

Figure 1. The linearity was analyzed by serially diluting (1:2, 1:4, 1:8, and 1:10) the mixed QC samples (MQC) and the unspiked pooled samples (P). The observed concentration was plotted against the reciprocal of the dilution.

loss in the stability of the CTX II epitope out to 4 freeze/thaw cycles. Differences in biological activity were identified when examining samples from 3 normal rested horses (111.40 ± 33.3 pg/ml), the same horses after 5 months race-training (158.18 ± 111 pg/ml), and those with naturally-occurring OC injuries (38.96 ± 12.91 pg/ml) (Figure 2).

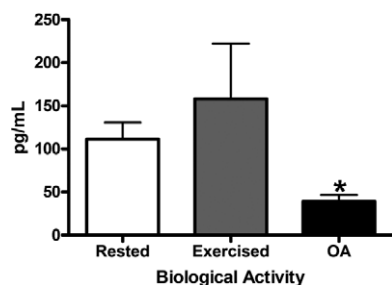


Figure 2. Differences in biological activity identified between samples from 3 horses before (Rested) and after 5 months race-training (Exercised) as well as 3 horses with OA. Significant differences between groups are represented as * $P < 0.05$ using a Kruskal-Wallis test.

Conclusions: The Serum Pre-Clinical Cartilaps[®] ELISA is a reproducible and valid assay for use with equine serum.

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OSTEOPROTEGERIN AND RECEPTOR ACTIVATOR OF NUCLEAR FACTOR κ B LIGAND IN SYNOVIAL FLUID AND SERUM IN PATIENTS WITH PRIMARY KNEE OSTEOARTHRITIS

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Purpose: to evaluate the relationship between OPG and RANKL in synovial fluid and serum in patients with primary knee osteoarthritis and disease severity as it is graded radiologically according to the system of Kallgren and Lawrence. To demonstrate the role of the RANKL/OPG system in the pathophysiology of primary knee osteoarthritis.

Methods: the study population included 37 patients (9 males, 28 females) with mean age 64.9 years (ranging from 50 to 80 years). Synovial fluid was aspirated from the affected joint during surgery where a total knee arthroplasty was performed. Blood samples were obtained from the same patients 1 to 2 hours before surgery. Anteroposterior weight bearing radiographs of the patients' knees were performed and osteoarthritic changes were graded according to the system of Kallgren and Lawrence.

Results: we found that OPG concentration in synovial fluid was significantly higher than in serum (257.5 ± 76.9 vs 49.2 ± 10.1 pmoles/l, $p < 0.0001$), indicating that the increase of OPG in

patients with knee OA is not systemic but rather localized in the affected joint. OPG levels were increased in synovial fluid in relation with the severity of knee OA and were significantly higher in patients with OA grade 4 than in those with grade 1 or 2. RANKL levels were found low in both synovial fluid and serum (1.5 ± 1.0 and 1.0 ± 1.0 pmoles/l respectively). Serum levels of OPG and RANKL did not correlate with the severity of knee OA. **Conclusions:** based on the fact that OA is characterized by progressive deterioration of the articular cartilage, the increase of OPG in synovial fluid of individuals with knee OA might reflect a compensatory response by chondrocytes or synovial fibroblasts to destabilization of the coupling between degradation and synthesis of articular cartilage. The increased concentration of OPG might thus serve to protect cartilage rather than be a cause of osteoarthritis.

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COMPARISONS OF ORIGINAL AND AUTHENTIC PHARMACEUTICAL AVOCADO AND SOYA UNSAPONIFIABLES VERSUS SOME ALLEGED NUTRACEUTICAL IMITATIONS BY GAS CHROMATOGRAPHY ANALYSIS

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Purpose: Avocado and Soya (A/S 1:2 w/w) unsaponifiables (ASU), developed and patented by Laboratoires Expanscience, are the components of a medicinal product found in numerous countries against degenerative osteoarthritis. The main ingredients are: 30% tocopherol, 25% sterols and 25% specific active molecules. The originality is based on A/S ratio and Avocado specific modified unsaponifiables obtained by chemical transformation of precursors. This modification by hemisynthesis constitutes one of the Laboratoires Expanscience know-how and is patented.

In vitro published studies showed effects of ASU on the anabolism (increase in the synthesis of collagens, proteoglycans and TGF β) and catabolism (inhibition of synthesis of MMPs, PGE2, and pro-inflammatory cytokines) of osteoarthritic chondrocytes. Moreover, clinical studies demonstrated the symptomatic effect of ASU combined to a reduction in NSAID administration. The pharmaceutical status requires perfect controls of the extraction process, of the hemisynthesis from precursors to converted molecules and an excellent between-batch reproducibility.

This is ensured by a strict control of the whole plant process: contractual partnerships with local subcontractors, expertise in vegetable lipid chemistry allowing an exact definition of the plant species extracted and the soils together with the application of GMP rules for the whole process from the plant starting material up to the pharmaceutical active ingredient.

Attracted by the scientific renown and success of pharmaceutical ASU, nutraceutical companies have attempted to develop imitations by suggesting more or less clearly a relation between the composition and even the activity of the original pharmaceutical.

Methods: An analytical study of 7 nutraceutical products (for human and veterinary uses) claiming the name ASU has been performed by gas chromatography methodology.

Results: The analytical study of the nutraceutical products versus the original and authentic ASU showed:

- The complete absence of specific molecules which are patented (obtained by transformation of avocado precursors) evaluated by GC assay of silyl derivatives on a 5%-phenylmethylpolysiloxane capillary column equipped with a cold on-column injector and a FID detector;
- A content of unsaponifiables of less than 25% measured by a method of the American Oil Chemistry Society N° Ca 6a-40;
- A lower sterol and tocopherol content of less than 15% and 5%