

EVs from the two autologous blood derived products PRP and the cell free alternative hyperacute (hypACT) serum in an inflammation model was investigated.

Methods: PRP and hypACT serum were generated by double or single centrifugation of whole blood. EVs were isolated from PRP and hypACT serum by differential ultracentrifugation and concentration and size of the EVs were determined by Nanoparticle Tracking Analysis (NTA). To generate an inflammation model, human primary monocytes were differentiated and activated into inflammatory (M1) macrophages and patient-derived OA chondrocytes were co-cultivated together with these M1 macrophages to mimic an inflammatory environment as present in OA. Culture medium was supplemented with EVs isolated from either PRP or hypACT serum and as control medium was supplemented with either FCS or EV-depleted FCS. Secretion of the inflammatory cytokines IL6, TNF α and IL1 β were measured by ELISA. mRNA expression of the cartilage specific genes SOX9, Collagen 2 and Aggrecan as well as of the matrix degrading enzymes MMP3 and MMP13 was assessed by RT-PCR. Protein expression of Nf κ B and COX2, proteins known to be involved in cartilage degradation during OA, was determined by Western Blot.

Results: In the presence of EVs isolated from blood derived products, secretion of inflammatory cytokines was strongly reduced within the inflammation model. In addition, expression of chondrogenic genes was upregulated when EVs were added to the medium. Furthermore, EVs could reduce the expression of the transcription factor Nf κ B and hence also of its target gene COX2.

Conclusions: Within an inflammation model, EVs from blood derived products harbor the potential to downregulate the secretion of inflammatory cytokines as well as the expression of proteins promoting extracellular matrix degradation. On the other hand, EVs can revert the osteoarthritic phenotype of patient-derived OA chondrocytes to a more hyaline one. Taken together, EVs from blood derived products can overcome the limits of using the whole blood derived product for cartilage regeneration while offering comparable biological effects. They can be seen as a minimally invasive injective biological approach for the treatment of OA.

165 BOSWELLIA SERRATA EXTRACT AND CURCUMIN INCREASE GDF15 PRODUCTION BY HUMAN PRIMARY OSTEOARTHRITIS CHONDROCYTES: A NEW MECHANISM OF ACTION

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Purpose: Boswellia serrata extract (BSE) and curcumin are used to relief symptoms in osteoarthritis (OA). This study aims to better understand the mode of action of these compounds on OA chondrocytes *in vitro*.

Methods: Therapeutic plasmatic concentrations of the different components of BSE correspond to an *in vitro* range from 25 to 100 μ g/ml of total BSE (100 μ g/ml of BSE corresponds to 9.2 μ M of 11-keto- β -boswellic acid (KBA), 5.2 μ M of acetylKBA, 22 μ M de α BA, 34 μ M de β BA, 4.4 μ M de acetyl α BA and 13.2 acetyl β BA), and between 2 to 10 μ M for bioavailability-increased curcumin. BSE (5-100 μ g/ml) and curcumin (0.04 to 4 μ g/ml corresponding to 0.1 to 10 μ M) were tested separately on primary chondrocytes from 3 OA patients. Lactate Dehydrogenase LDH, nitrite (NO₂), interleukin (IL)-6 and Growth Differentiation Factor (GDF)15 were quantified in 72h-treated supernatant using enzyme activity, Griess reaction and ELISAs, respectively.

Results: No mortality was observed at the tested concentrations. BSE and curcumin both decreased concentration-dependently NO₂ and IL-6 production, and increased GDF15 production. For NO₂ production, the decrease was observed from 0.2 μ g/ml of curcumin and 10 μ g/ml of BSE. For IL-6 production, the decrease was observed from 1 μ g/ml for curcumin and 10 μ g/ml for BSE. For GDF-15, the increase was observed from 2 μ g/ml for curcumin and 50 μ g/ml for BSE. Maximal effect was observed at 4 μ g/ml for curcumin: -67% NO₂ (p<0.0001), -71% IL-6 (p=0.0001) and +80% GDF15 (p<0.0001) and at 100 μ g/ml for BSE: -40% NO₂ (p=0.0003), -70% IL-6 (p=0.0003) and +73% for GDF15 (p=0.0017).

