synovial membrane from the control group was heavily infiltrated with inflammatory cells and blood vessels were abundantly found in the subintimal layer. In contrast, synovial membrane from the CS and ZO group had mild infiltration of inflammatory cells with fewer blood vessels.

Conclusions: Inflammatory changes in the synovial membrane from the experimentally induced joints were reduced following supplementation of CS and ZO extracts. This was evidenced by the histological changes. The significant increase in the PGP 9.5 immunoreactive nerve fibres in CS and ZO group shows that these extracts have anti-inflammatory properties. Apart from this CS significantly increases the PGP 9.5-immunoreactivity with huge reduction of inflammatory cells compared to ZO. This indicates that the active component in CS plays a major role in reducing the inflammatory cells and increasing the nerve fibres. Therefore, CS can be a better alternative in the treatment of osteoarthritis than ZO.

526

PROTECTION AGAINST LIPOPOLYSACCHARIDE-INDUCED CARTILAGE INFLAMMATION AND DEGRADATION PROVIDED BY A BIOLOGICAL EXTRACT OF MENTHA SPICATA

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Purpose: The purpose of the current study was to assess the anti-inflammatory and/or chondroprotective properties of Mentha spicata (MS), purpose-bred at the University of Guelph to express high levels of rosmarinic acid (RA), in LPS-stimulated cartilage explants.

Methods: RA content in MS leaves was determined by HPLC. MS was added to simulated gastric fluid and simulated intestinal fluid and incubated at 37°C (7% CO₂) for a total of 4 hours. Subsequently, pH was adjusted to 7.4 and liver microsomes from rat and NADPH were added in order to mimic metabolism of secondary plant metabolites by the liver and incubated for an additional 30 minutes. The resulting mixture was ultrafiltered (50 kDa) and an aliquot analyzed for RA and its primary hepatic metabolites (m-coumaric acid, caffeic acid, methyl rosmarinic acid and ferulic acid). A ‘blank’ simulated digest was made using the identical methodology but without including any MS. Cartilage explants (4mm diameter) were excised from the articulating surface of the intercarpal joint of 11 healthy pigs using a sterile 4mm dermal biopsy tool. Explants were cultured for a total of 28 days, all knees were harvested intact & histologically processed into 6 μm coronal sections yielding 15-20 sections per knee. The right knees of 10 week old 129S6/SvEv male mice were surgically induced to OA using the destabilization of the medial meniscus (DMM). Mice were treated with montelukast sodium in a preclinical mouse model of OA. Outcome measures were histological scoring methods performed on multiple sections in untreated- vs. montelukast-treated OA mice to assess potential chondroprotection and structure-modifying bone effects offered by montelukast.

Results: The mean maximal score for the montelukast-treated OA mice (Fig. 1) was reduced by 57%. For the first 48 h of culture, tissue culture media (TCM) contained no MS or LPS. From 24 - 96 h, TCM contained MS [0 (ie. ‘blank’), 8, 40, 80, 240, 400 μg/mL]. For the final 48 h, explants were exposed to an inflammatory stimulus (LPS; 0 or 3 μg/mL) in order to produce an inflammatory state in vitro. Samples were analyzed for PGE₂ (ELISA), IL-1 (ELISA), GAG (DMB), and NO (Griess Reaction). Data were analyzed using 2-way repeated measures ANOVA with respect to treatment and time. One-way RM ANOVA was used to detect changes in dependent variables over time within treatments. When a significant F-ratio was obtained, the Holm-Sidak post-hoc test was used to detect significantly different means. Significance was accepted when p<0.05.

Results: RA content of the dry leaves was 8.0% w/w. RA concentration in the biological extract was 49.3 μg/mL. Caffeic acid, ferulic acid, methyl RA and m-coumaric acid were identified in the biological extract, but were not identified in the undigested leaves. MS inhibited LPS-induced PGE₂ and NO at all doses tested. LPS-induced GAG release from explants at a dose of 80 μg/mL, but not at the lower or higher doses. LPS-induced IL-1 production was not affected by MS.

Conclusions: It is concluded that MS is an effective inhibitor of LPS-induced inflammation in porcine cartilage explants; this effect may be due to its high RA content. The relative contributions of RA and hepatic metabolites of RA are not known and are currently under investigation.

527

MONTELUKAST SODIUM AS A TREATMENT FOR EXPERIMENTAL OSTEOARTHRITIS IN MICE

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Purpose: Osteoarthritis (OA) is the most common form of arthritis, worldwide. The underlying causes of the disease are unknown however, the focal destruction of cartilage matrix and sclerosis of underlying bone are thought to be the two predominant factors involved in progression of osteoarthritic disease. The most effective treatment of OA remains the surgical removal and total replacement of the joint (TJR) in later stages of OA. Currently, no FDA-approved pharmacologic treatment of OA exists, particularly with regard to agents that interfere with the primary structural changes to cartilage and bone. The aims of the current study were to test one such disease-modifying OA drug (DMOAD) candidate, montelukast sodium in a preclinical mouse model of OA. Outcome measures were histological scoring methods performed on multiple sections in untreated- vs. montelukast-treated OA mice to assess potential chondroprotection and structure-modifying bone effects offered by montelukast.

Results: The right knees of 10 week old 129S6/SvEv male mice were surgically induced to OA using the destabilization of the medial meniscus (DMM). Mice were treated with montelukast sodium in a preclinical mouse model of OA. Outcome measures were histological scoring methods performed on multiple sections in untreated- vs. montelukast-treated OA mice to assess potential chondroprotection and structure-modifying bone effects offered by montelukast.

Results: The mean maximal score for the montelukast-treated OA mice (Fig. 1) was reduced by 57%. The mean summed score (Fig. 2) which represents a combined score of OA grade and stage was reduced by 43%. Direct morphometric measurements of MKS-treated OA (Fig. 3) showed a 42% reduction in average lesion area. Surprisingly, morphometric analysis of the subchondral bone area (Fig. 3) showed a 9% (medial) and 8% (lateral) decrease in OA-induced bony sclerosis with montelukast treatment. This latter finding was unexpected but could represent an...