Abstract

Biomarkers measured in equine synovial fluid are able to differentiate pathologic from exercise-induced changes in joint tissues. These biomarkers should be useful in diagnosing joint disease and monitoring treatment effects.

1. Introduction

Lameness, and more specifically joint disease, is a significant cause of loss of use of athletic horses. Consequently, lameness also has a large economic impact on the horse industry. Therefore, better methods of diagnosing and monitoring joint disease are always needed.

Biomarkers have been beneficial in assessing osteoarthritic (OA) patients in a number of species including humans, dogs, and horses. Questions have been raised concerning the ability of biomarkers to distinguish the pathologic changes of OA from normal metabolic alterations that occur secondary to exercise. Previous work suggests biomarkers used in human medicine can be useful in detecting joint pathology in naturally occurring cases of equine osteochondral fragmentations (chip fractures) [1]. Biomarkers specific for bone and cartilage turnover have been validated in equine synovial fluid and serum, although the effect of exercise alone on these biomarkers has not been assessed in a controlled study.

The objective of this study was to determine if synovial fluid biomarkers could differentiate subjects undergoing a strenuous exercise protocol from subjects with a solitary OA joint that were enrolled in a similar exercise protocol.

2. Materials and Methods

This study was an experimentally controlled, randomized block design that used 16 horses in an established model of OA [2]. Synovial fluid samples were collected weekly throughout the study period. All horses began a 21-day strenuous athletic exercise regimen after a 2-wk acclimatization period. On Day 22 of the study, arthroscopic surgery was performed on both mid-carpal joints of all horses. In eight horses, OA was induced in one of the mid-carpal joints. The other eight horses had a sham arthroscopic examination. Horses were rested for 14 days after surgery and then returned to the previous exercise regimen for 56 additional days. Synovial fluid was assessed for total protein concentration, white blood cell count (WBC), and levels of the inflammatory marker, prostaglandin E2 (PGE2). Additionally, biomarkers for aggrecan synthesis (CS-846), proteoglycan release (sGAG), type II collagen synthesis (CPII), type I and II collagen degradation (COL2 - 3/4C_short), and bone synthesis (osteocalcin) were estimated in the synovial fluids. Horses were assessed for lameness using the AAEP grading scale before initiation of the study and after termination of the study. Statistical analysis used both a mixed model analysis of variance and a discriminate analysis, with P < 0.05 considered significant.

3. Results

Induction of OA resulted in a significant increase in lameness within limbs containing OA joints (P < 0.01). All biomarkers increased significantly with exercise, although some plateaued during the study period. A significant difference in biomarker concentrations (non-OA versus OA) was observed for each biomarker at some time during the study. The most dramatic differences were seen with CS-846 (Fig 1) and PGE2 (two-fold increase within 7 days post-OA induction, P = 0.01). Levels of sGAG were significantly elevated (1.4-fold) in OA compared with control joints 14 days post-OA induction (P = 0.01). CPII was significantly elevated 21 days post-OA induction in OA compared with control joints and showed 1.7 fold differences (P < 0.01). COL2 - 3/4C_short was significantly elevated three-fold by 28 days post-OA induction in OA compared
with control joints (P = 0.0001). Bone synthesis (osteocalcin) was significantly elevated 1.4-fold 49 days post-OA induction in the OA compared with control joints (P = 0.01).

4. Discussion
This study demonstrated the ability of biomarkers to differentiate between strenuous exercise and OA in a solitary joint using an experimental model. Also observed was the ability to correctly identify the presence of OA based on the synovial fluid levels of a small number of biomarkers within 14 days of OA induction. The concentration of two markers, CS846 and PGE2, seemed to be very useful early in differentiating normal from OA joints. OA is induced in this model by creating an acute osteochondral fragment and leaving joint tissue debris in the synovial space. Because full thickness articular cartilage erosions are present in these experimental OA joints within 70 days post-surgery, it is worth noting that concentrations of biomarkers are likely to be higher than might be present in some naturally occurring forms of OA at a similar time post-injury. Based on the temporal differences in biomarkers noted in this study, alterations in aggrecan seem to be one of the earliest changes in articular cartilage pathology detectable with synovial fluid biomarkers followed by bone turnover. These results suggest that further investigation of biomarkers for the prediction and monitoring of OA is warranted and correlation of biomarker levels to traditional disease monitoring parameters is possible.

References
