P.102

Effects of cryopreserved human amniotic membrane on ischemic wounds

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Introduction

- Ischemic wounds are a very intractable disease. It is necessary to develop minimally invasive treatment of ischemic wounds for many patients cannot undergo revascularization surgery due to their severe comorbidity.
- Human amniotic membrane (hAM) has immunomodulatory, production various cytokines including VEGF and antibacterial effect, so they make it potentially useful as a wound dressing material.
- We found fresh hAM promotes the healing of ischemic wounds. However, cryopreserved hAM is more useful for clinical use.
- In this study, we examined wound healing promotion effect and angiogenesis promotion effect in order to reveal the efficacy of cryopreserved hAM as a wound dressing material for ischemic wounds.

Funding information

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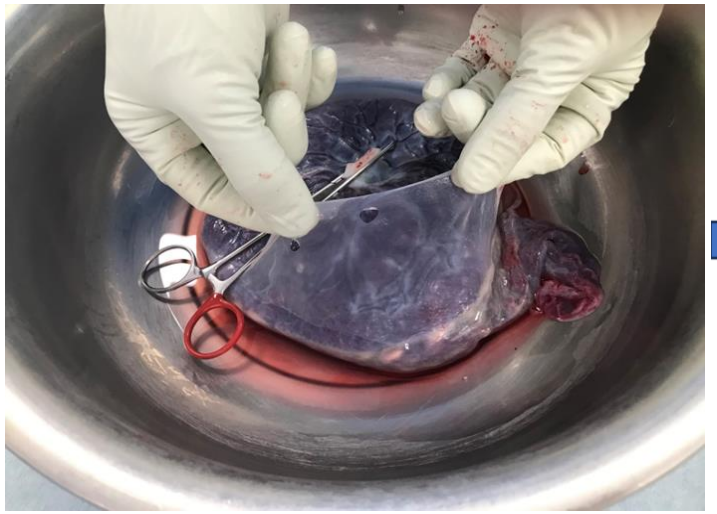
Method

hAM: collected from delivered placentas
(cesarean section at full term)

<Observation information>
period: 7 days
items: Wound area reduction
Microvessel density

