Randomized Double-blinded, Placebo-controlled Multi-center Trial of an Orally Administered Composition (Connectin®) for the Treatment of Canine Osteoarthritis

Abstract

Joint disorder affects 25% of the 77.5 million[1] dogs in the United States. Symptoms range from mild or situation induced to lameness and severe impact on the pet's, and their human's, quality of life. A randomized, double-blinded, placebo controlled, multi-center trial on a patented joint medicant **(Connectin®, In Clover, Inc.)** was completed. The primary endpoint was determination of statistically significant improvement in orthopedic examination scores and pressure plate measurements (Peak Ground Force and stance time) in the treated animals compared to the controls. The study ran eight weeks consecutively. This multi-center trial involved five different sites and followed Good Laboratory Practices (GLP).

Forty-six dogs completed the study. Twenty-two dogs were treated with the studied composition and twenty-four dogs were administered the placebo. There were thirteen males and eleven females in the placebo group. The test group was comprised of nine males and thirteen females. The mean ±SD age of the dogs in the placebo group was 9.79 ±3.43. The mean ±SD age of the dogs in the test group was 8.55 ±2.52.

Dogs of any breed, age or sex were included in the study if after the first examination osteoarthritis was diagnosed on orthopedic examinations and radiographs. Complete safety data were gathered including kidney function, liver function, hematology, physical parameters and owner assessment.

Conclusions: There were no significant changes in kidney function, liver function, hematology, physical parameters, and owner assessment between the test and placebo groups. Statistically significant improvement in orthopedic evaluation parameters were seen in the test group as compared to a placebo. The studied composition, Connectin, is statistically less symptomatic than the placebo for Lameness, Willingness to Hold Contra lateral Limb and Weight Bearing of the Affected Joint.

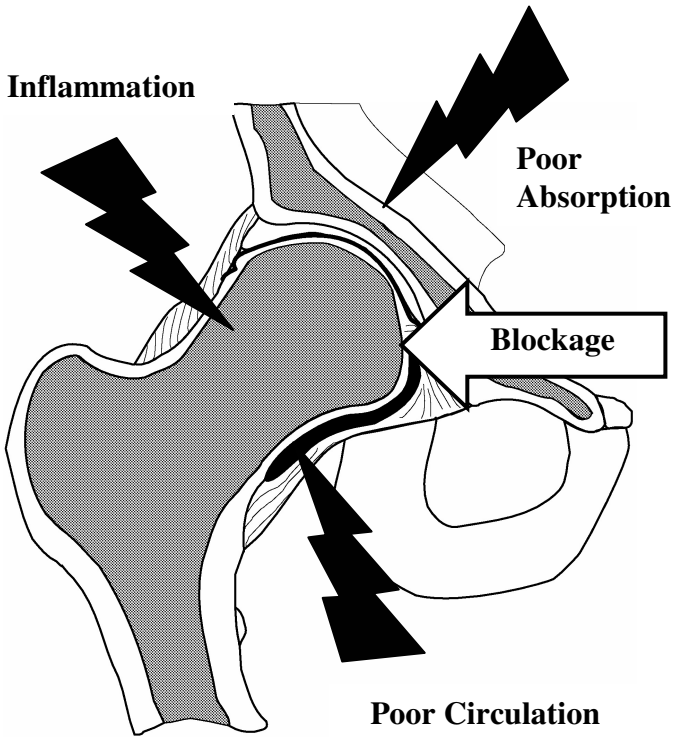
1. *2009-2010 AAPA National Pet Owners Survey, p. 57*

Discussion

In healthy conditions, articular cartilage forms a smooth surface between articulating bone ends to reduce the friction caused by movement. This friction is further reduced by the synovial fluid. Articular cartilage consists of chondrocytes and two major macro-molecules; i.e., collagen and proteoglycans, which are synthesized by and deposited around the chondrocytes. The chondrocytes also synthesize the synovial fluid which bathes the articular cartilage. The structural integrity of the articular cartilage is the foundation of optimal functioning of the skeletal joints in the hip, shoulders, elbows, hocks and stifles.

