Effectiveness of an Enzyme Inhibitor of Cartilage Breakdown by Force Plate Analysis in Cranial Cruciate Ligament Deficient Dogs

Summary: Force plate analysis was used to evaluate the effectiveness of oral administration of a synthetic MMP inhibitor administered to dogs with cranial cruciate ligament (CCL) transection. The researchers found that there was no effect of treatment by an MMP inhibitor on how the dog ambulated across the force plate. However, there was confirmation that there was decreased weight bearing post-transection when compared to baseline, verifying complete transection of the cranial cruciate ligament.

Osteoarthritis (OA) is a major cause of morbidity and expense for all species. Often OA is initiated by some sort of traumatic incident that may lead to instability of the joint. This creates inflammation within the joint, which leads to upregulation and activation of degradative enzymes, called matrix metalloproteinases (MMPs). These enzymes have the capability of degrading articular cartilage, leading to breakdown of the joint resulting in lameness. Research has been conducted for many years trying to control the progression of the disease as well as the morbidity it causes. A novel therapeutic approach to the treatment of OA is the use of a synthetic MMP inhibitor. These inhibitors bind to the MMPs so that they are unable to degrade the cartilage. Therefore, this therapy should reduce the degree of OA that develops over time, improving the patient morbidity and degree of lameness.

Many different animal models of OA have been used to mimic the early stages of OA in humans and dogs, in an attempt to be able to test different treatment methods. One of the most common models used is the cranial cruciate ligament (CCL) transection model in the dog. Transection of the CCL in the dog causes instability, leading to radiographic evidence of arthritis and ultimately results in damage to the articular cartilage and morbidity that causes lameness.

Force plate analysis has been used as an objective measurement of lameness, by allowing analysis of peak forces in three different planes (Figure 1). The peak vertical force (Z plane in Figure 1) is the most important because it allows an objective way to measure the amount of weight the dog is bearing on a given limb. In addition, a related component to the peak vertical force is the impulse area of that force applied to the plate. The impulse area represents the total force applied by the limb onto the plate over the entire contact time of the foot. Historically, this force and area have been successfully used in force plate analyses of CCL deficient dogs. In these studies, weight bearing improves over time, but does not return to normal. Therefore, this technology can be used to assess the efficacy of a therapeutic for osteoarthritis to see whether it reduces lameness so that weight bearing returns to normal. Dr. Troy Trumble, working with Dr. Clark Billinghurst and Dr. Wayne McIlwraith, evaluated the success of CCL transection, and the effectiveness of oral administration of a synthetic MMP inhibitor administered to dogs with transected cranial cruciate ligaments. Their hypotheses were that the peak vertical force and respective impulse area will significantly decrease at day 14 post-transection, when compared to baseline, but will not be different from baseline by day 126 post-transection in the treated dogs.

Thirty-nine mature male Walker Hounds had their right CCL transected arthroscopically. Nineteen of the dogs were administered an oral placebo (control group) and 20 dogs were administered the oral MMP inhibitor (treatment group) daily throughout the study, starting at 14 days post-transection. Dogs were trotted across the force plate by one handler prior to having the CCL transected (baseline), and then again at day 14, 28, 70, and 126 days post-transection. Since the force applied to the force plate is dependent upon the velocity of the dog, only speeds between 1.45 m/s and 2.05 m/s with less than 10% acceleration/deceleration were considered acceptable. In addition, in order for the trial to be considered acceptable, the dog had to strike the plate with its right forelimb followed by its right hindlimb. Any other combination of limbs striking the force plate was not acceptable. Each dog was trotted across the force plate a maximum of 75 times per trial period, and the first five acceptable trials were collected per dog per time period. If the dog was completely non-weight bearing on the right hindlimb, then the value entered for each trial was zero. Each force plate measurement (peak vertical force and impulse area) were examined separately. The five trials for each dog per measurement time