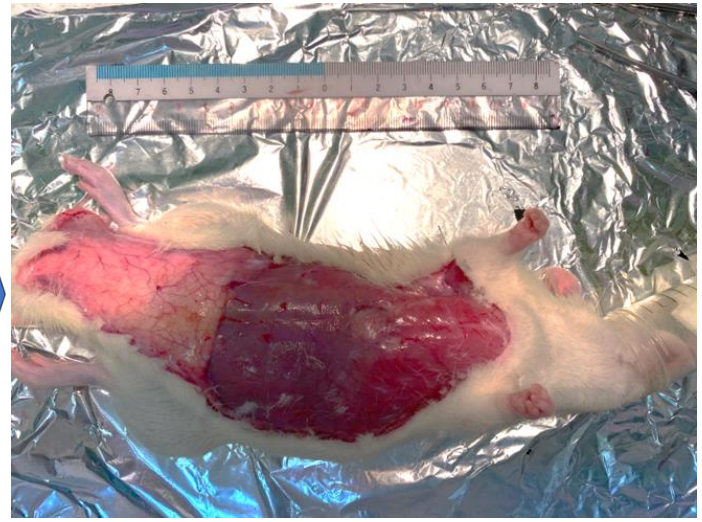


Cryopreserved (-80℃)
in Phosphate-buffered saline

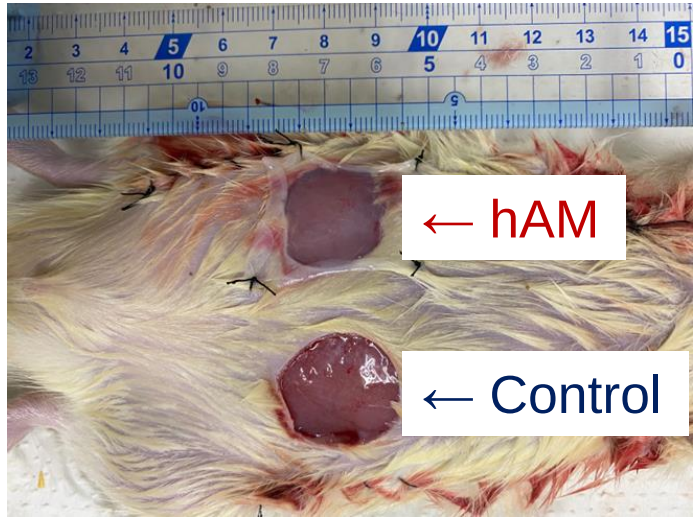
Applying within 6 hours after thawing

Rats: SD, male, nine weeks old, 300-350 g

Ligating the arteriovenous and nerve bundles of the inferior abdominal wall

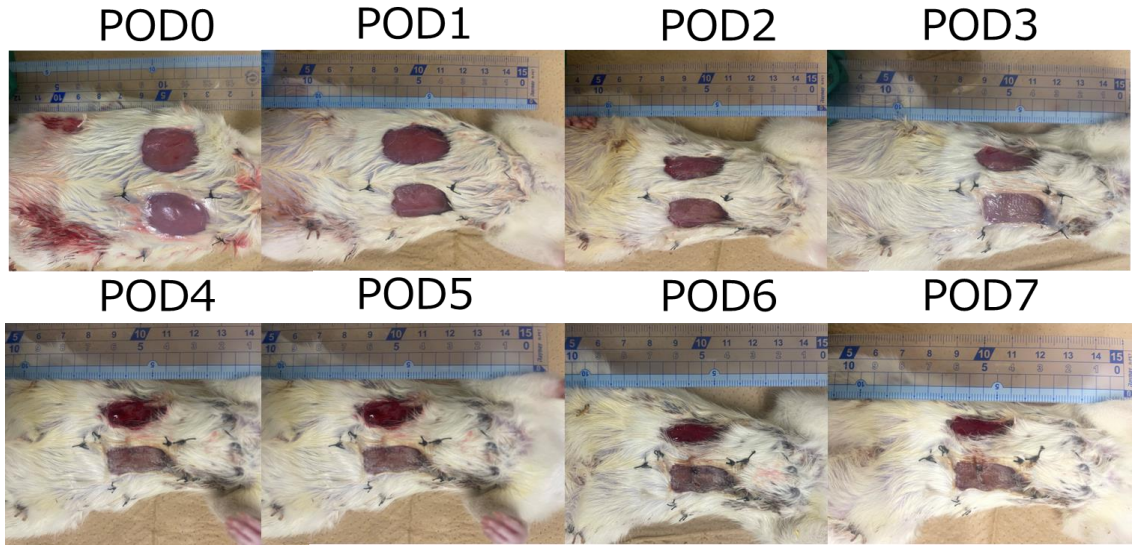


Creating a skin flap

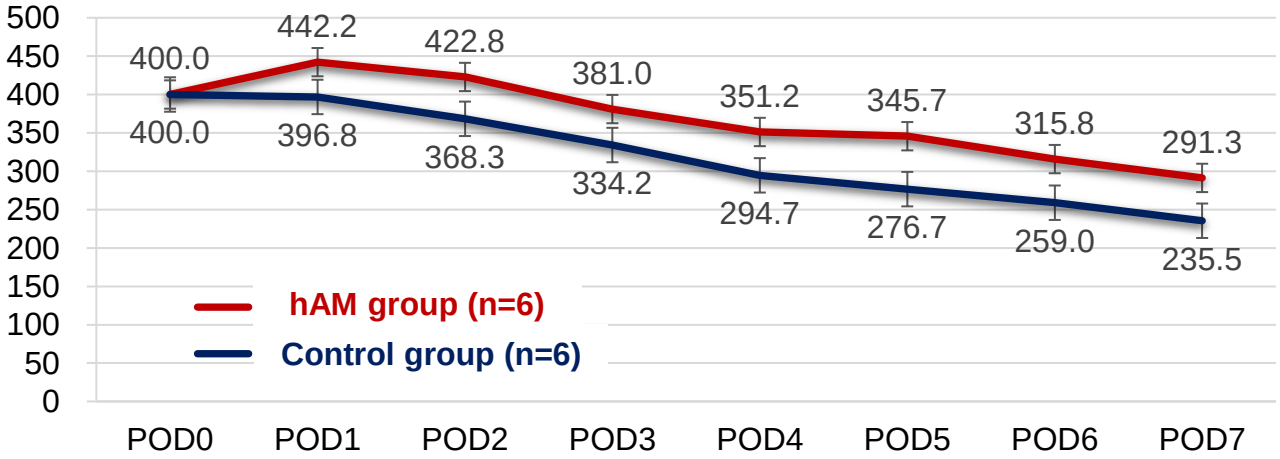


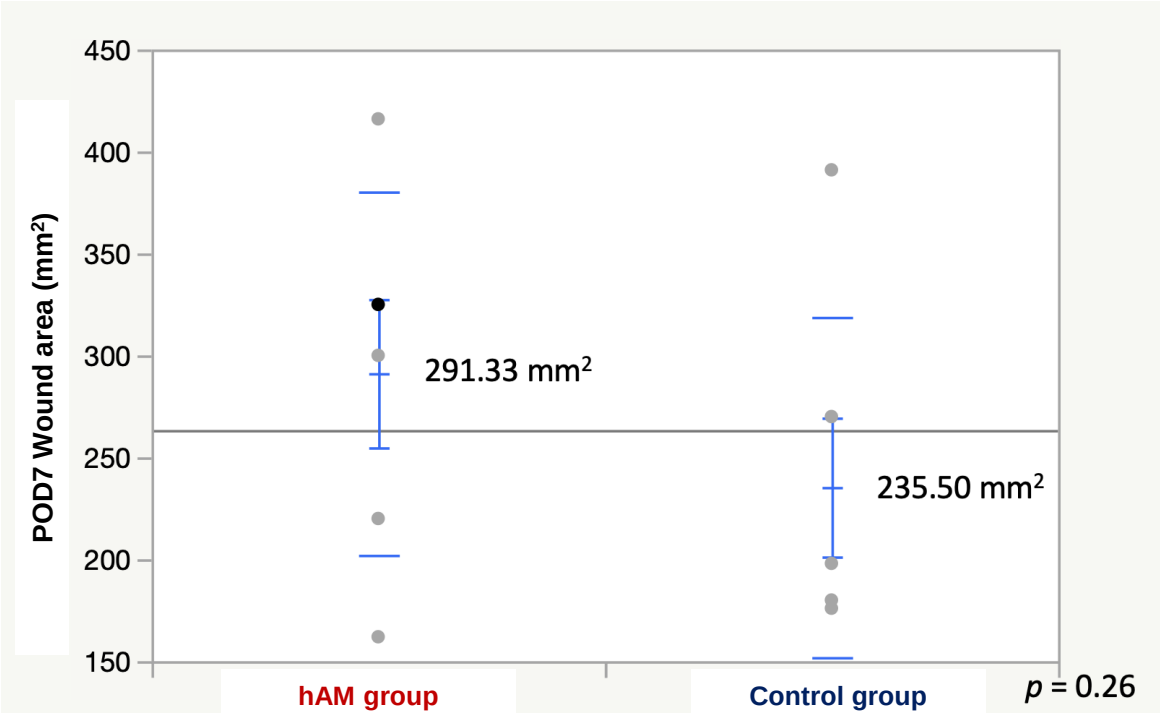
