4CYTE™ is a Vet Only product. This information is intended for the Veterinary Professional only.

4CYTE™ is an animal health product. Product specific research has been conducted by independent research facilities to gain insight into the mode of action of 4CYTE™. The summaries and conclusions sited are those of researchers, many of which have been peer reviewed and published. They should not be misconstrued as claims made by Interpath. Ultimately the Veterinarian needs to assess the merits of 4CYTE™ for use in-clinic.

Please contact Interpath with any questions and or for further information on any of the papers presented in this document.
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A randomised controlled masked clinical trial of a novel nutraceutical for treatment of osteoarthritis in the dog.

Melbourne University, Faculty of Veterinary Science (2012)

Sasha’s EQ reduces synovial fluid PGE₂ in horses with osteoarthritis: a clinical study

Wendy Pearson, Nathalie Cote, Pub Peding EVJ


W. Pearson, M. Lindinger. Equine Veterinary Journal (2009)41(1)00-00

Evaluation of inflammatory responses induced via intra-articular injection of interleukin-1 in horses receiving a dietary nutraceutical and assessment of the clinical effects of long-term nutraceutical administration.

Wendy Pearson, Michael W. Orth; Michael I. Lindinger: AMJVR, Vol.70, No.7,(2009)

Effects of simulated digests of Biota orientalis and a dietary nutraceutical on interleukin-1 induced inflammatory responses in cartilage explants

Wendy Pearson, Michael W. Orth; PhD; Niel A. Karrow, Michael I. Lindinger, AJVR, Vol 69, No.12, December (2008 )

Anti-inflammatory and chondroprotective effects of nutraceuticals from Sasha’s Blend™ in a cartilage explant model of inflammation


*Note Sasha’s EQ is the original term for 4CYTE™ Equine
Objective:
The objective of this study was to compare the efficacy of 4CYTE™ with a positive control NSAID carprofen, for the treatment of naturally occurring osteoarthritis in the dog. The primary hypotheses was that improvement in lameness scores were not different between treatments.

Summary:
A randomised controlled clinical trial was used to assess response to therapy. 82 dogs with clinical signs of osteoarthritis were enrolled in the study of which 16/82 (19.5%) were then excluded due to lack of radiographic evidence of osteoarthritis. The remaining 66 dogs completed the study. The main breeds represented were cross breeds 20/82 (24%), Labradors, Retrievers and Border Collies as the three main pure bred cases 22/82 (27%) and the remaining dogs were a variety of pure breeds. All dogs were over 6 months of age, from 10 to 50 kg and were enrolled from May 2011 to May 2012 in Victoria, Australia. The primary outcome was defined as the binomial “improvement versus no improvement” of Owner Lameness Score (OLS) at Day 28 compared to Day 0. Other outcomes were the improvement in OLS, Veterinary Lameness Score (VLS) and the Owner Mobility Scores (OMS) at Days 14 and 28. All outcomes used ordinal observation scales with masked assessment. Outcomes were evaluated for non-inferiority using the 90%CI of the difference between two independent proportions. The Hodges-Lehmann estimate of the median of the pairwise differences was also calculated from the ordinal ratings.

Key Findings:
• For dogs dosed with 4CYTE™ versus carprofen, OLS at day 14 improved for 10/29 (34%) dogs and 13/37 (35%) dogs, and at day 28 had improved for 14/29 (48%) and 13/37 (35%) respectively.
• 4CYTE™ was found to be non-inferior to carprofen at day 14 for the OMS, and non-inferior at day 28 for all three outcomes.
• The results support the conclusion that 4CYTE™ was at least as effective as carprofen in relief of pain and lameness resulting from osteoarthritis in adult dogs, i.e. clinical end-point bioequivalence was demonstrated.
A DIETARY NUTRACEUTICAL PRODUCT REDUCES SYNOVIAL FLUID PROSTAGLANDIN E2 IN HORSES WITH OSTEOARTHRITIS: A DOUBLE-BLIND, RANDOMIZED TRIAL.

Wendy Pearson, Nathalie Cote, and Marc Desjardins, (Publication Pending)
Presented at: 2013 American Association of Equine Practitioners Convention, Anaheim.

Objective:
The inclusion of DN (composed of New Zealand green lipped mussel (NZGLM), abalone (AB), marine cartilage (MC) and a proprietary lipid extract of Biota orientalis (Epiitalis®) in the diet of horses immediately following surgical removal of osteochondral fragment would reduce synovial fluid prostaglandin E2 (PGE₂), nitric oxide (NO) and glycosaminoglycan (GAG), while improving clinical signs of articular inflammation in these horses.

