Mechanical Injury Suppresses Autophagy Regulators in Cartilage Superficial Zone and Pharmacological Autophagy Activation Results in Chondroprotection

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INTRODUCTION
Mechanical injury induces cell death in cartilage and triggers a remodeling process that ultimately can manifest as osteoarthritis (OA). Autophagy is a process for turnover of intracellular organelles and molecules that protects cells during stress responses (1). This study determined whether autophagy is activated and has a protective function in the cartilage response to mechanical injury.

METHODS
Tissues: Articular cartilage explants were obtained from mature bovine knee joints (14-30 months).

Load apparatus and mechanical injury: Mechanical injury was applied with an Instron 8511 mechanical testing device (Instron, Norwood, MA). Each explant was centralized on a loading platform and a radially unconfined compressive load was applied through an impermeable stainless steel platen. In this study, after a small preload (0.1MPa) was applied for two minutes, a 40% strain was applied to the explants for 500ms. Control explants were placed in the loading apparatus but not loaded.

Autophagy markers: We used antibodies to the autophagy regulators ULK1, Beclin1 (Santa Cruz Biotechnologies) and LC3 (Abgent).

DISCUSSION
Mechanical injury to cartilage does not activate autophagy even suppresses ULK1, Beclin1 and LC3 expression by treating cartilage explants with rapamycin. This significantly reduced cell death at 48 hours (P < 0.05) in the superficial zone (Fig 2).

Mechanical injury also causes an increase of sGAG release into supernatants, which was significant at 48 and 96 hours compared to control explants without injury (P < 0.001). In the presence of rapamycin, the levels of sGAG in supernatants were significantly decreased at 48 and 96 hours after mechanical injury (P < 0.001).

REFERENCES

ACKNOWLEDGEMENTS
This study was supported by NIH grants AG07996 and AR AR058954. B. Caramés was supported by Postdoctoral Fellowship “Anxeles Alvariño”, Secretaría Xeral I+D+i, Xunta de Galicia, Spain.