Excising the abdomen skin with a diameter of 2.0 cm.
※including fascia

Result① Wound area reduction



Wound area: major diameter ∗ minor diameter
Covering agent: absorbent cotton and gauze

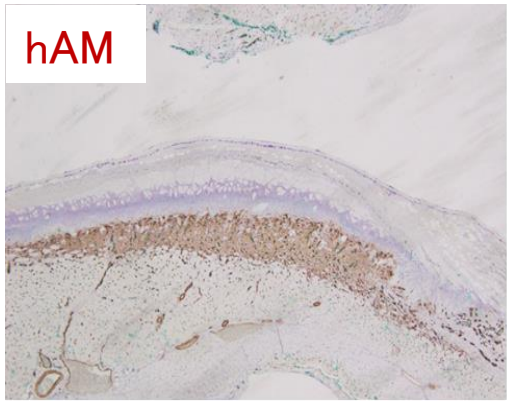




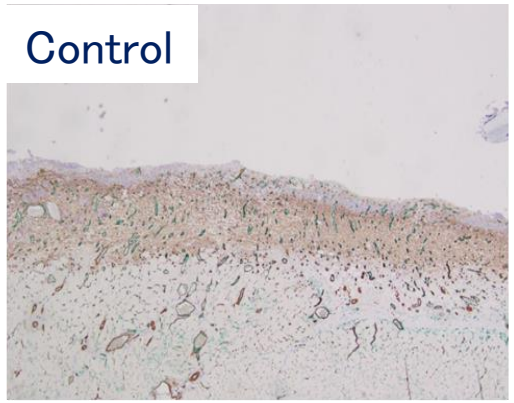
hAM group vs. control group; 291.3 vs. 235.5 mm^2, (p = 0.26)

⇒ No significant wound area reduction after seven days

Result② Microvessel density (MVD)



