

Synovial Fluid Lubricin Increases in Spontaneous Canine Cruciate Ligament Rupture

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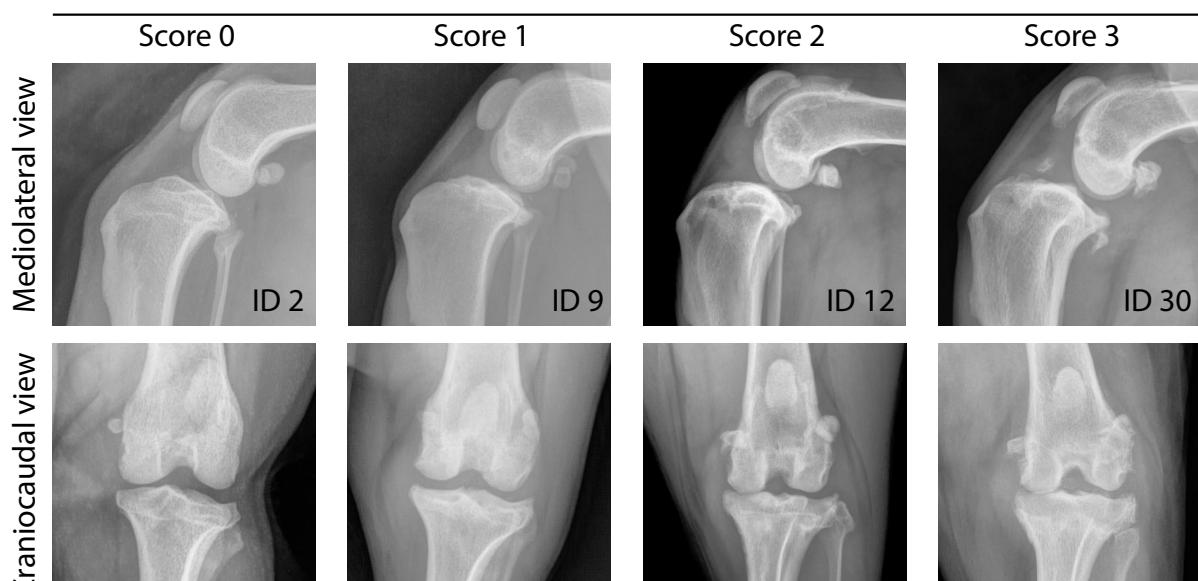
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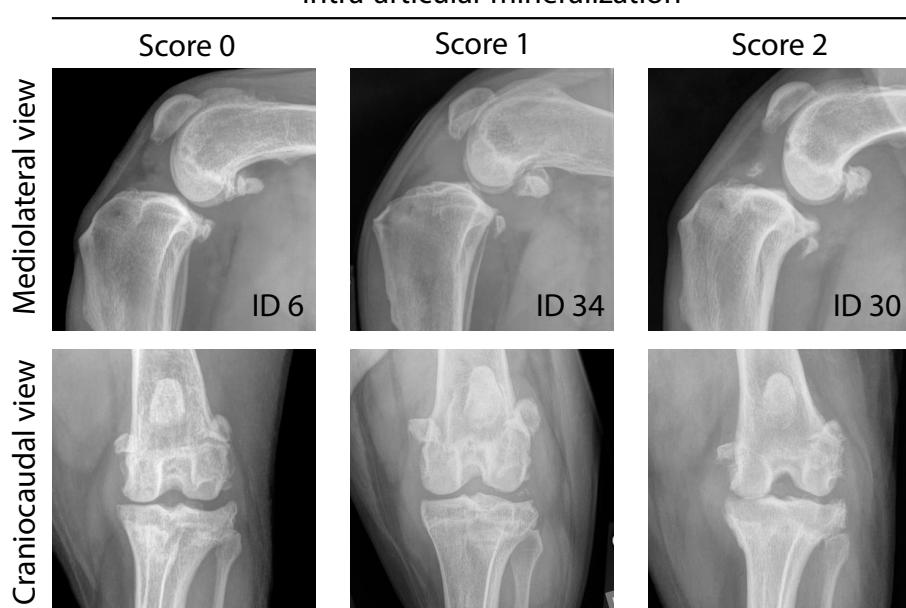
Phone: 607-253-3100

