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Timing of Injury in Human Thermal Burns

How old is a burn? The answer is obviously important to the forensic pathologist. To provide some guidelines to the pathologist, this study of thermal burns delineates the histologic changes in a group of people who survived from a few hours to 15 days.

Materials and Methods

The 81 well-documented necropsy cases selected for this study from the files of the Armed Forces Institute of Pathology had been reviewed previously by the staff of the Forensic Pathology Branch. In each case the clinical summary, report of autopsy, and microscopic slides were examined. The clinical classification for the distribution of the 81 cases by degree of burning included 3 persons with second-degree burns, 8 persons with third-degree burns, and 70 persons with combined second- and third-degree burns. We arranged the cases into groups according to days of survival, beginning with persons living for a few hours to one day and continuing with cases for each day through the seventh. Each group contained 3 to 15 cases. Because of special findings, we included two cases beyond the seventh day, one of a person surviving 10 days and the other 15 days (Fig. 1). The formalin-fixed, paraffin-embedded samples of burned skin and subcutaneous tissue were stained in hematoxylin and eosin, alcian blue, Verhoeff's elastin, and phosphotungstic acid-hematoxylin (PTAH) [*1*].

Since we were concerned with the evaluation and timing of vital reaction to injury by histopathologic methods, neither first-degree burns nor fourth-degree burns were included in this study. The clinical and pathologic classifications of the burns in the cases we used, based upon external examination of the extent of injury, were correlated with the microscopic findings to provide a suitable histologic classification. For the purpose of this study, coagulation of the epidermis and the upper portion of the dermis classified the burn as second degree; coagulation of all portions of the dermis and

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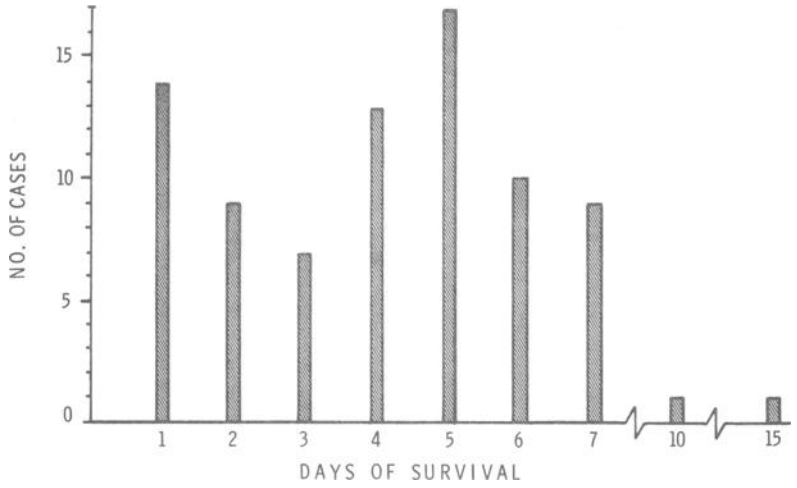

FIG. 1—*Graph showing number of cases by the length of survival (days).*

damage to the upper layers of the subcutaneous tissue classified the burn as third degree.

Microscopic Findings

Second-Degree Burns

The epidermis was either absent or edematous and had nuclear pyknosis and karyorrhexis. The uppermost dermal collagen fibers were coagulated. The earliest lesions (2 h and 4 h) showed these changes. The epithelial cells of the skin appendages often showed no change except for cytoplasmic vacuolization in some cases (Fig. 2). PTAH staining of the damaged collagen revealed deep-blue-violet fibers. The dermal fibroblasts and blood vessels appeared undamaged.

Polymorphonuclear neutrophils (PMNs) first appeared in the three 16-hour-old burns but were not seen in one of two 24-hour-old burns. In all burns 2 days or older, PMNs infiltrated between dermal collagen fibers, and by 5 to 7 days often formed a distinct border between the coagulated and viable areas (Fig. 3).

