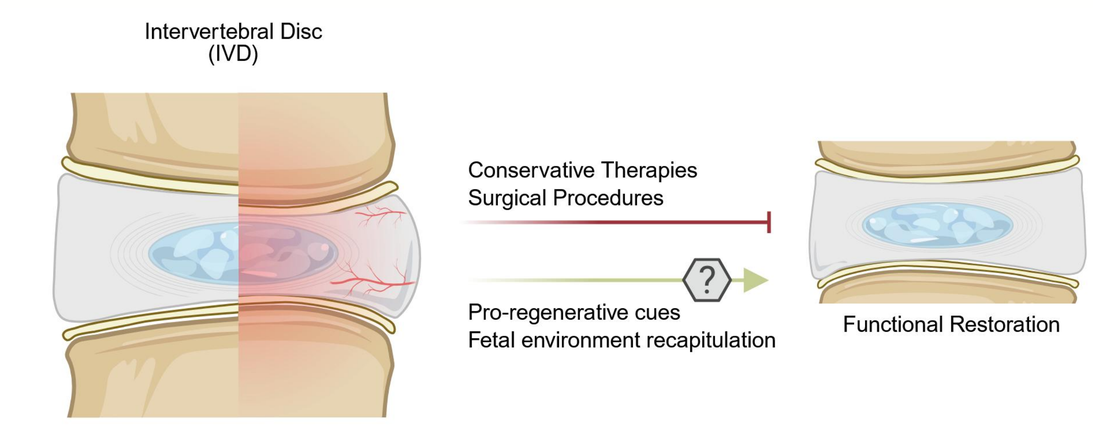
Rewinding time:
Fetal-inspired matrices for intervertebral disc regeneration

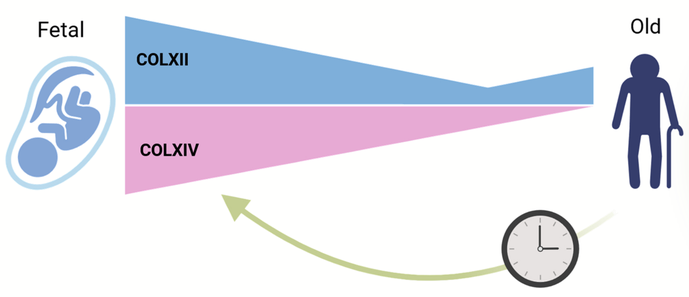
C. Milheiro, M.L. Moura, C. Cunha, J.P. Castro, P.B. da Silva, C.C. Barrias, F. Gärtner, S.M. Diaz-Prado, G.G.H. van den Akker, R.M. Gonçalves, M. Amendola, M.A. Barbosa, J. Caldeira

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BACKGROUND



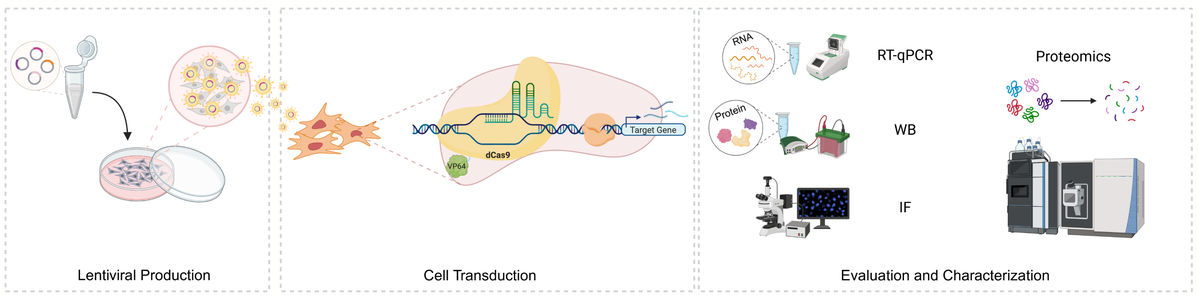
HYPOTHESIS



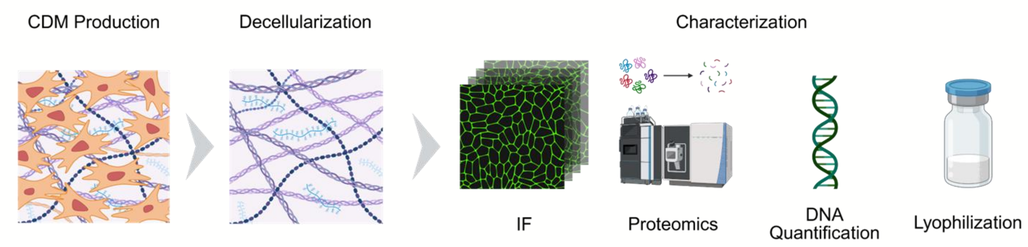
Promotion of IVD **regeneration** by recapitulation of **a fetal-like microenvironment** based on **tailored** cell-derived extracellular matrix (CDM) containing COLXIV and COLXII

