Table 1. Baseline characteristics of patients:

Variable	PRP group (n=16)	HA group (n=22)	NALJ group (n=13)	P
Age (years) (x±SD)	59.56±8.70	63.32±5.49	62.23±7.73	NS*
BMI (kg/m ²) (x±SD)	30.31±3.75	30.3±3.36	31.99±5.94	NS*
Female (n,%)	8(50)	12(54.5)	7(53.8)	NS**
KOA stage (n,%)				NS**
II	5(33.3)	11(50)	6(46.2)	
III	10(66.7)	11(50)	7(53.8)	
Knee treated (n,%)				NS**
Left	4(25)	12(54.5)	7(58.3)	
Right	12(75)	10(45.5)	5(41.7)	
VAS pain (x±SD)	6.81±2.14	6.73±1.55	6.54±2.07	NS*
WOMAC (x±SD)	48.56 ±13.31	48.68 ±16.09	57.69±18.98	NS*
OARSI (x±SD)	63.92 ±17.48	57.03 ±18.78	70.80±13.51	NS*

NS: not significant. statistical significance based on: *the simple ANOVA test and ** on the Xi-square test.

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AB0101 SECRETOME ANALYSIS OF CHONDROCYTES AND SYNOVIAL FIBROBLASTS IN OSTEOARTHRITIS: MODULATION BY VIP

Selene Pérez García¹, Valentina Calamia², Tamara Hermida Gómez², Mar Carrión¹, Raúl Villanueva Romero¹, Yasmina Juarranz¹, Francisco J. Blanco², Rosa P. Gomariz¹, on behalf of Bases moleculares y celulares de enfermedades inflamatorias/autoinmunes. Departamento de Biología Celular. Facultad de Biología, UCM. Grupo de Investigación en Reumatología, Instituto de Investigación Biomédica de A Coruña (INIBIC).. ¹Facultad de Biología, Universidad Complutense de Madrid (UCM), Departamento de Biología Celular, Madrid, Spain; ²Instituto de Investigación Biomédica de A Coruña (INIBIC), Servicio de Reumatología, A Coruña, Spain

Background: Osteoarthritis (OA) is a chronic, degenerative and multifactorial disease, and the main cause of pain and dysfunction among elder people. It is characterized by a progressive loss of function of synovial joints (1). The role of chondrocytes in this pathology has been widely studied (2), but other joint cells are also involved, including the synovial fibroblast (SF) (3-5). During joint destruction, inflammatory and degradative mediators are released by joint cells and from the extracellular matrix (ECM), including fibronectin degradation fragments (Fn-fs) (3, 4, 6). On the other hand, vasoactive intestinal peptide (VIP) exerts anti-inflammatory and immunomodulatory actions in several autoimmune and inflammatory disorders, including OA (3, 5). The study of the mediators released by joint cells and their modulation by pro- and anti-inflammatory mediators would be useful for the design of novel therapies for OA treatment.

Objectives: To analyse the mediators released from co-cultures of OA chondrocytes and SF, and to elucidate the effect of Fn-fs and VIP on these cells.

Methods: Human articular chondrocytes (HAC) and SF from 4 OA patients were provided by the Rheumatology Service at Complejo Hospitalario Universitario A Coruña. Isolated cells were recovered and plated in SILAC DMEM-Flex lacking Arginine and Lysine. In the case of medium and heavy media, isotope-labeled L-lysine and L-arginine were used. When 100% of labeling was reached, cells were put in co-culture and incubated in serum-free medium with or without Fn-fs (10⁻⁸M) or Fn-fs + VIP (10⁻⁸M) for 48h. Cell secretomes were separated on a 10% SDS PAGE gel. Gels were stained with Coomassie blue and the resulting lanes were cut into slices and subjected to in-gel digestion. Extracted peptide mixtures were desalted and concentrated via NuTip, subjected to liquid chromatography, using a Tempo nano LC equipped with a Sun Collect MALDI Spotter, and analyzed by MALDI-TOF/TOF. Identification of peptides and proteins and relative quantification were performed using Protein Pilot software (Sciex).

Results: Cell secretomes were analysed in 4 OA patients in duplicate. Database search (UniprotKB/Swissprot) and Venn's diagrams drawing tool (Venny 2.1.0) allowed us to identify 79 common proteins in the HACs-SF co-cultures. Among them, VIP was able to modulate 33 different proteins, significantly reducing 9 of them: CH3L1, PTX3, PGS2, MMP2, Complement C1R, Complement C3, TBA1, QSOX1, and CATB. These proteins include inflammatory and ECM proteins, proteases and complement system proteins among others, which play a main role in the OA pathogenesis.

Conclusion: VIP decreases inflammatory and degradative mediators in HAC-SF co-cultures, potentially slowing the progression of the disease and supporting its therapeutic role in osteoarthritis.

