

Short-term Preservation of Human Autografts

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Short-term storage of a patient's harvested skin is clinically desirable for numerous reasons. Previous experience in our center using a skin storage solution of saline with a high concentration of antibiotics resulted in poor graft viability and an unsatisfactory clinical outcome. This report defines an improved method of storage which allows longer storage time, yielding viable skin and results in subsequent graft acceptance on the patient.

Split-thickness autografts from patients were stored in: 1) saline + 10^4 units/ml penicillin and 0.005 gm/ml streptomycin, or 2) RPMI-1640 + 25 units/ml penicillin and 25 mcg/ml streptomycin, at 4°C . The pH range of the saline solution was 5.90–6.20, compared to 7.20–7.32 for the RPMI-1640 solution. The medium was changed every 3 to 4 days during the storage period. Before graft reapplication the autografts were rinsed with sterile saline.

Previous clinical results using the saline-antibiotic storage solution resulted in poor graft viability and no graft survival was noted on patients after 5 days of skin storage. In contrast 11/16 autografts which had been stored in the RPMI-1640 solution for 5 to 22 days (median, 11 days) were successful takes when regrafted to patients. Graft loss was observed in five cases due to the following reasons: inability to immobilize graft (one); poor vascular bed (two); and bacterial infections (two). These data are in agreement with results reported in a separate paper, demonstrating the effectiveness of RPMI-1640 as a storage medium for maintaining viable human skin grafts which were subsequently transplanted to athymic nude mice.

Our results indicate that with proper storage techniques viable autografts can be maintained to 22 days without resorting to freezing. The RPMI-1640 solution was clearly superior to the saline solution with medium pH being an important factor in skin storage. Improvements in skin storage at this center have reduced the number of surgical procedures for many patients and have resulted in a shortened time of wound closure in these cases.

Short-term storage of a patient's harvested skin is clinically desirable for numerous reasons. Previous experience in our center using a skin storage solution of saline with a high concentration of antibiotics resulted in poor graft viability and an unsatisfactory clinical outcome. This report outlines an improved method of storage which yields viable skin, with longer storage time, and results in subsequent graft survival on the patient.

Before this study, split-thickness autografts were stored in 100 ml of saline with 10^4 units/ml penicillin and 5.0 mg/ml streptomycin at 4°C .

Previous clinical results using this saline-antibiotic storage solution showed poor graft viability and no graft survival was noted on patients where skin was stored for more than 5 days.

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Studies in our laboratory demonstrated that using a tissue culture medium (Roswell Park Memorial Institute-1640 [RPMI-1640]) (2), with a low concentration of antibiotics added would lead to improved graft viability. The athymic nude mouse model was utilized as the recipient for stored human skin, which proved RPMI to be a superior skin storage media (1).

METHOD

Split-thickness autografts not exceeding 10×2 cm were stored in 100 ml RPMI-1640 with 25 units/ml penicillin and 25 mcg/ml streptomycin at 4°C (4). Media were changed twice weekly and also if the pH was outside the range of 7.2 to 7.4.

When there was a need for an autograft, the skin was removed from the storage medium, and prepared for the patient.

The protocol for banked skin applications was as follows: When the wound site was ready for grafting

1) The autograft was removed from the storage medium, a section was prepared for H + E stain, and the

remaining portion rinsed with room-temperature sterile saline.

2) The autograft was then placed on the previously prepared recipient bed using standard aseptic technique. The graft was secured with nylon net or with the use of adhesive strips.

3) The area was dressed with bulky wraps soaked in 0.5% silver nitrate solution.

4) Each day the graft was examined and redressed with silver nitrate.

5) On the fifth post-graft day, a visual evaluation was made by the clinical staff regarding take or no take. Grafts which had a viable and adherent surface area of 30% or greater were considered takes.

RESULTS

Thirty-two autografts were stored in RPMI-1640 for periods of up to 22 days. Twenty-four of 32 autograft applications were successful (Table I).

Of the 32 grafts applied, eight failed to survive. Two failures were on the same patient. Five failures were due

to infection, three of these from *Staphylococcus aureus* and two from *Enterobacter cloacae*. Two failures were from site complications because of the inability to properly immobilize the graft. One site was the axilla and one the inner thigh. The eighth failure was due to a poor vascular bed, on one lower extremity which later required amputation for vascular complications.

DISCUSSION

Our results indicate that with this storage technique, viable autografts can be maintained for up to 22 days without resorting to rapid freeze technique (3). The RPMI-1640 is clearly superior to saline. We believe maintaining physiological pH is an essential factor in successful skin storage. Improvements in skin storage at our center have reduced the number of surgical procedures for many patients and have resulted in shortening the time necessary for wound closure. This is a cost-effective means of reducing the length of hospitalization and total hospital cost.

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TABLE I

Days in Storage/Median	No. Grafted	No. Takes
3-10/7	16	12
11-15/11	11	9
16-20/17	3	2
22	1	1
28	1	0
Totals	32	24

Total no. grafts = 32.

Total no. patients = 24.

REFERENCES

1. Cram, A. E., Domayer, M. A., Shelby, J.: Human skin grafts transplanted onto nude mice: A model for determining the viability of banked skin. *J. Trauma*, **23**: 924-926, 1983.
2. Moore, G. E., Gerner, R. E., Franklin, H. A.: Culture of normal human leukocytes. *J.A.M.A.*, **199**: 519-524, 1967.
3. Ninneman, J. L., Fisher, J. C., Frank, H. A.: Clinical skin banking: A simplified system for processing, storage, and retrieval of human allografts. *J. Trauma*, **18**: 723-725, 1978.
4. Paul, J.: *Cell and Tissue Culture*, 4th ed. Edinburgh and London, Churchill, Livingston, 1973, pp. 135-136.