallmarks of senescence, including a pro-inflammatory secretome, expression of p21, increased β-galactosidase activity, and engagement of senescence pathways. FAP function is required for muscle cell health with changes to their phenotype potentially proving detrimental. In this respect, the transcriptomic landscape of IBM was also characterized by changes to the myogenic compartment demonstrating a pronounced loss of type 2A myofibers and a rarefaction of acetylcholine receptor expressing myofibers. IBM muscle cells also engaged a specific pro-inflammatory phenotype defined by intracellular complement activity and the expression of immunogenic surface molecules. Skeletal muscle cell dysfunction may be linked to FAP senescence by a change in the collagen composition of the latter. Senescent FAPs lose collagen type XV expression, which is required to support myofibers’ structural integrity and neuromuscular junction formation in vitro. Taken together, this study demonstrates an altered phenotypical landscape of muscle-resident cells and that FAPs, not myofibers, are the primary senescent cell type in IBM. Taken together, this study demonstrates an altered phenotypical landscape of muscle-resident cells and that FAPs, not myofibers, are the primary senescent cell type in IBM.

Disclosure
The authors declare no competing interests.