TISSUE ENGINEERING OF BOVINE PERICARDIAL TISSUE IN THE CIRCULATORY SYSTEM OF A YOUNG OVINE MODEL: COMMERCIAL VERSUS IN-HOUSE DECELLULARIZATION

*Botes L[1], **van den Heever JJ**[2], Laker L[2], Bester D[2], Dohmen PM[2&3], Smit FE[2]*

[1]Dept of Health Sciences, Central University of Technology, Bloemfontein, South Africa
[2]Dept of Cardiothoracic Surgery, University of the Free State, Bloemfontein, South Africa
[3] Department of Cardiovascular Surgery, Rostock University, Germany

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Introduction

- **Biological substitution material in reconstructive surgery**
  - Allografts/Homografts – Valves & Vasculature
  - Xenografts – Bovine Pericardium
- **General Requirements**
  - Versatility and Durability
  - Biocompatibility
  - Recellularization potential
- **Frequent complications**
  - Restenosis
  - Pseudoaneurysm formations
  - Calcification and fibrosis
  - Infections
- **Tissue engineering**
  - Decellularization to minimize immune response

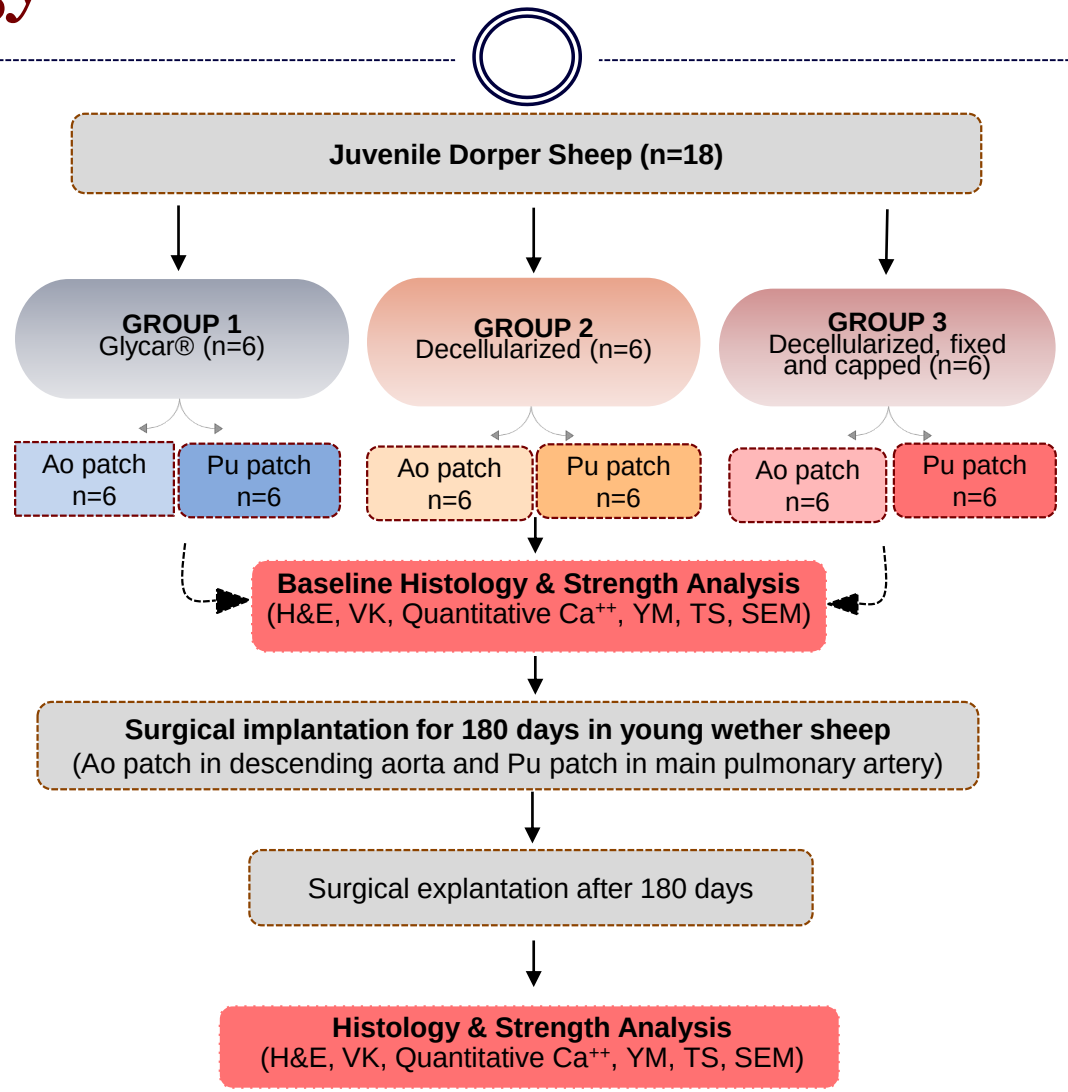
Aim of the Study

Compare the **Biological Interaction and Tissue Integrity** of

- *a Commercially available Bovine Pericardial patch (**Glycar**)*
- *In-house **Decellularized** Bovine Pericardium*
- *In-house **Decellularized + Fixed & Capped B**ovine Pericardium*