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AB0102 GENERATION OF OSTEOARTHRITIC MESENCHYMAL STROMAL CELL LINES

María Piñeiro-Ramíl^{1,2}, Rocio Castro-Viñuelas^{1,2}, Clara Sanjurjo-Rodríguez^{1,2}, Silvia Rodríguez-Fernández^{1,2}, Tamara Hermida Gómez², Francisco Javier de Toro-Santos^{1,2}, Francisco J. Blanco², Isaac Fuentes-Boquete^{1,2}, Silvia María Díaz-Prado^{1,2}. ¹Universidad de Coruña (UDC), A Coruña, Spain; ²Instituto de Investigación Biomédica de A Coruña (INIBIC), A Coruña, Spain

Background: Bone-marrow mesenchymal stromal cells (MSCs) are multipotent self-renewal adult cells with high potential to regenerate the damaged tissues in degenerative diseases such as osteoarthritis (OA). Nevertheless, their usefulness for osteochondral Regenerative Medicine research is hampered by their proneness to senescence when in vitro cultured. Currently, MSC lines available are scarce and present limitations regarding their differentiation capacities. In addition, there is none OA MSC line available for research on this disease.

Objectives: The aim of this study was to generate and characterize immortalized human OA and non-OA MSC lines for their use in osteochondral Regenerative Medicine research.

Methods: For the generation of the immortalized MSC lines, SV40 large T antigen (SV40LT) and GFP-fused human telomerase reverse transcriptase (hTERT) were used. Primary MSCs derived from two hip OA patients and one hip fracture patient without OA were transduced by spinoculation at 800 xg for 45 minutes. Transgene expression was induced by valproic acid. Nuclear expression of SV40LT and GFP was tested by immunofluorescence. Proliferation and senescence were investigated through calculation of population doublings (PDs) at each passage after immortalization and β-galactosidase staining after 100 PDs for each MSC line. Maintenance of MSC characteristics in immortalized MSCs was tested by analysis of CD29, CD44, CD73, CD90, CD105, CD34 and CD45 expression by flow cytometry and cell differentiation experiments. Multi-lineage differentiation potential was analysed histochemical, immunohistochemical and molecularly.

Results: Three MSC lines have been generated: two OA and one non-OA. As shown by immunofluorescence, SV40LT is expressed in the nucleoplasm of these cells, while GFP-fused hTERT is expressed in the nucleoli. A constant proliferation rate throughout subculturing in addition to β-galactosidase negative staining confirms that immortalized MSC lines do not senesce, unlike primary MSCs. Expression of CD29, CD44, CD73 and CD90 and lack of CD34 and CD45 was conserved in immortalized MSC lines, while CD105 expression was altered for transduction status and passage. Both OA and non-OA immortalized MSC lines maintain their multipotency (namely, osteogenic, chondrogenic and adipogenic differentiation capacity).

Conclusion: Both OA and non-OA MSCs are susceptible to immortalization by SV40LT and hTERT. For they increased lifespan combined with keeping of most

of primary MSC characteristics, these MSC lines are expected to be valuable tools for the research on Regenerative Medicine for OA as part of Tissue Engineering in vitro models.

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AB0103 A NOVEL PROTECTIVE ROLE OF GPNMB/Osteoactivin IN POST-TRAUMATIC OSTEOARTHRITIS

Asaad Al Adlaan¹, Nazar Hussein¹, Fatima Jaber¹, Favez Safadi^{1,2}. ¹Rootstown, Anatomy and Neurobiology, Musculoskeletal Research Group, Rootstown, United States of America; ²Akron Children's Hospital, Rebecca D Considine Research Institute, Akron Children's Hospital, Akron, United States of America

Background: Osteoarthritis (OA) is a chronic joint disease causes irreversible damage to the articular cartilage resulting in loss of joint function and subchondral bone remodeling. There is no cure for OA and currently the available treatment like pain management, and joint replacement surgery is used to treat this disease. GPNMB/Osteoactivin is a type-I transmembrane protein plays a vital role in osteogenesis and bone remodeling.

Objectives: However, the function of GPNMB/Osteoactivin in chondrogenesis and cartilage repair is unknown. Hence, in this study, we examined the role of GPNMB/Osteoactivin using post-traumatic osteoarthritis model and ex vivo chondrocytes cell culture and assessing the effects of GPNMB/Osteoactivin as anti-inflammatory factor in chondrocytes.

Methods: We first examined the expression GPNMB/Osteoactivin in normal cartilage. Next, we assessed the effects of recombinant GPNMB/Osteoactivin on mouse primary chondrocytes proliferation/viability and extracellular matrix markers.

Results: We found that GPNMB/Osteoactivin treatment did not have any effect on cell proliferation/viability, however, there was an increased in Collagen Type II and aggrecan expression in a dose- and time-dependent manner. These data suggest that GPNMB/Osteoactivin plays a role in cartilage maintenance. Next we determined whether GPNMB/Osteoactivin contributes to the OA disease progression. Human osteoarthritic damaged and undamaged cartilage harvested following knee replacement were used to examine GPNMB/Osteoactivin expression and found that GPNMB/Osteoactivin mRNA was 6-7-fold increased in damaged compared to undamaged cartilage. Similar results were obtained from mouse primary chondrocytes treated with IL-1 β and showed a significant increase (4-fold) in GPNMB/Osteoactivin mRNA compared to untreated control. Next, we evaluated the effects of GPNMB/Osteoactivin deficiency on the OA progression using the DMM model. GPNMB/Osteoactivin mutant (DBA/2J) mice showed significant increased in OARS1 scores compared to WT mice.