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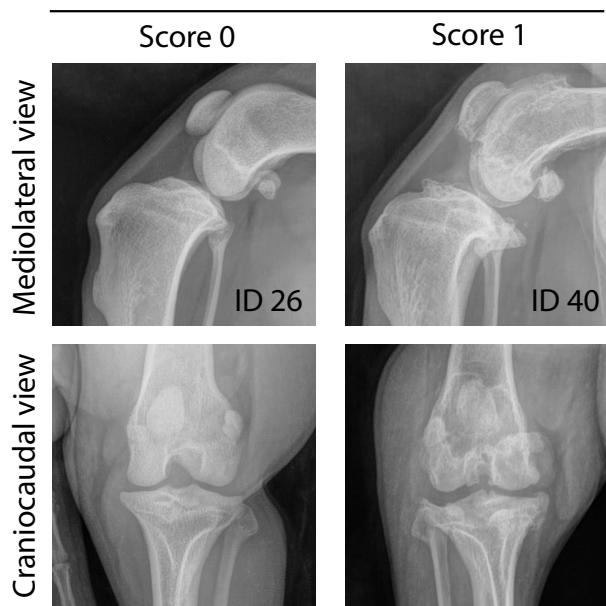
Global OA Score



Intra-articular mineralization



Caudal tibial shelf

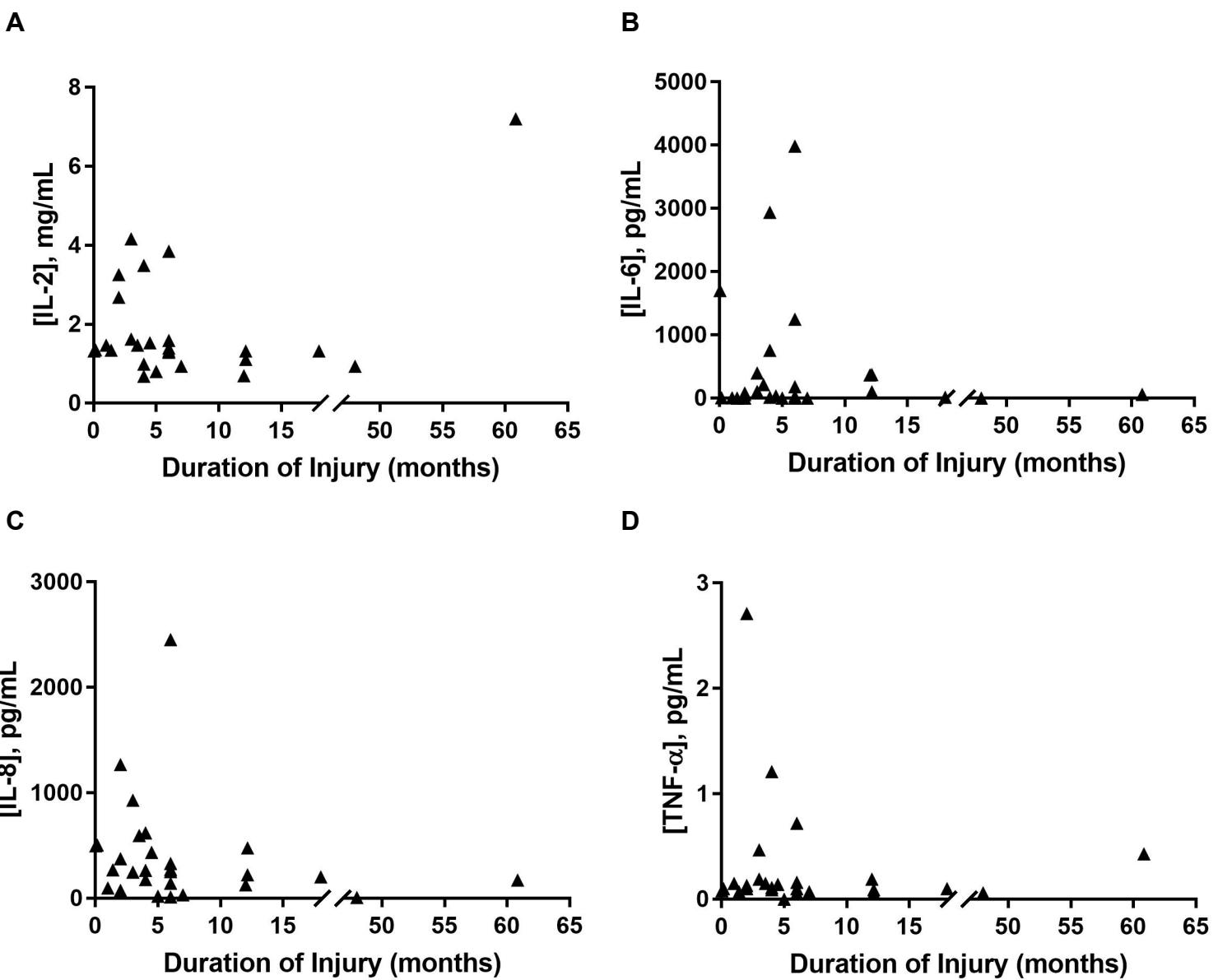


Supplemental Figure 1.