in the circulatory system of young sheep

Methodology



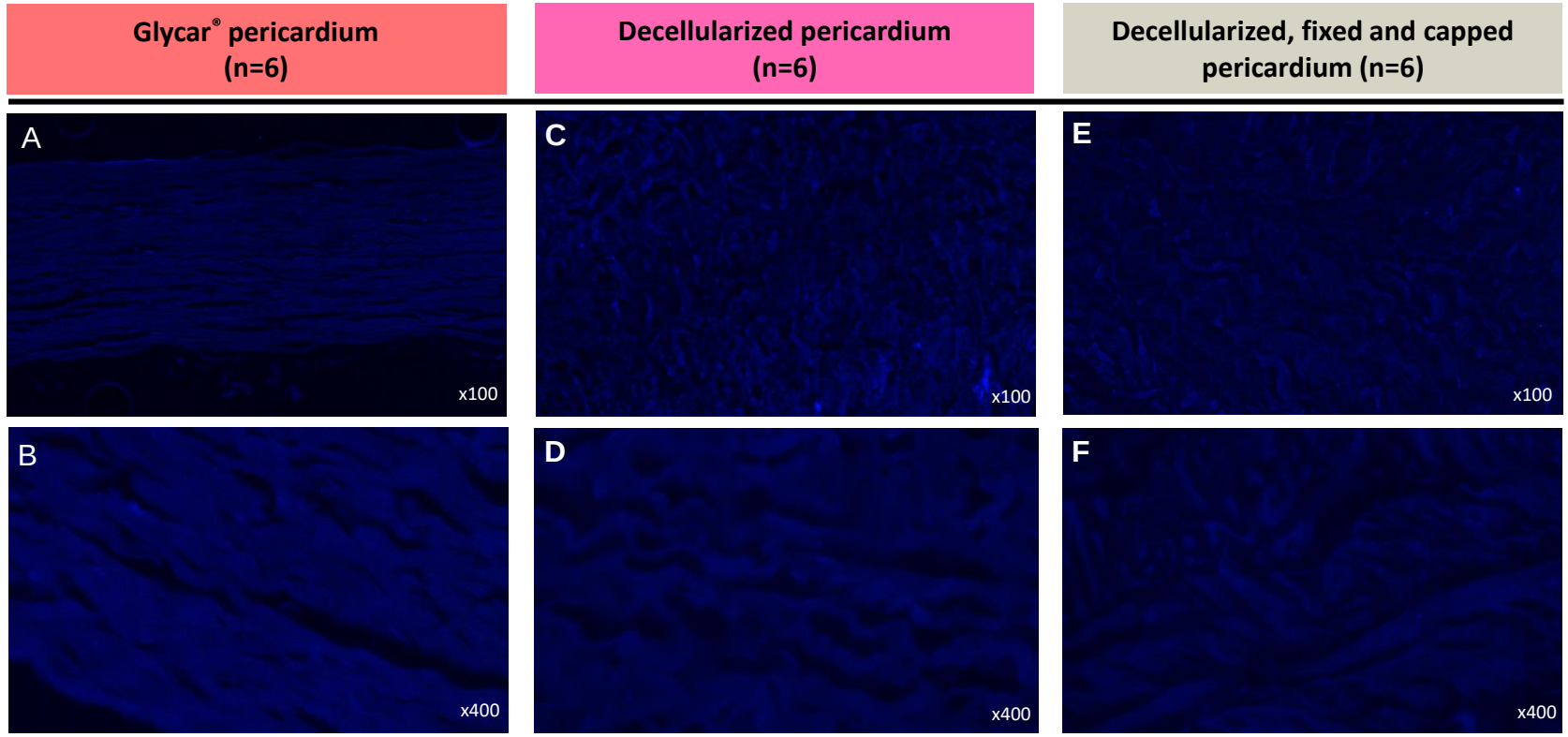
Methodology

Decellularization included cycles of:

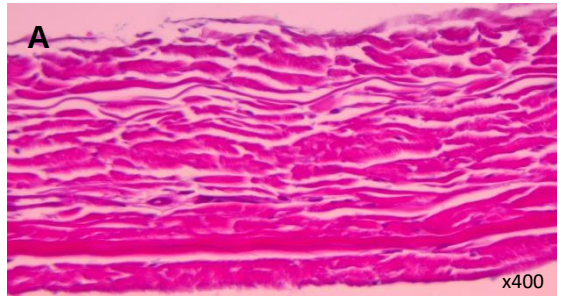
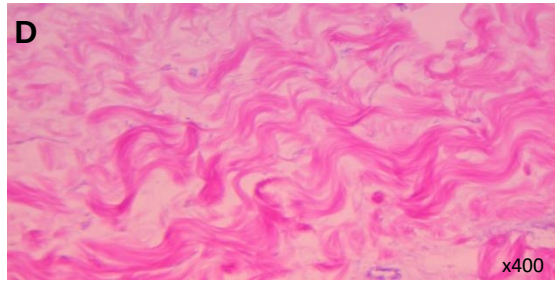
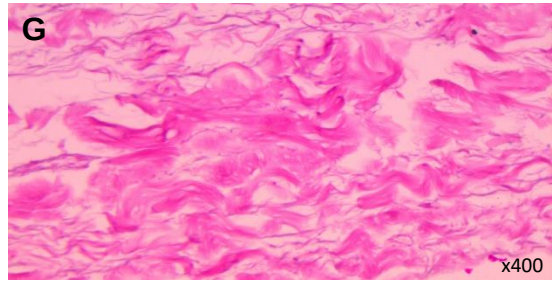
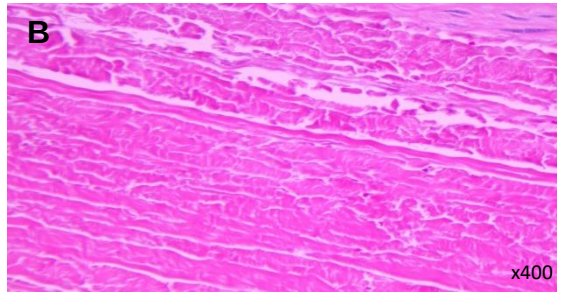
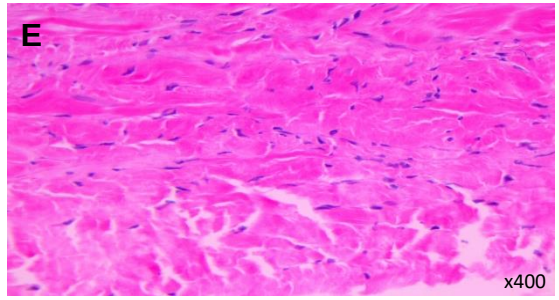
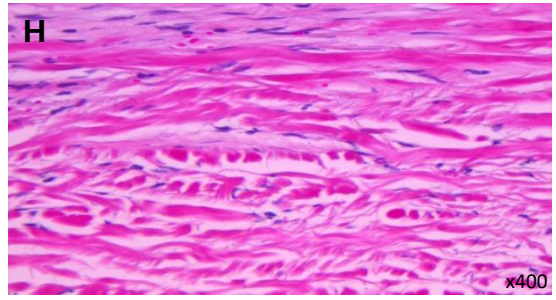
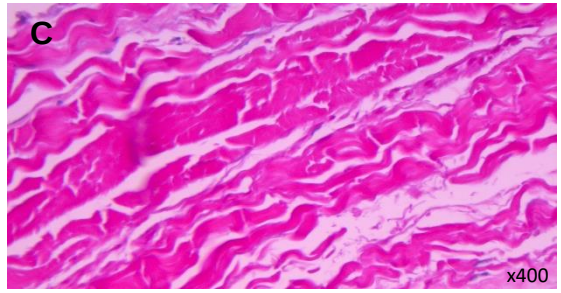
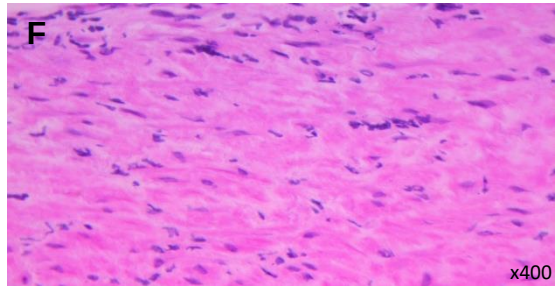
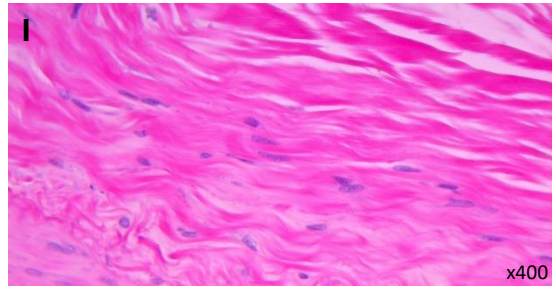
Hyper- and hypotonic solutions,
Detergents,
Washings, and
Sterilization and Storage (in A/B)

Tissue in Group 3 were additionally fixed with Glutaraldehyde(GA), capped with Propylene glycol and stored in Propylene oxide