Impaired function of skeletal joints will dramatically reduce mobility such as rising from sitting position or climbing and descending stairs. To maintain the structural integrity and the proper functioning of the articular cartilage, the chondrocytes constantly synthesize collagen and proteoglycans, the major components of the articular cartilage, as well as the friction-reducing synovial fluid. This constant synthesis of the macro-molecules and synovial fluid provides the articular cartilage with the repairing mechanism for most of the wearing caused by friction between the bone ends. However, it also leads to the constant demand for the supply of precursors, or building blocks, for the macromolecules and synovial fluid.

Lack of these joint precursors will lead to defects in the structure and function of the skeletal joints. This deficiency occurs often when activity levels are very high, or cartilage tissue has been traumatized *(Joint fig.1).*

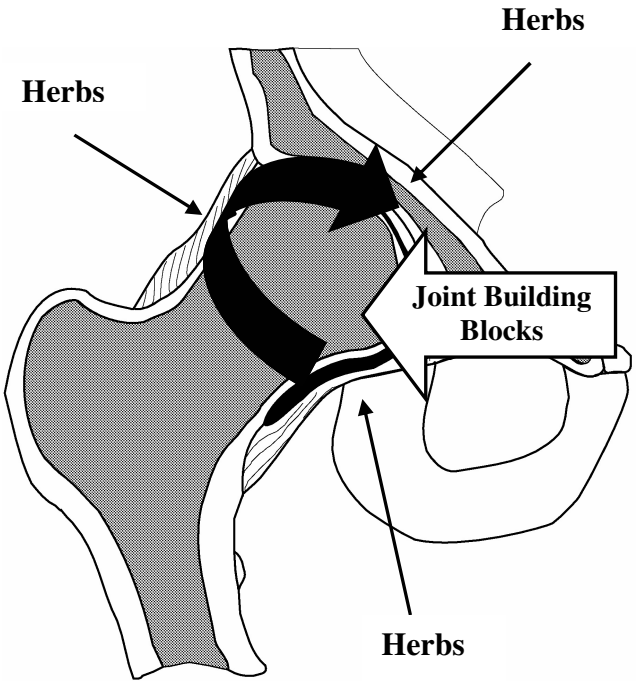


Joint Figure 1

An adequate supply of metabolic precursors or building blocks is thus paramount to replacement and repair of the constituents of skeletal joints, connective tissue and synovial fluid. Proteoglycans (or mucopolysaccharides) form the ground substance of cartilage, bone and joint fluid. Proteoglycans are comprised of proteins linked to glycosaminoglycans (GAGS). The building block GAG subunit of the proteoglycan in cartilage and bone is chondroitin sulfate. The building block GAG subunit of the proteoglycan in joint fluid is hyaluronic acid. Intercellular solutions of hyaluronic acid are viscous

and thus assist in lubrication of the joints of body skeleton. Hyaluronic acid is synthesized from the metabolic precursor, glucosamine.

The availability of glucosamine in cartilage tissue can be rate-limiting to the enzymatic step leading to the production of proteoglycans. Exogenous glucosamine serves to drive the biosynthetic pathway forward past the rate-limiting blockage point. Glucosamine serves as a substrate for a kinase enzyme which yields glucosamine-6-phosphate, the rate-limiting precursor in proteoglycan synthesis *(Patent fig.1).*



Joint Figure 2

Connectin is a combination of natural physiological metabolites and herbal phytochemicals. The components of Connectin act synergistically to provide sufficient sources of necessary metabolic precursors for repair and maintenance of connective tissues. This combination works synergistically to ensure the proper absorption of metabolic precursors; to diminish the inflammatory response in the affected area so that the connective tissue degradation process is halted and repair may be initiated. Additionally, the phytochemical combination stimulates the blood circulatory system which simultaneously enhances the delivery of the metabolic precursors to the affected areas and removes deleterious deposits in the affected areas *(Joint fig.2).* To encourage dosage compliance, flavorful palatability agents are used in the Connectin formulation. *(Patent fig. 2).*

Study Design

Connectin was tested in a randomized, double-blind, placebo controlled, multi-center study. Five sites were included in the study. Twenty four dogs were in the placebo group of the study and twenty two dogs were in the Connectin group. Treatment was given for 8 weeks consecutively.