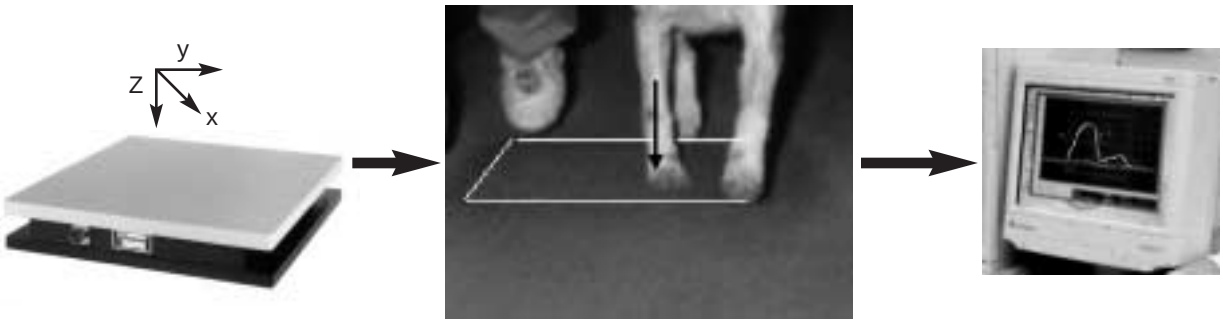

***Figure 1:** Photograph of the force plate (left) with the representative forces that can be measured. The force in the Z direction represents the amount of force applied to the plate when the dog bears weight on the limb, as it crosses the plate (outlined in the center). This force and respective contact area can be examined and analyzed via software on the computer (right).*

