Cardiac Regenerative Therapy for Dilated Cardiomyopathy in Doberman Pinchers with PDK4 gene mutation

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Cardiac Regenerative Therapy for Dilated Cardiomyopathy in Doberman Pinchers with PDK4 gene mutation

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Doberman Pinscher Dilated Cardiomyopathy

Dilated cardiomyopathy (DCM) is a common cardiac affliction in people and dogs. The Doberman Pinscher is a canine breed affected with DCM more than any other dog breed, accounting for at least 50% of all cases of canine DCM. They have a very high morbidity and mortality rate. The disease has been shown to be inherited as an autosomal dominant trait with incomplete penetrance.

Prior to knowing the specific genetic mutation in this breed, regenerative strategies used by our group of researchers included the use of stem cell therapy to promote repair in damaged myocardial tissue using rat and swine models. An improvement in the ejection fraction was noted up to 8 weeks after therapy (Figures 1 and 2).

Due to the encouraging results obtained with our rat and swine models we decided to study three different AAV vectors (AAV8, AAV9 and AAV2tm) to infect canine stem cells (Figures 3 and 4). After selecting AAV2tm as our vector of interest, we further evaluated its biodistribution and the safety of retrograde coronary sinus delivery method in normal dogs (Figures 5 and 6).

Next our group safely delivered stem cell therapy to 15 Dobermans affected with DCM via retrograde coronary sinus delivery. Median procedural time in these patients was 35 minutes and most dogs were discharged 24 hours following the procedure without complications (Figures 7 and 8).

Recently, a specific genetic mutation was identified in Doberman Pinchers with DCM, which encodes for a mitochondrial protein involved in energy metabolism (PDK4). Preliminary results of our current study on the role of PDK4 mutation indicate that Doberman pinschers with DCM and the PDK4 mutation have a lower basal mitochondrial oxygen consumption rate (OCR) compared to normal Dobermans. We have successfully used gene therapy with AAV2tm-cPDK4 to transfect skin fibroblasts of Dobermans carrying the genetic mutation to assess changes in the OCR. Preliminary results of this experiment have shown a trending in increase in OCR.

Future studies are aimed into assessing the global role of the PDK4 mutation in mitochondrial metabolism and creation of a PDK4 Knockdown cell line to compare results and eliminate the contribution of other genetic factors to the disease phenotype.

Modified Skeletal Myoblast Therapy for Cardiac Failure in Rattus and Porcine Models

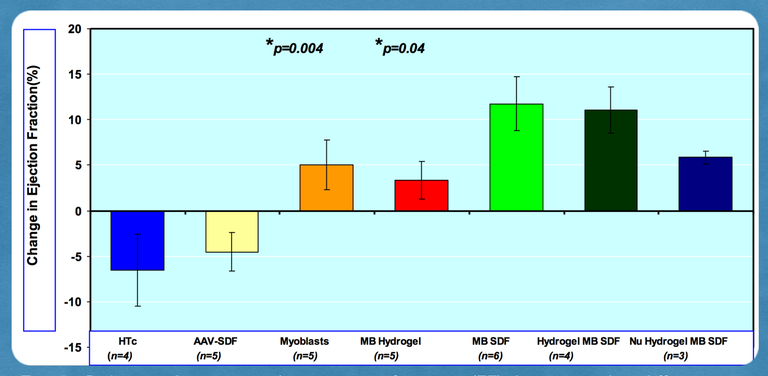


Fig 1. Relative changes in the ejection fraction (EF) between the different treatment groups in rats.

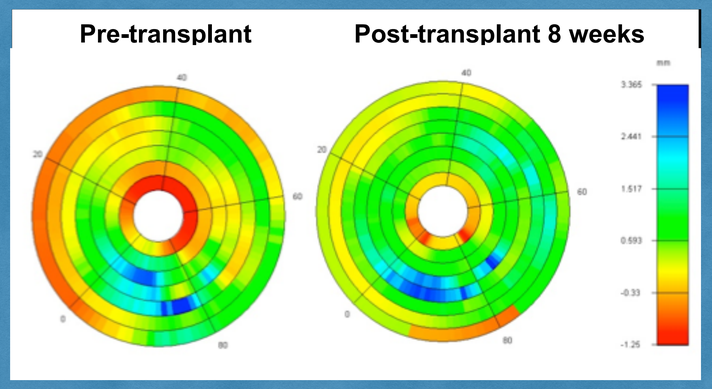


Fig 2. Bulls eye plot of the infarcted myocardium 3 weeks post-infarction and 8 weeks post-transplantation with myoblasts transduced with AAV1 SDF1.