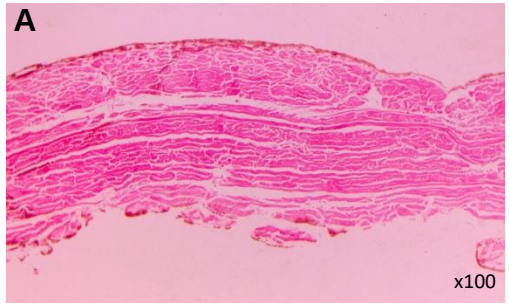
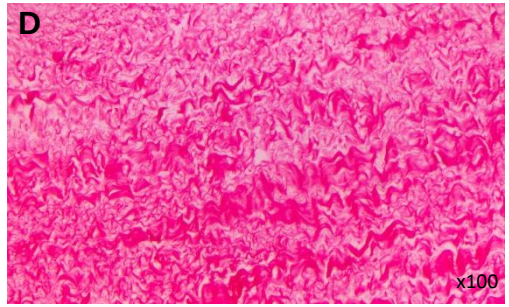
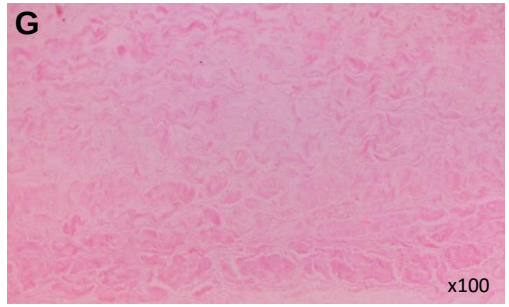
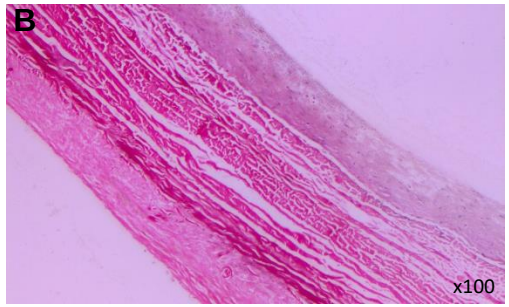
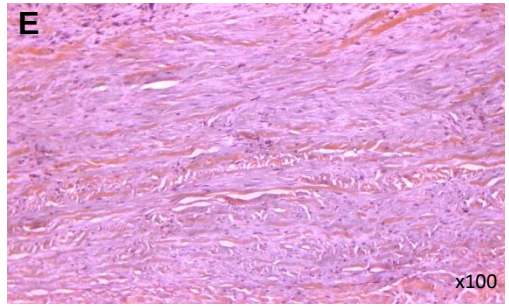
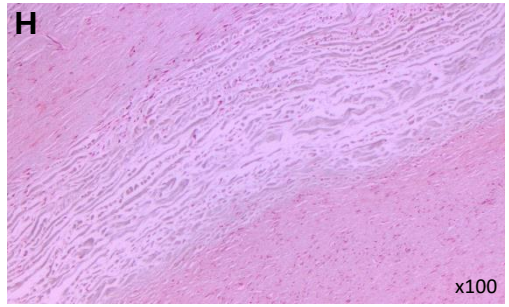
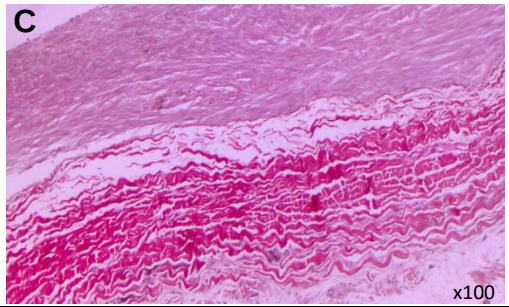
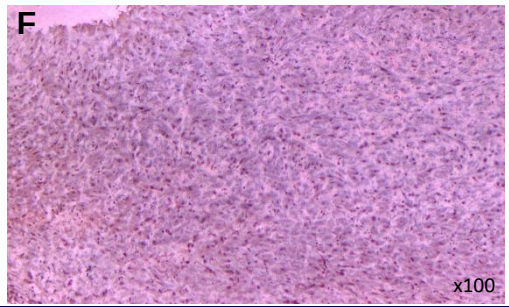
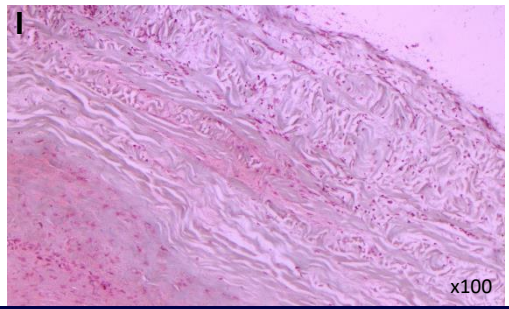
Results (Pre-implantation) - DAPI