METHODOLOGY

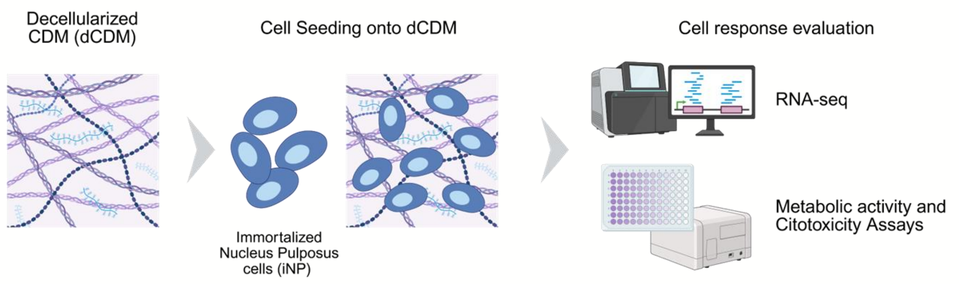
1. Establishment of a CRISPR activation system targeting *COL12A1* and *COL14A1*



2. Tailored cell-derived matrix (CDM) production, decellularization and characterization



3. Pro-regenerative effect assessment *in vitro*

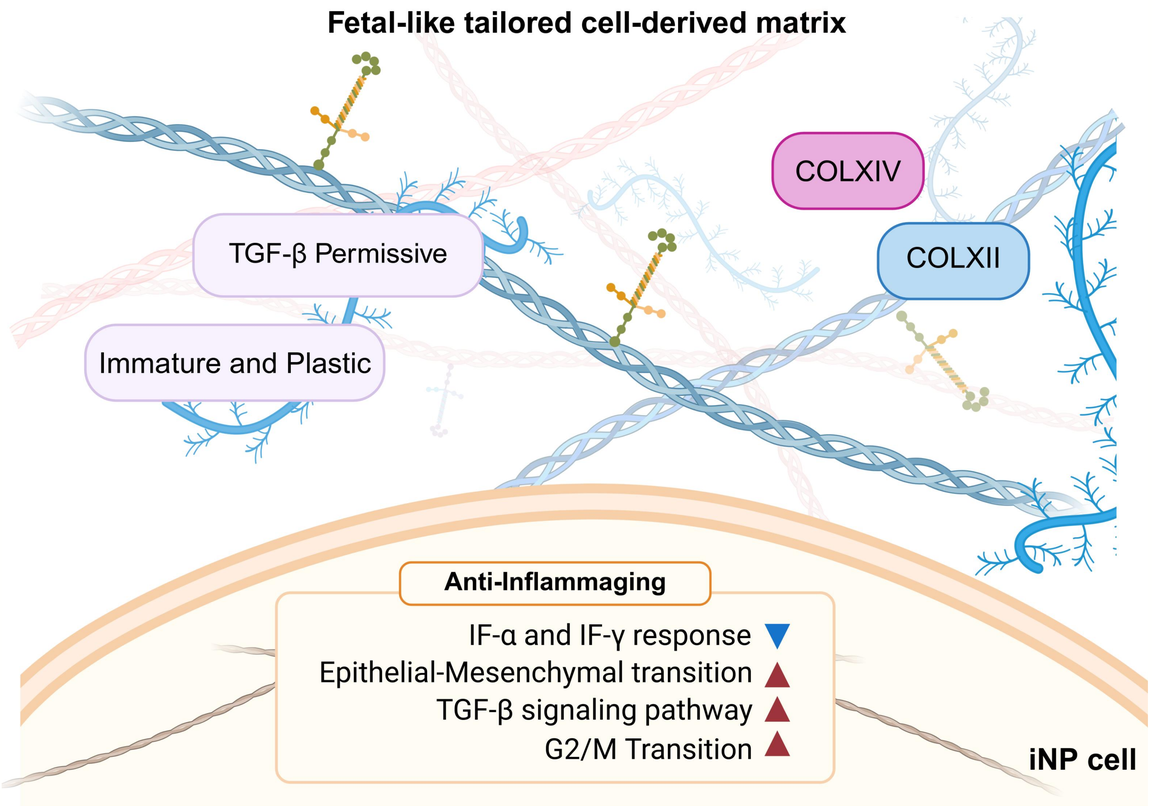


DATA OVERVIEW

CRISPR-dCas9 editing leads to **stable expression** of *COL14A1* or *COL12A1* by immortalized mesenchymal stem cells (iMSC)

Modified iMSC's **stromal markers and core proteomic signature** is retained

Tailored CDMs show **structural and biochemical** characteristics associated with fetal ECM



COL14-engineered CDMs overexpress both COLXIV and COLXII

iNP cells show an **anti-inflammaging** signature when seeded with tailored CDMs

Tailored CDMs show **biocompatibility** *in vivo*

CONCLUSION

In summary, we established a CRISPR activation system for *COL12A1* and *COL14A1*, enabling iMSCs to act as **biofactories** for fetal-inspired CDMs.

Proteomic analyses confirmed **fetal-like matrix tailoring** and revealed **anti-inflammaging** effects on human iNP cells.

In vivo studies demonstrated **biocompatibility**, supporting the use of this versatile and innovative approach for recapitulating the fetal microenvironment in intervertebral disc regeneration and other tissue repair strategies.

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