than the SDFT. Fibrillin −2 staining was found in the fascicles and IFM, whereas fibrillin −1 was mostly in the IFM. Both fibrillin types were more abundant in the positional CDET. In the IFM, the elastin formed a peri-cellular meshwork and amorphous collagen bridged between elastin fibres. In old tendons, the elastin appeared more disordered and fragmented, especially in the SDFT.

Discussion: The results suggest that specialised mechanical properties are achieved, in part, by differences in the elastic fibre type and distribution. The thick, elastin-containing fibres in the SDFT IFM allow high strains, whereas the positional CDET has more fascicle located, thin, stiffer, fibrillin fibres. Ageing is detrimental to both.

Acidic PH inhibits the stiffness response in human bone marrow mesenchymal stem cells (BM-MSCS)

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Introduction: Cells sense mechanical forces from their environment and convert these physical forces into biochemical signals in a process termed mechanotransduction. Importantly, changes in physical force do not occur in isolation, but in concert with chemical factors, a consideration often overlooked. Tissue extracellular pH (pHe) is an important environmental factor known to change during disease and ageing, and often coincident with changes in tissue stiffness. However, the influence of pH on the cellular mechano-response has received little attention. Here, we assessed the proteomic stiffness-response of human BM-MSCs (a cell type often chosen for direct tissue implantation as a therapeutic) cultured on type I collagen coated polyacrylamide gels at extracellular pH (pHe) akin to healthy (pH 7.1), moderately (pH 6.8) and severely (pH 6.5) degenerate musculoskeletal tissues.

Materials and Methods: Human BM-MSCs isolated from hip joints were expanded on tissue culture plastic and transferred to type I collagen-coated polyacrylamide gels of different stiffness (2 and 25 KPa). Following 3 days of culture in medium at pH 7.1, 6.8 or 6.5, cells were trypsinised and processed for label-free mass spectrometry. The peptide abundances were quantified and mapped to proteins using MaxQuant. Differential expression, gene ontology enrichment and reactome pathway analysis were used to assess the systemic response of BM-MSCs to substrate stiffness at different pH.

Results: At pH 7.1 and 6.8, BM-MSCs were stiffness responsive, increasing proteins in pathways related to regulation of matrix and collagen synthesis. Proteins involved in metabolic pathways, including electron transfer chain and ATP synthesis were downregulated. Differential expression analysis revealed an increase in collagen types I, III and V, and SPARC. Decreases in the metabolic proteins FTL and SOD2 were also observed. At pH 6.5, the mechano-response was inhibited (10 downregulated proteins including TNC and FGL) with no significantly enriched pathways identified.

Discussion: The mechano-response of BM-MSCs is stiffness and pH-dependent, which has implications for tissue engineering and regenerative medicine (BM-MSCs often suggested for implantation into degenerate tissues with complex physiochemical environments). Further studies are required to elucidate the altered mechanotransduction pathways, which could lead to discovery of novel targets for cell and tissue engineering.

Glaucomatous trabecular meshwork cell-derived extracellular matrix influences differentiation of healthy adult trabecular meshwork stem cells

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Introduction: The trabecular meshwork (TM) is an essential tissue in the eye with a complex 3D structure that facilitates aqueous humour outflow and filtration. In POAG patients, TM cellularity decreases along-side an altered and increased accumulation of extracellular matrix (ECM). The resulting disruption to aqueous humour outflow and rise in intraocular pressure leads to retinal ganglion cell dysfunction and irreversible blindness. Adult TM progenitor cells (PET) can contribute to the proliferation response seen with externally induced damage but do not do so with increasing age and POAG. We investigate the influence of glaucomatous TM derived ECM on the differential ability of PET.

Materials and Methods: Primary TM cells (pTM) were cultured from dissected human donor TM tissue (one donor, aged 28 years old). Primary cultures were