

# Regenerative Medicine for Partial Cranial Cruciate Ligament Injuries in **Agility Dogs**



By Sherman O. Canapp, DVM, MS, CCRT, DACVS, DACVSMR and Brittany Jean Carr, DVM, CCRT  
Photos courtesy of VOSM except where noted

Cool is a 5-year-old Border Collie from Washington. Cool presented for evaluation of left hind limb lameness/offloading. About a month prior to presentation, Cool was noted to be offloading the left hind limb after playing outside. He was taken to his primary care veterinarian where radiographs were performed and appeared unremarkable. The lameness resolved with strict rest but reappeared a few days later. Cool was referred to both a surgeon and a neurologist. Cool had an MRI of his stifles and lumbar spine, which revealed a mild partial disruption of the cranial cruciate ligament of the left stifle joint with mild intracapsular soft tissue swelling and bone edema. Cool was taken to multiple specialists, all of whom recommended surgical intervention (TPLO). Owners declined aggressive surgical intervention for the mild cranial cruciate ligament (CCL) injury, and instead contacted VOSM regarding the use of stem cell therapy for this injury. Cool was continued on strict crate rest and flown to VOSM for further evaluation.

## Physical Examination and Diagnostic Findings

At initial presentation, Cool had a mild left hind limb lameness and shortened stride at a trot. He would slightly offload the left hind limb at a stance. On physical exam, Cool had mild effusion in the left stifle. Very mild instability was noted in the left stifle, consistent with an early partial tear.

Initial Gait4Dog objective gait analysis revealed a decreased pressure on the left pelvic limb as compared to the right pelvic limb. On radiographs there was mild effusion confirmed in the left stifle.

Cool's left hind limb offloading/lameness was attributed to an early partial CCL rupture. A left stifle arthroscopy was recommended to confirm the CCL injury and further evaluate the left stifle joint.

A left stifle arthroscopy was performed and showed mild disruption of the craniomedial band of the CCL (~10% tear). Mild synovitis (joint inflammation) was noted. The remainder of the cranial cruciate and caudal cruciate ligaments were within normal limits as was the meniscus.

## CCL Injury in the Agility Dog

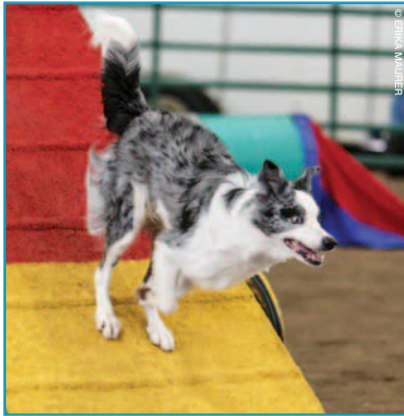
CCL injury is the most common stifle (knee) injury in dogs. A rupture of this ligament causes pain, lameness, and arthritis formation. Instability may lead to injury of other structures in the knee (the meniscus). Diagnosis can often be accomplished through physical examination and minimally invasive diagnostic modalities. VOSM's specialists commonly recommend an objective gait analysis (such as Gait4Dog System) to identify and quantitate lameness and various gait parameters. Gait analysis is repeated at every recheck to allow for an objective measure of response to treatment. Minimally invasive diagnostic modalities such as stifle arthroscopy and MRI are used not only to confirm a diagnosis, but also grade the level of injury. Depending on the grade of injury, conservative management with regenerative medicine and rehabilitation therapy may be an option. In Cool's case, a left stifle arthroscopy was used to confirm the partial cranial cruciate ligament tear and grade the injury. Since Cool had changes consistent with an early tear (~10%), regenerative medicine was offered.

## Stem Cell Collection and Treatment

Under fluoroscopic guidance bone marrow was collected from Cool's femur. The stem cells were isolated from the bone marrow sample (BMAC) in VOSM's regenerative medicine laboratory using the validated CRT machine. Blood was also collected from Cool for processing of platelet rich plasma (PRP) using the validated CRT machine. The stem cells and PRP were combined and injected into Cool's stifle using fluoroscopic guidance.

## Regenerative Medicine Therapy for the Agility Dog

Regenerative medicine therapy has become increasingly popular in both human and veterinary medicine for multiple disease processes. Recent studies have shown regenerative medicine to be efficacious in managing numerous orthopedic conditions,



Cool Ross competing at the US Open.

including osteoarthritis and soft tissue injuries (tendon and ligament injuries). Preliminary studies in rats, humans, and dogs have shown regenerative medicine therapy to be efficacious for treating cranial cruciate ligament injury (Pitts J 2015, Mifune Y *et al* 2013, Oe K *et al* 2011).