Objective

To determine the efficacy of Connectin in dogs compared to a placebo in a clinical setting.

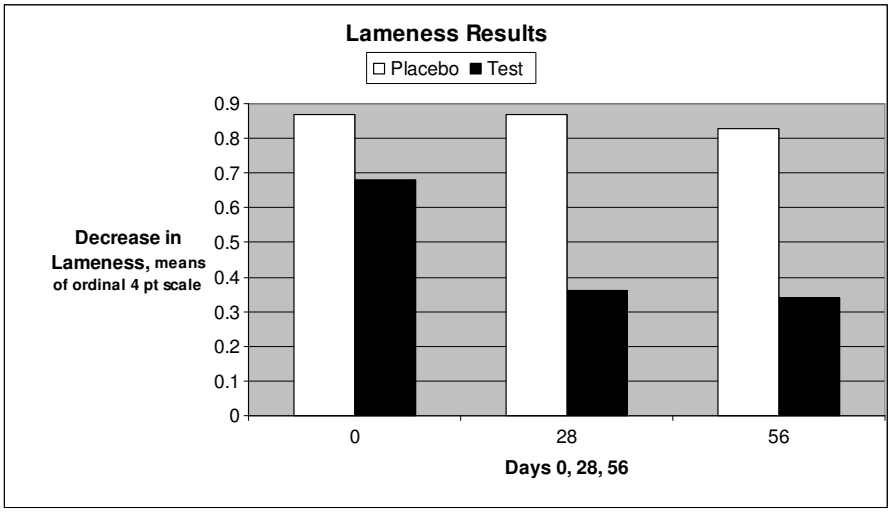
Evaluation Schedule

Between **Days - 30 to 0**: The Investigator interviewed the owner and obtained history of the dog and conducted physical examination, lameness evaluation, pressure plate analysis, radiograph, and collected blood samples.

Days 1-56: Daily oral supplementation.

Days 7, 14, 21, 35, 42 and 49: Owner completed Weekly Owner Assessment Form.

Graph 1



Days 28 and 56: Owner and dog visited the Investigator to repeat the physical examination, lameness evaluation and pressure plate analysis. Blood samples were collected.

Inclusion criteria

Dogs of any breed, age or sex were included if after a first examination osteoarthritis in the elbow or hip was diagnosed based on orthopedic examinations and radiographs.

Criteria for efficacy

Primary endpoints

Connectin was considered effective if there was a statistically significant improvement in lameness evaluation parameters including clinical orthopedic examination scores, and pressure plate measurements (Peak Ground Force and stance time) in the treated animals compared to the controls.