Summary:
A dietary nutraceutical product (DN) has previously demonstrated prophylactic PGE₂-inhibitory effects in interleukin-1 stimulated cartilage explants and in horses with experimentally-induced articular inflammation. The current study was undertaken in order to evaluate the therapeutic effect of including DN in the diet of horses with articular inflammation and/or cartilage damage secondary to osteochondral fragmentation of the carpal or metacarpal joints.

Fifteen horses presenting at an equine hospital for surgical removal of an osteochondral fragment of the carpal or metacarpal joints were included. Horses received DN (0 or 21 g/day) for 42 days beginning immediately after surgery. Synovial fluid pre and post supplementation was analyzed for PGE₂, GAG, and NO. Radiographs and lameness assessments were also obtained.

Key Findings:
- Synovial fluid PGE₂ was significantly reduced from baseline in horses receiving DN, but not in those horses treated with surgery alone.
- Data supports previously published experimental evidence of an inhibitory effect of DN on synovial fluid PGE₂.
- DN may be a useful post-surgical treatment for elevated PGE₂ in horses with surgical removal of osteochondral fragments.
Summary:
Nutraceuticals are increasingly applied to the management of equine arthritis and joint disease, particularly those based upon glucosamine in horses, which appeared more than 25 years ago. It was not until 1992 that isolate studies began to be reported. Since that time, 15 in-vivo papers have been published in the equine literature, usually on products already commercially available and often seeking evidence for efficacy.

These studies demonstrate an encouraging trend to manufacturers of these products investing in research, but most do not meet a quality standard that provides sufficient confidence in the results reported. This review discusses the entirety of published in-vivo research on glucosamine-based nutraceuticals (GBN) for horses, including Cosequin, Cortalax, Synequin, Sasha’s EQ (4CYTE™ Equine), Myristol, chondroitin sulphate, glucosamine sulphate and glucosamine hydrochloride; and considers experimental limitations of this research along with their impact on interpretation of results.

A quality score was calculated for each paper according to preset quality criteria. A minimum quality standard of 60% was set as the threshold for confidence in interpretation of results. Of the 15 papers reviewed, only 3 met the minimum quality standard.

Key Findings:
• It is concluded that the quality of studies in this area is generally low, prohibiting meaningful interpretation of the reported results.
• 4CYTE™ Equine (Sasha’s Equine) has obtained the highest score (68% - see table 3 Supp1) according to these predetermined criteria.
• In particular, the strength of the 4CYTE™ Equine research is that:
  • Control horses fed a placebo diet were included in both studies (safety study and induced inflammation study).
  • A safety study was done for 84 days at 7 times the maintenance dose.
  • Synovial (joint) fluid was assessed after 14 days of 4CYTE™ Equine supplementation.
• Intra-articular inflammation was induced and the effect of 4CYTE™ Equine fed horses was measured compared to placebo fed horses.
• The important thing to realise here is that the research done on 4CYTE™ Equine focused on objective outcome measures (blood tests, urine tests, evaluation of GAGs and enzymes in synovial fluid, joint circumference etc.), rather than the subjective assessment (lameness, flexion tests etc.) of an evaluator.
Objective:
To evaluate inflammatory responses induced via intra-articular recombinant human interleukin-1 (IL-1) treatment in horses receiving a dietary nutraceutical (DN; composed of mussel, marine cartilage, abalone and Epitaleis®) and assess the clinical effects of long-term DN administration.

Summary:
In a previous in-vitro study involving the DN, a simulated digest of the product significantly inhibited the inflammatory response of equine cartilage explants to exogenous IL-1. To correlate those in-vitro data with in-vivo data, IL-1 was injected into inter-carpal joints of horses to simulate an inflammatory challenge in the present study (of 22 healthy horses).

12 horses were fed 0, 15, 45 or 75mg of DN (3 horses/treatment) daily for 84 days. General health and clinic pathologic variables were monitored at intervals. Ten other horses received 0 or 15mg of DN/d (5 horses/treatment) for 29 days. One inter-carpal joint in each horse was injected twice with IL-1, (10 and 100ng on days 0 and 1, respectively) and the contralateral joint was similarly injected with saline solution. Synovial fluid prostaglandin E2, (PGE2) sulfated glycosaminoglycan (GAG), nitric oxide (NO), protein concentrates and leukocyte counts were analysed before and at intervals after injections.