### What Are Stem Cells and PRP?

Stem cells are the body's progenitor cells from which all other cells are derived. Recent studies have shown that stem cells can regenerate and heal injured tissue, decrease inflammation, stimulate new blood supply to support healing, activate resident stem cells, create a scaffold for healing tissue, protect cells from death, and break down scar tissue. Stem cells can be obtained from numerous sources from a patient's own body (autologous adult-derived mesenchymal cells). The most common places to harvest adult-derived mesenchymal cells are either from the patient's bone marrow or adipose (fat) tissue. Both bone marrow-derived and adipose-derived stem cells have the ability to differentiate into cartilage, bone, tendons, and ligaments. Recent studies have shown that bone marrow-derived stem cells and adipose-derived stem cells are equally viable and effective. Since recent studies have also shown that stem cells act synergistically with PRP to improve tissue healing, stem cells are combined with PRP prior to injection.

PRP is an autogenous (self-derived) fluid concentrate composed primarily of platelets and growth factors. PRP is made by processing a patient's own blood sample. The goal is to obtain the highest concentration of platelets and growth factors, while removing the other components of the blood such as the red and white blood cells, which can cause pain and inflammation.

### How Are Stem Cells and PRP Collected and Processed?

Bone marrow-derived stem cells are obtained under brief anesthesia or heavy sedation. A needle is inserted into the medullary cavity (center of the bone) and the cells

are aspirated with a syringe. The sample is then processed and prepared for injection. Similarly, adipose-derived stem cells are collected under a brief anesthesia. While there are numerous locations where fat may be collected, recent studies suggest that the falciform ligament (fat located inside the abdominal cavity) yields the highest quantity and quality of adipose-derived stem cells. A small incision is made along the cranial abdominal area to obtain the fat.

Once the sample is obtained, it is then processed. Both bone-marrow derived stem cells and adipose-derived stem cells can be processed either on-site or shipped to a university for processing, culturing, and banking for future use.

To make PRP, a sample of blood is obtained from the patient, mixed with an anticoagulant, and processed either manually by spinning it in a centrifuge to separate its components (centrifugation) or through an automated system. This process concentrates the platelets as well as growth factors, amplifying healing properties. PRP is added to the stem cell sample immediately prior to injection.

### Regenerative Medicine Therapy for CCL Injury

Regenerative medicine therapy for partial cranial cruciate ligament injuries at VOSM consists of PRP and stem cell therapy. Together PRP and stem cells act synergistically to regenerate tissues, increase blood supply, and break down scar tissue formation, replacing it with regenerated tissue. VOSM has the capability to offer PRP, bone marrow-derived stem cell concentrate (BMAC), culture expanded bone marrow-derived stem cell (BMSC), adipose-derived stromal vascular fraction stem cells (SVF), and culture-expanded adipose-derived stem cells (ADSC).

For Cool, bone marrow derived stem cells were harvested from Cool's proximal left femur and blood was collected for in-house preparation of regenerative progenitor cells (stem cells) and PRP. An intra-articular injection of stem cells and PRP was performed in Cool's left stifle.





### Inclusion Criteria for Stem Cell Therapy for a Partial CCL

There are strict inclusion criteria for stem cells to be considered for the treatment of a partial CCL based on current literature and VOSM's data. In order to be considered, the partial tear must be confirmed on arthroscopic evaluation. If there is a 50% tear or less (of the cranio-medial band) stem cells may be considered. For greater than a 50% tear a TPLO would be recommended as there is not enough scaffold for regeneration.

VOSM has been treating CCL partial tears with stem cells since 2008. As evidence based medicine and objective outcome measures are of top priority at VOSM the majority of cases have had a follow-up second look arthroscopy to confirm CCL healing and an

objective gait analysis to confirm soundness. In addition, since the majority of the cases treated are performance and sporting dogs we have continued to follow their performance and return to successful competition. VOSM is in the process of publishing a retrospective study on the return to agility following stem cell therapy for partial CCL injury using the VOSM return to agility grading scale. So far, the data shows a significant amount of cases were able to completely heal following stem cell treatment (based on second look arthroscopy and objective gait analysis) and return to full function and competition.

