Fig. 2. (Case 1, patient No. 1) The patient suffered flame burns over 11% of TBSA. On day 5, the wound was tangentially excised and two days later a part of the wound on the left calf was treated with one 30-cm² RHPS graft; the rest was covered with xenografts. The area covered with RHPS clinically healed within five days, while the area covered with xenografts had to be autografted. (A) Day 9. A contrasting difference can be seen between the RHPS-grafted area two days after treatment (arrow) and the rest of the wound treated with xenografts. (B) Day 16. Healed area under RHPS (a biopsy (arrow) was taken seven days after grafting, on day 14). (C) Histological examination seven days (day 14) after grafting showed well-stratified epidermis (1) of variable thickness and irregular rete ridges. A thin layer of very loose cellular connective tissue (2) is formed on the original lower corium that consists of densely collageneous bundles (3). Strips of putative older haemorrhages (4) can be observed in the loose, newly formed connective tissue. The dried RHPS is still attached to the epidermis (5).

Fig. 3. A 3-year-old boy (Case 2, patient No. 13) was scalded on 30% of TBSA. On day 4, a part of the deep dermal wound on the trunk was covered after surface dermabrasion with RHPS and Aquagel without any healing effect. The wound was treated with silver sulphadiazine cream. (A) Day 8 after the injury. (B) On day 8, the lower part of the wound was tangentially excised and covered with RHPS. The figure shows the grafted area (arrow) on day 10, two days after RHPS application. (C) Day 13. Five days after RHPS application the graft was peeled off, leaving a new epidermis underneath. (D) The second RHPS graft was applied on day 13 on the wound prepared by deep dermabrasion – the figure shows the grafted area (arrow) two days after treatment, on day 15. (E) It healed four days after RHPS application. The upper third of the wound which was covered with RHPS on day 17 (too late) had to be autografted on day 19. (F) Six months later, the lower two thirds of the wound showed no hypertrophic scarring in comparison with the upper third.

Fig. 4. A 1-year-old girl (patient No. 18) was scalded on 30% of the body surface. On day 4 after the accident, the deep dermal burn on the left upper extremity was tangentially excised and covered with RHPS. Five days later (day 11), the wound was healed. (A) The wound on the day of admission (day 1), (B) the grafted area (red) on day 6, two days after RHPS grafting, (C) the healed wound on day 18, (D) after 1 year, the burn wound covered with RHPS did not show any hypertrophic scarring.

Case 3 (Fig. 4, patient No. 9): A 1-year-old girl suffered deep dermal and full-thickness scalds over 30% of her body surface. On day 4 after the accident the deep dermal burn on the left upper extremity was tangentially excised (4% TBSA) to obtain a clean, bleeding wound bed. The excised area was immediately covered with RHPS, which stopped bleeding. Five days later the wound was healed. No hypertrophic scarring developed in this area in the course of one year. All other areas were full-thickness burns and had to be autografted.

<u>Case 4</u> (patient No. 5) demonstrates healing of a wound prepared by early excision/RHPS grafting compared to a non-excised wound. A 10-month-old boy suffered deep dermal scalds over 26% of the body surface. On day 3 after the accident the wounded area on the right arm (100 cm²) was tangentially excised and covered with RHPS, and a smaller wound (12 cm²) was covered with Aquagel. The left arm, which was evaluated to be analogically scalded, was treated partially with xenografts and partially with silver sulfadiazine cream. The right arm healed five days after excision (day 8 after the accident), while the burn on the left arm deepened and had to be autografted on day 17.

<u>Case 5</u> (patient No. 21): A 2-year-old girl suffered scalds on 28% of the body surface. On day 2 a deep dermal wound on the left forearm prepared by surface dermabrasion was covered with an RHPS graft. No

healing effect appeared in the course of eight days and the wound had to be autografted. From day 2 the wound on the left arm was treated with silver sulfadiazine cream. On day 6 the epidermis started to dissolve; a limited area was dermabraded down to capillary bleeding (deep dermabrasion) and covered with RHPS. This wound healed in four days. On day 8, another part of the wound was dermabraded to the level of capillary bleeding and treated with RHPS, but it was already converted to a full-thickness wound and had to be autografted. This case demonstrates the importance of time and method of preparation of the wound bed.

