Figure. 1. RCGD423 prevents articular cartilage degradation in a rat model of osteoarthritis.

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EFFECT OF INTRAARTICULAR INOCULATION OF MESENCHYMAL STEM CELLS IN DOGS WITH HIP OSTEOARTHRITIS BY MEANS OF OBJECTIVE FORCE PLATFORM GAIT ANALYSIS. CONCORDANCE WITH NUMERIC SUBJECTIVE SCORING SCALES

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Purpose: Intensity of pain is difficult to accurately assess in dogs. Veterinarians assess the severity of pain in their patients using scoring systems based on several signs, including patient vocalization, activity level, degree of lameness, and reaction to manipulation. However, all of these signs are subjective and may be influenced by a variety of external factors. Subjective pain assessment scales have been widely used for assessing lameness in response to pain, but the accuracy of these scales has been questioned. For this reason, the objective of this study was to evaluate the concordance between subjective measures of pain as scales (Bioarth and visual analog scale (VAS)), and objective measures of pain as the force platform (peak vertical force, PVF, maximal force applied during stance phase; and vertical impulse, VI, total force applied over time), of limb function in dogs of the same breed that had osteoarthritis (OA) lameness due to bilateral hip dysplasia and were treated with autologous mesenchymal stem cells (MSCa)
Methods: To carry out this clinical study, ten adult, client-owned Presa Canario dogs (6 males, 4 females) with lameness and pain attributed to OA associated with hip dysplasia were included. A control group consisted of 5 healthy dogs of the same breed. The sample size was selected based on the availability of the subjects of the same breed, same pathology and similar degree of severity in order to achieve a study group with maximal homogeneity. None of the dogs were forced to perform physical activity. The dogs were first treated with MSCa. Then, at basal and month 1, 3 and 6,the potential lameness improvement was evaluated with Bioarth and VAS. These data were compared with similar data collected using a force platform with the same animals during the same period, basal,1, 3 and 6 months after treatment.
Results: The body weight of the enrolled dogs ranged from 46 to 65.2 kg (mean ± SD: 51.21 ± 5.48 kg), and ages were 4 – 9 years (mean: 5.6 ± 2.3 years). Walking speed of both healthy (control) and diseased groups of dogs was 1.6 ± 0.5 m/s. No significant differences in walking velocity was observed between dogs ($P = 0.08$). The F test for intraclass correlation showed that concordance in pain/lameness scores between the 2 measuring methodologies was not significant (P value ≥ 0.9213; 95 % confidence interval, −0.56, 0.11) (Table 1). Although subjective pain assessment showed improvement after 6 months, force platform data proved those same animals had returned to the initial lameness state (Figure 1 and 2).
Conclusions: Pain is an emotional response to a painful stimulus and is difficult to reliably determine in a nonverbal animal. Pain of the locomotor system can usually be detected by a certain disability to support weight; in other words, lameness is the expression of pain. MSC therapy significantly improved limb function in dogs with hip OA, but the duration of the improvement was inferior to 6 months post treatment. Subjective evaluation of gait correlates poorly to objective measures of limb function. For this reason, subjective evaluation of gait should be interpreted cautiously as an outcome measure.

