INTRODUCTION

Bone undergoes reformation in response to the demands placed on it during exercise (modeling) and injury (remodeling). Bone alkaline phosphatase (BAP) is an early osteoblastic activity biomarker that has been identified in serum from humans with osteoporosis as well as osteoarthritis (OA) [1,2]. Recently, synovial fluid BAP levels were identified to positively correlate with the radiographic osteophyte scores post cranial cruciate ligament transection (CrCL) in dogs, and were increased in horses with osteochondral fragmentation [3,4]. Synovial fluid BAP has the potential to be a useful biomarker for the detection of bone changes that occur in response to exercise as well as injury.

METHODS

Synovial fluid was collected from the carpal joints and blood was collected from the jugular vein of 14 normal mature Thoroughbred horses. Samples were initially collected after a 2-month period of pasture rest (Week 0), and then horses underwent regular treadmill training for 5 days a week thereafter for 9 months. Serum and synovial fluid were then collected on weeks 6, 12, 32, and 38 following commencement of exercise. In addition, serum was collected from the jugular vein and synovial fluid was collected from the carpal joints from 16 horses that presented for arthroscopic debridement of osteochondral fragmentation. BAP concentrations were measured in the synovial fluid and serum using a commercially available immunoassay (Metra BAP, Quidel Corporation, San Diego, CA). A one-way ANOVA was used to compare the differences between means for each measurement period as well as for arthroscopy samples in the synovial fluid and serum respectively. Tukey’s post-hoc test for multiple comparisons was performed to distinguish significant differences between groups. A value of P<0.05 was considered significant.

RESULTS

Synovial Fluid – BAP concentrations in the synovial fluid increased significantly from baseline (week 0) over time when horses were subjected to regular treadmill exercise (P<0.0001). In addition, synovial fluid BAP concentrations from horses with osteochondral fragmentation that underwent arthroscopy were significantly elevated from baseline (P<0.001), week 6 (P<0.001), 12 (P<0.001), and 32 (P<0.05), but were not significantly different than week 38 samples (Figure 1A). Serum – BAP concentrations in the serum decreased significantly from baseline (week 0) at week 12 of regular treadmill exercise (P=0.0001). In addition, serum BAP concentrations from horses with osteochondral fragmentation that underwent arthroscopy were significantly decreased compared to baseline (P<0.001), week 6 (P<0.001), 32 (P<0.05), and 38 (P<0.001), but were not significantly different than week 12 samples (Figure 1B).

DISCUSSION

BAP concentrations have been shown to increase in the synovial fluid from horses that have sustained an osteochondral fragment injury, and was hypothesized to be due to early changes in the exposed subchondral bone [4]. In addition, in a cruciate transection model of OA in dogs, increased synovial fluid BAP concentrations were shown to be from an osteous source within the joint [3]. In our study, horses that presented for removal of osteochondral fragments from their carpal joints also had elevated synovial fluid BAP concentrations compared to normal rested horses (baseline, week 0). However, the carpal synovial fluid BAP concentrations also increased from baseline over time in normal horses subjected to regular (5 days/week) exercise. Interestingly, by 38 weeks after beginning the treadmill exercise protocol, the synovial fluid concentrations in the normal horses were no different from those that had osteochondral fragmentation. This may indicate that examination of BAP in the synovial fluid has the potential to be used as a predictive biomarker of bone modeling/remodeling that may indicate impending injury in exercising horses. This may indicate microdamage that is occurring to the subchondral bone that could lead to subsequent osteochondral fragmentation or sclerosis. Future work would need to be performed to determine this. To our knowledge, this is also the first report indicating increases in synovial fluid biomarker concentrations as a result of exercise in the horse.

Other studies have demonstrated increases in synthetic and degradative biomarkers in the serum of horses undergoing an exercise protocol [5-6]. The results of our study indicate that there is actually a decrease in the serum BAP concentrations of BAP with exercise as well as when horses have osteochondral fragments. This finding is in agreement with the results from a cruciate transection model in dogs where there was a significant decrease over time [3]. This decrease in the serum BAP values cannot be directly explained but may be due to differences in osteoblastic activity relative to bone modeling/remodeling. In other words, during exercise, osteoblastic activity would be expected to increase rapidly as the bone models to withstand the increased loads placed on it. It would be expected that at some stage, the bone reaches a stage where this process slows down, which may be why the levels decrease at 12 weeks after the beginning of exercise. In addition, the BAP concentrations may be decreased in the serum from horses with osteochondral fragmentation because the osteoblastic activity has to lag behind the osteoconstrictive activity. Another potential is that BAP may undergo further proteolytic processing in the blood making it so that the immunoassay cannot detect it.

To our knowledge, this is the first report of increases in BAP concentrations in the synovial fluid and decreases in the serum of horses in response to exercise and injury.

Figure 1. Concentrations (U/L) of BAP in the synovial fluid (A) and serum (B) from normal horses subjected to regular treadmill exercise over 0, 6, 12, 32, and 38 weeks as well as from horses with osteochondral fragments removed via arthroscopy. Like letters represent no significant differences.