Conclusions: At therapeutic plasmatic concentrations, BSE and curcumin decreased the production of NO₂ and IL-6, two inflammatory mediators. Furthermore, BSE and curcumin enhanced GDF-15 production, an anti-inflammatory growth factor. GDF15 was first identified as Macrophage inhibitory cytokine-1 or NSAID-activated gene-1 (by a prostanoid-independent manner), and is known as a regulator of

inflammatory, cell repair and apoptosis pathways. GDF-15 has proapoptotic and anti-tumorigenic activity *in vitro* and *in vivo*. It could represent a new pathway explaining the beneficial effects of BSE and the curcumin on synovium inflammation and cartilage degradation.

166 EVALUATING CYTOKINES AS BIOMARKERS FOR TRAPEZIO-METACARPAL OSTEOARTHRITIS

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Purpose: Osteoarthritis at the base of the thumb (Trapeziometacarpal Osteoarthritis (TMOA) is a prevalent, degenerative condition leading to pain, deformity and disability. Patients with TMOA have difficulty grasping, writing, and performing other activities necessary for working and daily living. End-stage TMOA is treated with arthroplasty, and little information exists pertaining to long term outcomes of intervention. Non-operative management demonstrates only short-term symptomatic alleviation, and no approved disease modifying drugs exist to treat or delay this condition. A key issue in this patient population is that radiographic disease severity is typically discordant with patient reported pain, illustrating the need for objective markers to detect the disease earlier, as well as predict who will progress more quickly to end-stage disease, and who would benefit most from surgical treatment. Our study seeks to characterize the biochemical profile of TMOA patients in order to establish new diagnostic and prognostic tools, and elucidate molecular mechanisms driving TMOA progression.

Methods: To determine the biochemical profile of TMOA patients we have compared plasma and synovial fluid of patients with symptomatic TMOA undergoing surgical (n=37) or non-surgical management (n=35), and followed them up to 1-year post-surgery using targeted inflammatory cytokine panels (Bio-Plex Pro™ Human Cytokine 27-plex assay). We have also collected radiographic (Eaton-Littler), anthropometric and longitudinal pain (VAS, T ASD, quickDASH) and functional (key pinch, grip strength) data to evaluate relationships between structure, pain, and systemic and local cytokine expression.

Results: Our data demonstrate no significant differences in age, sex, BMI, or radiographic score between surgical and non-surgically managed patients. Patients undergoing surgery have significantly higher pain as determined by quick DASH (54.5 \pm 18.3 vs 35.2 \pm 20.2, p<0.0001), Visual Analog Scale (70.8 \pm 16 vs 51.1 \pm 27.2, p<0.001), and Trapeziometacarpal Arthrosis Symptoms and Disability Questionnaire (60.4 \pm 17.6 vs 43.1 \pm 19.6, p<0.001). Systemically, these patients can be distinguished by differing levels of Interleukin-7, Granulocyte Macrophage Colony Stimulating Factor, Interleukin 1 A Receptor Antagonist, Monocyte Chemoattractant Protein 1, and Interferon Gamma, (p<0.05) at baseline. Additionally, 1-year follow-up of surgical patients indicate that four cytokines: Eotaxin, Interleukin 7, Interleukin 4, and Granulocyte Colony Stimulating Factor significantly change 6-months post-surgery and maintain this change 1 year post surgery (p<0.05). Interestingly, our data indicate that regardless of surgical status, a subset of patients with an "inflammatory" phenotype exists.

Conclusions: Overall, our study has demonstrated that circulating cytokines are capable of distinguishing TMOA disease severity, and has identified promising targets that have potential to be used as prognostic markers after validation in other cohorts. We have also identified a cluster of patients who self-segregate based on a group of inflammatory cytokines. Further evaluation of this patient subset may help develop an approach to personalized medicine among this population.

167 DISTINCT GALECTIN PROFILE IN INFLAMMATION-MEDIATED CARTILAGE DAMAGE

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Purpose: Galectins (Gal) are a family of carbohydrate binding proteins that serve as regulators of fundamental numerous biological processes