

Conditioned medium produced by mesenchymal stromal cells against joint disorders: content characterization and effects in increasingly complex models of osteoarthritis

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OBJECTIVE: The vesicular and soluble components of cell-free products must be comprehensively characterized to unravel potential therapeutic targets. In this context, we characterized ASC-conditioned medium (CM) and tested its effects in counteracting osteoarthritis (OA) drawbacks.

MATERIALS and METHODS: CM was obtained from human primary ASCs maintained in serum-free medium for 72h. After concentration, the content was analysed using biochemical, spectroscopical and omics approaches. NTA and TEM gave a punctual description of its vesicular fraction. In vitro effects were measured in three OA models: 2D-chondrocytes, *ex-vivo* punches of exfoliated cartilage and osteochondral explants, all stimulated with inflammatory cytokines. In addition, CM chemotactic properties were also assessed.

RESULTS: CM characterization emphasised a high EVs content, confirming the preservation after the concentration process, as well as a multitude of soluble factors. Several of these bioactive molecules were positively associated with anti-catabolic and immunomodulatory pathways. These findings were confirmed through the significative reduction of MMP activity in the different OA models, and the chemotactic action on THP-1 cells.

DISCUSSION: The wide range characterization of the CM highlighted its rich composition of soluble and EV-embedded effectors, including proteins and bioactive lipids, suggesting its potential use in the context of OA and, eventually, other inflammatory pathologies.