Conclusion: Taken together, these data are the first to report a role for GPNMB/Osteoactivin in cartilage maintenance and protection against the progression of OA.

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AB0104 ARE POLLUTANTS A NEW RISK FACTOR FOR OSTEOARTHRITIS? RESULTS FROM A SYSTEMATIC LITERATURE REVIEW

Camille Deproux¹, Alice Courties¹, Jean-Baptiste Fini², Marie-Stéphanie Clerget-Froidevaux², Barbara Demeneix², Francis Berenbaum¹, Jérémie Sellam¹, Karine Louati¹. ¹St Antoine Hospital, Paris, France; ²Museum National d'Histoire Naturelle, Paris, France

Background: Prevalence of knee osteoarthritis (OA) has doubled since the mid-20th century. Interestingly, this increase cannot be explained solely by longer life expectancy and the obesity epidemics (1). Therefore, other environmental factors known to have increased in the recent years could be part of the explanation.

Objectives: We performed a systematic literature review (SLR) to summarize the existing knowledge about associations between OA and pollutants.

Methods: Pubmed database was used to identify studies reporting data on OA and pollutants in humans (examples of MeSH terms: "Polychlorinated Biphenyls (PCB)" or "Lead"). Abstracts from international congresses were reviewed for the past 2 years, combined with manual curation. Studies were classified in epidemiological clinical studies, pollutants assessments in *ex vivo* OA joint tissues, and *in vitro* effect on human chondrocyte.

Results: As of January 15, 2019, 193 potentially relevant articles were screened. Among them, 14 were selected. After manual curation, a total of 21 full text articles were analyzed. Ten articles reported epidemiological clinical studies. Four articles underlined the link between PCB and OA: in the 3 articles reporting association with past exposure to PCB, the most robust one showed that risk of OA was significantly increased, at least in men (OR 4.1 [95%CI: 1.8-11.2] for men and 1.3 [95%CI: 0.8-2.3] for women). In the 4th study, the highest serum concentration of PCB was associated with a higher risk of arthritis: maximal adjusted OR (aOR) was 3.2 [95%CI: 1.6-6.7], $p < 0.01$. However, sensitivity analysis of OA patients did not find any significant association in this study. Two articles focused on serum levels of perfluorooctanoate (PFOA) and perfluorooctane sulfonate (PFOS): aOR to have OA in the PFOA highest serum level quartile was 1.42 [95%CI: 1.26-1.59], $p = 0.00001$ and 1.55 [95%CI: 0.99-2.43]. Data were conflicting for PFOS: aOR to have OA in the PFOS highest serum level quartile was 1.77 [95%CI: 1.05-2.96], $p < 0.05$ and 0.76 [95%CI: 0.68-0.85], $p = 0.00001$. Two articles showed an association between serum lead levels and knee OA in the general population. In Korea, the risk of radiographic OA was increased in the highest quartile of lead serum level compared to the lowest one (aOR 1.90 [95%CI: 1.09-3.32] for men and 1.81 [95%CI: 1.17-2.77] for women). In the United States, risk knee radiographic OA correlates with increased lead serum levels. In the last study, household air pollution induced by the cooking heat source was examined: compared to electricity, the risk of OA was increased with the use of liquids (kerosene/paraffin: aOR 1.73 [95%CI 1.33-2.25]) or solid (coal, wood: aOR 1.73 [1.32-2.27]), (agriculture/crop: aOR 2.0 [1.47-2.72]). Lead and zinc accumulation was reported in 7 articles: 4 of them analyzed the concentration in OA joint obtained during arthroplasty and showed highest concentration in cartilage versus bone. In another, lead serum levels were associated with biomarkers of joint tissue metabolism. Two studies with X-ray fluorescence on cadavers found a high accumulation of lead and zinc in the articular cartilage tidemark. Four studies assessed the *in vitro* effect of pollutants on human chondrocytes: the viability of chondrocytes was reduced in presence of PCB, gold, silver, fluoric acid and bisphenol A.

Conclusion: This SLR suggests a possible link between OA and pollutants but the designs of the studies, the populations investigated and the type of pollutants were still too heterogeneous and limited to conclude definitively that pollutants represent a new environmental risk factor for OA. However, this SLR highlights a critical need for novel epidemiological, clinical and basic research studies in order to identify other potential environmental factors in OA.

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AB0105 THE EFFECT OF TROMBOSIT RICH PLASMA APPLICATION ON DEGENERATED CARTILAGE: IN VITRO EXPERIMENTAL STUDY

Kevser Arapoğlu, Ömer Faruk Şendür, Serhan Sakarya. ADU, PİrandM, İZMİR, Turkey

Background: Osteoarthritis is the most common problem among musculoskeletal system disorders. Platelet-rich plasma (PRP) has been suggested to be