4CYTE™

Research Summary

Type	Researcher	Objective	Protocol	Results
In-Vitro	**Pearson et al Guelph University 2007**	Using an integrated simulated digestion protocol with ultra-filtration and cartilage explants to determine the anti-inflammatory and chondroprotective properties of New Zealand Green Lipped Mussel (NZGLM), Marine Cartilage (MC) and Abalone (AB) (this is SASHAS BLEND formulation).	• Each nutraceutical was artificially digested before being applied individually to porcine cartilage explants. • Explants were then stimulated with IL-1 to deliver an acute inflammatory response. • Explants were conditioned with nutraceuticals and indomethacin (Non-Steroidal Anti-inflammatory Drug (NSAID)).	• MC & NZGLM significantly inhibited IL-1 induced Prostaglandin E2 (PGE2) and GAG release. Both actives also improved cartilage cell viability. • AB was an effective inhibitor of IL-1 induced Nitric Oxide (NO) production but had no effect on cartilage cell viability. • Indomethacin significantly inhibited PGE2, had no effect on NO and had an anti-proliferative (negative
In-Vitro	**Pearson et al Guelph University 2008** Published AJVR 2008	To test the hypothesis that simulated digests of Biota Orientalis (BO) and a dietary nutraceutical (DN; composed of NZGLM, MC, AB & **EPIITALIS®**) inhibit PGE2, NO and Glycosaminoglycan (GAG) production in IL-1 stimulated porcine cartilage explants.	• Each nutraceutical was artificially digested before being applied individually to porcine cartilage explants. • Explants were then stimulated with IL-1 to deliver an acute inflammatory response. • Explants were conditioned with nutraceuticals and indomethacin (NSAID).	• Combination of the 4 ingredients vs individual ingredients significantly inhibited PGE2 and NO. • At 24/48 hours PGE2 inhibition by **4CYTE™** was comparable to Indomethacin (NSAID). • Biota Oil (**EPIITALIS®**) increased chondrocyte cell viability (proliferation) whereas Indomethacin suppressed PGE2, but increased NO and GAG leaching (chondrocyte death).
In-Vitro	**Pearson et al Guelph University 2009** Published AJVR 2009	To evaluate inflammatory responses induced via intra-articular recombinant human IL-1 treatment in horses receiving a dietary nutraceutical (**4CYTE™**; composed of NZGLM, MC, AB & **EPIITALIS®**) and assess the clinical effects of long-term **4CYTE™** administration. STUDY SIZE: 10 horses (5/ treatment & 5/placebo control)	• Horses were fed their allocated diet (treatment or control) for the entire duration of the experiment (29 days). Diet feeding was commenced 14 days prior to administration of an intraarticular injection of IL-1. Horses were monitored and evaluated at intervals until day 15. Samples of synovial fluid and jugular venous blood were collected from each horse on days −14, 0 (immediately prior to administration of IL-1-baseline), 1 (immediately prior to administration of IL-1), 1.3 (ie, 8 hours after the second IL-1 injection), 2, 4, 8, and 15.	• No side effects at up to 5x daily dose. • At Day 1.3, **4CYTE™** fed horses had significantly lower levels of synovial fluid PGE2 and GAG concentration levels compared with Control. • Joint circumference measurements in **4CYTE™** fed horses were significantly improved compared to Control. • Clinical trial suggests that administration of **4CYTE™** may be useful in preventing inflammation associated with naturally occurring arthritis and Degenerative Joint Disease (DJD).
Clinical	**Pearson et al Guelph University Milton Equine Hospital 2013**	It was hypothesized that inclusion of **4CYTE™** in the diet of horses immediately following surgical removal of osteochondral fragment would reduce synovial fluid PGE2, NO and GAG, while improving clinical signs of articular inflammation in these horses.STUDY SIZE: 13 horses (7/ treatment & 6/ placebo control)	• Thirteen horses presenting at an equine hospital for surgical removal of an osteochondral fragment of the carpal or metacarpal joints were included. • Horses received **4CYTE™** (0 or 21 g/day) for 42 days beginning immediately after surgery. Synovial fluid pre- and postsupplementation was analyzed for PGE2, GAG, and NO. • Radiographs and lameness assessments were also obtained.	• Horses receiving **4CYTE™** had significantly reduced levels of PGE2 and GAG post surgery vs surgery alone horses. • Surgery alone horses' PGE2 did not reduce from post-surgery through to Day 42. • PGE2 plays a central regulatory role in initiation and progression of arthritis and continues to be the primary pharmacological target for treatment of pain and inflammation associated with pre-existing arthritis. • There was no effect on NO as horse cartilage seems to produce a barely detectable level of NO (Species specific).