Third-Degree Burns

Damage extended through all layers of the skin and often reached the subcutaneous tissue. Many of the nuclei were shrunken, with or without elongation (Fig. 4). The burning coagulated the dermis throughout, with homogenization and swelling of the fibers. Verhoeff's elastin stain demonstrated reduction in the numbers of elastic fibers and infrequent brown discoloration of collagen fibers. As with second-degree burns, PTAH stained the damaged collagen a deep bluish violet (Fig. 5).

When the upper layer of the subcutaneous tissue was coagulated the nuclei shrank and sometimes disappeared. The cytoplasm of the fat cells rarely showed complete necrosis but sometimes appeared foamy. Some vessels contained coagulated blood, and focal perivascular hemorrhages were seen.

The onset of dermal inflammatory response varied, and delay was common. By the

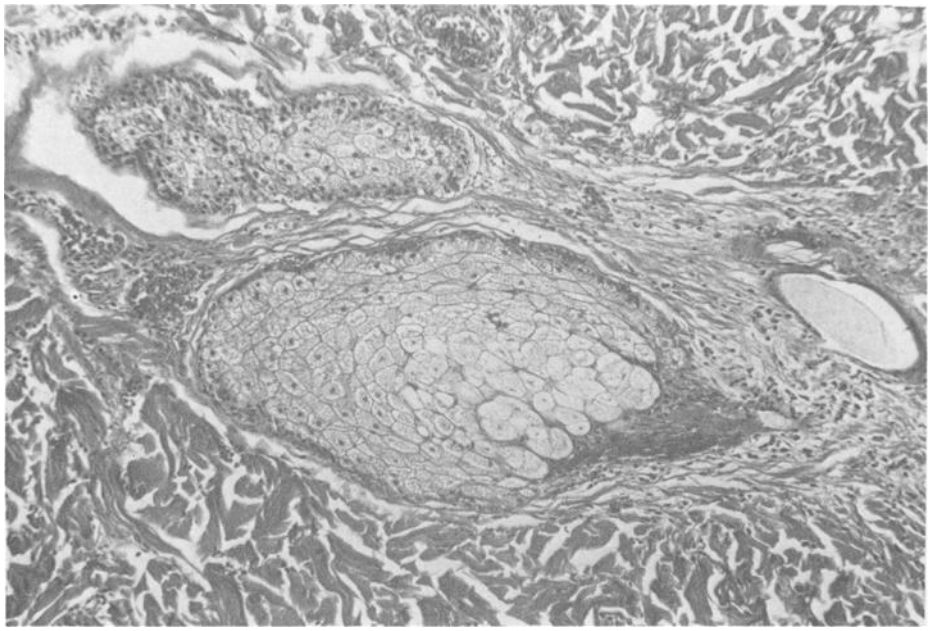

FIG. 2—*Second-degree burn, 18 hours old. Vacuolization is seen in some cells of the sebaceous gland, but in general the skin appendages are well preserved (hematoxylin and eosin; original magnification ×105; AFIP Neg. 70-11185).*