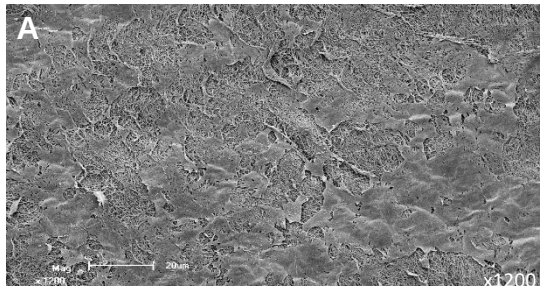
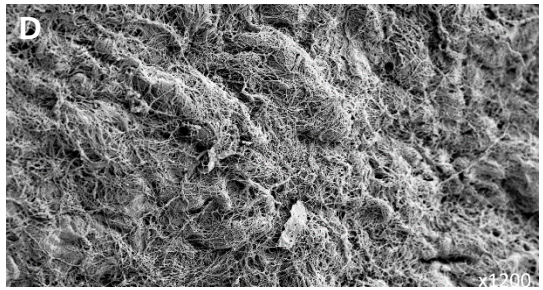
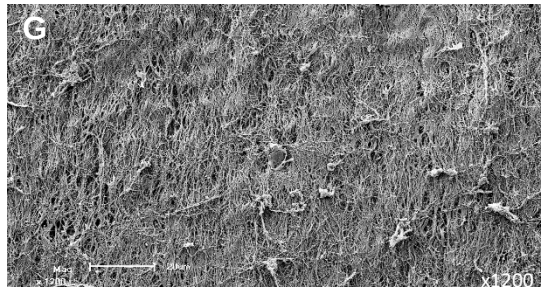
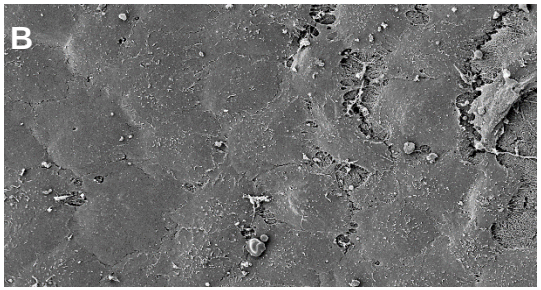
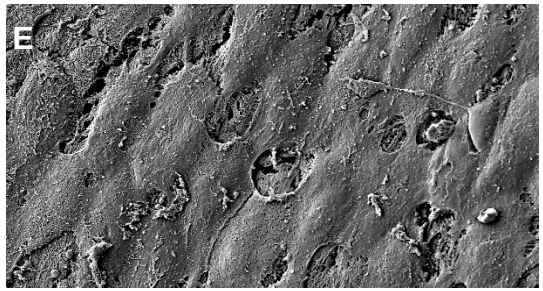
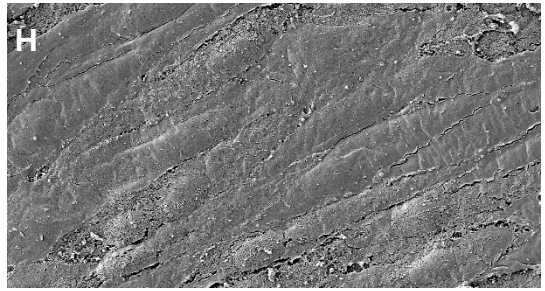
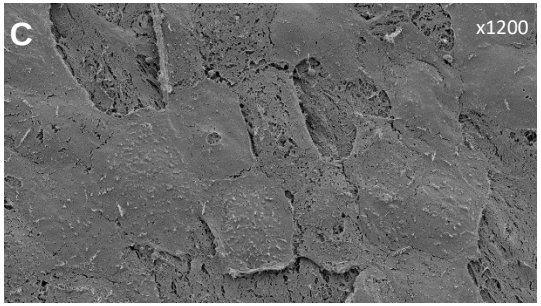
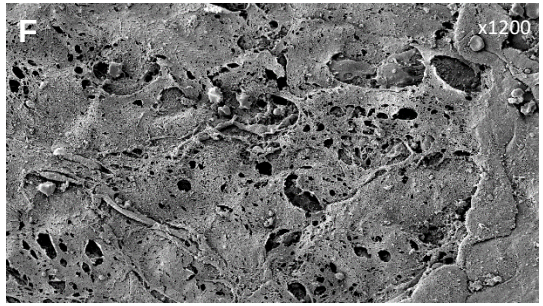
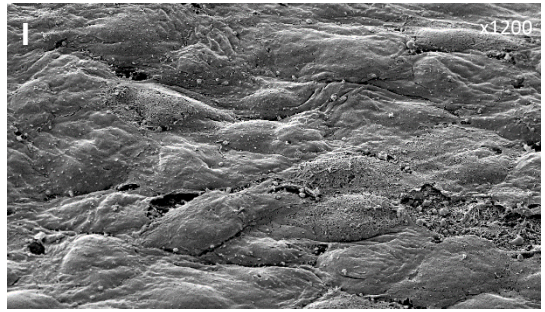
Results (Post-implantation) – H&E

