

Tissue engineering for cartilage repair: pre-clinical trial evaluation methods

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- We presented methods for evaluation of a scaffold-free cell delivery system made from mesenchymal stromal cells in a translational study that allows further clinical trial studies of safety and efficacy using good manufacturing practices for human usage.



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- Background: A chondral defect is a limiting condition that may cause worsening in quality of life and economic burden due to the cost of immediate treatment and losses in work productivity. Additionally, if left untreated, knee disorders may progress to osteoarthritis, a degenerative and debilitating joint disease characterized by pain and functional impairment. The predictable sources and relatively easy handling of mesenchymal stromal cells (MSCs) that have immune-modulatory properties and the ability to differentiate into chondroblasts and osteoblasts are helpful for tissue engineering and treatment of cartilage injuries. We aim to present method tools to evaluate cartilage repair by tissue engineered treatments in a translational and pre-clinical large animal model.

METHODS

- Experiment:** This controlled experimental study with fourteen miniature pigs tested a scaffold-free Tissue Engineering Construct (TEC) derived from dental pulp and synovial MSCs for cartilage therapy. Total thickness cartilage defects were performed in both posterior knees. The defect was left empty in one of the knees, and the other received the TEC.

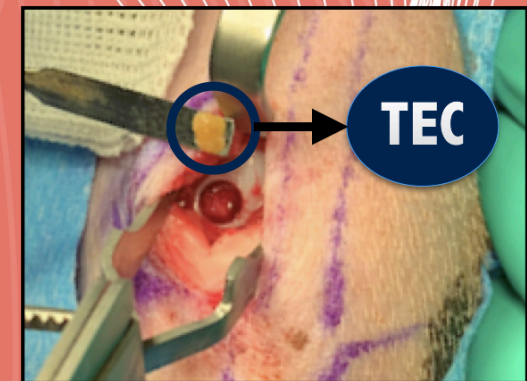
14 mini-pigs (BR1)



28 Surgeries



14 chondral defect
(defect group)



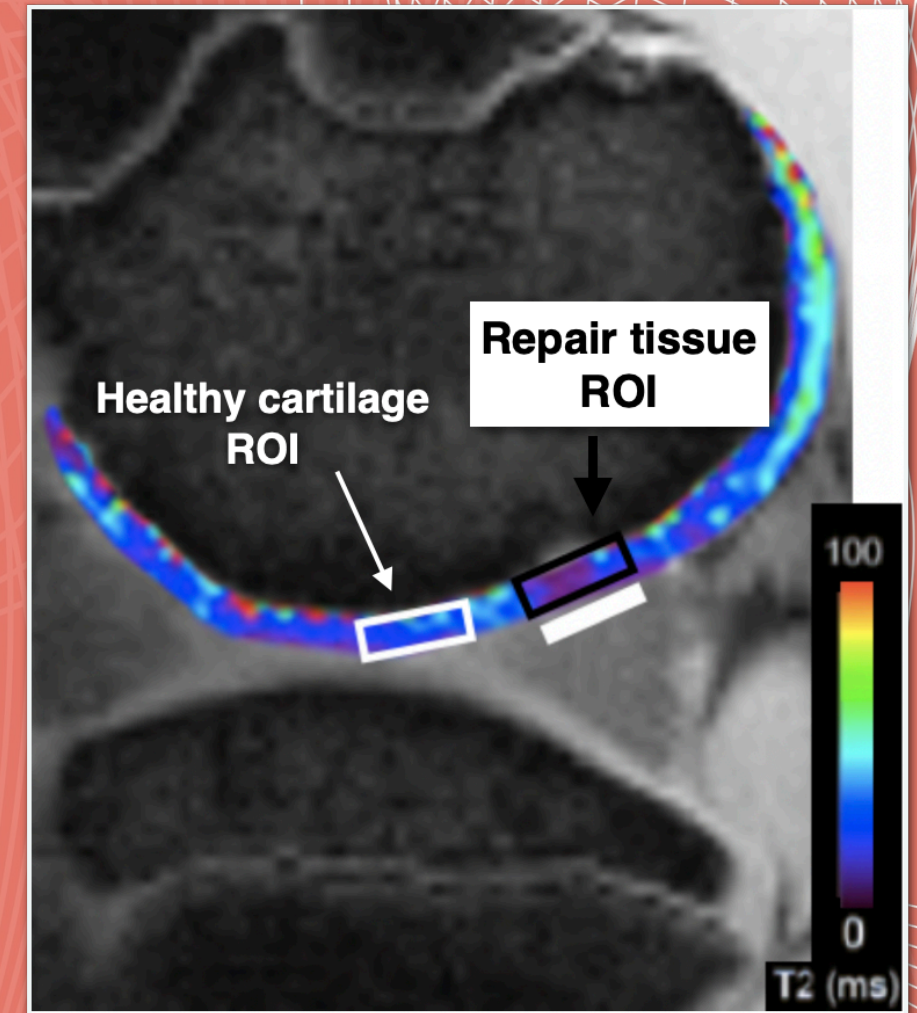
14 chondral defect+TEC
(experimental group)