Clinical orthopedic examinations

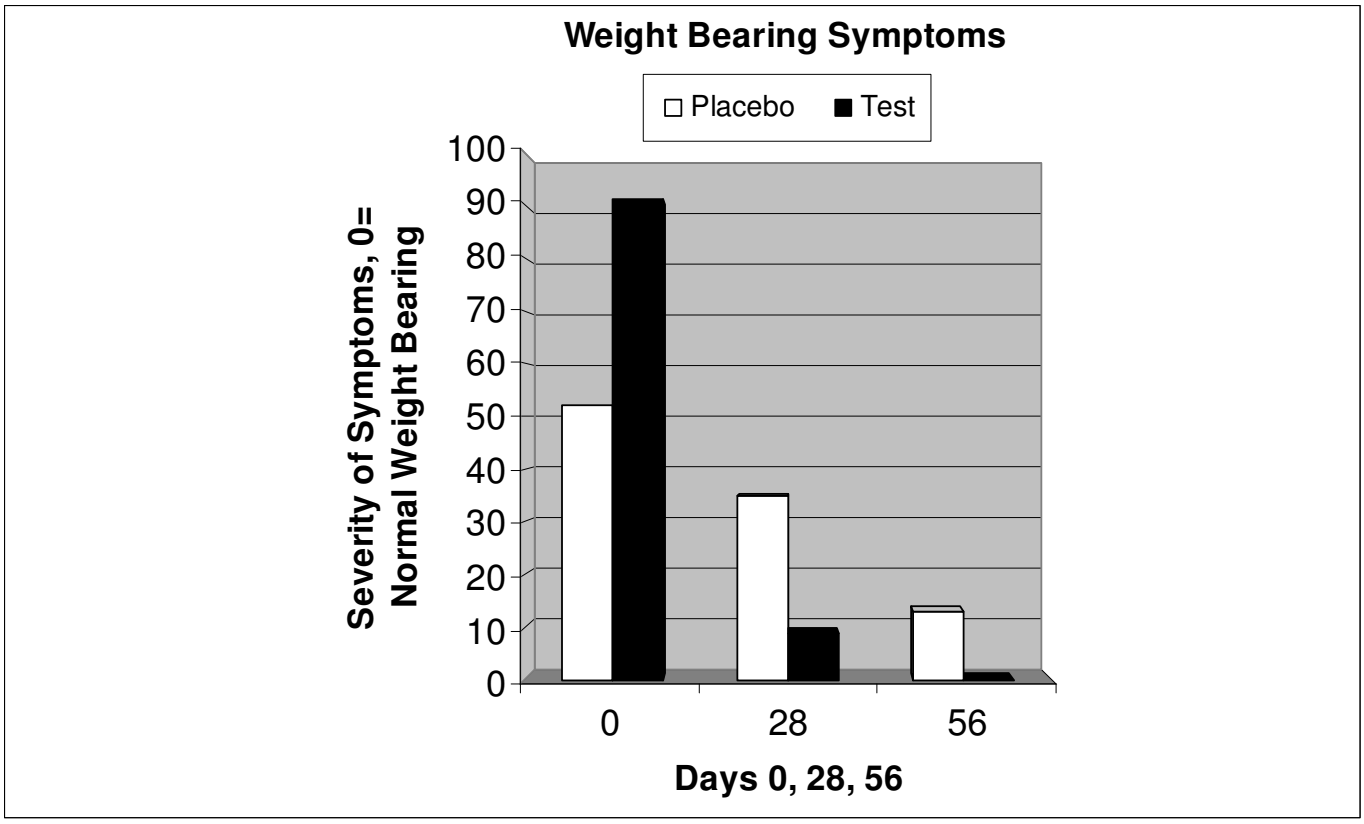
The Investigator prior to selection and on Day 28 and 56 of treatment performed a graded lameness examination including Weight Bearing, Lameness and Willingness to Hold Contra Lateral Limb.

At the time of the lameness examination, the dogs' gaits also were examined using pressure plate analysis. The dogs were lead by leash at an approximately constant velocity over a Footscan™ pressure plate. A 3-m zone, including the pressure plate, was calibrated for video based on quantification of the dog's velocity. Peak ground force, stance times, time to peak braking, and time topeak propulsive force were recorded for each animal and standardized to that animal's velocity. One investigator made all the pressure plate measurements.

Statistical Methodology

Mann-Whitney U, Kruskal-Wallis and stratified variants thereof, and Fisher's Exact, ANOVA, ANCOVA, Shapiro-Wilk, Kolmogorov-Smirnov, Cramer-von Mises, Anderson-Darling.

Statistical Programs - SAS and GenMod.



Graph 2

Conclusion

Statistically significant improvement in orthopedic evaluation parameters were seen in the Connectin group as compared to a placebo. There were no significant changes in kidney function, liver function, hematology, physical parameters, and owner assessment between the test and placebo groups. The studied composition, Connectin, is statistically less symptomatic than the placebo for Lameness *(Graph 1)*, Willingness to Hold Contra lateral Limb and Weight Bearing of the Affected Joint *(Graph 2)*.

Acknowledgements

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