	Glycar® pericardium (n=6)	Decellularized pericardium (n=6)	Decellularized, fixed and capped pericardium (n=6)
Baseline			
Ao Explant			
Pu Explant			

Results (Post-implantation) – von Kossa

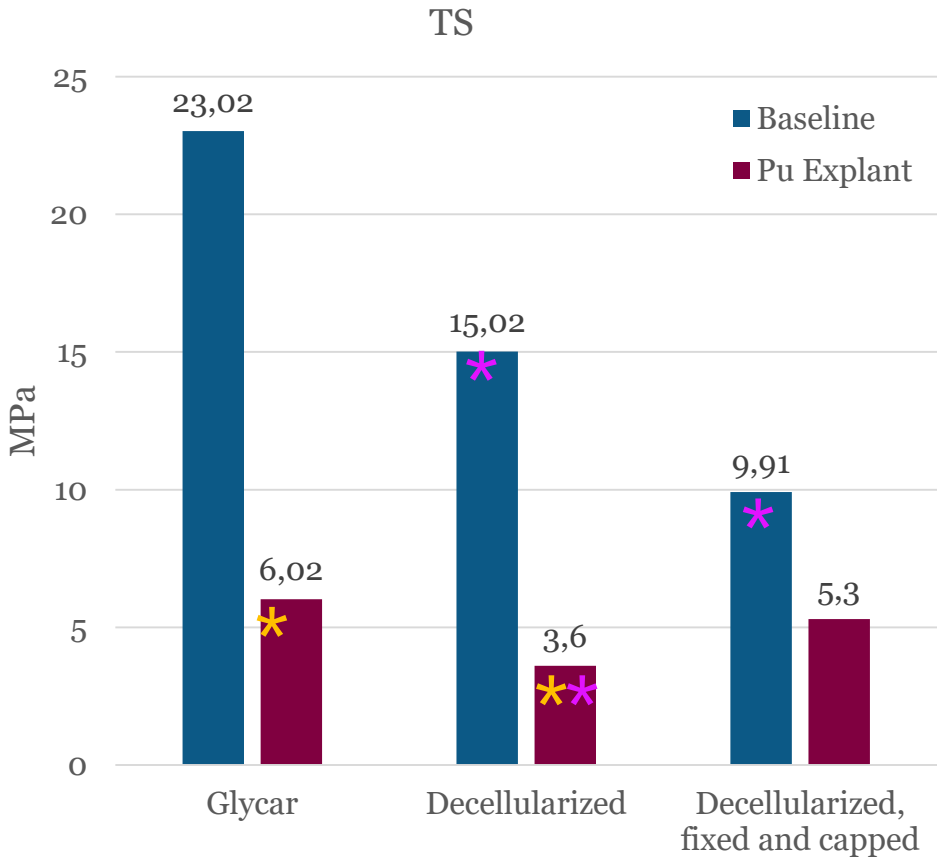
	Glycar® pericardium (n=6)	Decellularized pericardium (n=6)	Decellularized, fixed and capped pericardium (n=6)
Baseline			
Ao Explant			
Pu Explant			