METHODS

The tissue repair from both side were morphologically assessed with 3D MOCART score from magnetic resonance imaging using the 3D-DESS sequence, and compositional assessment was carried out based on T2 mapping technique.

3D MOCART score		
Category	Item	Points
Defect fill	0%	0
	0–25%	3
	25–50%	5
	50–75%	10
	75–100%	15
	100%	20
	100–125%	15
	125–150%	7
Cartilage interface	150–200%	3
	>200%	0
	Complete	10
	Demarcating border	8
	Defect visible < 50%	3
Bone interface	Defect visible > 50%	0
	Complete	10
	Partial delamination	5
Surface	Complete delamination/delamination of periosteal flap	0
	Intact	10
	Damaged < 50% depth	5
	Damaged > 50% depth	0
Structure	Homogeneous	10
	Inhomogeneous	5
	Cleft formation	2
	Absence of repair tissue	0
Signal intensity	Normal (identical to adjacent cartilage)	10
	Nearly normal (slight areas of signal alteration)	5
	Abnormal (large areas of signal alteration)	0
Chondral osteophytes	Absent	5
	<50% of chondral thickness	3
	>50% of chondral thickness	0
Integrity of subchondral bone plate	>75%	10
	50–75%	8
	25–50%	5
	0–25%	3
	0%	0
Subarticular spongiosa	Intact	10
	Granulation tissue	8
	Sclerosis	8
	Cyst	5
	Granulation tissue and sclerosis	5
	Granulation tissue and cyst	2
	Sclerosis and cyst	2
	Granulation tissue, sclerosis and cyst	0
Adhesions	Absent	3
	Yes	0
Effusion	Absent	2
	Yes	0
Total points		100



MOCART 3D score.

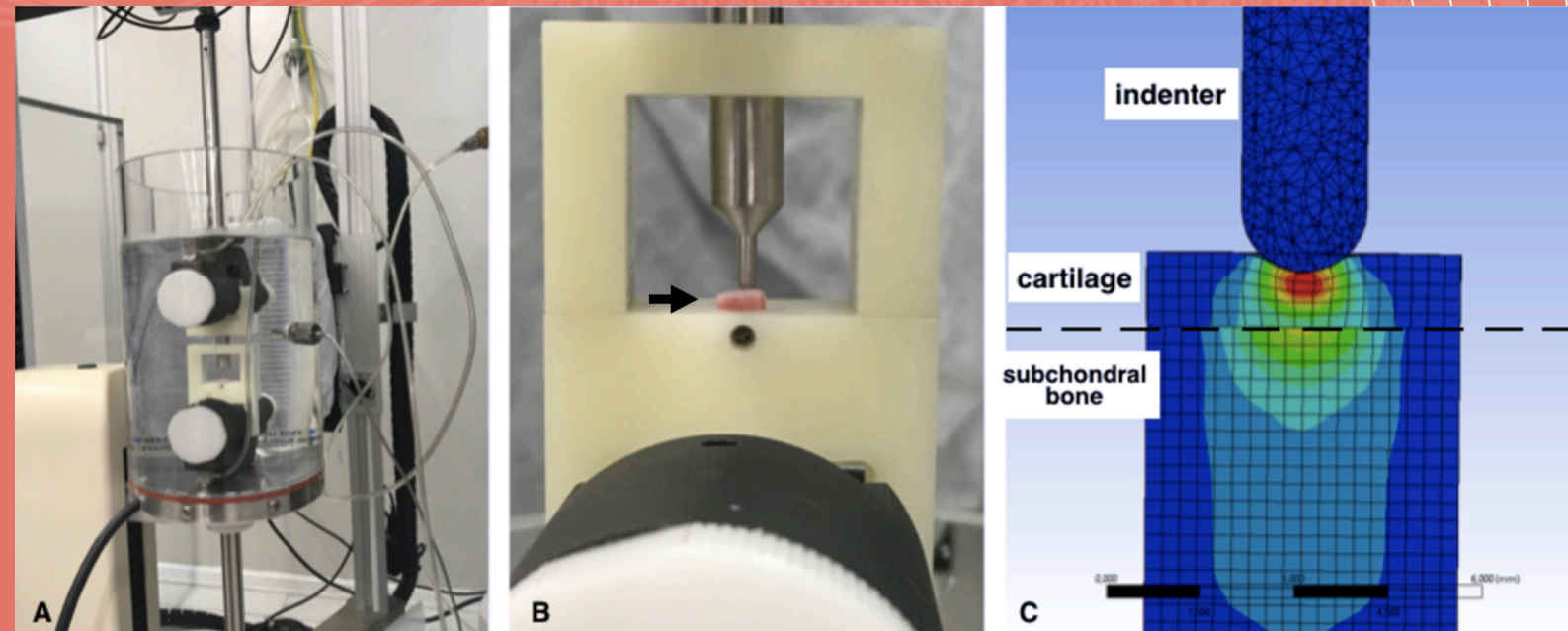
(Welsch et al. 2009)

