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Influence of short-term storage at 4 °C, -20 °C or -80 °C on the quality of decellularized porcine oviductal scaffolds

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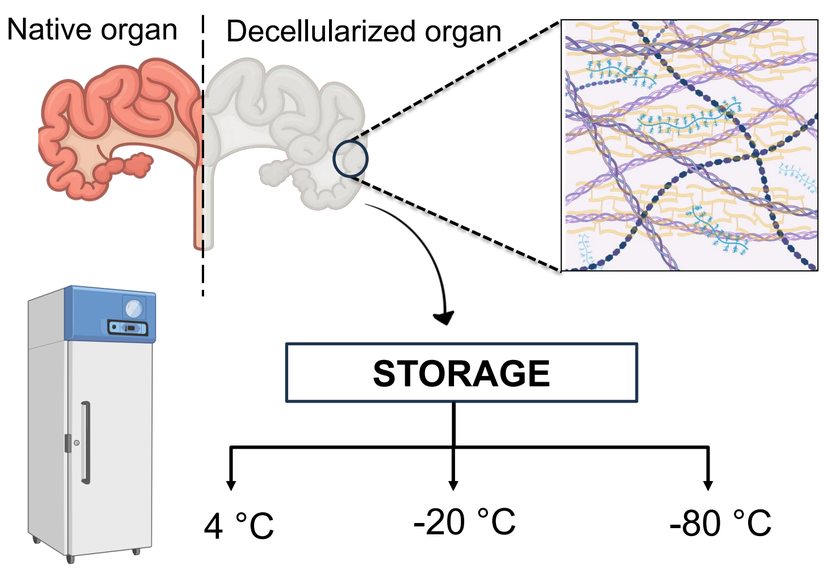
INTRODUCTION & AIM

Decellularization is a process that allows the isolation of the extracellular matrix (ECM), yielding biocompatible, low-immunogenic scaffolds that retain key components and 3D structure.



APPLICATIONS

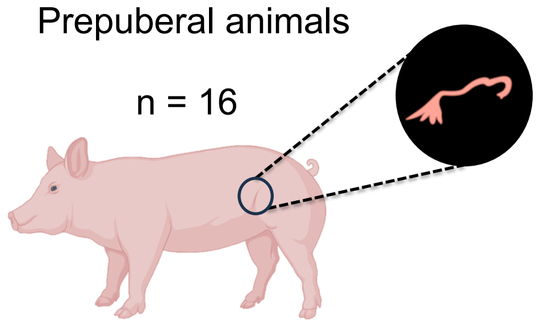
- Tissue regeneration
- Improvement of assisted reproductive technologies (ART)

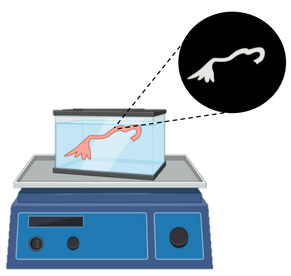
The effect of storage temperature on scaffold quality remains unclear.

The aim of this study was to evaluate the short-term effect of storage temperature (4 °C, -20 °C, -80 °C) on the quality of decellularized porcine oviduct scaffolds.

METHODS

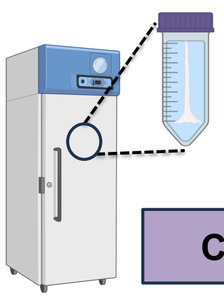
1) Obtaining porcine oviducts
Prepuberal animals
n = 16





2) Decellularization
Immersion-agitation method [1]

3) Storage
Four experimental groups (n = 4 per group)

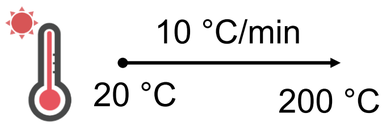


- **Control**: Freshly decellularized
- **4 °C**, **-20 °C**, **-80 °C**: Samples stored for 1 month

4) Scaffold evaluation

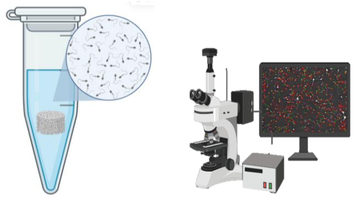
Thermal stability

Differential scanning calorimetry to evaluate thermal stability



Biocompatibility

Co-incubation with porcine spermatozoa (3h, 38 °C)