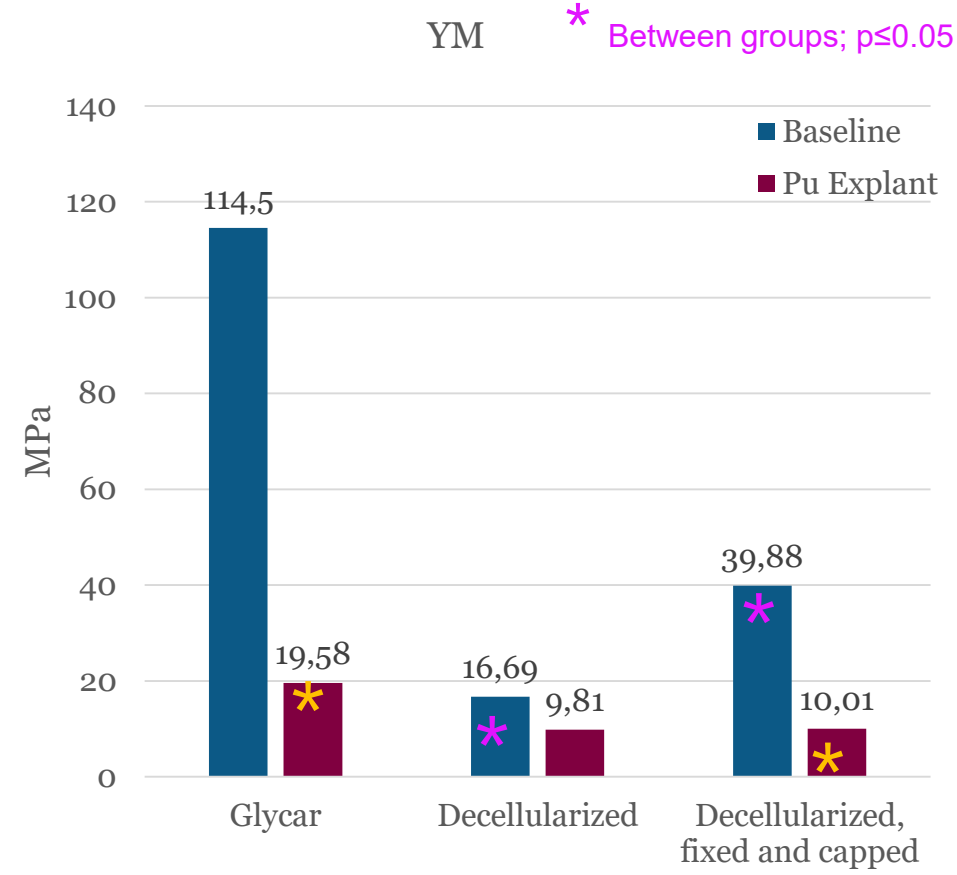
Results (180 days explantation) - SEM

	Glycar® pericardium (n=6)	Decellularized pericardium (n=6)	Decellularized, fixed and capped pericardium (n=6)
Baseline			
Ao Explant			
Pu Explant			