4CYTE™

Type	Researcher	Objective	Protocol	Results
Clinical	**Benker et al Ocala Equine Medical Centre 2014**	To establish the clinical efficacy of rates of **4CYTE™ EPIITALIS® FORTE** oral gel application in horses with symptoms of joint inflammation and osteoarthritis. SAMPLE SIZE: 10 Horse trial (5 horses/treatment loading for 14 days, 5 horses/ treatment loading for 28 days).	• The following assessment measures utilized at pre-treatment, after Day 14, Day 28 and Day 42 were: Veterinary Lameness Score (0 -4) adapted from AAEP, Joint Circumference Measurement, Handler Mobility Score (completed by handlers), and Heat and Swelling. • Horses in the study all had consistent lameness, significant heat and swelling, significant reduction in their range of movement.	• Results consistent between Day 0 and Day 28 across both groups (no benefit to loading with **4CYTE™ EPIITALIS® FORTE** for 28 days compared to 14 days) • Lameness – at Day 14 was absent or difficult to observe in 8 horses, at Day 42 only one horse had subtle limp. • Range of Movement – at Day 14, 8 horses were showing full range of movement and at Day 42 only one horse did not have full range. • Heat and Swelling – at Day 14, 8 horses had nominal to absent levels and at Day 42 only 1 horse had any symptom. • At conclusion the 1 horse that did not improve across all assessment measures was found to have adhesions on his digital sheath.
Clinical	**Whittemet al Melbourne University 2013** Published AVJ 2021	To compare the efficacy of **4CYTE™** canine granules with the Reference Product (carprofen) as a positive control non-steroidal antiinflammatory agent (NSAID), for the treatment of naturally occurring osteoarthritis radiographically evident in the dog. SAMPLE SIZE: 66 Dogs	• Randomised, masked, positive controlled, parallel group, intention clinical trial. • Three outcome measures were utilized to assess the efficacy of both groups: Owner Lameness Score (OL), Veterinary Lameness Score (VL) and Owner Mobility Score (OM). • Results will be determined off scores taken at Day 14, Day 28, between Day 14 and 28, and finally improvement from baseline at any time point.	• **4CYTE™** found to be statistically non-inferior to the control (carprofen) at Day 14 for the OLS, and at Day 28 **4CYTE™** was found to be statistically non-inferior for all three outcomes (OLS, OM, & VL) • These results strongly support the conclusion that **4CYTE™** is not inferior to carprofen, and that **4CYTE™** can be declared as bioequivalent to carprofen by end-point efficacy.
Clinical	**T Beths et al Melbourne University** Published AVJ 2020	To assess the efficacy of a new nutraceutical **4CYTE™ EPIITALIS® FORTE**, containing as a stand alone, a proprietary plant seed oil extract, in dogs with naturally occurring OA	• Pilot study assessed 46 dogs using subjective Helsinki Chronic Pain Index (HCPI) and objective (TPI%) measures over 28 days. • Day 0, dogs completed 3 gait assessments over the walkway and HCPI was completed by the owner. • Day 28, dogs completed 3 more gait assessments over the walkway and owners completed the HCPI questionnaire	• • 74% of dogs registered TPI% improvement • 71% of dogs on concurrent OA medication, also improved on both outcomes • 94% of dogs that demonstrated TPI% improvement, also showed improvement in HCPI scores • 93.5% of dogs improved their HCPI scores

Type	Researcher	Objective	Protocol	Results
Clinical	**K.Seabaugh et al Colorado State University** *Published Frontiers in Veterinary Science 2022*	To evaluate the efficacy of **EPIITALIS®** in horses with surgically induced osteoarthritis by assessment of clinical, biochemical histological, and imaging end points	16 horses with surgically induced osteoarthrits in one joint and sham operation in the contralateral limb were randomised on day 0 to receive 2.5ml of **EPIITALIS®** or placebo. • Synovial fluid samples takenon day 0 then weekly from day 14 • Radiographs taken Day 0, 14 and 70, MRI taken day 7 and 70 • Lameness assessed day 0 then weekly from day 14 • GAG levels taken day 0 then every other week from day 14 • Tissue assessment taken day 0 and day 70.	Significant reduction in PGE2 to baseline by week 5 in **EPIITALIS®** treated horses compared to placebo Significant reduction in White Blood Cell count in **EPIITALIS®** treated horses compared to placebo Signifcant radiographic severity reductions observed with **EPIITALIS®** treated horses 63% lower than placebo. Joint effusion the only common abnormality between the two groups.
Clinical	**P. Mitchell et al**	To explore the clinical efficacy of **EPIITALIS®** in patients with knee pain due to osteoarthritis (OA)	235 patients with x-ray diagnosed K&L II/III knee OA and knee pain ≥60 on a 100-point VAS scale were randomised to receive **EPIITALIS®** high, medium, low dose or matching placebo for a total of 56 days • VAS pain and mWOMAC were assessed at baseline and on days 0,14, 28 and 56. • SF-36 Quality of Life assessments were completed at baseline and on day 56 • OMERCT-OARSI responder index was calculated on day 56	• By day 56, patients in all three **EPIITALIS®** treatment groups exhibited significantly greater reductions in VAS pain compared to patients taking placebo. • On day 56, the OMERCT-OARSI responder index reached approximately 80% for each **EPIITALIS®** group, compared to 10.9% in the placebo group • The SF-36 total score as well as each of the eight SF-36 sub-domain scores were all significantly increased (baseline to day 56) for all three doses of **EPIITALIS®** compared to the placebo group • The mWOMAC total score as well as each of the three mWOMAC sub-domain scores (pain, stiffness, function) were all significantly decreased (baseline to day 56) for all three doses of **EPIITALIS®** compared to the placebo group. • The results provide strong support for the use of **EPIITALIS®** as a safe and efficacious intervention to treat patients with clinical knee OA