Comparative morphological and histological study between the implantation of skin auto grafts and skin sheep hetero grafts in rabbits

Amir Ibrahim Towfek
Veterinary Medicine College, AL- Qadisiya University

Abstract:
This study was designed to compare the morphology and vascularization between the implantation of skin autografts of rabbits and skin sheep heterografts in rabbits.

The morphological and histological results indicated that skin autografts was more benefit than skin sheep heterografts due to the differences of the type of animals and formation of antibodies against the foreign antigens.

Introduction:
Skin grafts can be used in skin necrosis following surgery, car accidents and burn lesions. The skin could be propagated in vitro and the million of cells thus could be cultured and grown used as autografts (1). All cutaneous wounds heal by two independent processes, contraction and epithelialization (2).

Wound contraction reduces the perimeter by centripetal movement of dermis & epidermis and occurs as long as the contractive forces of the wound myofibroblasts exceed the reactive forces of the surrounding skin on the wound edge (2).
The average healing time of autografts was 39.8 days (3). The tissue morphology and the vascular changes of skin xenografts from the rats to the rabbits were studied by (4), the study showed the early vascularization was demonstrated in the autografts while the xenografts were incapable of becoming vascularized and exhibited necrosis on the third day. The xenografts underwent a course of white graft rejection which probably contributed to rejection.

The blood circulation in the full-thickness skin autografts was first detected one day after grafting (5), (6). To and fro moderate irregular blood circulation begins in the graft and modification of the preexisting vascular pattern of the graft takes place after circulation begins (5).

There was some increase in number and aborization of small vessels in the established grafts, although the vascular pattern was essentially similar to that of unoperated skin (6).

The host did react to a heterograft application as evidenced by antibody production, the interface reaction with the breakdown of foreign protein could cause host sensitization with out any actual vascularization (7), (9).

The aims of this study was to compare the clinical and histological morphology and vascularization of skin autografts and skin heterografts of sheep in rabbits.

**Material and methods:**

This study was done on (30) Iraqi local rabbits, their weights ranged (1.100 – 1.800) kg. The rabbits were divided randomly into three equal groups;

1- Skin autografts group (10) rabbits.
2- Skin heterografts group (10) rabbits.
3- Control group (10) rabbits.

All rabbits had the same management, feeding and was injected sodium hydrocortisone (10 mg/kg B. W.) intramuscularly daily for two weeks, to inhibit the immune system and prevent rejection of the grafts.

Group No. 1:

The left and right flanks of each rabbit was prepared surgically and were anaesthetized by combination of xylazine (3 mg/kg B. W.) and ketamine (25 mg/kg B. W.) intramuscularly.

A full-thickness skin piece (5x5 cm.) was excised from the right flank region and was put in sterilized gauze saturated with sterilized normal saline to implant in the left flank which also excised piece, at the same size of right flank region.

The grafts was fixed by polypropylene suture no. 1 at simple
interrupted pattern. The site of graft cleaned daily and had procaine penicillin powder for two weeks.

Group No. 2:
Piece of sheep skin (5x5) sq.cm. which collected from slaughterhouse in the same day of grafting, was put in sterilized neutral physiologic ringers solution (prepared 24 hours before grafting and preserve at (4c).
The skin sheep heterografts was fixed in the left flank region of the rabbits as the same matter in group (1), fig. (1).

Group No. 3:
All the rabbits prepared surgically and were anaesthetized as in groups (1) & (2). Pieces of skin (5x5) sq.cm. was excised from the left flank region but without graft implantation, treated as open wound by procaine penicillin powder only for two weeks.

Post-surgical care:
All rabbits was injected by procaine penicillin (10.000 –20.000 i.u./kg B.W.) intramuscular daily for 5 days. Also was injected by sodium hydrocortisone (5 mg / kg B.W.) I/M daily for one week.

**Histo-pathological examination**
Skin biopsies was taken (1, 2, 4) weeks after graft implantation and fixed by neutralized formalin solution 10% and embedded in paraffin. Multiple sections were cut by micrometer from each specimen, stained with haematoxylin and eosin and examined by light microscope (Olympus) x 40.

**Results:**
The results of morphologic examination of (8) rabbits from group 1 (two rabbits was died due to pneumonia and coccidiosis) showed the healing time was 35 + 1.5 days and there was no rejection of autografts.
Some grafts showed already on the 5th post-operative day slight hardening in part of grafts borders, reddening specially at the junction between the grafts and the recipient pad and shrinkage of autografts after three days. After (6) weeks there was hair growth in skin of rabbit with different directions.

The results of morphologic examination of (9) rabbits from group 2 (one rabbit was died due to coccidiosis) showed no healing at the junction of the grafts and recipient pad and there is complete rejection of the skin sheep heterografts which separate from recipient body with pus formation. There was no shrinkage of heterografts pieces.

The histo-pathological examination of group 1 showed good growth of fibroblasts proliferation, granulation tissue growth specially within two weeks, slight hy-
perimia and slight inflammatory cells accumulation fig. (2), (3). Vascularization, severe hyperemia in the recipient pad borders with accumulation of inflammatory cells fig. (4), (5).

Fig(1): Fixation of skin sheep heterograft
Fig(2): C.S of auto graft after 2 weeks:

Haematoxylin and eosin stain × 40

1. Granulation tissue
2. Fibroblasts
3. Blood vessel
Fig(3): C.S of auto graft after 4 weeks

Hematoxylin and eosin stain x40

1. Fibro blasts proliferation
2. Blood vessel
Fig(4): C.S of skin sheep heterograft after 2 weeks

Hematoxylin and eosin stain x40

1. Oozing of plasma outside the blood vessels
2. Hemorrhage
Fig(5): C.S of skin sheep heterograft after 4 weeks
Hematoxylin and eosin stain x40
1. Oedema
2. Hemorrhage
3. Necrosis
Discussion:
The results showed good healing with autografts as mentioned by (3),(10), there was slight reaction observed clinically and histopathologically in any of the rabbits.
The recipient pad was slightly redness which return to normal after five days, due to the surgical interference. The autografts borders success to heal with no any signs of rejection.
The histopathologic examination showed fibroblasts proliferation in the junction between the autografts and the recipient pad and the granulation tissue heal normally even there was slight inflammatory cells, these results also mentioned by (1),(4).
These results indicated that the immune response to the autografts was slight and this method was the best helpful method to treat many surgical cases like burns, car accidents, skin necrosis following surgery.
While the results of skin sheep heterografts showed that they failed to heal, the grafts separate from the recipient body even they were fixed very well by stitches. The redness of the recipient pad continued for more than one month with severe pus formation.
The histopathologic examination showed severe inflammatory cells accumulation which effect on fibroblasts proliferation and the granulation tissue growth.
The main factor for any healing was the blood supply, in this study of heterografts there was no actual vascularization which lead to failing of healing.
The interface reaction, with the breakdown of foreign protein could cause host sensitization without any actual vascularization (7).
The immune response to nonspecific and altered tissue antigens in soft tissue had been described (7),(8). So the skin sheep heterografts as antigens coated by specific antibodies which interfered with subsequent cell-mediated immune reactions, and that explained the type of donor animal was very important factor for heterografts implantation.
Nothing works as well as patients own skin (10).

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