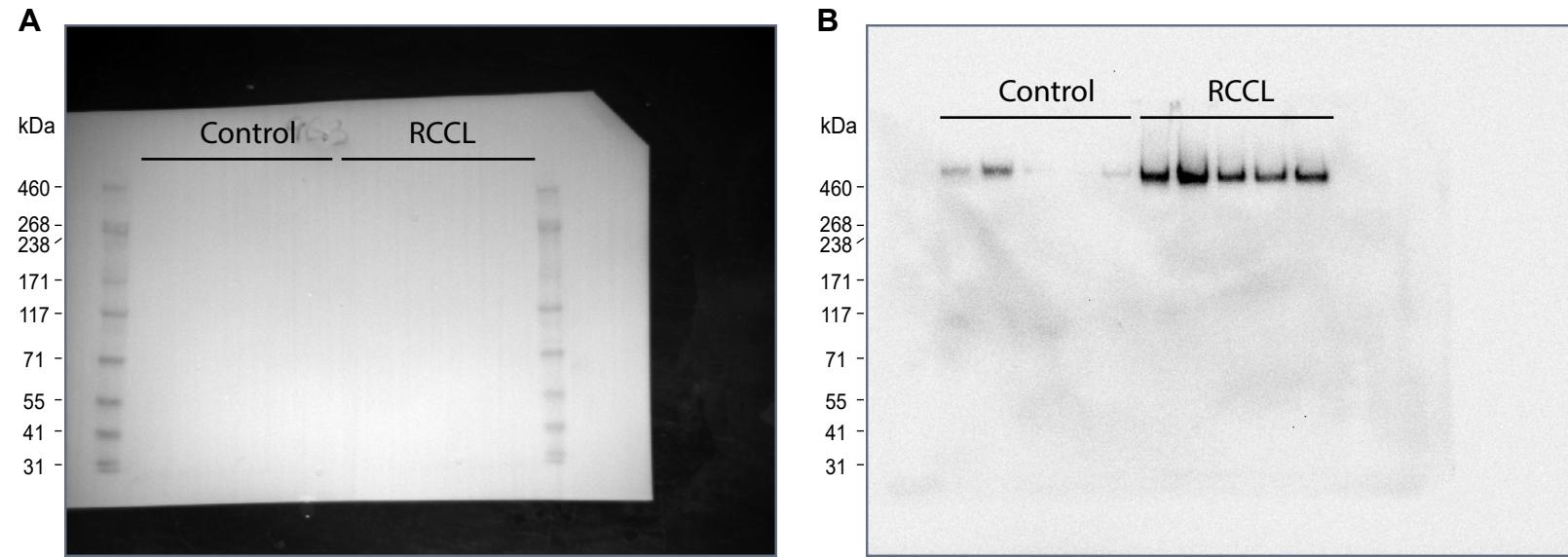
Paired mediolateral and craniocaudal radiographs from individual canine patients representing the range of possible scores for: global OA (0-3), intra-articular mineralization (0-3) and caudal tibial shelf (0-1).