Conlon T, et al. Modified Skeletal Myoblast Therapy for Cardiac Failure Using AAV SDF1 in Rattus and Porcine Models. 28th Ann Ped Sci Day, University of Florida, May 13, 2010.

Evaluation of three AAV vectors to infect Canine MSCs*

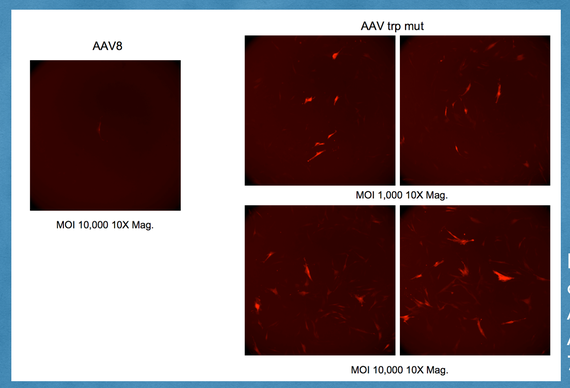


Fig 3. Microscopic images of CnMSCs transduced with AVV8-mCherry and AAV2tm-mCherry 72 hours post infection.

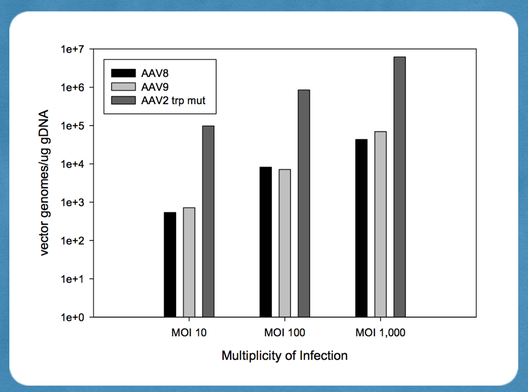


Fig 4. Comparison of vector genomes/ug gDNA and MOI between AAV8, AAV9 and AAV2tm.

Unpublished data*

AAV2tm-MSC-ferritin Biodistribution and Safety Study in Beagle Dogs

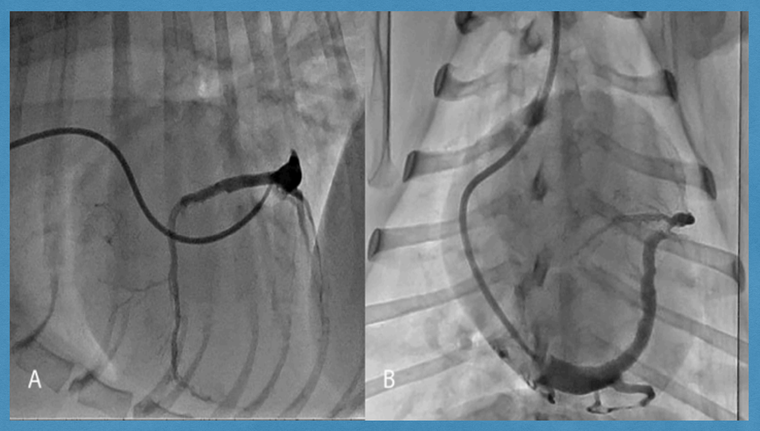


Fig 5. Fluoroscopic images obtained during contrast injection into the coronary sinus delineating the site of cell delivery during MSC injection.