### Rehabilitation Therapy Post Regenerative Medicine Therapy

Post-injection restrictions and rehabilitation therapy are crucial to obtain a complete recovery. The dogs should be restricted from heavy activity and confined to an x-pen, crate, or small area when unattended for 12 weeks. Leash walks are allowed and increase in duration over the 12-week period. Custom hinged functional stifle braces may be considered for 12 weeks if there is gross



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PK 12928  
**Cool**  
Owner: Lisa Ross

Tested on: 4/30/2013 2:21:05 PM

Doctor: Sherman Canapp, DVM, MS, DACVS

Age Gender 5 M Left (H/F) L/EG Right (H/F) Shoulder Hip

Breed: **Border Collie**

Velocity: 222.8 \*  
Reach (L/R): -3.8 / -2.9

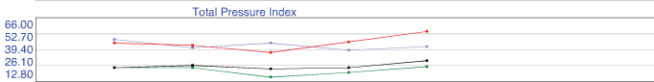
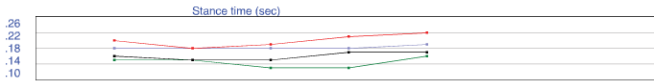
	Stance time(sec)	Stance %GC (%)	Total Pres. Index	TPI %	Stride Vel(vised)	Step Length(cm)	Stride Length(cm)	Step/Stride Ratio
Left Front	.18(0.00)	41.55(1.13)	43.0(3.54)	31.8 %	226.9	48.58(1.10)	96.4(2.40)	50.4%
Right Front	.20(0.02)	45.60(1.34)	45.2(6.50)	33.4 %	223.7	47.25(2.56)	95.0(1.59)	49.7%
Left Hind	.14(0.01)	32.03(2.36)	21.8(3.77)	16.1 %	223.1	47.95(2.10)	96.1(2.41)	49.9%
Right Hind	.16(0.01)	35.85(0.80)	25.4(2.79)	18.8 %	223.2	47.50(1.37)	95.3(3.30)	49.8%

Symmetry Ratio			
Front/Hind	1.27	1.28	1.87
Left/Right	0.89	0.90	0.92
Left Front/Right Front	0.90	0.91	0.95
Left Hind/Right Hind	0.88	0.89	0.86
Left Front/Left Hind	1.29	1.30	1.97
Right Front/Right Hind	1.25	1.27	1.78
Left Front/Right Hind	1.13	1.16	1.69
Right Front/Left Hind	1.43	1.42	2.07

Indicates a front load when comparing any front to hind or indicates left loading when comparing any left to a right

Indicates a hind load when comparing any front to hind or indicates right loading when comparing any left to a right



Initial Gait4Dog objective gait analysis revealed a decreased pressure on the left pelvic limb as compared to the right pelvic limb.

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8897

PK 12928  
**Cool**  
Owner: Lisa Ross

Tested on: 8/21/2013 9:17:37 AM

Doctor: Sherman Canapp, DVM, MS, DACVS

Age Gender 5 M Left (H/F) L/EG Right (H/F) Shoulder Hip

Breed: **Border Collie**

Velocity: 197.9 \*  
Reach (L/R): -3.3 / -3.2

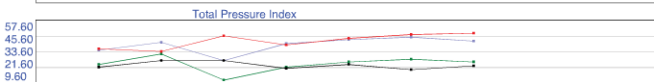
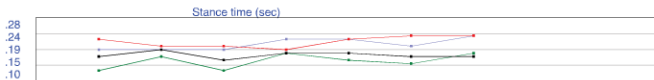
	Stance time(sec)	Stance %GC (%)	Total Pres. Index	TPI %	Stride Vel(vised)	Step Length(cm)	Stride Length(cm)	Step/Stride Ratio
Left Front	.21(0.02)	45.75(2.25)	39.0(6.14)	30.3 %	200.1	44.36(1.34)	87.9(2.67)	50.5%
Right Front	.21(0.02)	47.13(2.84)	42.0(5.63)	32.6 %	194.7	43.08(2.45)	87.5(3.15)	49.2%
Left Hind	.16(0.02)	34.42(2.97)	24.3(6.26)	18.9 %	201.3	44.27(1.67)	88.3(1.62)	50.1%
Right Hind	.17(0.01)	38.73(1.69)	23.4(2.76)	18.2 %	199.1	43.61(0.96)	88.0(2.16)	49.6%

Symmetry Ratio			
Front/Hind	1.27	1.27	1.70
Left/Right	0.97	0.93	0.97
Left Front/Right Front	1.00	0.97	0.93
Left Hind/Right Hind	0.94	0.89	1.04
Left Front/Left Hind	1.31	1.33	1.60
Right Front/Right Hind	1.24	1.22	1.79
Left Front/Right Hind	1.24	1.18	1.67
Right Front/Left Hind	1.31	1.37	1.73

Indicates a front load when comparing any front to hind or indicates left loading when comparing any left to a right

Indicates a hind load when comparing any front to hind or indicates right loading when comparing any left to a right



At 90 days post stem cell therapy, Gait4Dog objective gait analysis revealed an even pressure, stride length, and step length on the left hind limb as compared to the right.

instability on palpation (cranial drawer or thrust in flexion), or if the owners are unable to fully restrict and control the dog. It can be challenging to obtain a perfect custom brace fit for some dogs and bracing may not be possible even if recommended or attempted.

