Hydrogen sulphide synthesis is impaired in osteoarthritic chondrocytes from diabetic patients and in vitro in cells exposed to high glucose stress

Vaamonde-García C[(1-2)], Burguera EF[(3-4)], Blanco FJ[(3)], Meijide Failde R[(1)]

[(1)]Cell Therapy and Regenerative Medicine Group, Department of Medicine, University of A Coruña, A Coruña, Spain
[(2)]Rheumatology Research Group, INIBIC A Coruña, Spain
[(3)]Tissue Engineering Unit, Rheumatology Research Group, INIBIC-Hospital Universitario A Coruña, A Coruña, Spain
Elena.Fernandez.Burguera@sergas.es

Background: A growing number of findings support the hypothesis that type 2 diabetes is an independent risk factor of osteoarthritis (OA). However, the mechanisms underlying the connection between both diseases remain unclear. Hydrogen sulfide (H2S) plays an important role in the pathogenesis of diabetes and its complications. In relation, we and other authors have observed a protective impact of H2S induction on activation of pathological pathways in the chondrocyte (1).

Objectives: In this study we examined the modulation of H2S levels in osteoarthritic chondrocytes from diabetic (DB) or nondiabetic (non-DB) patients subjected or under glucose stress, in order to elucidate whether impairment in H2S-mediated signaling could participate in the establishment of diabetes-related OA.

Methods: Chondrocytes were isolated from OA cartilage of diabetic (DB) or non diabetic (non- DB) patients. T/C28a2 and primary human chondrocytes were stimulated w/o IL-1β (5 ng/mL) under a normal (5.5 mM; NG) or a high (25 mM; HG) glucose environment. Gene and protein expression of enzymes involved in H2S synthesis (cystationine γ-liase [CSE], cystationine β- synthase [CBS], and 3-mercaptopyruvate sulfurtransferase [3-MT]) and HO-1 were assessed by RT-qPCR and WB, respectively. To determine the involvement of H2S in catabolic pathways activated by HG in chondrocytes, NaSH and GYY 4137 (500 μM), a fast and slowreleasing H2S donor respectively, were employed.

Results: Fresh isolated chondrocytes from OA cartilage of diabetic patients showed lower levels of H2S synthesizing enzymes (CSE, CBS and 3-MT) than those of non-DB patients (Figure 1). In relation, chondrocytes T/C28a2 exposed to HG stress expressed lower mRNA and protein levels of these 3 enzymes after 3

ISSN: 0214-2813
DOI: 10.23853/bsehm.2018.0603

days of incubation compared to those incubated in NG conditions (0.41- fold and 0.83-fold [CSE], 0.42-fold and 0.66-fold [CBS], and 0.52-fold and 0.79-fold [3-MT] for mRNA and protein expression, respectively; n=6, $p<0.05$). IL-1β also attenuated the gene and protein expression of CBS elicited by chondrocytes incubated in NG (0.47-fold and 0.86-fold, respectively; n=6, $p<0.05$). Additionally, the expression of pro- inflammatory chemokine IL-8 induced by IL-1β was significantly higher in chondrocytes under HG than NG condition (6-fold; n=5, $p<0.05$); whereas protein levels of heme oxygenase 1, an anti-inflammatory enzyme, were reduced in HG exposed chondrocytes (0.77-fold; n=6, $p<0.05$). GYY 4137 and NaSH co-treatment recovered HO-1 expression and reduced IL-8 levels in chondrocytes under IL-1β + HG conditions. Furthermore, similar results were registered in primary human chondrocytes from OA cartilage.

Conclusions: The results indicate a reduction of H2S synthesis as a critical feature involved in hyperglucidic-mediated dysregulation of articular chondrocytes. The impairment of H2S signaling could participate in the mechanisms underlying the predisposition to OA development in diabetic individuals and may open new opportunities for treating patients with a diabetes-related OA phenotype.

Reference: 1. Burguera EF, Vela-Anero A, Magalhães J, Meijide-Faílde R, Blanco FJ. Effect of hydrogen sulfide sources on inflammation and catabolic markers on interleukin 1β-stimulated human articular chondrocytes. Osteoarthritis Cartilage. 2014 Jul;22(7):1026-35.

Acknowledgements: The present work was supported by contrato posdoctoral Xunta (ED481D2017/023)