x1200 x1200 x1200

Results (180 days explantation) – TS & YM

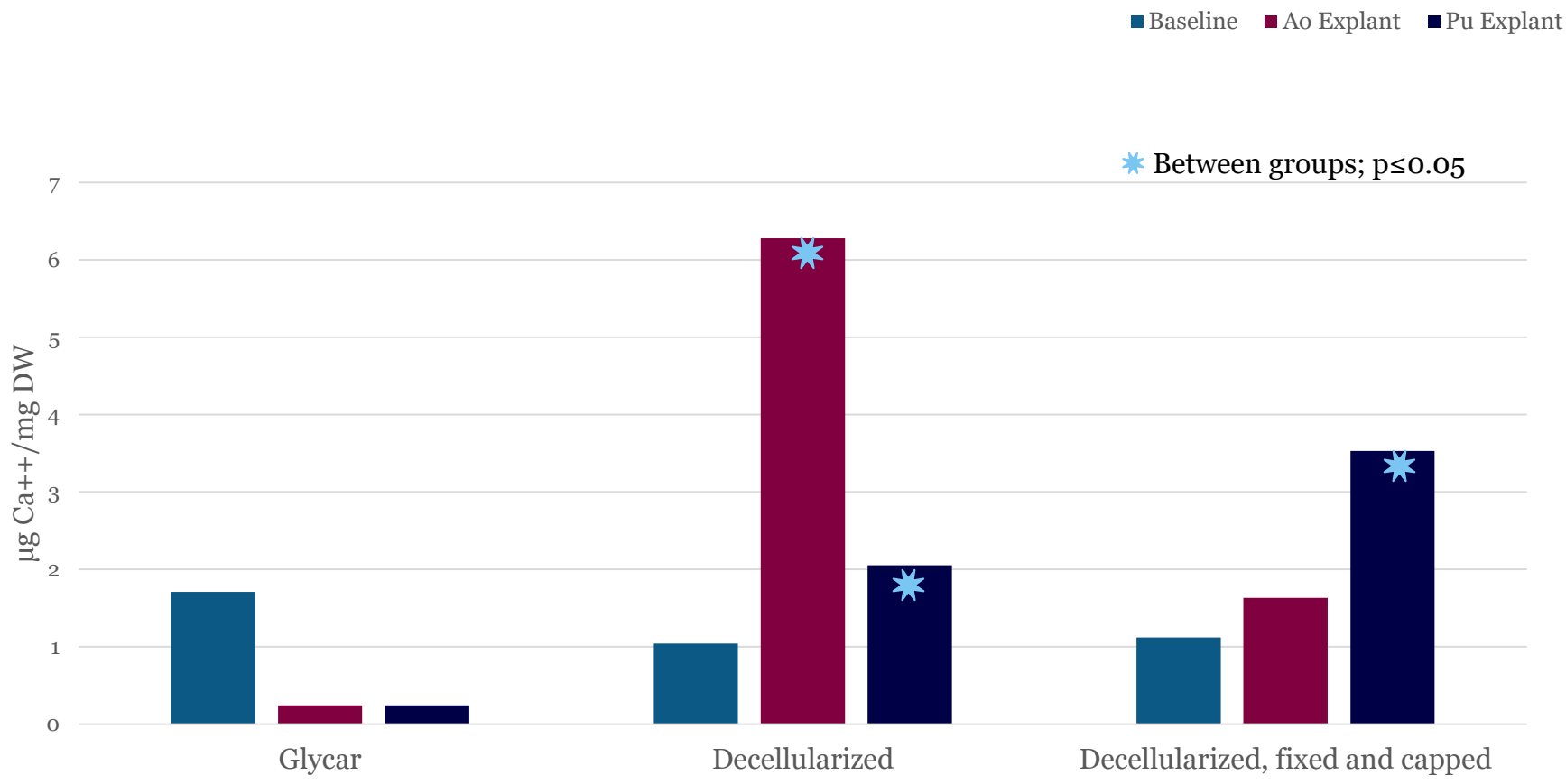




* Within group; p≤0.05

* Between groups; p≤0.05

Results – Quantitative Ca^{++}



Conclusion

- Unfixed decellularized pericardial tissue does not appear inferior to other commercial products regarding mechanical properties after implantation in the circulatory system of young sheep.
- Decrease in YM of explanted tissues indicated that the tissue became more pliable.
- Uniform host cell ingrowth creates the potential for tissue regeneration, growth potential and reduction in calcification and early degeneration.
- Elevated calcium levels in explants from group 2&3 might be attributed to presence of intracellular calcium in host cell ingrowth.

Thank You
Dankie

T: 051 401 9111 info@ufs.ac.za **www.ufs.ac.za**

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Results (180 days explantation) – TS & YM

Variable	Glycar® (n=6) (Median ± SD)			Decellularized (n=6) (Median ± SD)		
	Baseline	Explant (Pu)	Baseline vs. Explant	Baseline	Explant (Pu)	Baseline vs. Explant
YM (MPa)	114.50±40.41	19.58±17.18	**p=0.0027***	16.69±9.87 **p=0.0001***	9.81±3.55 **p=0.0604**	**p=0.2228**
TS (MPa)	23.02±4.72	6.02±0.96	**p=0.0004***	15.02±3.25 **p=0.0238***	3.60±1.28 **p=0.0144***	**p=0.0004***

*- Significant (p<0.05)(SD = Standard Deviation; Pu = Pulmonary; YM = Youngs modulus; TS = tensile strength)

Variable	Glycar® (n=6) (Median ± SD)			Decellularized, fixed and capped (n=6) (Median ± SD)		
	Baseline	Explant (Pu)	Baseline vs. Explant	Baseline	Explant (Pu)	Baseline vs. Explant
YM (MPa)	114.50±40.41	19.58±17.18	**p=0.0027***	39.88±28.46 **p=0.0150***	10.01±11.41 **0.1442**	**p=0.0121***
TS (MPa)	23.02±4.72	6.02±0.96	**p=0.0004***	9.91±2.53 **p=0.0001***	**5.30**±1.40 **p=0.2567**	**p=0.1051**

*- Significant (p<0.05)(SD = Standard Deviation; Pu = Pulmonary; YM = Youngs modulus; TS = tensile strength)

Results (180 days explantation) – Ca++

Table 3.2 Quantitative calcium analysis of baseline and explanted aortic and pulmonary pericardial patches: Glycar® and Decellularized groups

Variable	Glycar® (n=6) (Median ± SD)				Decellularized (n=6) (Median ± SD)			
	Baseline	Explant (Ao)	Explant (Pu)	Baseline vs. Explant	Baseline	Explant (Ao)	Explant (Pu)	Baseline vs. Explant
Ca^{++}	1.71±1.12	1.04±12.74	1,12±0.41	-	0.24±0.16 **p=0.3757**	6.26±6,17 **p=0.0074***	1,63±7,60 **p=0.0067***	-
Baseline vs. explant (Ao)	-	-	-	**p=0.2289**	-	-	-	**p=0.0003***
Baseline vs. explant (Pu)	-	-	-	**p=0.4576**	-	-	-	**p=0.0015***

*- Significant ($p<0.05$)(SD = Standard Deviation; Pu = Pulmonary; Ca = Calcium)

Table 5.2 Quantitative calcium analysis of baseline and explanted aortic and pulmonary pericardial patches: Glycar® and decellularized, fixed and capped groups

Variable	Glycar® (n=6) (Median ± SD)				Decellularized, fixed and capped (n=6) (Median ± SD)			
	Baseline	Explant (Ao)	Explant (Pu)	Baseline vs. Explant	Baseline	Explant (Ao)	Explant (Pu)	Baseline vs. Explant
Ca^{++}	1.71±1.12	1.04±12.74	1,12±0.41	-	0.24±0.07 **0.3604**	**2.05**±15.76 **0.0866**	**3.53**±3.86 **0.0054***	-
Baseline vs. ex-plant (Ao)	-	-	-	**p=0.2289**	-	-	-	**p=0.0020***
Baseline vs. ex-plant (Pu)	-	-	-	**p=0.4576**	-	-	-	**p=0.0007***