

Evaluation of Pentosan Polysulfate

Summary: This was one of the first clinical studies of pentosan polysulfate (PPS) as an antiarthritic agent in horses. The researchers found that PPS treated the cause of osteoarthritis by significantly slowing or halting cartilage fibrillation and by increasing aggrecan synthesis has provided evidence to classify sodium pentosan polysulfate as a structure-modifying osteoarthritis drug, or SMOAD.

The performance of athletic horses depends greatly upon their legs, and the integrity of their joints, bones, tendons, and ligaments. A common disorder that affects the equine limb is osteoarthritis.

Osteoarthritis (OA), also known as Degenerative Joint Disease, involves pathologic changes in the joint structures, including the articular cartilage covering the joint surfaces, the subchondral bone beneath the articular cartilage, the synovial fluid inside the joint, and the synovial lining of the soft-tissue joint capsule. As cartilage is lost, the joint space narrows and the underlying subchondral bone remodels to form small bone growths, called “bone spurs” or osteophytes. These changes cause pain and inflammation due to chronic wear without protective cartilage.

Until recently, OA has been managed with drugs that treat the symptoms rather than the underlying cause of the disease. This symptomatic relief has been accomplished by using analgesics for pain relief, as well as steroids and nonsteroidal anti-inflammatory drugs (NSAIDs) to reduce inflammation and to provide pain relief in some cases. Recently, researchers of osteoarthritis have turned to the possibility of structure-modifying osteoarthritis drugs (SMOADs), which could treat the cause of the disease. One possibility of a SMOAD is pentosan polysulfate, which has recently undergone a clinical trial by the Orthopaedic Research Center at Colorado State University.

For over 30 years, pentosan polysulfate (PPS) has been used in Australia and Europe as an antithrombotic-antilipidemic agent, to prevent blood clots and fatty buildup. Clinical studies of pentosan polysulfate as an antiarthritic agent have been done in humans, dogs, sheep, rabbits, rats, and chickens, but this is one of the first studies in horses. Sodium pentosan polysulfate (NaPPS) was first approved as an injectable treatment for osteoarthritis in dogs in Australia in 1986, and then was also approved in New Zealand, Finland, the United Kingdom, Canada, and Ireland. PPS is approved for use in humans in most European Economic Community

countries, the Scandinavian countries, South Africa, and Australia.

Pentosan polysulfate is derived from beechwood hemicellulose, unlike other osteoarthritis drugs, such as hyaluronan, glycosaminoglycan polysulfate ester, and chondroitin sulfate, which come from animal or bacterial sources. The drug can be formulated as a sodium salt (NaPPS) or a calcium derivative (CaPPS), and can be administered orally, subcutaneously, intramuscularly, intravenously, or intraarticularly. CaPPS is absorbed orally and subcutaneously much better than NaPPS. The past 30 years of use have shown pentosan polysulfate to be a very safe drug with few side effects and low toxicity.

Proteoglycans are important for the ability of cartilage to withstand compression, as occurs whenever the leg bears weight. Osteoarthritis causes proteoglycans to be lost from the extracellular matrix, the supporting material surrounding the cells of articular cartilage. This process could be caused by a decline or change in the activity of chondrocytes, the cells of cartilage, and/or an increase in catabolism, the breakdown of components of cartilage. Both sodium pentosan polysulfate and glycosaminoglycan polysulfate ester (PSGAG), another antiarthritic drug, have been shown to stimulate the synthesis of proteoglycans. However, only NaPPS increased the amount of proteoglycan incorporated into the extracellular matrix, while PSGAG did not. In addition, PPS can increase proteoglycan production without the help of the cytokine interleukin 1. PPS also preserves the normal ratio of the components of cartilage.

A major component of the fluid of a synovial joint is hyaluronan, or hyaluronic acid. Hyaluronan is produced by synoviocytes or synovial fibroblasts, the cells of the joint capsule, and is important for the lubrication of articular cartilage. Osteoarthritis causes a decrease in the amount of hyaluronan produced, as well as a decrease in its molecular weight, the combination of which reduces its lubricating ability. Studies have shown that pentosan polysulfate increases both the synthesis

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and the molecular weight of hyaluronan produced in osteoarthritic joints. In contrast, glycosaminoglycan polysulfate ester (PSGAG) was not effective in accomplishing this goal of the treatment of osteoarthritis.