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GLUCOSE METABOLISM DURING MESENCHYMAL STEM CELL CHONDROGENESIS

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Purpose: Human mesenchymal stem cells (hMSCs) are a key cell source for cartilage tissue engineering applications. Despite the potential, cartilage tissues from hMSCs fall short in their biochemical and biomechanical properties compared to those of the native tissue. hMSCs use glycolytic metabolism for ATP production during chondrogenesis; local availability of glucose can therefore govern the properties of the resultant cartilage tissue. The primary objective of this research is to investigate glucose metabolism during hMSCs chondrogenesis. A secondary objective of this research is to explore whether glucose metabolism can be used as a criterion to predict the success/failure of the resultant tissue.
Methods: Chondrogenic aggregates were formed from hMSCs over a 21-day period using standard procedures. To determine the effect of local glucose concentration, the aggregates were cultured in four different chondrogenic media containing four different glucose concentrations (1, 2, 3, and 4.5 g/l). Further, to evaluate the effect of continuous versus discrete feeding modes, media were changed more frequently (every 24 hours) or less frequently (every 48 hours). Glucose levels were monitored daily. Size of the constructs, and DNA, glycosaminoglycan (GAG) and hydroxyproline (HYP) contents were determined at day 7, 14 and 21. Histology was performed at similar time intervals.
Results: Glucose consumption rate increased during chondrogenesis; the rate was the highest in the last week of 21-day culture. Glucose levels had an impact on cumulative glucose consumption. For less frequent media changes, glucose levels 2, 3 and 4.5 g/l led to similar cumulative consumption, whereas 1 g/l led to significantly lower consumption (Fig. 1 Left). For more frequent media changes, 4.5 g/l treated aggregates had significantly increased consumption. This is followed by 2 and 3 g/l treated aggregates. 1 g/l treated aggregates had the least cumulative consumption (Fig. 1 Right). HYP content increased with glucose levels, and with the frequency of media change (Fig. 2). GAG content followed a similar trend (data not shown). More importantly, however, the biochemical properties were directly related to the cumulative consumption of glucose; aggregates with enhanced consumption of glucose had improved GAG and HYP.

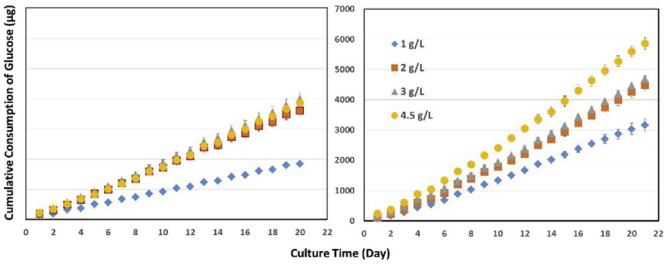

Fig. 1. Cumulative consumption of glucose of chondrogenic aggregates as a function of culture time. hMSC aggregates were treated with differentiation media of different glucose levels (1, 2, 3, and 4.5 g/l) and media were changed at two different frequencies (24 and 48 hours).

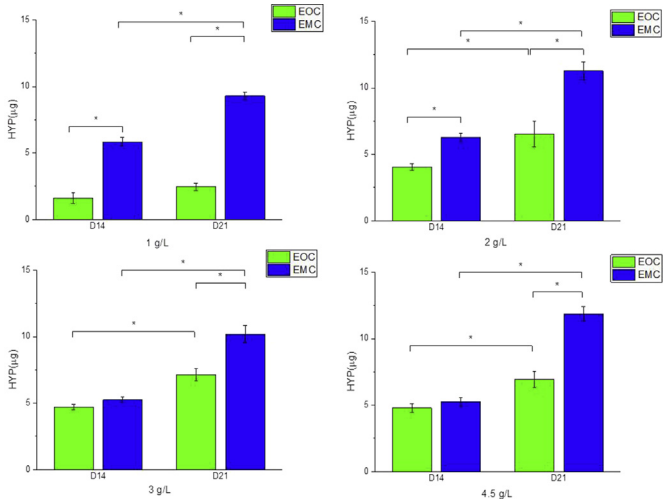

Fig. 2. Hydroxyproline (HYP) content of chondrogenic aggregates exposed to different glucose levels (1, 2, 3, and 4.5 g/l). HYP content of aggregates harvested at Day 14 and Day 21 are shown for two media change frequencies (EOC refers to media change every 48 hours and EMC refers to media change every 24 hours).

Conclusions: Glucose is an important nutrient during chondrogenesis. Lower glucose levels which can arise due to diffusional limitations can significantly affect the biochemical properties of the resultant tissue. A