Key Findings:
• Administration of the DN (up to 75g/d) to horses for 84 days did not induce any adverse effects.
• Results suggested that administration of the DN may be useful in preventing inflammation associated with arthritis and degenerative joint disease in horses.
• Intra-articular injection of the IL-1 caused increased concentration of synovial fluid PGE2 and GAGs consistent with inflammation and cartilage degradation associated with arthritis.
• Feeding of DN inhibited the increase of PGE2 (54.6% decrease) and GAG induced by the intra-articular injection of IL-1 highlighting DN anti-inflammatory and chondroprotective properties as also demonstrated in in-vitro studies.
• Feeding DN lead to a decrease in the circumference of joints swollen by the intra-articular administration of IL-1 consistent with inhibition of increased joint effusion seen in cases of arthritis.
Objective:
To test the hypothesis that simulated digests of Biota orientalis (Epiitalis®) and a dietary nutraceutical (DN; composed of mussel, marine cartilage, abalone, and BO seed lipid extract) inhibit prostaglandin E2 (PGE2), nitric oxide (NO), and glycosaminoglycan (GAG) production in interleukin-1 (IL-1) stimulated cartilage explants.

Summary:
The University of Guelph conducted a series of experiments evaluating the effects of Epiitalis® on cartilage inflammation and degradation induced by interleukin-1 (IL-1). Research models applied in this study were designed to promote in-vitro conditions consistent with the desired context of use of Epiitalis®: i.e. that the product was to be administered orally; the target tissue was articular cartilage.

Key Findings:
• Results of the present study supported the hypotheses that Epiitalis® and DN modulate response of articular cartilage explants to exogenous IL-1 without adversely affecting viability or metabolism of chondrocytes.
• The use of indomethacin as a reference anti-inflammatory drug provided an allopathic benchmark against which the effects of these 2 nutraceuticals could be compared.
• Simulated digests of NZGLM, SC, and abalone (the 3 constituents of the DN other than Epiitalis®) are known to have anti-inflammatory or cartilage-sparing effects in IL-1 stimulated cartilage explants. The data obtained in the present study indicated that the oil from the seed of BO also partially inhibits IL-1 dependent PGE2 production by cartilage explants.
• The heterogeneous composition of the DN (i.e., NZGLM, SC, abalone, and BO) resulted in significantly improved NO - and PGE2- inhibiting properties, compared with the effects of the individual ingredients alone at the same dose. This synergistic effect may result from each ingredient influencing divergent PGE2- or NO-synthetic pathways.
• In the present study, cell viability subsequent to IL-1 challenge was significantly increased by Epiitalis® sim and DN sim. These increases were not a result of reduced apoptosis, as might have been predicted from the NO inhibitory properties of the DN, because the increases in viability resulted from an increase in staining of live cells but not a decrease in staining of dead cells.
• The DN sim, perhaps through provision of Epiitalis® sim (which had almost equal effect on cell viability), may produce an agonist effect on the prostanoid receptor involved in chondrocyte proliferation, resulting in an increase in live-cell staining.
**Objective:**
The objective of this study was to evaluate the anti-inflammatory and chondroprotective effects of the New Zealand green lipped mussel (NZGLM), abalone (AB), and marine cartilage (SC) using a cartilage explant model of inflammation.

**Summary:**
New Zealand green lipped mussel (NZGLM), abalone (AB), and marine cartilage (SC) are extensively used for treatment of and/or as preventatives for arthritis, despite a relative paucity of scientific evidence for efficacy. This research integrated a simulated digestion protocol with ultrafiltration and cartilage explants to generate new information on the anti-inflammatory and chondroprotective properties of NZGLM, SC and AB.

Each nutraceutical was artificially digested using simulated gastric and intestinal fluids. Each filtrate was applied individually to cartilage explants before the explants were stimulated with interleukin-1 (IL-1) to induce an acute inflammatory response. Media were collected daily for 48hrs and analyzed for prostaglandin E2 (PGE2), glycosaminoglycan (GAG) and nitric oxide (NO), and cartilage tissue was differentially stained to determine the relative proportion of live and dead cells.

**Key Findings:**
- SC and NZGLM significantly inhibited IL-1 induced PGE2 synthesis and IL-1 induced GAG release, and AB was an effective inhibitor of IL-1 induced NO production.
- The three test nutraceuticals affect at least three major pathways involved in the catabolic cycle of arthritis and may prove important treatments and/or preventatives for the pain and degradation associated with this condition.
- The methodology and results describe a useful model for evaluating dietary nutraceuticals in-vitro.