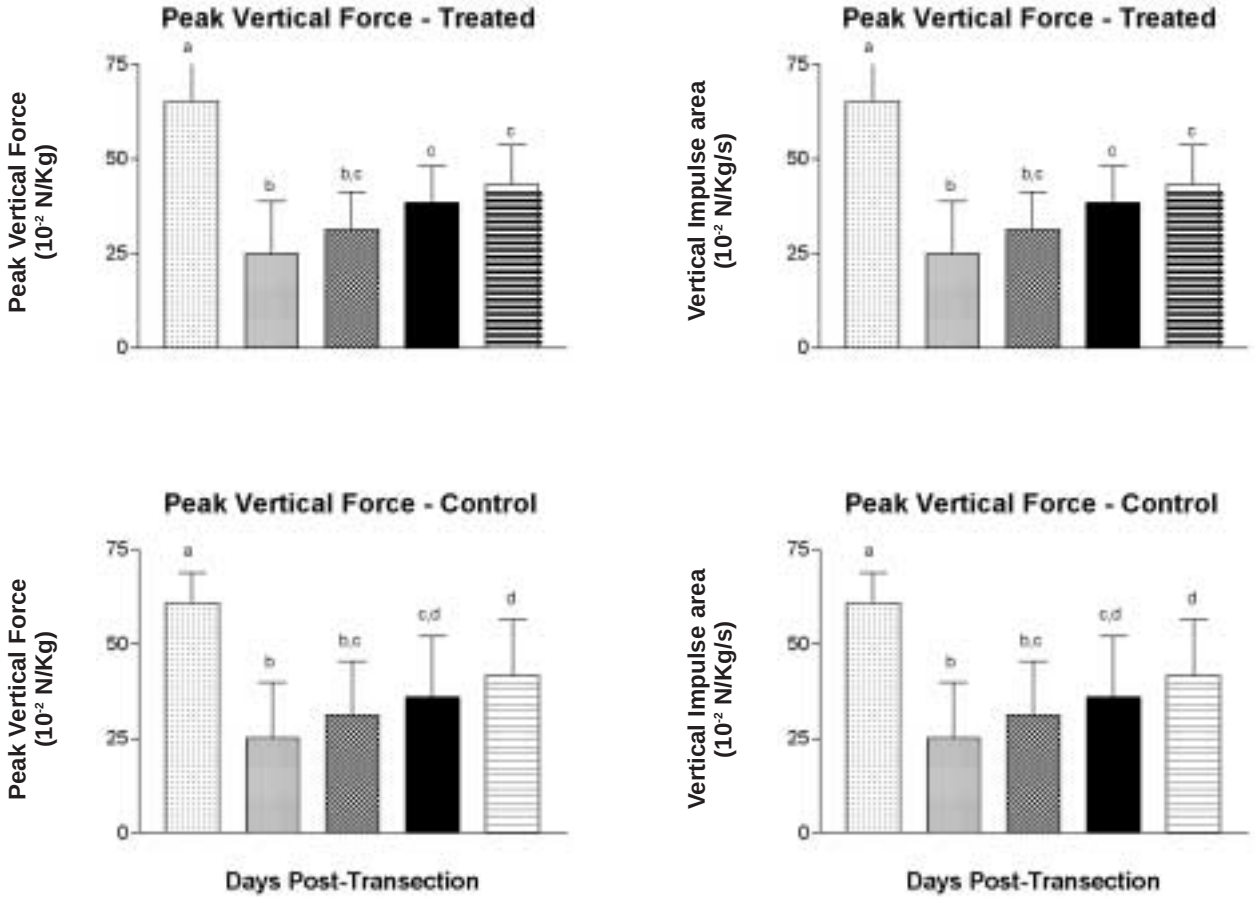

***Figure 2:** Mean peak forces (left) and impulse area (right) for the cranial cruciate deficient limb in treated (top) and control (bottom) dogs at measurement periods prior to (day 0) and after (days 14, 28, 70, and 126) CCL transection. MANOVA analysis was applied to compare treated dogs to control dogs, as well as post-transection to baseline values with significance set at P<0.05. Variation is represented by the standard error of the mean (SEM), and different letters represent significant statistical differences between measurement periods. For treated and control dogs, there is a statistically significant decrease (p<0.0001) in peak force and the respective impulse area from baseline for all measurement periods post-CCL transection. In addition, there is no statistical difference between treated and control dogs with respect to the peak force or impulse area applied to the force plate from the CCL deficient limb.*

period (day 0, 14, 28, 70, and 126) were averaged for each force plate measurement. The means for each group (control vs treated) were tested for each measurement period using a multivariate test followed by univariate multiple comparison of means.

Examination of the treatment means (control vs treated) for each measurement period (day 0, 14, 28, 70, and 126), for both the peak vertical force and the impulse area demonstrated no significant differences between the two treatment groups (Figure 2). However, examination of the means for the control and treated dogs over the measurement periods, demonstrated that for each force plate measurement, the means decrease by at least 50% by 14 days post-transection (Figure 2). The means then slowly increase with each measurement period, but never get back to baseline values. This effect is significant ($p<0.0001$) for all force plate measurements, indicating successful transection of the cranial cruciate ligament.

The results of this study support the researchers' hypothesis that there is a significant decrease in the peak vertical force and impulse area at the day 14 post-transection measurement period when compared to baseline. This verifies that the cranial cruciate ligament was successfully transected due to the instability created in the acute period, making the limb more painful to the dog during use; therefore, putting less weight on the limb when walking. Also, the increases in the percentage of force applied and contact area of the foot over the remaining measurement periods are reasonable, since over time the stifle will become more stable, despite the potential increase in cartilage damage. In other words, the comfort gained from the added stability will most likely override the pain from cartilage damage. This conclusion is a plausible explanation for why our second hypothesis was negated. In other words, the peak vertical force and impulse area at day 126 post-operatively were significantly different from baseline for control and treatment dogs, indicating no treatment effect. The potential protection of cartilage degradation supplied by the MMP inhibitor most likely is not enough to make the dog that much more comfortable beyond what is supplied by the added stability of the stifle over time. In conclusion, there was no effect of treatment by an MMP inhibitor on how the dog ambulated across the force plate. However, there was confirmation that there was decreased weight bearing post-transection when compared to baseline, verifying complete transection of the cranial cruciate ligament.

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