Results of healing of differently prepared wound beds

In our previous study we have shown that healing of DDB under RHPS (often applied like xenografts on wounds prepared by surface dermabrasion) varied and was in some cases unsuccessful. On the other hand, fresh donor sites always healed in one week. The main goal of the present study was to find conditions (timing and wound bed preparation) for the optimum healing effect of RHPS that could result in preventing conversion. We show the results obtained in 22 patients (45 wounds) prepared either with tangential excision, early

Table 2. Wound healing after surface dermabrasion

| Pt. | Sx | Age | Burn type | %TBSA burned | Day of application | Type of cover | Area grafted (cm ²) | No. of applications | Wound prepa- ration | Healing /days |
|-----|----|-----|--------------|-----------------|--------------------|---------------|---------------------------------------|---------------------|---------------------------|------------------|
| 17 | M | 4 | Scald | 15 | 7 | RS | 40 | 1 | SD | Y/4 |
| 18 | F | 2 | Scald | 9 | 5 | RS | 75 | 1 | SD | Y/3 |
| 19 | M | 2 | Scald | 8 | 5 | RS | 40 | 1 | SD | N |
| 3 | M | 14 | Scald | 13 | 4 | RS | 60 | 1 | SD | N |
| 12 | M | 3 | Scald | 30 | 4 | RS | 40 | 1 | SD | N |
| 20 | M | 9 | Scald | 8 | 6 | RS | 40 | 1 | SD | N |
| 21 | F | 2 | Scald | 28 | 2 | RS | 40 | 1 | SD | N |
| 22 | M | 3 | Scald | 10 | 5 | RS | 40 | 1 | SD | N |
| 18 | F | 2 | Scald | 9 | 5 | Xe | 60 | 1 | SD | Y/3 |
| 17 | M | 4 | Scald | 15 | 7 | Aq | 40 | 1 | SD | Y/4 |
| 3 | M | 14 | Scald | 13 | 4 | Xe | 50 | 1 | SD | N |
| 6 | M | 6 | Scald | 25 | 4 | Xe | 40 | 1 | SD | N |
| 5 | M | 1 | Scald | 26 | 3 | Xe | 50 | 1 | SD | N |
| 12 | M | 3 | Scald | 30 | 4 | Aq | 30 | 1 | SD | N |
| 19 | M | 2 | Scald | 8 | 5 | Aq | 40 | 1 | SD | N |
| 5 | M | 1 | Scald | 26 | 3 | cream | 200 | 1 | SD | N |

Pt - patient, Sx - sex, RS - RHPS, Xe - xenograft, Aq - Aquagel, SD - surface dermabrasion, Y/n - healed in n days, N - not healed

Table 3. Wound healing after deep dermabrasion

| Pt. | Sx | Age | Burn type | %TBSA burned | Day of application | Type of cover | Area grafted (cm ²) | No. of applications | Wound prepa- ration | Healing /days |
|-----|----|-----|--------------|-----------------|--------------------|---------------|---------------------------------------|---------------------|---------------------------|------------------|
| 12 | M | 3 | Scald | 30 | 13 | RS | 45 | 1 | DD | Y/4 |
| 13 | M | 8 | Scald | 12 | 8 | RS | 50 | 1 | DD | Y/3 |
| 13 | M | 8 | Scald | 12 | 8 | RS | 40 | 1 | DD | Y/5 |
| 21 | F | 2 | Scald | 28 | 6 | RS | 50 | 1 | DD | Y/4 |
| 12 | M | 3 | Scald | 30 | 17 | RS | 45 | 1 | DD | N |
| 21 | F | 2 | Scald | 28 | 8 | RS | 40 | 1 | DD | N |

Pt - patient, Sx - sex, RS - RHPS, Xe - xenograft, Aq - Aquagel, DD - deep dermabrasion, Y/n - healed in n days, N - not healed

The most rapid healing with RHPS that could even prevent conversion into full-thickness wounds has been achieved after early tangential excision. Alternatively, deep dermabrasion can be used with some delay in healing. Surface dermabrasion is not suitable for this purpose.

Discussion

Commonly used biological or synthetic covers do not promote fast epithelization of donor sites and deep dermal burns. Janzekovic found that for a fast, efficient therapy and prevention of further deepening and hypertrophic scarring, dermal burns may be grafted with thin split-thickness skin autografts, resulting in shorter hospital stay and pain reduction (Janzekovic, 1970). Also human epidermal keratinocytes, cultured by the technique described by Rheinwald and Green (Rheinwald and Green, 1975; Green et al., 1979), have resulted in

an important advance in the therapy of DDB. Nuñez-Gutiérrez et al. (1996) showed that with an integral combined therapy consistently utilizing cultured allokeratinocyte sheets for donor sites and deep dermal burns (and cultured autografts when needed), the total hospital stay of extensively burned patients is shortened by 30–40%. The disadvantage of cultured keratinocyte sheets, however, is their fragility, hypersensitivity, complicated preparation and difficult manipulation.

In order to overcome these problems, we have developed recombined human/pig skin (Matoušková et al., 1993). The RHPS shows higher mechanical resistance permitting easy handling, optimal adhesiveness, as well as a haemostatic effect and noticeable relief of pain. All these effects are accompanied by an apparently increased level of resistance to infection (Matoušková et al., 1997).