(Example of T2 mapping ROI selection)



METHODS

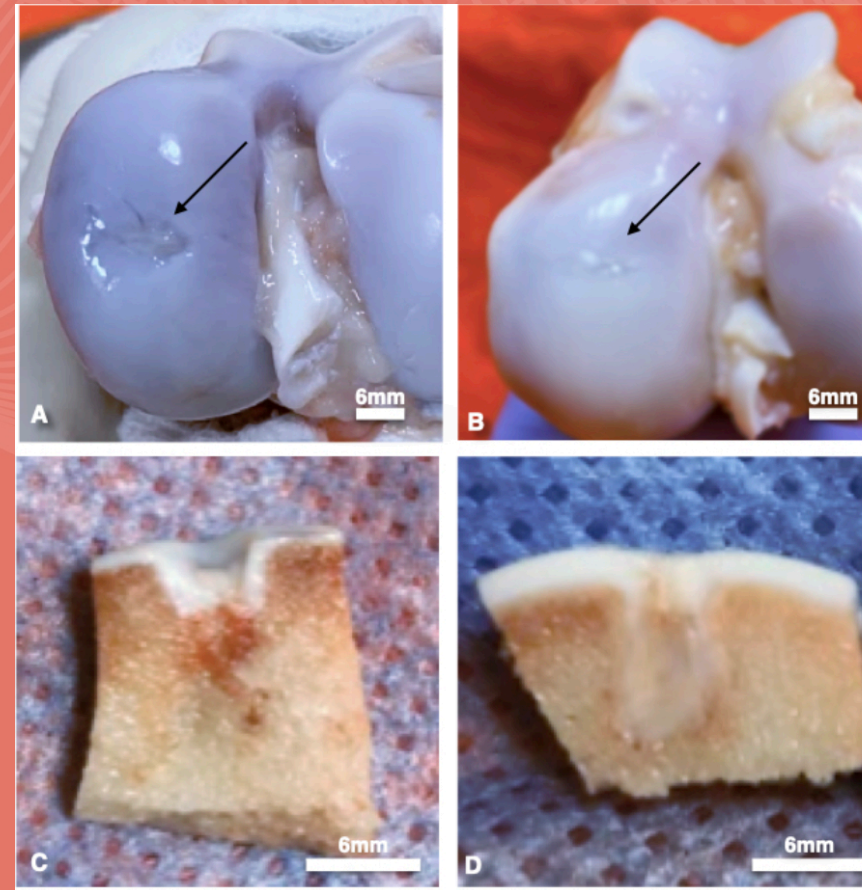
- The **mechanical evaluation** was made from indentation and maximum compression tests, and the Finite Element model was used to simulate and characterize properties of the osteochondral block.



Indentation test set up. Structure immersed in saline solution (0.9% NaCl) at a temperature of 36°C. Sample placed on a support with the cartilage part facing up and positioned on the equipment

METHODS

- The osteochondral specimens were fixed for **histopathology**, decalcified, submitted to standard histological processing, sectioned, and stained with hematoxylin & eosin. The sections stained for immunohistochemical detection of collagen types were digested with pepsin and chondroitinase and incubated with antibodies against them.



Osteochondral specimens from control group (A and C) and Treatment group (B and D). The osteochondral blocks (C and D) were submitted to histological processing.



RESULTS

- At six months after surgery, there were no complications with the animals and the MRI, histological, immunohistochemical and biomechanical evaluations proved to be viable and qualified to differentiate good quality chondral repair from inadequate repair tissue.



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- **Conclusions:** The proposed methods were viable and capable to correctly evaluate the defect filled in with TEC containing mesenchymal cells after six months of follow-up on a large animal model for articular cartilage restoration.



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THANK YOU

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