40x



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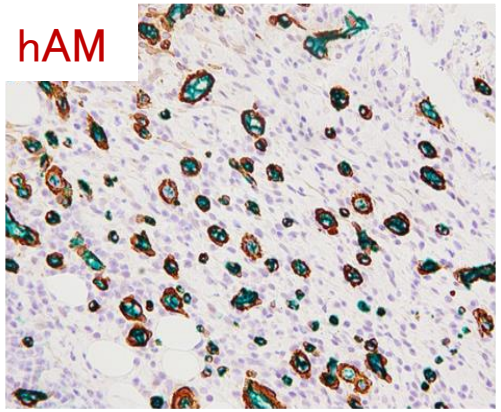
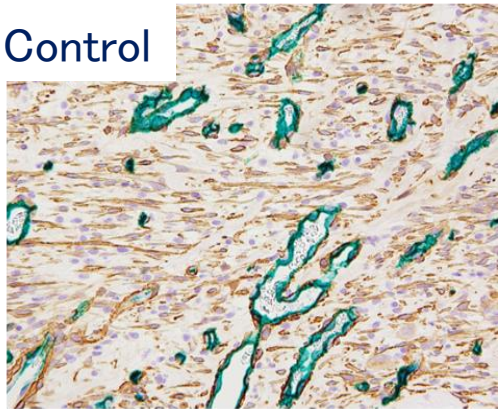


100x



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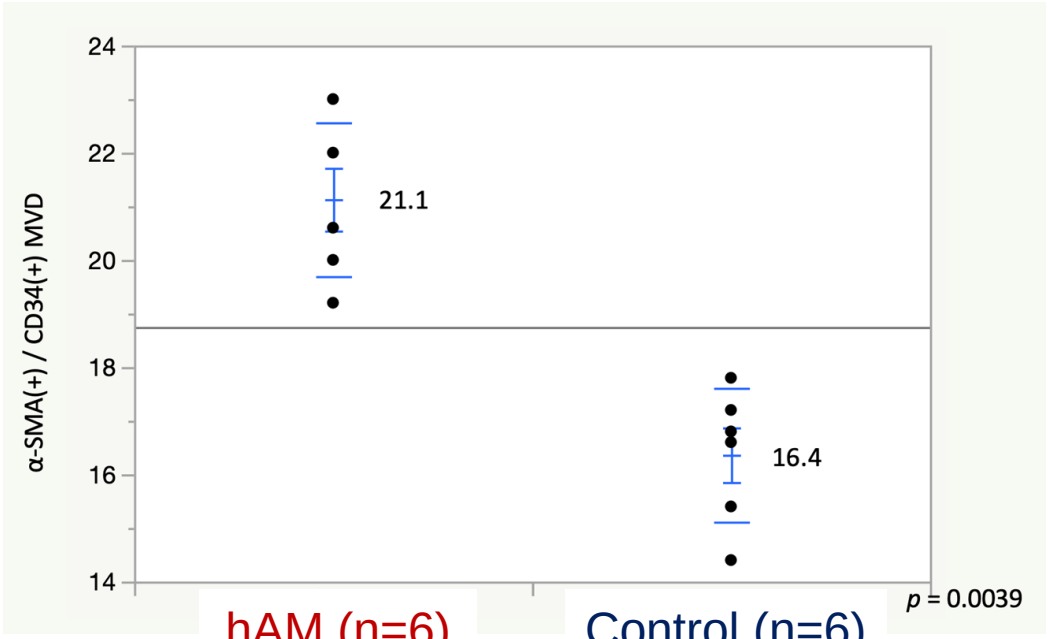
MVD :an index of angiogenesis measured by double staining with anti-α-SMA and anti-CD34 antibodies. We compared the density of the number of blood vessels and the wound area



hAM group vs. control group; 21.1 vs. 16.4 mm^2, (**p = 0.0039**)

⇒The mean value of MVD was significantly higher in the hAM group than in the control group

Discussions

- Cryopreserved hAM promoted angiogenesis but did not reveal wound-healing effects.
- This results seemed to be caused that hAM were damaged by cryopreservation with PBS.
- Adding glycerol to PBS may prevent the damage to the amniotic epithelial cells.

Gholipourmalekabadi M, et al; Burns. 2020;46(6): 1254-1271.

- In addition, it is also necessary to assess the effects of the oxygenation status of the wound area.

Conclusions

- This study showed the angiogenesis-promoting effects of cryopreserved hAM but did not reveal wound-healing effects.
- The mechanism of the loss of wound healing effect through hAM cryopreservation requires further investigation.