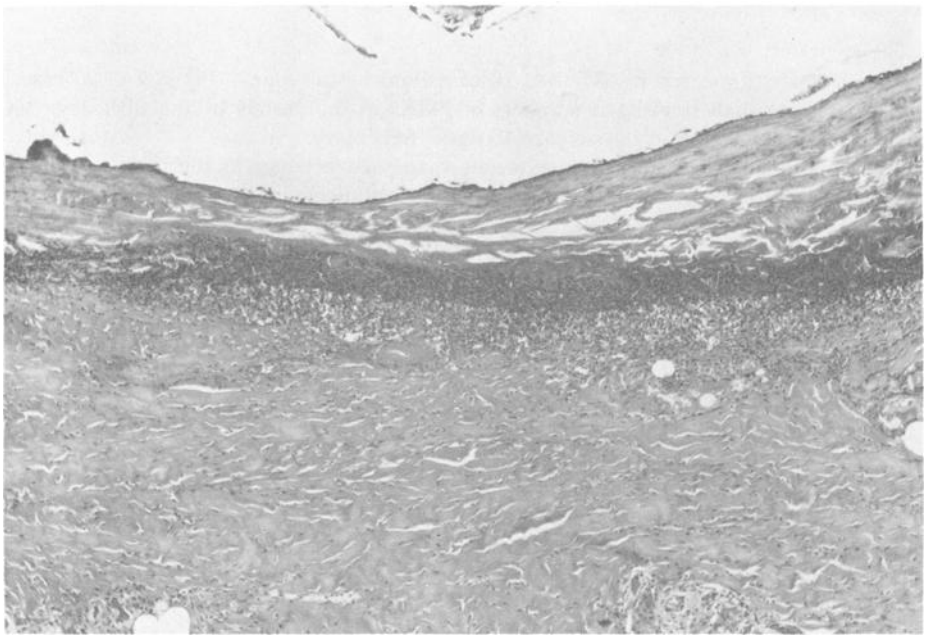

FIG. 3—*Second-degree burn, 6 days old. Demarcation zone of leukocytes at the border between the damaged and the viable dermis (hematoxylin and eosin; original magnification ×70; AFIP Neg. 70-11186).*

FIG. 4—*Third-degree burn, 1 hour old. Swollen and homogeneous collagen fibers of the dermis. Pyknotic nuclei in the skin appendages (hematoxylin and eosin; original magnification ×165; AFIP Neg. 70-11188).*

first day, PMNs appeared in only 1 of 10 cases and by the fourth day in 7 of 23 cases. Almost all cases had significant numbers of PMNs in the dermis by the fifth day, but they still had not appeared in one case 15 days after injury.

Inflammation began in the subcutaneous tissue sooner than in the dermis. All five cases by the second day showed PMNs in the septa of subcutaneous adipose tissue, while only one case showed PMNs in the dermis. After two days, inflammation occurred in the adipose tissue of 37 out of 45 cases (82 percent).

Hypermia of the subcutaneous tissue was neither as frequent nor as easy to evaluate as inflammation. Fibroblastic activity occurred in only three cases: in two persons surviving 5 days, and in the third, 10 days. In these three cases the epidermis and dermis showed no signs of regeneration.

There was no relationship between the onset of inflammation and the total area of skin surface burned. Extensive burns did not delay inflammation.

Discussion

Many texts of forensic medicine describe inflammation as the most reliable sign of antemortem trauma. The inflammatory response usually begins very shortly after injury with arteriolar contraction, followed shortly by vascular dilatation. As the vessels dilate, the blood flow slows, and white blood cells, predominantly PMNs, begin to marginate along the capillary walls. The PMNs then migrate through the capillary walls into the injured tissue. The presence of significant numbers of neutrophils in the tissue is the best sign that the injury has taken place before death, since a flow of blood is required. In

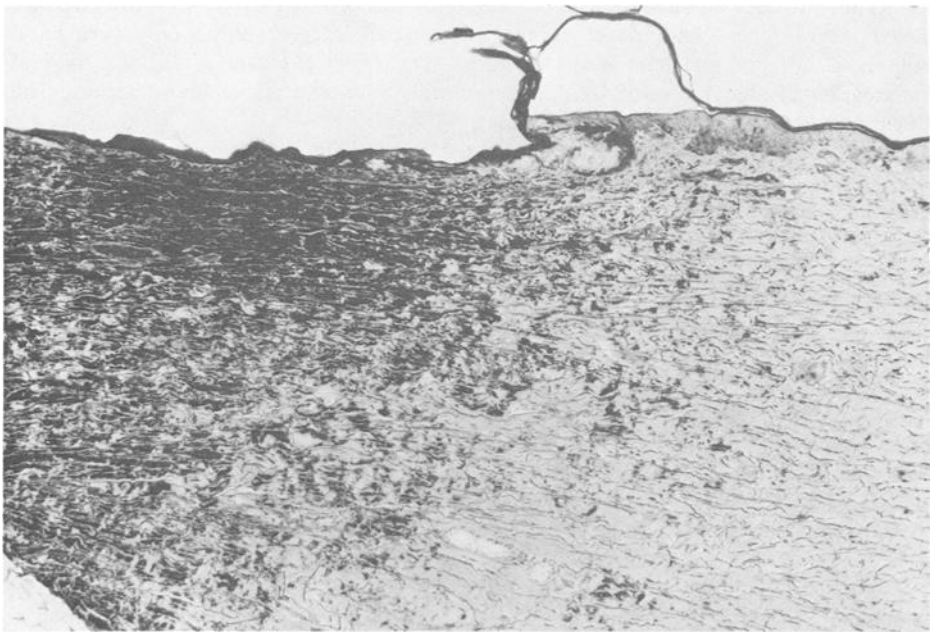

FIG. 5—*Third-degree burn, 6 days old. Dark blue collagen fibers in the burned region* (left). *No inflammation visible (Phosphotungstic acid-hematoxylin (PTAH); original magnification ×70; AFIP Neg. 70-11192).*