Weekly rehabilitation therapy, beginning 2 weeks postoperatively, is recommended for 12 weeks. Rehabilitation therapy helps to speed healing by decreasing post-operative inflammation and swelling, building muscle mass, increasing range of motion, and improving overall comfort. Therapy sessions included manual physical therapy, standard isometric exercises, gentle passive range of motion (PROM), and class III-b laser therapy. Class 4 lasers, ultrasound therapy and shock wave therapy are not recommended post stem cell injection as these modalities may harm the cells or decrease the ability of healing. NSAIDs are also not recommended as they can block the positive inflammatory response needed for appropriate healing. Rehabilitation therapy is performed weekly in conjunction with an at-home exercise program. Cool did not have access to an adequate rehabilitation therapy facility, so his owner diligently performed home exercises under the guidance of VOSM.

**Case Summary**

On 90-day post-operative physical examination, Cool was fully weight bearing in the left hind limb at a trot. Stifle palpation revealed no joint effusion in either stifle. There was no instability present in the left stifle joint. Gait4Dog objective gait analysis revealed an even pressure, stride length, and step length on the left hind limb as compared to the right. Arthroscopic exploration of the left stifle revealed that the cranial cruciate ligament was intact and the region that previously had showed disruption has healed. An area of neovascularization (new blood supply to the injured site of the CCL) was noted. The previously noted joint inflammation (synovitis) had resolved. Cool was slowly returned to activity over the course of 8 weeks and has since returned to full function. Cool returned to agility competition approximately 11 months after his injury and won a national competition to earn



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his spot on the AKC/USA World Team exactly one year after his stem cell/PRP treatment. Cool is currently over 2 years out and competing at his pre-injury level. Here's a video of him competing 12 months after therapy: <https://youtu.be/wL6yVyR-A1Y>.

Note: If you subscribe to the printed copy of the magazine, the easiest way to view the video is to go to the Clean Run magazine YouTube channel (<http://www.youtube.com/user/CleanRunMagazine>) and select the December 2015 play list.

### Conclusion

In conclusion, it appears based on the reports in animals and humans as well as our data at VOSM that stem cell therapy/PRP combination may be a viable option for agility dogs with partial CCL injuries. A definitive diagnosis and strict inclusion criteria, validated canine stem cell and PRP system, and regimented post-injection rehabilitation therapy must be included when considering this treatment. 🐾

Dr. Sherman O. Canapp, Jr. completed a combined Doctor of Veterinary Medicine/Master of Science at Kansas State University, an internship in small animal medicine and surgery at the University of Missouri, and followed up with a three-year residency in small animal surgery at the University of Florida. Dr. Canapp currently practices orthopedic surgery and sports medicine at Veterinary Orthopedic & Sports Medicine Group in Annapolis Junction, Maryland, where he is chief of staff. Dr. Canapp has earned diplomate status from both the American College of Veterinary Surgeons and the American College of Veterinary Sports Medicine and Rehabilitation. His primary focus and research is in arthroscopy, regenerative medicine, and sports medicine. Dr. Canapp lectures nationally and internationally and is a consultant for numerous organizations and medical companies.

Dr. Brittany Jean Carr is currently a rehabilitation therapist and American College of Veterinary Sports Medicine and Rehabilitation resident at Veterinary Orthopedic and Sports Medicine Group (VOSM) in Annapolis Junction, Maryland. She earned her B.S. in biology from Furman University in Greenville, South Carolina. She then attended the Virginia-Maryland Regional College of Veterinary Medicine at Virginia Tech in Blacksburg, Virginia. During her time in veterinary school, she served as president of the student chapter of the American Veterinary Medical Association and was honored with the College of Veterinary Medicine Outstanding Senior Award. After earning her DVM, Dr. Carr completed a small animal rotating internship at the Animal Specialty Group in Los Angeles, California and surgical internship at VOSM. Dr. Carr's research studies include objective gait analysis, regenerative medicine, and return to sport following cranial cruciate ligament injuries.



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