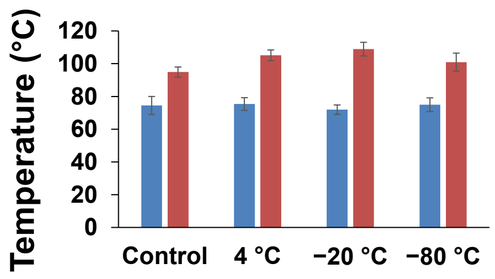
Microbiology

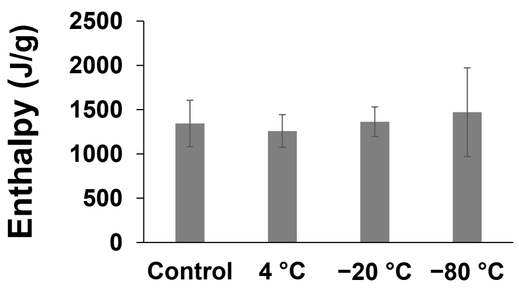
Analysis of TAB, TC and SRC

TAB: Total aerobic bacteria; TC: Total coliform; SRC; sulfite-reducing clostridia

RESULTS & DISCUSSION

Thermal stability

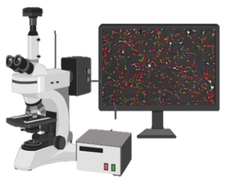




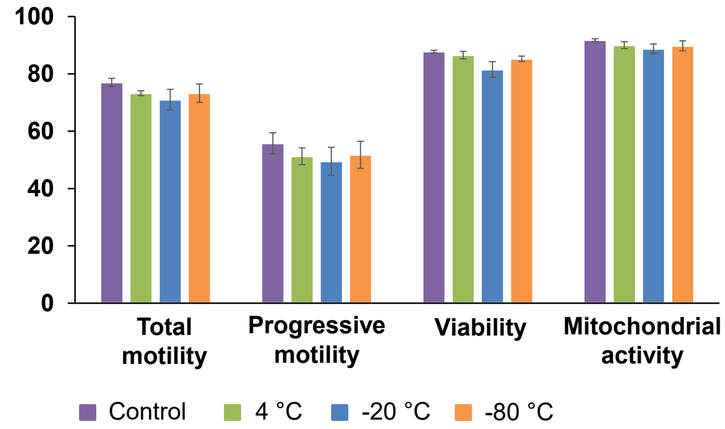
Tonset: temperature at which protein denaturation begins.
Tpeak: temperature at which the maximum denaturation peak occurs.

Storage temperature did not affect the protein stability

Biocompatibility



Computer-Assisted Sperm Analysis (CASA)



Storage temperature did not affect the scaffold biocompatibility

Microbiology

Groups	TAB (CFU/mL)	TC (CFU/mL)	SRC (CFU/mL)
Control	$4.73 \times 10^5 \pm 1.09 \times 10^{5\ a}$	$6.03 \times 10^3 \pm 5.98 \times 10^3$	0
4 °C	$6.97 \times 10^6 \pm 2.64 \times 10^{6\ b}$	0	0
-20 °C	$6.20 \times 10^6 \pm 5.29 \times 10^{5\ b}$	0	0
-80 °C	$2.23 \times 10^6 \pm 8.95 \times 10^{5\ ab}$	0	0

*Different superscripts letters (a, b) denote statistically significant differences ($p < 0.05$)
TAB: Total aerobic bacteria; TC: Total coliform; SRC; sulfite-reducing clostridia

Storage temperature influenced TAB count

CONCLUSION

Overall, these results demonstrate that decellularized oviductal scaffolds exhibit good stability after one month of storage. Nevertheless, bacterial growth can compromise scaffold quality, and storage at -80°C is the most effective way to prevent contamination.

REFERENCES

[1] Martínez-López C et al. *Theriogenology*. 2025;231:36–51.