burns, the infiltration of PMNs is delayed for several hours [2,3]. This delay increases with increased severity of the burn. Shock delays the infiltration even more.

Degenerative changes such as epidermal and dermal coagulation and fragmentation of the dermal elastic tissue occur even before inflammation, but their presence before death is more difficult to prove. Regeneration through the formation of granulation tissue and proliferation of epidermis is an obvious sign of antemortem reaction to injury, but it occurs much later and contributes only to the timing of older burns.

In experimental animals some early reactions to burning have been demonstrated by histochemical studies for enzymatic reactions [2,4]. In human skin the reaction is slower than in small animals [4]. The formalin fixation of tissue in our cases obviated the detection of any enzymatic changes.

Other authors [3,5,6] found microscopic changes in the skin and adipose tissue similar to our observations. In second-degree burns, epidermal edema, nuclear pyknosis, and karyorrhexis typically occurred. Nuclear elongation [3] was observed in the epidermis, skin appendages, and collagen fibers.

Third-degree burning coagulated the entire dermis, with swelling and homogenization of the collagen fibers, skin appendages, and blood vessels. Often the upper layer of the subcutaneous tissue was coagulated. Blue-violet staining of the collagen fibers with the PTAH method has been previously reported [2,7]. This change, along with the darker brown staining of the fibers by Verhoeff's method, indicates alteration of the collagen. These alterations also occur post mortem [2].

The inflammatory reaction differed between the second- and third-degree burns, so these burns are considered separately. In second-degree burns, PMNs infiltrated the

dermis in all cases 16 hours or older except for one 24-hour-old case. This response lagged several hours behind those reported in second-degree burns of experimental animals [2,3,8] and of man [4,6]. In burns five days or older, a distinct zone of demarcation divided the coagulated upper dermis from the viable lower dermis. The subcutaneous adipose tissue remained intact.

In third-degree burns, the inflammatory response in the dermis occurred later. Two cases, 7 and 15 days old, still showed no signs of inflammation. Subcutaneous inflammation in third-degree burns developed sooner than in the dermis. By the first day, PMNs were seen in two out of four cases and by the second day appeared in over 70 percent of the cases. This is a valuable sign of antemortem burning. Therefore one should always include subcutaneous tissue in samples of the skin from burns.

We feel that impairment of the circulation explains the long delay of inflammation. This occurs locally from coagulation of the blood vessels and systemically as a result of circulatory shock.

In third-degree burns no viable vessels remained in the dermis, while in second-degree burns a zone of inflammation separated the bloodless, coagulated upper portion of the dermis from the viable lower portion. Coagulation of blood vessels also occurred in the subcutaneous tissue of third-degree burns. The inflammatory cells infiltrated deep in the fat and not in the areas adjacent to the dermis where the vessels were damaged.

Summary

Eighty-one fatalities from thermal burns were analyzed to relate the pathologic changes in the skin and subcutaneous tissue to the time of injury. Clinical complications were compared with the time of injury, but there was no significant correlation with any of these specific complications.

Histologically, inflammation was the most distinct antemortem reaction. Polymorphonuclear neutrophils usually migrated into the dermis of second-degree burns by 16 h. In third-degree burns, dermal inflammation lagged several days, but the subcutaneous tissue became inflamed sooner, usually by the second day.

In order to estimate the time of injury by burns, it is first necessary to establish the degree of the burn, since the progression of inflammatory reaction differs. Sections of both the skin and subcutaneous tissue, especially in third-degree burns, are required to make an estimate of the time of injury.

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