Degenerative joint disease (DJD), commonly known as arthritis, occurs naturally at a high rate in horses, resulting in loss of performance ability, early retirement and significant cost to horse owners. Observed lameness, and tissue imaging are the standard diagnostics for arthritic conditions. However, by the time these tools can confirm a diagnosis, significant and possibly irreversible damage to the joint has already occurred. Conventional therapies, such as non-steroidal anti-inflammatory drugs (NSAIDS) and corticosteroids can be harmful when administered continually. Nutritional therapies may offer a safe and effective substitute that could be used for extended periods of time without adverse health effects. Tissue biomarkers are molecules released into the bloodstream during tissue breakdown or remodeling. It has been proposed that monitoring their concentrations may be useful for the diagnosis and prognosis of joint disease, as well as providing a means to monitor effectiveness of therapy. The first objective of this study was to establish variation in the biomarkers in individual horses and among horses over time. Concentrations of biomarkers were then evaluated for correspondence to lameness. The second objective was to assess the effectiveness of Steadfast® Equine, a dietary supplement designed to maintain the health and growth of joints and hooves, in horses with naturally acquired lameness. Results support the usefulness of biomarkers for the detection of equine lameness, determining the degree of pathology, and evaluating the effects of therapy. However, Equine® Steadfast administered over 84 days failed to produce effects on lameness score, affected area, or biomarker concentrations. Further investigation in horses with defined DJD and radiographs throughout disease progressions is indicated to refine parameters correlating biomarker concentration to lameness in horses with DJD.