Normal articular cartilage is avascular, or without a blood supply, and is dependent on the underlying bone for blood. Osteoarthritis causes a reduction in the vascularity of the bone beneath the cartilage, and also causes deposits of the clotting factor fibrin and lipids in these vessels, which can clog them. PPS reduces these clots through its antithrombotic-antilipidemic activity and also encourages the regrowth of the endothelial cells lining these vessels. This improved blood supply increases the nutrition of the bone and cartilage cells, which improves their metabolic activity and also provides relief from the pain caused by the reduced blood flow.

Osteoarthritis causes components of the articular cartilage to be degraded and released into the synovial fluid, which results in an immunological response in the form of inflammation of the synovium, or joint capsule. These degraded cartilage components are attacked by antibodies and complement, which starts a complement cascade resulting in an increase in the degradation of the articular cartilage. Pentosan polysulfate binds to and inhibits the actions of leukocytes, or white blood cells, in order to provide an anti-inflammatory effect.

Osteoarthritic articular cartilage loses proteoglycan due to both reduced production and increased catabolism, or degradation, of proteoglycan. This process is caused by many enzymes and cytokines from chondrocytes, synoviocytes, and leukocytes. One of these enzymes, which increases in amount in osteoarthritic joints, is stromelysin (MMP-3), which is a matrix metalloproteinase that degrades components of the cartilage matrix, including aggrecan, type II collagen, and link protein. Pentosan polysulfate inhibits the activity and decreases the levels of MMP-3. Cytokines such as interleukin 1 (IL-1) and tumor necrosis factor α (TNF- α) promote articular cartilage degradation in osteoarthritis by reducing the synthesis of proteoglycan and collagen type II, and by increasing the release of matrix metalloproteinases. These two cytokines are counteracted by a third cytokine, interleukin-6 (IL-6), which inhibits IL-1 and TNF α and supports cartilage repair. Although PPS does not affect IL-1, it does decrease the production of

TNF α and increase IL-6 synthesis. Thus pentosan polysulfate reduces cartilage degradation by directly and indirectly affecting these inflammatory mediators.

All of these previous studies show that pentosan polysulfate has a beneficial effect on osteoarthritic joints. Although devoid of analgesic activity, PPS can provide symptomatic relief of osteoarthritis by correcting the pathological imbalances of the disease.

The current clinical study of sodium pentosan polysulfate was undertaken by Dr. David Frisbie, Ellen Ricky, Dr. Louise Southwood, Dr. Wayne McIlwraith and Heather Colhoun of the Orthopaedic Research Center at Colorado State University. This study involved eighteen horses, who underwent surgery to create a chip fracture defect in their carpal joint and then were exercised on a treadmill five days a week, the combination of which has been documented to create osteoarthritis. They received four weekly intramuscular injections of either sodium pentosan polysulfate or the control article, or placebo. Since the study was blinded, the researchers did not know which horses received the pentosan polysulfate injections and which received the control article, and thus the researchers' observations were unbiased.

Various parameters were used to determine the efficacy of the drug. Lameness exams were performed weekly on each horse and included carpal flexion tests to determine the amount of lameness due to pain of bending the joint and joint effusion scores to determine the amount of swelling in and around the joint. Ground reaction forces were recorded using a Tekscan mat at two, six, and ten weeks after surgery. Blood and synovial fluid were also collected weekly. The blood was used to look for serum markers of osteoarthritis, including collagen II α 1(2) epitope 846 of chondroitin sulfate, and carboxy propeptides of type II procollagen, as well as the coagulation factors of platelet count, prothrombin time, and activated partial thromboplastin time. The synovial fluid was also analyzed for the same three osteoarthritis markers, as well as for routine parameters, including total protein, white blood cell count, color, clarity, and mucin clot. Radiographs, or x-rays, were taken prior to surgery and on day 71 post operation. At necropsy, 72 days after surgery, the legs were dissected and the carpal joint abnormalities such as cartilage lesions, synovial adhesion, and the

appearance of the chip were described. The synovial membrane and joint capsule underwent histological preparation and were scored in the areas of cellular infiltration, intimal hyperplasia, subintimal edema, subintimal fibrosis, and vascularity. Histological preparation allows tissues to be evaluated under the microscope. Sections of articular cartilage from both damaged and undamaged sites were also prepared for histological evaluation. Additional articular cartilage was frozen to allow the possibility of analysis for proteoglycan content, if deemed necessary. The most important factors in this study were the lameness exams, routine synovial fluid analysis, osteoarthritis markers in blood serum and synovial fluid, and the histological evaluation of the synovial membrane and cartilage.