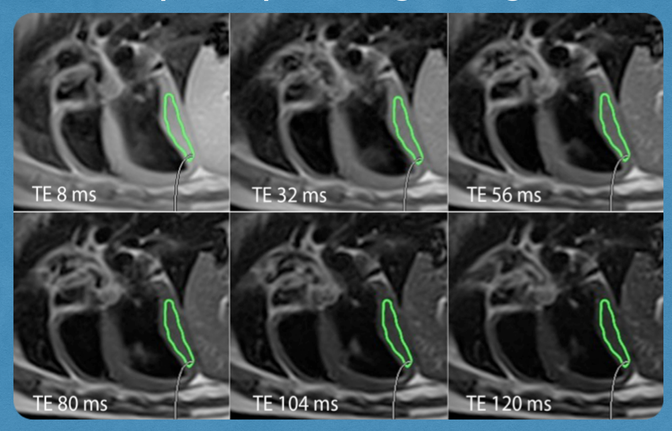
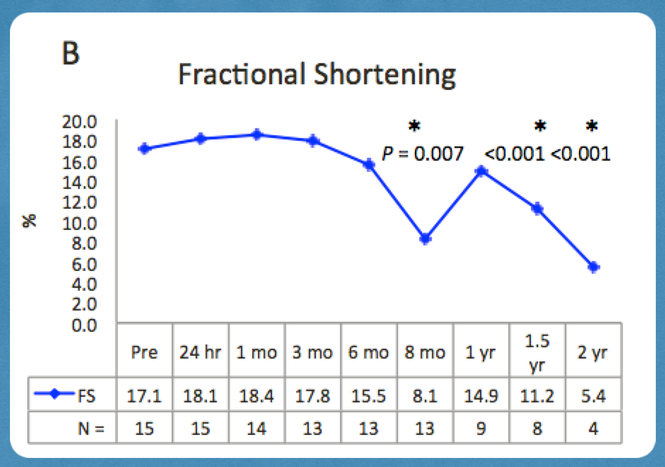


Fig 6. Parasagittal MRI images of the heart with a region of interest drawn to include the left ventricular free wall used to calculate T2 relaxation time.

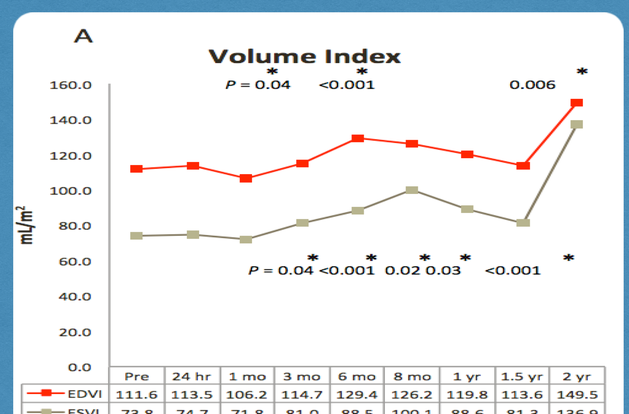
Sosa, I., Estrada, A. H., Winter, M. D., *et al.* (2012) Biodistribution of retrograde coronary sinus delivery of mesenchymal stem stems: A Pilot Study. North American Veterinary Regenerative Medicine Association Conference. Savannah, GA, USA

Delivery of AAV2tm-SDF1-MSC to Dobermans with DCM



	Pre	24 hr	1 mo	3 mo	6 mo	8 mo	1 yr	1.5 yr	2 yr
FS	17.1	18.1	18.4	17.8	15.5	8.1	14.9	11.2	5.4
N =	15	15	14	13	13	13	9	8	4

Fig 7. Mean ejection fraction (%) of all dogs from the day prior to stem cell implantation (pre) to two years post stem-cell implantation.



	Pre	24 hr	1 mo	3 mo	6 mo	8 mo	1 yr	1.5 yr	2 yr
EDVI	111.6	113.5	106.2	114.7	129.4	126.2	119.8	113.6	149.5
ESVI	73.8	74.7	71.8	81.0	88.5	100.1	88.6	81.3	136.9
N =	15	15	14	13	13	13	9	8	4

Fig 8. Mean end-diastolic volume indexes (red) and mean end-systolic volume indexes (green) from the day before stem-cell implantation (pre) to 2 years post stem-cell implantation

Pogue, B, et al. Stem-cell therapy for dilated cardiomyopathy: a pilot studying evaluating retrograde coronary venous delivery. JSAP (2013), 54, 361-366.