PROFILING THE HISTONE LANDSCAPE IN OSTEOARTHRITIS: ENRICHMENT OF HISTONE VARIANT H3.3 AND ITS ASSOCIATED POST-TRANSLATIONAL MODIFICATIONS

Keywords: Genetics/Epigenetics, Osteoarthritis, Cartilage

C. Núñez Carro1, V. Calamia2, P. Fernández Puente3, M. Blanco-Blanco1, P. Quartaña Diaz4, T. Hermida Gómez4,5, C. Ruiz-Romero6,7, F. J. Blanco6,7, M. C. De Andres1. 1Instituto de Investigación Biomédica de A Coruña, Unidad de Epigenética - Grupo de Investigación de Reumatología (GIR), A Coruña, Spain; 2Instituto de Investigación Biomédica de A Coruña, Unidad de Proteómica - Grupo de Investigación de Reumatología (GIR), A Coruña, Spain; 3Centro de Investigaciones Científicas Avanzadas (CICA), Universidad de A Coruña (UDC), A Coruña, Spain; 4Centro de Investigación Biomédica en Red de Bioingeniería, Biomateriales y Nanomedicina, CIBER-BBN, Madrid, Spain; 5Grupo de Investigación de Reumatología y Salud (GIR-S), Departamento de Fisioterapia, Medicina y Ciencias Biomédicas, Facultad de Fisioterapia, Universidad da Coruña (UDC), A Coruña, Spain

Background: Nucleosomes, the basic structural unit of chromatin, are composed of DNA wrapped around an octameric core of histone proteins. The latter is usually comprised by canonical histones H2A, H2B, H3 and H4[1]. These can be replaced by histone variants, which provide structural and functional changes to the nucleosome[2]. Histones are prone to undergo post-translational modifications (PTMs) that can modify chromatin conformation to induce changes in gene expression[3]. Recent evidence supports that epigenetic mechanisms, such as DNA methylation and non-coding RNA modifications, are involved in the pathogenesis of osteoarthritis (OA). Nevertheless, histone modifications mainly remain an unexplored epigenetic mechanism in OA.

Objectives: The aim of the present study was to identify, characterize and compare the histone profile in human articular cartilage samples from normal (N) and OA patients to provide novel insights into the role of histones in the development of OA.

Methods: Histones were extracted from 3 N and 3 OA cartilage samples following a protocol based on acid extraction previously described by our group. Prior to mass spectrometry (MS) analysis, samples were reduced with dithiothreitol and alkylated with iodoacetamide. After an overnight digestion with trypsin, peptides were desalted via StageTips. Samples were injected on a nanoElute LC and alkylated with iodoacetamide. After an overnight digestion with trypsin, peptides were analyzed by LC-MS/MS. The most detected PTMs are among the most studied, “PTM” and “Label-free” modules from PEAKS were employed.

Results: Our histone extraction protocol successfully enriched the samples for subsequent MS analysis. The most detected PTMs are among the most studied, including methylation, acetylation, ubiquitination, phosphorylation, deamidation, biotinylation, citrullination, and ADP-ribosylation. PTMs were found in core histones H2A, H2B, H3, H4 and linker histone H1. Moreover, PTMs were detected in some of their variants. Strikingly, we found that the histone variant H3.3 is significantly increased in OA cartilage compared to N cartilage (2.67×104 ± 9.56×103 vs. 3.11×103 ± 1.71×103) (Figure 1 A). Further H3.3 analysis showed three differentially abundant PTMs between OA and N (Figure 1 B). Acetylation of lysine 18 (K18ac) was more abundant in OA cartilage. Monomethylation of K79 (K79me1) and dimethylation of K36 (K36me2) was also more abundant in OA cartilage.

Conclusion: The histone variant H3.3, which associates with decondensed states of chromatin and transcriptionally active sites, is upregulated in human OA cartilage. Furthermore, we identified three H3.3-associated PTMs that are differentially expressed in OA and N cartilage. These data provide new insights into the epigenetic landscape of OA and could contribute to the search for novel therapeutic targets to treat the disease.

REFERENCES:

Disclosure of Interests: None Declared. DOI: 10.1136/annrheumdis-2023-eular.2339

ACKNOWLEDGEMENTS: NIL.

Intra-articular injection of Stigmasterol loaded nanoparticles is effective in inhibiting joints destruction in osteoarthritis rat model

Keywords: Animal models, Osteoarthritis, Cartilage

J. H. Jung1, B. Y. Kim2, S. W. Nam3. 1Korea University College of Medicine, Internal Medicine, Seoul, Korea, Rep. of (South Korea); 2Gangneung Asan Hospital, Internal Medicine, Gangneung, Korea, Rep. of (South Korea); 3Yonsei University Wonju College of Medicine, Internal Medicine, Wonju, Korea, Rep. of (South Korea)

Background: Stigmasterol is the most abundant plant sterols and its chemical structure is very close to cholesterol. Stigmasterol inhibits pro-inflammatory and matrix degradative mediators typically involved in cartilage degradation through inhibition of the nuclear factor kappa-B (NF-κB) pathway and attenuates chondrocyte injury induced by interleukin-1β (IL-1β). Mesoporous silica nanomaterials (MSNs) have been proven to be promising nanoparticles due to their superior loading capability, multitudinous functionalization, and biocompatibility. Since

Acknowledgements: NIL.

Disclosure of Interests: None Declared. DOI: 10.1136/annrheumdis-2023-eular.2339

Figure 1. (A) Histone variant H3.3 abundance in N vs OA. Data were analyzed using Student’s t-test and plotted as mean ± SEM. *p <0.05. Log2 fold change >1 or <1. (B) Heatmap of histone variant H3.3 PTMs in N vs OA.