A clinical study is empirically evaluated by whether or not the data is statistically significant, which is indicated by a p-value less than .05. There were four major statistically significant findings. First of all, there was a significant difference between the affected limbs, in which chips had been created, and the unaffected limbs, in the parameters of lameness score, flexion score, effusion score, total protein of synovial fluid, the osteoarthritis markers of carboxy propeptides of type II procollagen (equine collagen) and epitope 846 of chondroitin sulfate in synovial fluid, radiographic scores of enthesiophyte formation (bone remodeling at the site of a ligament, tendon, or joint capsule attaching to the bone) and subchondral lysis (death of bone cells located underneath the articular cartilage), hemorrhage and full thickness articular cartilage erosion scores at necropsy, vascularity and cellular infiltration scores of synovial membrane histology, and histological cartilage damage scores. These findings indicate that the surgical model was successful in creating osteoarthritis in all of the horses in the study. The clinical signs of lameness, loss of joint flexibility, and joint effusion indicate that there was pain and inflammation associated with osteoarthritis. The radiographic evidence of enthesiophyte formation and subchondral lysis is also a sign of osteoarthritis that can be observed clinically. The erosion of articular cartilage indicates traumatic injury and the progression of osteoarthritis. Secondly, following a natural log transformation of the data, there was a significantly higher level of epitope 846 of chondroitin sulfate in the blood of horses treated with pentosan polysulfate, compared to horses treated with the control article. The level of epitope

846 indicates the level of aggrecan synthesis, which is an important molecule in the health of articular cartilage. Increases in the level of epitope 846 in the blood and synovial fluid have previously been interpreted as a response to repair the damage created by osteoarthritis. Since the level of epitope 846 was elevated in the synovial fluid from both the affected and unaffected limbs of horses treated with pentosan, it is more likely that this is a systemic response of the entire body to increase aggrecan synthesis in reaction to the drug, rather than a response of the joint to the damage created by the disease. Thirdly, in the histological analysis of the synovial membranes, there was significantly higher cellular infiltration in the affected limbs of PPS-treated horses, in comparison to the affected limbs of control horses. This finding is considered to be a questionable result. Finally, histological evaluation revealed significantly less articular cartilage fibrillation in the affected limbs of horses that received PPS, versus horses that received the control article. Articular cartilage fibrillation, or local fragmentation of the cartilage along the planes of the collagen fibrils, occurs due to traumatic injury as well as the progression of osteoarthritis.

In addition, there were several strong trends, which were close to being statistically significant. Histologically, there was a strong trend, with a p-value of .062, for decreased cartilage damage in the affected joint of horses treated with pentosan polysulfate. Another strong trend, with a p-value of .063, showed improvement in the overall average histological score, which was a combination of all of the histological parameters measured, in the affected limbs of PPS-treated horses, compared to the affected limbs of control horses. The observation that the factors of blood coagulation, including platelets, prothrombin time, and activated partial thromboplastin time, appear not to be affected by treatment with pentosan polysulfate indicates that blood clotting in the entire body is not affected by this dose of the drug. Therefore, this potential negative side effect has apparently been avoided, at least at the time it was measured in this study, which was 7 days after treatment.

In conclusion, this surgical model successfully created osteoarthritis, even though there was not an evident increase in the concentration of white blood cells in the synovial fluid to indicate inflammation and there was not a difference in glycosaminoglycan concentration and synthesis, as noted in previous

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models. This study has provided evidence to classify sodium pentosan polysulfate as a structure-modifying osteoarthritis drug, or SMOAD. In support of this statement, PPS has treated the cause of the disease by significantly slowing or halting cartilage fibrillation and by increasing aggrecan synthesis, as evidenced by the elevated level of epitope 846 of chondroitin sulfate, in order to provide the materials necessary for cartilage health. No negative side effects to PPS were discovered in this study. However, pentosan polysulfate may not be sufficiently potent to demonstrate a clinical effect at the current dose of 3 mg/kg.

This study has demonstrated that pentosan polysulfate has a beneficial therapeutic effect. Therefore, further study is indicated. These studies could increase the dose frequency and also evaluate the drug in clinical models, in which the horses studied have osteoarthritis that occurred naturally. Further study could find the appropriate use of pentosan polysulfate to treat the cause of osteoarthritis.

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