| | PJ vs. HR | HR vs. DG | PJ vs. DG |
|---------------------------------------|------------------|------------------|------------------|
| Global OA score | 0.659 | 0.674 | 0.312 |
| Joint effusion | 0.736 | 0.567 | 0.502 |
| Osteophytosis | 0.712 | 0.712 | 0.700 |
| Intra-articular mineralization | 0.493 | 0.396 | 0.393 |
| Medial buttress | 0.433 | 0.570 | 0.474 |
| Caudal tibial shelf | 0.692 | 0.665 | 0.479 |

Supplemental Table 1. Linear weighted Kappa interrater agreement for canine radiographic osteoarthritis scoring. Strength of agreement was interpreted with kappa values: 0-0.2 being poor, 0.21-0.40 being fair, 0.41-0.60 being moderate, 0.61-0.80 being good, and 0.81-1.00 being very good for interrater agreement between scores for three co-authors, including the second (DG), sixth (PJ) and final co-author (HR).



Supplemental Figure 2. Synovial fluid A) IL-2, B) IL-6, C) IL-8, and D) TNF- α concentrations plotted as a function of injury duration in dogs with RCCL ($n = 28$).



Supplemental Figure 3. Anti-lubricin western blots of synovial fluid from 5 randomly selected control and RCCL dogs. A) Bright field image of standard ladders (HiMark™ Pre-stained Protein Standard, cat. #LC5699, Invitrogen), 5 µL/lane, B) chemiluminescence, exposure time: 15 s; sample loaded:10 µL/lane.

| Cytokine/Chemokine | Lubricin | Duration of Injury |
|--------------------|--------------|--------------------|
| IL-2 | -0.23 (n.s.) | -0.31 (n.s.) |
| IL-6 | -0.24 (n.s.) | -0.02 (n.s.) |
| IL-8 | -0.11 (n.s.) | -0.39 (n.s.) |
| TNF- α | -0.25 (n.s.) | -0.17 (n.s.) |

Supplemental Table 2. Correlation of inflammatory cytokine/chemokine concentrations, lubricin concentrations and injury duration in dogs with RCCL (n = 28). Data reported as Spearman's ρ , n.s.: not significant, Spearman's test, $\alpha = 0.05$.

| | Control | RCCL |
|---------------------------|-------------------------------|--|
| Number | N = 9 | N = 30 |
| Age | 3.3 - 7.6 yrs, mean = 5.5 yrs | 2.6 – 11.0 yrs, mean = 6.2 yrs |
| Sex | Intact male | 13 neutered male 14 spayed female 2 intact male 1 intact female |
| Breed | Beagle | 1 Bernese Mountain Dog 1 Boxer 1 Bulldog, English 1 Cane Corso 2 English Springer Spaniel 1 German Shepherd 3 Golden Retriever 9 Labrador Retriever 1 Mastiff 6 Mixed Dog 1 Pembroke Welsh Corgi 2 Rottweiler 2 Staffordshire Bull Terrier |
| Duration of injury | N/A | 0 - 60.8 months, mean = 8.4 months |

Supplemental Table 3. Demographics of the RCCL and control groups